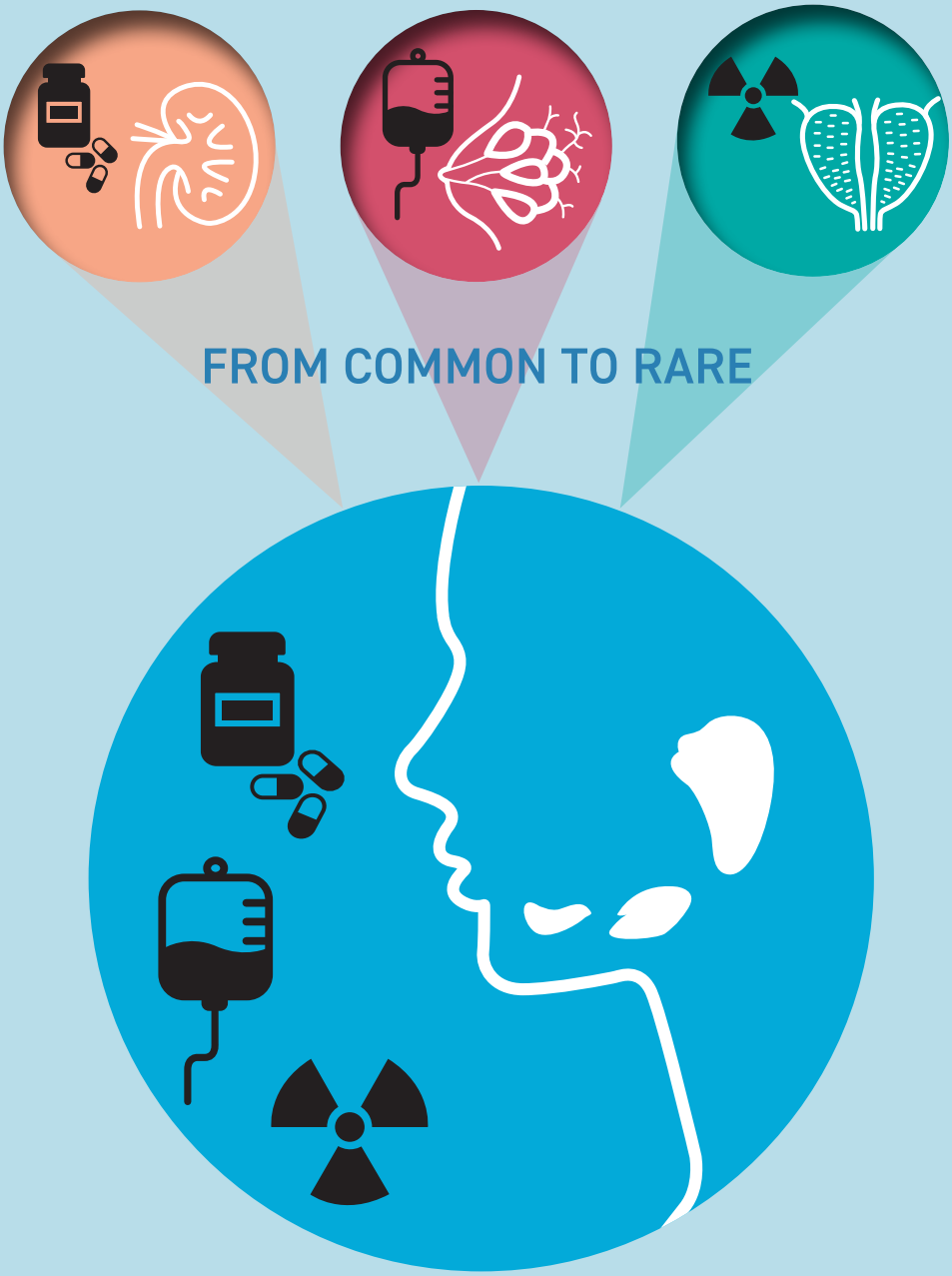
PALLIATIVE SYSTEMIC THERAPY FOR SALIVARY GLAND CANCER



Maike Uijen

PALLIATIVE SYSTEMIC THERAPY FOR SALIVARY GLAND CANCER

FROM COMMON TO RARE

Maike José Maria Uijen

COLOFON

The research presented in this thesis was carried out at the Radboud University Medical Centre.
Research institute: Radboud Institute for Health Sciences
Departments: Medical Oncology and Medical Imaging (section: Nuclear Medicine)

The clinical studies were financially supported by:
- The Dutch Cancer Society (KWF): Grant number: 11807-2018-2, Assigned Funder: Alpe d'HuZes
- The Dutch Salivary Gland Cancer Patient Network (Patiëntenvereniging Speekselklierkanker)
- The Adenoid Cystic Carcinoma Research Foundation (ACCRF)
- The Radboud Oncology Fund (ROF)
- Ipsen Pharmaceuticals

ISBN: 978-94-6423-990-4

Cover & lay out: Bregje Jaspers | ProefschriftOntwerp.nl

Print: ProefschriftMaken | www.proefschriftmaken.nl

Uijen, M.J.M.
Palliative Systemic Therapy for Salivary Gland Cancer

Thesis, Radboudumc, Nijmegen, the Netherlands

PALLIATIVE SYSTEMIC THERAPY FOR SALIVARY GLAND CANCER

FROM COMMON TO RARE

PROEFSCHRIFT

Ter verkrijging van de graad van doctor
aan de Radboud Universiteit Nijmegen
op gezag van de rector magnificus prof. dr. J.H.J.M. van Krieken,
volgens besluit van het college voor promoties
in het openbaar te verdedigen op

vrijdag 18 november 2022
om 10.30 uur precies

door

Maike José Maria Uijen

geboren op 5 september 1993
te Ubbergen

Promotoren:
Prof. dr. C.M.L. van Herpen
Prof. dr. J. Nagarajah (Universität Duisburg – Essen, Duitsland)
Prof. dr. M. Gotthardt

Manuscriptcommissie:
Prof. dr. K. Grünberg (voorzitter)
Prof. dr. R.P. Takes
Dr. W.V. Vogel (NKI-AVL)

CEPIT COPIAM CEPA

CONTENTS

Chapter 1

General Introduction and outline of this thesis.

GENERAL INTRODUCTION

The Salivary Glands

The salivary glands are small organs located around the oral cavity that produce saliva. This saliva is transported through the salivary ducts into the oral cavity where it enables the swallowing of food by lubricating it. The presence or even the thought of food is a strong stimulus for the secretion of saliva, which occurs unconsciously through activation of the automatic nervous system. In addition, saliva also plays a vital role in speech and the protection of the oral mucosa and teeth. Furthermore, food digestion begins in the mouth even before it reaches the stomach since saliva contains the enzyme amylase which is involved in the digestion of carbohydrates (1).

There are three well-known major salivary glands; the parotid glands, the submandibular glands, and the sublingual glands (figure 1), which are all present on both sides of the face. Additionally, there are 600-1000 minor salivary glands that are spread throughout the oral cavity (2).

Interestingly, although almost unthinkable in the 21st century, researchers have potentially discovered a fourth pair of major salivary glands in 2020. These are located posteriorly in the nasopharynx and were named the tubarial glands (3). Yet, the discovery of these tubarial glands is still debated, as some researchers argue that these are not a new pair of major glands, but rather an accumulation of minor salivary glands (4).

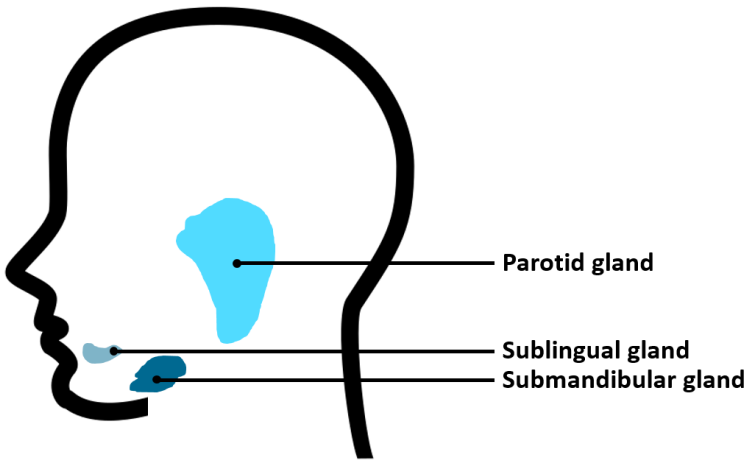


Figure 1: Salivary glands

Salivary Gland Disorders

Salivary gland pathology can be divided into *nonneoplastic* or *neoplastic* disease. Common nonneoplastic diseases of the salivary glands include sialolithiasis (salivary gland stones), sialadenitis (salivary gland infection, most often caused by bacteria, but mumps is a well-known viral cause), and salivary gland cysts (tiny fluid-filled sacs) (5).

Neoplastic disease of the salivary glands can be further classified into benign tumours or malignant tumours (cancer). The vast majority (70-85%) of salivary gland tumours arise in the parotid gland and about 75%-90% of these parotid gland tumours are benign. Tumours in the submandibular, sublingual, and minor salivary glands occur less frequently, but the proportion of malignant tumours in these locations is higher (6-9). Approximately 25-35% of the submandibular tumours and 85% of the sublingual tumours are malignant (7-13).

A pleiomorphic adenoma is the most common type of benign salivary gland tumour, and accounts for roughly 65-70% of the benign salivary gland tumours (8, 14). Although classified as benign, surgical resection of a pleomorphic adenoma is advised, since there is a chance (≈10% over 10 years) of transformation into a malignancy. A Warthin tumour is the second most common type of benign salivary gland tumour, accounting for 30% of all benign salivary gland tumours (8). Malignant transformation of a Warthin tumour is extremely rare. Furthermore, there are other forms of benign tumours that have a much lower incidence (combined they make up less than 5% of all benign salivary gland tumours), such as a basal cell adenoma and a myoepithelioma.

Malignant salivary gland tumours (salivary gland cancer) are histologically diverse, and significant advances have been made in the histopathological classification of salivary gland cancer in recent decades. The first WHO classification of salivary gland tumours, published in 1972, distinguished 7 subtypes of salivary gland cancer (15). The most recent WHO classification of salivary gland tumours, issued in 2017, distinguishes 22 different subtypes (shown in table 1) (16).

Table 1: Histopathological subtypes of Salivary Gland Cancer

WHO classification 1972 **Salivary gland cancer subtypes**	**WHO classification 2017** **Salivary gland cancer subtypes**
1. Mucoepidermoid tumour	1. Mucoepidermoid carcinoma
2. Acinic cell tumour	2. Adenoid cystic carcinoma
3. Adenoid cystic carcinoma	3. Acinic cell carcinoma
4. Adenocarcinoma	4. Polymorphous adenocarcinoma
5. Epidermoid carcinoma	5. Clear cell carcinoma
6. Undifferentiated carcinoma	6. Basal cell adenocarcinoma
7. Carcinoma in pleomorphic adenoma (malignant mixed tumour)	7. Intraductal carcinoma
	8. Adenocarcinoma, not otherwise specified
	9. Salivary duct carcinoma
	10. Myoepithelial carcinoma
	11. Epithelial-myoepithelial carcinoma
	12. Carcinoma ex pleomorphic adenoma
	13. Secretory carcinoma
	14. Sebaceous adenocarcinoma
	15. Carcinosarcoma
	16. Undifferentiated carcinoma
	17. Large cell neuroendocrine carcinoma
	18. Small cell neuroendocrine carcinoma
	19. Lymphoepithelial carcinoma
	20. Squamous cell carcinoma
	21. Oncocytic carcinoma
	22. Sialoblastoma *(uncertain malignant potential)*

Adapted and edited from: Thackray *et al*. Histological Typing of Salivary Gland Tumours. World Health Organization, Geneva, 1972 and El-Naggar *et al*. World Health Organization Classification of Tumours of Head and Neck. IARC, Lyon, 2017.
Displayed in the same order as the original source.
Abbreviation: WHO: World Health Organization.

Salivary Gland Cancer (SGC)

SGC has a relatively low incidence; each year approximately 2 per 100.000 people are diagnosed with this disease (17). Therefore, it is considered a rare cancer based on the RARECARE definition (<6/100,000/year) (18). The average age of diagnosis is 64 years (19). Due to the rarity of SGC, relatively little is known about the risk factors. While smoking and alcohol are well-established risk factors for other types of head and neck cancer (especially squamous cell carcinoma), most studies showed no or limited association between smoking or alcohol consumption and SGC (20-25). Prior exposure to ionizing radiation is the only well-known

risk factor for developing SGC (25-28). Furthermore, certain occupations (e.g. plumber, metal worker, and painter) may present a higher risk due to work-related exposure to chemicals (25, 28).

Symptoms

The primary symptoms of SGC patients depend on the site of occurrence. In general, a mass or swelling may be observed. Swelling in an affected parotid gland may lead to facial paralysis, due to the anatomical relationship of the parotid gland and the facial nerve, while in an affected minor salivary gland in the mouth this may lead to difficulty swallowing or opening the mouth. Ultimately, the diagnosis will be made with a combination of cytology, imaging, and histology. Furthermore, determining the SGC subtype is crucial, as biological behaviour differs between subtypes and, as a result, prognosis and treatment differ as well.

Treatment

Surgery is the cornerstone for the treatment of salivary gland cancer (figure 2), often supplemented by postoperative radiotherapy in case of certain SGC subtypes or specific tumour features e.g. positive resection margins or lymph node metastases (29). A large proportion of patients will be cured of SGC after this treatment. But unfortunately, in a proportion of patients the disease will recur. This can occur as a local recurrence, as regional lymph node metastases in the neck, or as distant metastases. In most cases, the disease can no longer be cured at this stage. Only patients with regional lymph node metastases may receive a lymph node neck dissection with curative intent.

For patients who can no longer be cured, the focus of treatment is on preventing or treating disease-related symptoms (palliative care) and prolonging life. Two SGC subtypes where a local recurrence or metastases often develop are adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC). **Palliative systemic treatment for these two SGC subtypes (ACC and SDC) is the main focus of this thesis.**

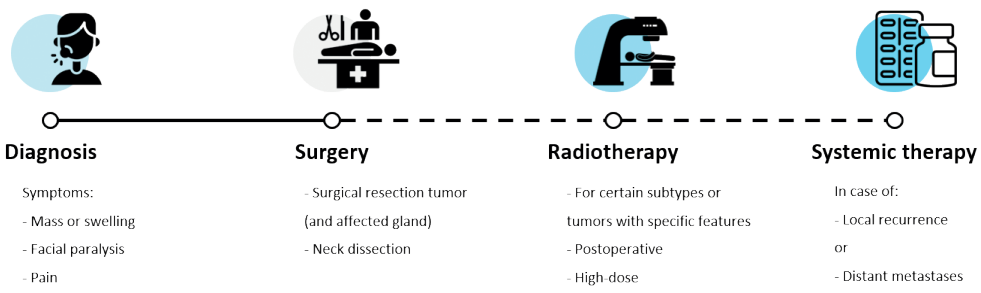


Figure 2: Overview of SGC treatment

Adenoid Cystic Carcinoma (ACC)

ACC is one of the most common SGC subtypes. It is the most common SGC subtype arising in the submandibular, sublingual, and minor salivary glands (e.g. at the tongue base or maxillary sinus) (9). Apart from the salivary glands, approximately one-third of ACC cases develop from other secretory glands throughout the body such as secretory glands of the breast, trachea, lung or Bartolin's gland (in women) (30-33). The male-to-female ratio of ACC disease is about 1:1.5 (16).

A typical feature of ACC disease is perineural spread (tumour growth along nerves) (34). Perineural spread complicates resection-free margins during surgery and is associated with a higher risk of local recurrence and metastatic disease (35). Furthermore, ACC is generally known as an indolent tumour, although some subgroups of patients may show a more aggressive disease course (e.g. NOTCH1 mutated ACC tumours (36)). Additionally, of the three basic growth patterns that can be observed during pathology assessment (i.e. tubular, cribriform, and solid), a solid growth pattern correlates with more aggressive ACC (37).

ACC has a low probability of lymphatic spread, but a high rate of haematological spread. Approximately 60% of all ACC patients will develop a local recurrence and/or distant metastases over time (38). In some ACC patients, recurrences or metastases may even occur 10 years after the initial treatment (late-onset disease recurrence) (39). Overall, distant metastases are most often observed in the lungs (77%), followed by bone (35%), pleura (22%), and liver (12%), but can occur in many other organs (36). Due to the indolent tumour growth of ACC the 5-year survival rates (approximately 89%) are deceptively encouraging. However, at longer time intervals the aggressiveness of ACC becomes apparent (40). For patients with metastatic ACC, the median overall survival is about 3 years (41).

Palliative systemic treatment in recurrent or metastatic ACC

Due to the indolent tumour growth and often asymptomatic metastases in ACC, specific considerations should be given to when to initiate systemic treatment (table 2).

In patients with slow-growing oligometastases (≤5 metastases), that developed >3 years after initial curative treatment, localized therapy (i.e. surgery or radiotherapy) should be considered (29).

In 2011, Laurie *et al.* published a systematic review that summarized systemic therapy options in locally recurrent or metastatic ACC disease (41). They concluded chemotherapy to be the best first-line treatment option, either with single agents such as mitoxantrone, vinorelbine, or epirubicin (response rates 10-15%) or with a combination of several agents, preferably a combination of cisplatin and anthracycline (example: cyclophosphamide plus doxorubicin plus cisplatin [CAP] showed response rates of 25%). Prior to 2011, none of the clinical studies

on other cytotoxic, hormonal or targeted agents showed encouraging results. Since then, several phase II trials showed hopeful results with anti-angiogenic tyrosine kinase inhibitors in ACC patients. Lenvatinib showed a response rate of 16% in 32 ACC patients (42) and apatinib showed the most promising results, with a response rate of 47% in 59 ACC patients (43).

Table 2: Specific considerations for initiating systemic treatment in recurrent or metastatic ACC

Important considerations systemic treatment in ACC patients	Explanation
Is localized therapy an option?	Patients with indolent growing oligometastases occurring >3 years after curative treatment might achieve local disease control with local ablative treatments (e.g. metastasectomy or stereotactic radiation therapy).
Do the side effects of systemic therapy outweigh current disease burden?	Several ACC patients, especially those with lung metastases, may have a limited disease burden. Some patients can remain asymptomatic for years. Therefore, active surveillance can be considered for patients with a limited disease burden.
Is rapid progressive disease expected?	Certain tumour characteristics make rapid progression more likely (e.g. NOTCH 1 mutation, solid growth pattern).
Critical evaluation of stable disease in clinical trials.	Due to the indolent tumour growth in many ACC patients, stable disease in clinical trials may reflect the natural behaviour of ACC rather than treatment efficacy.

Salivary Duct Carcinoma (SDC)

SDC in one of the most aggressive SGC subtypes and comprises approximately 4-10% of all SGC (16, 44-46). The male-to-female ratio of SDC is 3:1 (47, 48). Histopathologically, SDC shows resemblance to high-grade ductal carcinoma of the breast (16). SDC has a high likelihood of either lymphatic spread or haematological dissemination. At disease presentation, most patients (49-72%) already have regional lymph node involvement (47-49). Additionally, distant metastases are present at diagnosis in around 4-10% of the patients or arise relatively soon in the course of the disease (median time of 16 months after diagnosis until the presence of distant metastases) (47-50).

In total, around half of all SDC patients will develop distant metastases, most often in the lungs (54%), bones (46%), lymph nodes (≈40%), liver (≈25%) and brain (18%) (47, 51). The aggressiveness of SDC is evident in the poor median survival rates: 3.1-4.3 years, according to the largest SDC cohort studies (47, 48). In SDC patients with recurrent or metastatic disease, median overall survival is only 5 months when best supportive care is given (52), highlighting the need and urgency for effective systemic treatment.

Palliative systemic treatment in recurrent or metastatic SDC

Currently, prospective studies on the efficacy of chemotherapy in SDC tumours are lacking. To date, studies on chemotherapy for recurrent or metastatic SGC patients included all SGC subtypes (thus only a small proportion were SDC patients) or included only ACC patients (due to the relatively high incidence of this subtype). In the last years, 3 prospective studies have been published which evaluated the efficacy of targeted therapy in patients with recurrent/metastatic SDC and all showed promising results (53-55).

Since 78-96% of the SDC tumours are androgen receptor-positive (47, 56, 57), Fushimi *et al.* evaluated the effect of androgen receptor blockade in patients with recurrent or metastatic androgen receptor-positive SGC (54). Thirty-six patients, of which 96% were SDC patients, were treated with a combination of leuprorelin acetate (LHRH analogue) and bicalutamide (androgen receptor antagonist) and responses were observed in 42% of patients. As a logical next step, based on treatments in prostate cancer, Locati *et al.* explored the efficacy of second-line androgen receptor blockade with a combination of abiraterone (CYP17-inhibitor) and an LHRH agonist in SGC patients who previously had progression on first-line androgen receptor blockade (55). Of the 24 SGC patients (including 19 SDC patients), 5 patients responded (21%).

The human epidermal growth factor 2 receptor (HER2) is another interesting target for systemic therapy in SDC patients. Approximately one-third of all SDC tumours are HER2-positive (47, 56, 57). Takahashi *et al.* evaluated the combination of docetaxel and trastuzumab (anti-HER2 monoclonal antibody) in HER2-positive SDC patients (53). Of the 57 SDC patients, 40 patients (70%) responded to this anti-HER2 treatment.

Need for new treatment options for recurrent/metastatic ACC and SDC

As described above, relatively limited systemic treatment options exist for both ACC and SDC. SDC is an aggressive tumour that calls for further treatment options. And while ACC is often an indolent tumour, it is also generally considered a relentless tumour as almost all patients with recurrent or metastatic ACC disease will die from this disease. Therefore, new systemic treatment options for recurrent/metastatic ACC and SDC are necessary.

Challenges in Rare Cancer Research and Improving the Prognosis of Rare Cancers

More research is required to explore the efficacy of other systemic treatments. However, research in rare cancers is generally more challenging than in more common cancers due to several reasons (58):

- there is less pre-clinical research to build upon;
- there are limited patient numbers and thus it is more difficult to recruit patients in clinical trials;
- there is less publicity (due to rareness) and therefore less research funding.

Centralization and international collaborations are the two pillars we might build on to overcome these challenges (59). In the Netherlands head and neck oncological care (including salivary gland cancer) is centralized in 8 head & neck oncological centres and 6 affiliated head & neck oncological centres (60). In recent years, the Radboudumc (Nijmegen, the Netherlands) has evolved to a tertiary (expert) referral and research centre for salivary gland cancer. This is illustrated by the recent doctoral theses of Dr. Eline Boon and Dr. Wim van Boxtel (61, 62).

International collaboration is increasingly accomplished in collaborations such as EURACAN (the European Reference Network for all rare adult solid cancers) launched in 2017 and IRCI (International Rare Cancer Initiative).

In 2018 a report of the Netherlands Comprehensive Cancer Organisation (IKNL) showed that the survival rate of patients with rare cancer was significantly worse than that of patients with common cancers (63). Furthermore, this report also showed that the prognosis of more common cancers improved in the last two decades, while the prognosis of rare cancers showed only a minor improvement.

From Common to Rare

Given these challenges for research in rare cancers, it seems logical to learn and derive from research in more common cancers. Of all new anti-cancer drugs or drug combinations, ultimately only a small proportion shows favourable results in large clinical trials and is subsequently registered and applied in clinical care. This is due to the different stages of research into new anti-cancer drugs. Unfortunately, at each stage, several drugs prove ineffective or too toxic. For example, results from preclinical research often do not translate to results in the clinical setting (64). Because the number of SGC patients is small, only a

limited number of new agents or drug combinations can be investigated for this rare cancer. Therefore, extra care must be taken in deciding which treatments to investigate in SGC. One logical way of achieving this, is to learn from research performed in common cancers by electing to investigate treatments in rare cancers which have already proven to be effective in common cancers.

Funding

Partly because of the findings of the IKNL report from 2018 (mentioned above), the Dutch Cancer Foundation (KWF) aims to assist in closing this gap regarding survival between common cancers and rare cancers. The research presented in this thesis was funded by a grant from the Dutch Cancer Foundation (KWF) and the American Adenoid Cystic Carcinoma Research Foundation (ACCRF). Furthermore, our research team received funding from the Dutch Salivary Gland Cancer Patient Network (Patiëntenvereniging Speekselklierkanker), which was often collected through creative initiatives from patients and/or their relatives, and donations for salivary gland cancer projects to the Radboud Oncology Fund (Radboud Oncologie Fonds).

Kindly note that the Dutch Salivary Gland Cancer Patient Network was closely involved in the research described in this thesis, and the researchers provided semi-annual updates to the members on the progress and results of their research.

OUTLINE OF THIS THESIS

The main aim of this thesis is to investigate new systemic treatments for patients with recurrent or metastatic adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC).

Part 1: Systemic therapy for Salivary Gland Cancer
First, in **Chapter 2** we summarize the systemic therapy options for recurrent or metastatic SDC. Such a complete overview had previously been published for ACC (Laurie *et al.* 2011 *Lancet Oncol)*, but for SDC an overview via a broad systemic search was lacking.

In **Chapter 3** the results of a phase II clinical trial of cabozantinib (a tyrosine kinase inhibitor) are described. This study included 3 cohorts: ACC patients, SDC patients, and patients with other salivary gland cancer subtypes.

In **Chapter 4** we describe retrospective results of anti-HER2 treatment in recurrent or metastatic HER2-positive SDC patients. Apart from the combination of docetaxel, trastuzumab and pertuzumab as first-line anti-HER2 therapy, several patients were also treated with ado-trastuzumab emtansine (antibody-drug conjugate) as second-line anti-HER2 treatment. Furthermore, we searched for potential predictive biomarkers in this patient cohort.

Part 2: PSMA radioligand therapy for Salivary Gland Cancer
In **Chapter 5** a relatively new treatment modality is introduced: prostate-specific membrane antigen (PSMA) targeted radioligand therapy. This treatment has mainly been investigated for prostate cancer patients. In this chapter, we give a summary of the developments of PSMA targeted radioligand therapy, describe the current knowledge of PSMA expression in other solid cancers, including salivary gland cancer, and provide a perspective on broader clinical implementation of this treatment modality for other cancers than prostate cancer.

In **Chapter 6** the first results are presented of an ongoing phase II pilot study of PSMA targeted radioligand therapy for recurrent or metastatic ACC and SDC patients.

In **Chapter 7** the protocol of an ongoing imaging study is described. In prostate cancer, hormone therapy can result in an increased PSMA expression of the tumour cells. We aim to investigate whether hormone therapy also increases PSMA expression in SDC tumours. This is evaluated by repeated PSMA PET imaging.

Part 3: Increasing general knowledge of Salivary Gland Cancer
In **Chapter 8** the results of a collaboration between colleagues from the Department of Otorhinolaryngology and Head and Neck Surgery (Radboudumc) is presented. Due to the rarity and diversity of salivary gland tumours, histopathological diagnosis of these tumours

can be challenging. Through the Nationwide Pathology Network of the Netherlands (PALGA) we evaluated the frequency and outcomes of pathology consultations and revisions of major salivary gland tumours in routine clinical practice.

In **Chapter 9** we report on two SDC patients who were occupationally exposed to chromium VI (a carcinogen) and discuss a potential relationship between this exposure and the occurrence of their SDC tumour.

Chapter 10 describes nine SDC patients with cutaneous lymphangitis carcinomatosa. This manifestation of advanced SDC disease is specific for this subtype of salivary gland cancer. To date, however, little has been reported on this disease manifestation in SDC disease.

Chapter 11 describes the summary of this thesis and **Chapter 12** provides a general discussion of this thesis and suggestions for future research.

REFERENCES

1. Holsinger F.C., Bui D.T. (2007) Anatomy, Function, and Evaluation of the Salivary Glands. In: Myers E.N., Ferris R.L. (eds) Salivary Gland Disorders. Springer, Berlin, Heidelberg.
2. de Paula F, Teshima THN, Hsieh R, Souza MM, Nico MMS, Lourenco SV. Overview of Human Salivary Glands: Highlights of Morphology and Developing Processes. The Anatomical Record. 2017;300(7):1180-8.
3. Valstar MH, de Bakker BS, Steenbakkers R, de Jong KH, Smit LA, Klein Nulent TJW, et al. The tubarial salivary glands: A potential new organ at risk for radiotherapy. Radiother Oncol. 2020.
4. Guntinas-Lichius O, Ihrler S, Freesmeyer M, Gühne F, Kluge R, Bräuer L, et al. [Is there a new salivary gland? - Rather not!]. Laryngorhinootologie. 2021;100(1):12-4.
5. Rice DH. SALIVARY GLAND DISORDERS: Neoplastic and Nonneoplastic. Medical Clinics of North America. 1999;83(1):197-218.
6. Hugo NE, McKinney P, Griffith BH. Management of tumors of the parotid gland. Surg Clin North Am. 1973;53(1):105-11.
7. Bradley PJ, McGurk M. Incidence of salivary gland neoplasms in a defined UK population. British Journal of Oral and Maxillofacial Surgery. 2013;51(5):399-403.
8. Pinkston JA, Cole P. Incidence rates of salivary gland tumors: results from a population-based study. Otolaryngol Head Neck Surg. 1999;120(6):834-40.
9. Spiro RH. Salivary neoplasms: overview of a 35-year experience with 2,807 patients. Head Neck Surg. 1986;8(3):177-84.
10. Eveson JW, Cawson RA. Salivary gland tumours. A review of 2410 cases with particular reference to histological types, site, age and sex distribution. J Pathol. 1985;146(1):51-8.
11. Li LJ, Li Y, Wen YM, Liu H, Zhao HW. Clinical analysis of salivary gland tumor cases in West China in past 50 years. Oral oncology. 2008;44(2):187-92.
12. Tian Z, Li L, Wang L, Hu Y, Li J. Salivary gland neoplasms in oral and maxillofacial regions: a 23-year retrospective study of 6982 cases in an eastern Chinese population. International journal of oral and maxillofacial surgery. 2010;39(3):235-42.
13. Aegisdottir AL, Tryggvason G, Jonsdottir AM, Jonasson JG. Salivary gland tumours in Iceland 1986-2015: a nationwide epidemiological analysis over a 30-year time period. Apmis. 2020.
14. Valstar MH, de Ridder M, van den Broek EC, Stuiver MM, van Dijk BAC, van Velthuysen MLF, et al. Salivary gland pleomorphic adenoma in the Netherlands: A nationwide observational study of primary tumor incidence, malignant transformation, recurrence, and risk factors for recurrence. Oral oncology. 2017;66:93-9.
15. Thackray AC, Sobin LH. Histologic typing of salivary gland tumours. Geneva, Switzerland: WHO; 1972.
16. El-Naggar AK CJ, Grandis JR, Takata T, Slootweg PJ. WHO Classification of Head and Neck Tumours. 4th ed: WHO; 2017.
17. Boukheris H, Curtis RE, Land CE, Dores GM. Incidence of carcinoma of the major salivary glands according to the WHO classification, 1992 to 2006: a population-based study in the United States. Cancer Epidemiol Biomarkers Prev. 2009;18(11):2899-906.
18. Gatta G, van der Zwan JM, Casali PG, Siesling S, Dei Tos AP, Kunkler I, et al. Rare cancers are not so rare: the rare cancer burden in Europe. European journal of cancer (Oxford, England : 1990). 2011;47(17):2493-511.

19. American Cancer Society. Cancer Statistics Center. [Available from: http://cancerstatisticscenter.cancer.org.] Accessed on 05-10-2021.
20. Alvi S, Chudek D, Limaiem F. Parotid Cancer. In: StatPearls [Internet] Treasure Island (FL): StatPearls 2020.
21. Hayes RB, Bravo-Otero E, Kleinman DV, Brown LM, Fraumeni JF, Harty LC, et al. Tobacco and alcohol use and oral cancer in Puerto Rico. Cancer Causes & Control. 1999;10(1):27-33.
22. Muscat JE, Wynder EL. A case/control study of risk factors for major salivary gland cancer. Otolaryngol Head Neck Surg. 1998;118(2):195-8.
23. Spitz MR, Fueger JJ, Goepfert H, Newell GR. Salivary gland cancer. A case-control investigation of risk factors. Arch Otolaryngol Head Neck Surg. 1990;116(10):1163-6.
24. Pan SY, de Groh M, Morrison H. A Case-Control Study of Risk Factors for Salivary Gland Cancer in Canada. Journal of Cancer Epidemiology. 2017;2017:4909214.
25. Radoï L, Barul C, Menvielle G, Carton M, Matrat M, Sanchez M, et al. Risk factors for salivary gland cancers in France: Results from a case-control study, the ICARE study. Oral oncology. 2018;80:56-63.
26. Mayne SM, DE.; Winn, DM. Cancer Epidemiology and Prevention. New York: Oxford University Press; 2006. Cancers of the Oral Cavity and Pharynx; p. 674-696.
27. Land CE, Saku T, Hayashi Y, Takahara O, Matsuura H, Tokuoka S, et al. Incidence of salivary gland tumors among atomic bomb survivors, 1950-1987. Evaluation of radiation-related risk. Radiat Res. 1996;146(1):28-36.
28. Horn-Ross PL, Ljung BM, Morrow M. Environmental factors and the risk of salivary gland cancer. Epidemiology. 1997;8(4):414-9.
29. Geiger JL, Ismaila N, Beadle B, Caudell JJ, Chau N, Deschler D, et al. Management of Salivary Gland Malignancy: ASCO Guideline. Journal of Clinical Oncology. 2021;39(17):1909-41.
30. Arpino G, Clark GM, Mohsin S, Bardou VJ, Elledge RM. Adenoid cystic carcinoma of the breast. Cancer. 2002;94(8):2119-27.
31. Moran CA, Koss MN, Suster S. Primary adenoid cystic carcinoma of the lung. A clinicopathologic and immunohistochemical study of 16 cases. Cancer. 1994;73(5):1390-7.
32. Azar T, Abdul-Karim FW, Tucker HM. Adenoid cystic carcinoma of the trachea. The Laryngoscope. 1998;108(9):1297-300.
33. Copeland LJ, Sneige N, Gershenson DM, Saul PB, Stringer CA, Seski JC. Adenoid cystic carcinoma of Bartholin gland. Obstet Gynecol. 1986;67(1):115-20.
34. Paes FM, Singer AD, Checkver AN, Palmquist RA, De La Vega G, Sidani C. Perineural spread in head and neck malignancies: clinical significance and evaluation with 18F-FDG PET/CT. Radiographics. 2013;33(6):1717-36.
35. Vrielinck LJ, Ostyn F, van Damme B, van den Bogaert W, Fossion E. The significance of perineural spread in adenoid cystic carcinoma of the major and minor salivary glands. International journal of oral and maxillofacial surgery. 1988;17(3):190-3.
36. Ferrarotto R, Mitani Y, Diao L, Guijarro I, Wang J, Zweidler-McKay P, et al. Activating NOTCH1 Mutations Define a Distinct Subgroup of Patients With Adenoid Cystic Carcinoma Who Have Poor Prognosis, Propensity to Bone and Liver Metastasis, and Potential Responsiveness to Notch1 Inhibitors. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2017;35(3):352-60.
37. van Weert S, van der Waal I, Witte BI, René Leemans C, Bloemena E. Histopathological grading of adenoid cystic carcinoma of the head and neck: Analysis of currently used grading

systems and proposal for a simplified grading scheme. Oral oncology. 2015;51(1):71-6.

38. Spiro RH. Distant metastasis in adenoid cystic carcinoma of salivary origin. Am J Surg. 1997;174(5):495-8.

39. Matsuba HM, Spector GJ, Thawley SE, Simpson JR, Mauney M, Pikul FJ. Adenoid cystic salivary gland carcinoma: A histopathologic review of treatment failure patterns. Cancer. 1986;57(3):519-24.

40. Adenoid Cystic Carcinoma: Statistics [Available from: https://www.cancer.net/cancer-types/adenoid-cystic-carcinoma/statistics] Accessed on 5-10-2021.

41. Laurie SA, Ho AL, Fury MG, Sherman E, Pfister DG. Systemic therapy in the management of metastatic or locally recurrent adenoid cystic carcinoma of the salivary glands: a systematic review. The Lancet Oncology. 2011;12(8):815-24.

42. Tchekmedyian V, Sherman EJ, Dunn L, Tran C, Baxi S, Katabi N, et al. Phase II Study of Lenvatinib in Patients With Progressive, Recurrent or Metastatic Adenoid Cystic Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(18):1529-37.

43. Zhu G, Zhang L, Li R, Dou S, Yang W, Zhang C. Phase II trial of apatinib in patients with recurrent and/or metastatic adenoid cystic carcinoma of the head and neck: Updated analysis. Journal of Clinical Oncology. 2018;36(15_suppl):6026-.

44. Bjorndal K, Krogdahl A, Therkildsen MH, Overgaard J, Johansen J, Kristensen CA, et al. Salivary gland carcinoma in Denmark 1990-2005: a national study of incidence, site and histology. Results of the Danish Head and Neck Cancer Group (DAHANCA). Oral oncology. 2011;47(7):677-82.

45. McHugh JB, Visscher DW, Barnes EL. Update on selected salivary gland neoplasms. Arch Pathol Lab Med. 2009;133(11):1763-74.

46. Luukkaa H, Klemi P, Leivo I, Koivunen P, Laranne J, Mäkitie A, et al. Salivary gland cancer in Finland 1991–96: an evaluation of 237 cases. Acta Otolaryngol. 2005;125(2):207-14.

47. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.

48. Gilbert MR, Sharma A, Schmitt NC, Johnson JT, Ferris RL, Duvvuri U, et al. A 20-Year Review of 75 Cases of Salivary Duct Carcinoma. JAMA otolaryngology-- head & neck surgery. 2016;142(5):489-95.

49. Jayaprakash V, Merzianu M, Warren GW, Arshad H, Hicks WL, Jr., Rigual NR, et al. Survival rates and prognostic factors for infiltrating salivary duct carcinoma: Analysis of 228 cases from the Surveillance, Epidemiology, and End Results database. Head & neck. 2014;36(5):694-701.

50. Villepelet A, Lefevre M, Verillaud B, Janot F, Garrel R, Vergez S, et al. Salivary duct carcinoma: Prospective multicenter study of 61 cases of the Reseau d'Expertise Francais des Cancers ORL Rares. Head & neck. 2019;41(3):584-91.

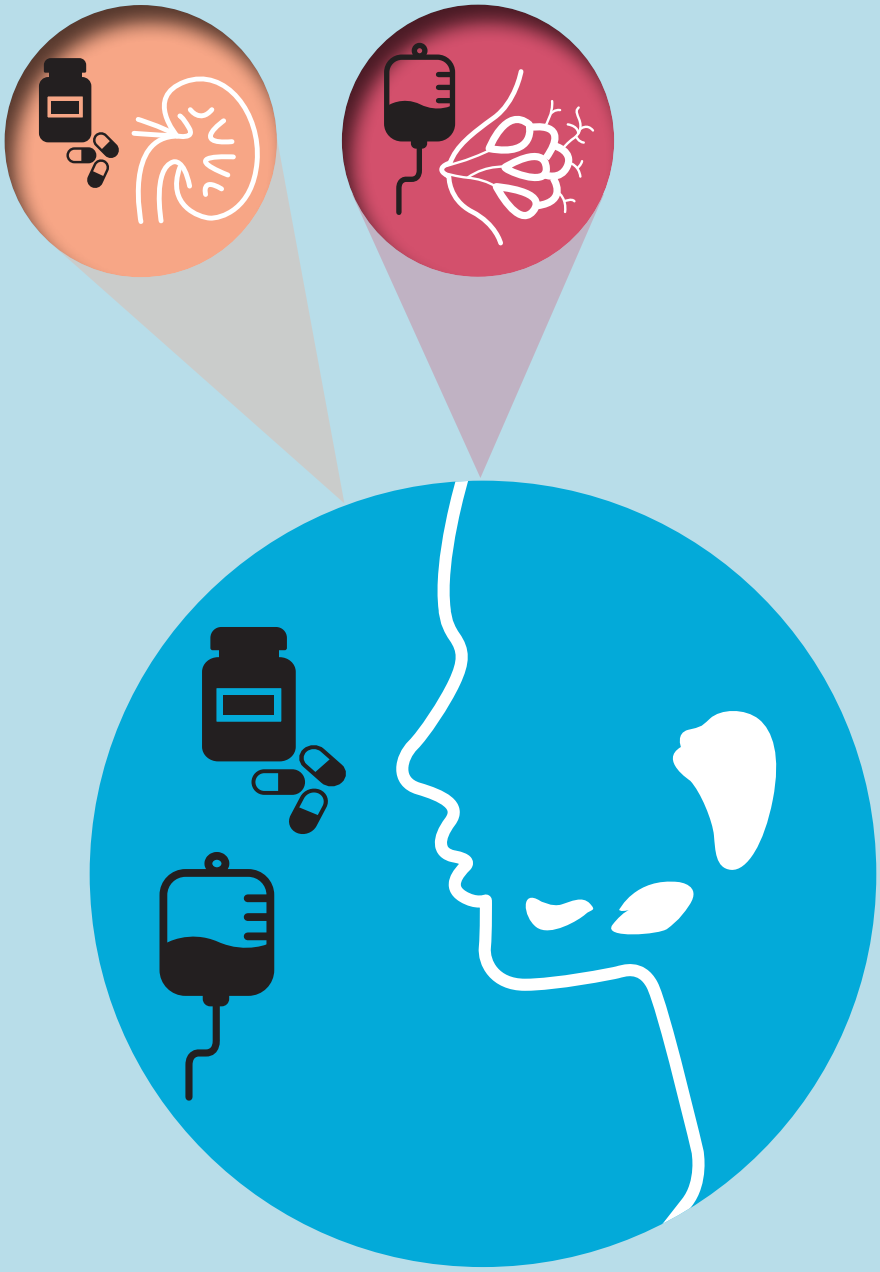
51. Johnston ML, Huang SH, Waldron JN, Atenafu EG, Chan K, Cummings BJ, et al. Salivary duct carcinoma: Treatment, outcomes, and patterns of failure. Head & neck. 2016;38 Suppl 1:E820-6.

52. Boon E, van Boxtel W, Buter J, Baatenburg de Jong RJ, van Es RJJ, Bel M, et al. Androgen deprivation therapy for androgen receptor-positive advanced salivary duct carcinoma: A nationwide case series of 35 patients in The Netherlands. Head & neck. 2018;40(3):605-13.

53. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct

Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.

54. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.
55. Locati LD, Cavalieri S, Bergamini C, Resteghini C, Colombo E, Calareso G, et al. Abiraterone Acetate in Patients With Castration-Resistant, Androgen Receptor–Expressing Salivary Gland Cancer: A Phase II Trial. Journal of Clinical Oncology.0(0):JCO.21.00468.
56. Schmitt NC, Kang H, Sharma A. Salivary duct carcinoma: An aggressive salivary gland malignancy with opportunities for targeted therapy. Oral oncology. 2017;74:40-8.
57. Takase S, Kano S, Tada Y, Kawakita D, Shimura T, Hirai H, et al. Biomarker immunoprofile in salivary duct carcinomas: clinicopathological and prognostic implications with evaluation of the revised classification. Oncotarget. 2017;8(35):59023-35.
58. Griggs RC, Batshaw M, Dunkle M, Gopal-Srivastava R, Kaye E, Krischer J, et al. Clinical research for rare disease: opportunities, challenges, and solutions. Mol Genet Metab. 2009;96(1):20-6.
59. Gatta G, Capocaccia R, Botta L, Mallone S, De Angelis R, Ardanaz E, et al. Burden and centralised treatment in Europe of rare tumours: results of RARECAREnet-a population-based study. The Lancet Oncology. 2017;18(8):1022-39.
60. Dutch Head and Neck Society (NWHHT) [Available from: https://www.nwhht.nl/centra/.] Accessed on 6-10-2021.
61. Boxtel Wv. Precision Medicine in Salivary Gland Cancer Patients [Doctoral thesis]. Nijmegen: Radboud University Nijmegen, Radboud Institute for Health Sciences 2020.
62. Boon E. Rare Cancers of the Head and Neck [Doctoral thesis]. Nijmegen: Radboud University Nijmegen, Radboud Institue for Health Sciences; 2019.
63. Integraal Kankercentrum Nederland (IKNL). *Kankerzorg in beeld - Zeldzame kanker.* 2018.
64. Hait WN. Anticancer drug development: the grand challenges. Nature Reviews Drug Discovery. 2010;9(4):253-4.



Part 1

Systemic therapy for Salivary Gland Cancer

Chapter 2

Systemic therapy in the management of recurrent or metastatic salivary duct carcinoma: a systematic review.

Maike J.M. Uijen, Gerben Lassche, Adriana C.H. van Engen-van Grunsven, Yuichiro Tada, Gerald W. Verhaegh, Jack A. Schalken, Chantal M.L. Driessen, Carla M.L. van Herpen

Cancer Treatment Reviews, 2020. ***89****, 102069.*

ABSTRACT

Background

Salivary duct carcinoma (SDC) is an aggressive subtype of salivary gland cancer. Approximately half of SDC patients will develop recurrences or metastases. Therapeutic palliative therapy is therefore often needed. The majority of SDC tumours express the androgen receptor (AR) and one-third express human epidermal growth factor receptor 2 (HER2), both are potential therapeutic targets. The aim of this paper is to systematically review and summarize the evidence on systemic palliative therapy for SDC and to provide treatment recommendations.

Materials and methods

Electronic libraries were systematically searched with a broad search strategy to identify studies where SDC patients received systemic therapy. Due to the rarity of SDC no restrictions were placed on study designs.

Results

The search resulted in 2014 articles of which 153 were full-text analysed. Forty-five studies were included in the analysis, which included in total 256 SDC patients receiving systemic therapy. Two phase II trials primarily including SDC patients were identified. The majority of the studies were case series or case reports, resulting in an overall low quality of available evidence. Based on studies including ≥5 SDC patients, objective responses to HER2 targeting agents were observed in 60-70%, to AR pathway agents in 18-53% and to chemotherapy in 10-50%.

Conclusion

For AR- or HER2-positive SDC, agents targeting these pathways are the cornerstone for palliative treatment. Regarding chemotherapy, the combination of carboplatin combined with a taxane is best studied. Regarding other targeted agents and immunotherapy evidence is anecdotal, limiting the formulation of treatment recommendations for these antineoplastic agents.

INTRODUCTION

Salivary duct carcinoma (SDC) is one of the 22 subtypes of salivary gland cancer (SGC), and has histological and immunohistochemical similarities with high grade intraductal and ductal carcinoma of the breast (1). SDC is most prevalent in the parotid gland and approximately one-third of the cases arises from a pleomorphic adenoma (carcinoma ex pleomorphic adenoma) (2, 3). SDC comprises approximately 4-10% of all SGC and distinguishes itself from many of the other subtypes by its often very aggressive behaviour (4-7). The latter is reflected in the median overall survival rate after diagnosis, which ranges between 48 and 79 months (2, 8, 9). In the case of resectable disease, patients will initially be treated with curative intent consisting of complete resection of the primary tumour often combined with a lymph node neck dissection, and in most cases followed by postoperative radiotherapy. At presentation, many patients already have regional lymph node involvement (49-72%), with a median number of 4 tumour positive lymph nodes (range 0-97), which negatively affects the overall survival (2, 3, 9). Additionally, distant metastases are present at diagnosis in around 4-10% of the patients or arise relatively briefly in the course of the disease (median 16 months until presence of distant metastases) (2, 3, 9, 10).

In total, around half of all patients will develop distant metastases, mostly in the lungs (54% of patients with distant disease), bones (46%), lymph nodes (approximately 40%), liver (approximately 25%) and brain (18%) (2, 11). In patients with recurrent or metastatic disease median overall survival is only 5 months when best supportive care is given (12). These numbers show the urgent unmet need for palliative systemic treatment in these patients.

Potential targets for systemic therapy are the androgen receptor (AR) and human epidermal growth factor 2 receptor (HER2). SDC show positivity for these receptors in approximately 78-96% and 29-46% of the cases, respectively (2, 13, 14). Described mutations include mutations in *TP53* (53-68%), *PIK3CA* (18-26%) and *HRAS* (16-23%), although not yet all of these mutations are druggable. The mutational landscape of SDC, with an overall tumour mutational burden of 1.7 mutations/megabase, might reveal other potential targets for systemic therapy. Overall, potentially actionable genetic alterations are present in 61% of the cases (13, 15, 16). Within the immunological microenvironment of SDC other clues might be present that rationalize immunotherapy treatment. For instance, 30-60% of SDC shows immunohistochemical positivity for the programmed death ligand 1 (PD-L1) (17, 18).

The rarity of SDC hampers performance of clinical trials. This limits the available evidence on the most effective treatment strategy, even though approximately half of all SDC patients will be considered for palliative systemic therapy during the course of their disease.

Therefore, the aim of this paper is to systematically summarize and review the available evidence on the treatment outcome of palliative systemic therapy in SDC patients, and to make treatment recommendations based on the findings.

METHODS

Search strategy

Articles were identified by conducting a search of the following electronic databases: PubMed (MEDLINE), Embase and the Cochrane library. The last search was conducted on the 29th of September 2019. The full search strategy can be found in supplementary tables 1 and 2. No restriction was placed on the year of publication. All article types and study designs were included. This includes experimental clinical trials and observational data (case series and case reports), since this is recommended for systematic reviews of rare diseases (19). All relevant studies were selected, regardless of the language of the article.

In addition, several clinical trial registries (clinicaltrials.gov, WHO International Clinical Trials Registry Platform, EU clinical trial register, ISRCTN and Australia and New Zealand Clinical Trial Registries) were searched for relevant studies or ongoing trials. A backward citation was performed by manually checking the reference lists from articles and reviews that were deemed relevant. Forward citation of the articles yielded from this search strategy was tracked using Web of Science.

Inclusion criteria

Studies were considered eligible for inclusion if at least one patient with incurable locally advanced, recurrent or metastatic (R/M) SDC was treated with systemic therapy as the main therapy. See table 1 for further details on the inclusion criteria.

Exclusion criteria

Articles were excluded if systemic therapy was administered as adjuvant or neoadjuvant treatment. Articles describing the treatment of SDC of non-salivary gland origin were excluded. Studies reporting on SDC patients besides other types of SGC were only included if the results of the subgroup of SDC patients were reported separately, unless the proportion of SDC patients exceeded 80% or if the treatment outcome of the SDC group could otherwise be determined (e.g. if no responses were observed in the entire study population). Studies in which systemic treatment was given in combination with local therapy (e.g. radiotherapy) were excluded.

Studies were assessed for eligibility independently by two reviewers (MU, GL). Data extraction was performed by one reviewer and integrally verified by a second. Disagreements were resolved by consensus or by consulting a third reviewer if deemed necessary (CvH).

Table 1: PICO search strategy

Population	Patients with incurable locally advanced salivary duct carcinoma, or incurable locoregional recurrences of salivary duct carcinoma or metastasized salivary duct carcinoma.
Intervention	Systemic therapy (e.g. chemotherapy, immunotherapy, hormonal therapy or targeted therapy)
Comparison	Not applicable
Outcome	Any of the following; - objective responses: e.g. objective response rate (complete and partial responses) or stable disease or duration of response (e.g. progression free survival) or - subjective responses (pain relief, symptom improvement) or - survival data (median progression free survival, overall survival).

Analysis

A meta-analysis was not possible due to heterogeneity in study designs, treatment types and outcomes of interest, so a narrative analysis was conducted.

Quality of evidence

Broad inclusion criteria and heterogeneity of included study designs limited the use of validated risk of bias tools. Bearing the principles of the GRADE approach in mind, an estimation of the overall quality of evidence was made (20).

RESULTS

The search strategy resulted in 2014 hits up to the 29th of September 2019. The abstracts of these 2014 studies were screened for eligibility, yielding 153 manuscripts. Full-text screening of these manuscripts resulted in 45 included studies (figure 1): nine phase II trials, one phase I trial, six case series and twenty-nine case reports (12, 21-62). No phase III studies were identified. In these 45 studies, in total 256 SDC patients were included who received systemic therapy for R/M disease. Several patients received multiple lines of therapy. All prospective studies and retrospective studies including ≥5 SDC patients are summarized in tables 2 and 3, respectively. The additional studies (mainly case reports) can be found in supplementary table 3.

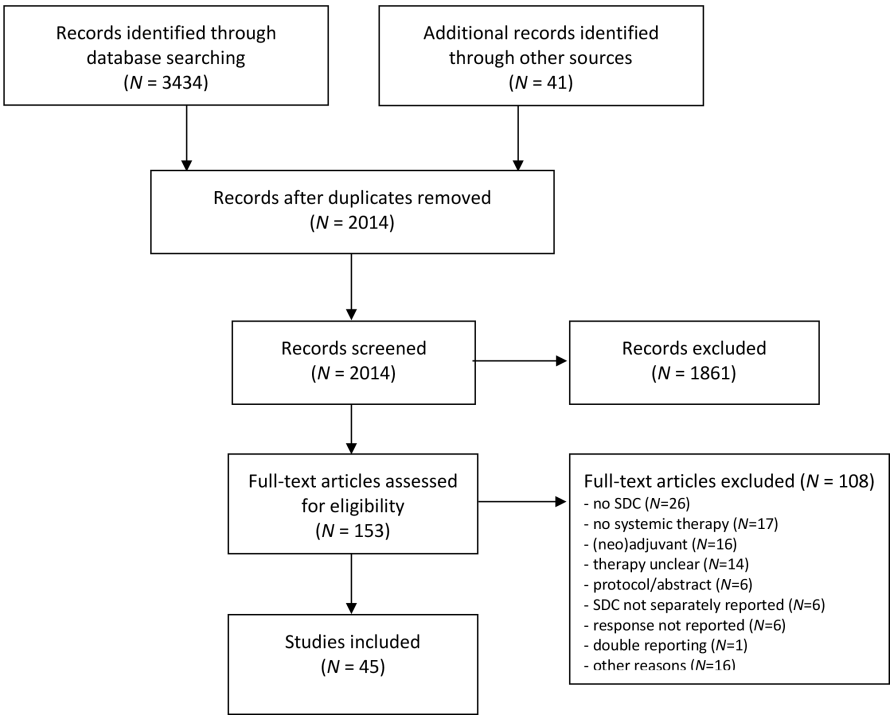


Figure 1: Flow diagram of search strategy

Hormonal therapy

One phase II trial studied the efficacy of combined androgen blockade (CAB) with leuprorelin acetate and bicalutamide in 36 patients (of which 64% with metastatic disease and 36% with unresectable locally advanced or locoregional recurrent disease) with advanced AR-positive SGC (94% SDC) (29). The results were not reported separately for the SDC patients. Objective responses occurred in 42% of the patients (complete response (CR): 11%, partial response (PR): 31%). The clinical benefit rate, defined as CR, PR, or stable disease (SD) ≥24 weeks, was 75% with a median progression-free survival (PFS) of 8.8 months and overall survival (OS) of 30.5 months (no survival data in historical cohort with other treatments reported).

Three case series reported on a total of sixty AR-positive SDC patients that received androgen deprivation therapy (ADT) (12, 45, 59). Patients were treated with either monotherapy (luteinizing hormone-releasing hormone (LHRH) analogues or the AR antagonists: enzalutamide or bicalutamide), or CAB (LHRH analogue and bicalutamide). In the largest of these studies, objective response was seen in 18% of the patients, all PR (12). In the other two studies, objective responses were seen in 53% and 50% (45, 59). Only the largest study reported the clinical benefit rate and compared survival to a best supportive care group.

The clinical benefit rate (CR, PR or SD) was 50%. The median OS in ADT treated SDC patients was 17 months, compared to 5 months in the best supportive care group. Additionally, this study reported on eleven SDC patients that received second-line ADT (LHRH analogue, either as monotherapy, or combined with bicalutamide and/or a 5-alfa-reductase-inhibitor), after progression on first-line ADT. The ten evaluable patients showed no objective response, but six patients had SD (60%) with a median duration of 9 months (12).

Furthermore, use of ADT in SDC was described in case reports in a total of twelve patients (23-26, 31, 43, 53, 57, 58, 62). Positive results were described in six patients (1 CR, 2 PR, 1 SD ≥6 months, 1 clinical improvement, 1 response on positron-emission tomography [PET] imaging). In addition, one patient was treated with a combination of hormone therapy and chemotherapy; this patient received bicalutamide, leuprolide and paclitaxel, which resulted in PR that was ongoing at 6 months (63).

HER2 targeted therapy

Fifty-seven SDC patients, of which 86% had distant metastases, were treated with the combination of trastuzumab and docetaxel in a phase II study including HER2-positive SDC patients (54). Objective responses were seen in 70% of the patients (14% CR and 56% PR) with a clinical benefit rate of 84% (CR, PR and SD ≥24 weeks). PFS and OS were 8.9 months and 39.7 months, respectively.

Another study (case series) evaluated the combination of trastuzumab with paclitaxel and carboplatin and in five HER2-positive SDC patients, all with distant metastases (42). The objective response rate was 60% (1 CR and 2 PR) with a median duration of response of 18 months.

In case reports, a total of twelve patients received a combination of trastuzumab with docetaxel/paclitaxel (22-24, 27, 30, 34, 38, 48, 56). Two patients had CR (17%) and eight patients had PR (67%). Duration of responses varied between 3 and 32 months.

Additionally, six patients received a combination of trastuzumab, docetaxel/paclitaxel and carboplatin, which led to 3 PR and 3 CR; little was reported on the duration of the responses (27, 35, 52, 55, 60, 64).

The combination of trastuzumab and pertuzumab in combination with different types of chemotherapy was given in six SDC patients (23, 50, 53, 58). The combination of trastuzumab and pertuzumab with chemotherapy (*N*=6) led to 3 CR, 1 PR, 1 SD and 1 response on PET imaging. Some durations were still ongoing but ranged from 3-17 months.

HER2 targeted therapy was given as monotherapy in three SDC patients. One patient with a parapharyngeal lymph node metastasis received trasGEX (second-generation monoclonal antibody of trastuzumab) in a phase I trial and achieved CR, without progression at 53 months follow-up (28). Two patients received trastuzumab monotherapy, leading to 1 CR, with ongoing duration at 18 months, and 1 SD for 5 months (40, 48). Additionally, four case reports described SDC patients treated with trastuzumab-emtansin (T-DM1) (22, 23, 53, 58). Two patients achieved PR (duration: 8 and 14 months), the other reports mentioned a clinical response of 12 months and an ongoing CR based on PET imaging at 29 months of follow-up.

Targeted therapy (other targets than AR and HER2)

Five prospective targeted therapy trials, which included SDC patients, focused on tyrosine kinase inhibitors (21, 32, 33, 37, 44). In all these studies, SDC patients comprised ≤10% of the total study population. No case series were identified reporting on targeted therapy in SDC patients, only several case reports.

BRAF:

The efficacy of vemurafenib was examined in a basket study for solid tumours with BRAF mutations (32). This study included one SDC patient, which achieved CR lasting for 8 months. Furthermore, one case report described the treatment combination of dabrafenib and trametinib in one SDC patient with a BRAF V600E mutation (43). The patient showed marked improvement of osseous metastases, but progression occurred at 13 months.

EGFR:

Two phase II trials in SGC patients studied the effect of EGFR inhibitors (gefitinib, lapatinib) (21, 33). In both trials no objective responses were observed in the entire study population. The trial of gefitinib (*N*= 37) included three SDC patients. In the non-adenoid cystic carcinoma (ACC) cohort (total of 18 patients) four patients did have SD ≥9 months, but it is unclear if these were SDC patients (33). None of the four SDC patients treated with lapatinib had SD >6 months (21). In a case report, lapatinib resulted in a complete resolution of skin lesions, with progression after 18 months (22).

VEGFR:

The effect of sorafenib and nintedanib (VEGFR inhibitors) was studied in two phase II trials in SGC patients (37, 44). Of the two SDC patients in the trial with sorafenib, one had a PR. The other patient did not have an objective response (44). No objective responses were observed in the nintedanib trial. In the only SDC patient in this trial SD was achieved for 7.3 months (37).

Other targets:

In one case report treatment with cabozantinib for two *NCOA-RET* gene fusion-positive SDC patients was evaluated. Both patients experienced clinical improvement with no specification

of the duration. One case report mentioned treatment with a combination of trastuzumab, lapatinib and bevacizumab, leading to a PR in a single SDC patient. Besides one asymptomatic bone metastasis treated with radiation, the patient had no signs of further progression at 25 months (27). The combination of temsirolimus and bevacizumab was given to two SDC patients; one patient showed a visual response of skin lesions, and the other patient had a PR for 3 months (51). One study reported on different targeted therapy approaches in three separate patients (supplementary table 3) (47). Three PR were observed: one in a patient treated with a combination of BRAF- and MEK-inhibitors, one in a patient treated with a PI3K-inhibitor, and one in a patient treated with TORC1/2 inhibitor with durations of 5, 12 and 3.7 months, respectively.

Chemotherapy

In total, three SDC patients received chemotherapy in prospective clinical trials (39, 41). Two received CAP (cyclophosphamide, doxorubicin and cisplatin) and one patient was treated with gemcitabine and cisplatin. CAP resulted in one PR and one SD; gemcitabine combined with cisplatin resulted in PR. Little was reported regarding the duration of the responses (table 2).

Three case series reported on the effect of chemotherapy in SDC patients. A total of 40 SDC patients were treated with chemotherapy in these studies. In two studies patients were treated with a combination of carboplatin and a taxane (docetaxel/paclitaxel) (46, 49). The combination of carboplatin and paclitaxel (*N*=18) resulted in objective responses in 39% of the patients, and the combination of carboplatin with docetaxel (*N*=12) resulted in objective response in 50%. One study reported on the use of several different chemotherapeutics in 10 SDC patients, mainly platinum-based regimes (59). One patient (10%) achieved CR (treatment schedule unclear), but there were no other objective responses reported.

Additional SDC case reports mentioned the effect of different combinations of chemotherapy in seven patients (24, 30, 43, 48, 61). Three out of these seven patients had PR (one on CAP, one with cisplatin+vinorelbine, one with cisplatin+docetaxel). In addition, in one case report first-line treatment of cisplatin and 5-fluorouracil combined with cetuximab was given, leading to a CR that lasted 3 months. As second-line cisplatin and 5-fluorouracil (5-FU) were replaced by tegafur-gimeracil-oteracil potassium (which also contains a 5-FU prodrug), which led to SD ongoing at 7 months (36).

Immunotherapy

One case report describing the use of immunotherapy in a SDC patient was identified. This patient received nivolumab as second-line therapy (43). Dosage and efficacy were not reported; the patient stayed on therapy for 3 months and treatment was discontinued due to severe fatigue.

Quality of evidence

Using the GRADE approach on the gathered evidence of systemic therapy in SDC led to low or very low quality of evidence, as was expected in this rare disease. This is mainly due to study designs and methodology of the studies. There were no sound prospective studies with an appropriate control group. Therefore, most studies used PFS and OS as surrogate endpoints to indicate increased overall survival benefit. Although the rarity of SDC limits efficient patient accrual, 2 prospective trials have been performed in which a large proportion of included patients were SDC patients. These studies did not predefine the sample size (29, 54). Earlier performed studies on SGC generally included a more heterogeneous groups of SGC patients, limiting their usefulness to draw conclusions on efficacy in each included subtype (indirectness of evidence).

The efficacy of systemic therapy strategies that have been examined in larger studies (>10 patients) shows consistency in effect size. ADT shows objective response rates of 42% in the only prospective study that has been performed, which is comparable to the 50% and 53% response rate reported in retrospective studies. Trastuzumab combined with taxane chemotherapy showed objective responses in 70% of the patients in retrospective data, which is comparable to the 84% when combining the objective responses reported in several case reports. The combination of carboplatin with taxane chemotherapy shows comparable results in two retrospective studies, with objective responses of 39% and 50%. Efficacy of other targeted therapy approaches remains unclear, as evidence is anecdotal and mainly derived from single case reports.

Table 2: Prospective studies in which SDC patients were included

Study	Design	Patient characteristics	Drugs	Only SDC	N of SDC patients (% of total number)	Disease stage*	Prior systemic therapy*	Response*	Median Survival (PFS, OS)*	Remarks
Hormone therapy										
Fushimi et al.(29)	Phase 2	SGC AR+	leuprorelin + bicalutamide	N	34 (94%)	NR	NR	NR	NR	Results total study (*N*=36): CR: 4 (11%) PR: 11 (31%) SD: 16 (44%) PD: 5 (14%) ORR: 42% [26%-59%] CBR: 75% [58%-88%] (includes SD>24 weeks) PFS: 8.8 mo, OS: 30.5 mo
HER2 targeted therapy										
Takahashi et al.(54)	Phase 2	SDC HER2+	docetaxel + trastuzumab	Y	57 (100%)	LR: 8 (14%) DM: 49 (86%)	chemo: 20 (35%) ADT: 3 (5%)	CR: 8 (14%) PR: 32 (56%) SD: 14 (25%) PD: 3 (5%)	PFS: 8.9 mo OS: 39.7 mo	ORR: 70% [57%-82%] CBR: 84% [72%-93%] (includes SD>24 weeks) 14 pts received other treatment after 6 cycli.
Fiedler et al.(28)	Phase 1	Solid tumours HER2+	trasGEX	N	1 (3%)	LR: 1 (100%)	NR	CR: 1 (100%)	PFS: 53 mo†	
Targeted therapy (other targets than AR and HER2))										
Agulnik et al.(21)	Phase 2	SGC EGFR+ and/or erbB2+	lapatinib	N	4 (10%)	NR	NR	NR	NR	No objective responses in trial SD>6 mo: 13/40 pts, but no SDC.
Jakob et al.(33)	Phase 2	SGC	gefitinib	N	3 (8%)	NR	NR	NR	NR	No objective responses in trial Non-ACC cohort: 4 pts SD >9 mo, unclear if SDC.
Locati et al.(44)	Phase 2	SGC	sorafenib	N	2 (5%)	NR	NR	PR: 1 (50%)	NR	Other SDC patient unclear if PD or SD.
Hyman et al.(32)	Phase 2 basket	Solid tumours BRAF V600 mutation	vemurafenib	N	1 (1%)	NR	NR	CR: 1 (100%)	PFS: 8 mo	
Kim et al.(37)	Phase 2	SGC	nintedanib	N	1 (5%)	NR	chemo: 1 (100%)	SD: 1 (100%)	PFS: 7.3 mo OS: 10.1 mo†	No objective responses in trial.

Table 2: Continued

Chemotherapy									
Licitra et al.(41)	Phase 2	SGC	CAP	N	2 (9%)	LR: 1 (50%) DM: 1 (50%)	NR	PR: 1 (50%) SD: 1 (50%)	PR: PFS: 6 mo, OS: 12 mo SD: PFS: NR, OS: 16 mo
Laurie et al.(39)	Phase 2	SGC	gemcitabine + cisplatin	N	1 (3%)	NR	NR	PR: 1 (100%)	NR

Abbreviations: AR: androgen receptor, CAP: cyclophosphamide, doxorubicin and cisplatin, CBR: clinical benefit rate, CR: complete response, DM: distant metastases, EGFR: epidermal growth factor receptor, erbB2+: refers to HER2 positivity, HER2: human epidermal growth factor receptor 2, LR: locoregional, mo: months, N: no, NR: not reported, ORR: overall response rate, OS: overall survival, PD: progressive disease, PFS: progression free survival, PR: partial response, pts: patients, SD: stable disease, SDC: salivary duct carcinoma, SGC: salivary gland cancer, trasGEX: second generation monoclonal antibody of trastuzumab, Y: yes.

*only SDC patients are reported

† (response) ongoing at time of report

Table 3: Retrospective studies in which ≥5 SDC patients were included

Study	Design	Patient characteristics	Drugs	Only SDC	N of SDC patients (% of total number)	Disease stage*	Prior systemic therapy*	Response*	Median Survival (PFS, OS)*	Remarks
Hormone therapy										
Boon et al.(12)	Case series	SDC AR+	bicalutamide +/- goserelin	Y	35 (100%)	LR: 2 (6%) DM: 33 (94%)	no	CR: 0 (0%) PR: 6 (18%) SD: 11 (32%) PD: 17 (50%)	PFS: 4 mo OS: 17 mo	Evaluable: 34/35 pts CBR: 50% Median PFS for pts with PR or SD: 11 mo. 11 pts later received second-line ADT (goserelin +/- bicalutamide +/- 5-ARI) Evaluable pts: 10/11 pts. SD: 6 (60%) with median PFS of 9 mo. PD: 4 (40%) CBR: 60%
Viscuse et al.(59)	Case series	SGC AR+	leuprolide +/- bicalutamide or bicalutamide or enzalutamide	N	17 (85%)	NR	no	CR or PR: 9 (53%)	NR	Study included 35 pts, 20 received ADT (17 SDC), 14 chemo (10 SDC) See row below.
Locati et al.(45)	Case series	SGC AR+	bicalutamide + triptorelin	N	8 (47%)	DM: 8 (100%)	chemo: 3 (38%)	CR: 2 (25%) PR: 2 (25%) SD: 3 (37.5%) PD: 1 (12.5%)	NR	Duration responses: CR: 11 and 39 mo PR: 6 and 7 mo SD: 8, 10 and 23 mo
HER2 targeted therapy										
Limaye et al.(42)	Case series	SDC HER2+	paclitaxel + trastuzumab + carboplatin	Y	5 (100%)	DM: 5 (100%)	chemo: 1 (20%)	CR: 1 (20%) PR: 2 (40%) SD: 0 (0%) PD: 2 (40%)	DoR: 18 mo	Study also reports on 8 pts in adjuvant setting.

Table 3: Continued

Chemotherapy										
Nakano et al.(46)	Case series	SGC	paclitaxel + carboplatin	N	18 (47%)	NR	chemo: 2 (11%)	Objective response: 7 (39%)	NR	Objective response was not further specified.
Okada et al.(49)	Case series	SGC	carboplatin + docetaxel	N	12 (50%)	NR	yes: 9 (75%)	CR: 2 (17%) PR: 4 (33%) SD: 3 (25%) PD: 3 (25%)	PFS: 8.0 mo OS: 32.6 mo	Disease stage NR, but target lesions: LR: 3 (35%) DM: 8 (67%)
Viscuse et al.(59)	Case series	SGC	chemo	N	10 (71%)	NR	no	CR: 1 (10%) PR: 0 SD: NR	NR	Different chemo combinations were given. SD: 3/14 pts, unclear if SDC

Abbreviations: ADT: androgen deprivation therapy, AR: androgen receptor, CBR: clinical benefit rate, CR: complete response, DoR: duration of response, DM: distant metastases, HER2: human epidermal growth factor receptor 2, LR: locoregional, mo: months, N: no, NR: not reported, OS: overall survival, PFS: progression free survival, PR: partial response, pts: patients, SD: stable disease, SDC: salivary duct carcinoma, SGC: salivary gland cancer, Y: yes, 5-ARI: 5-alfa-reductase-inhibitor.

*only SDC patients are reported.

DISCUSSION

Main findings

In this systematic review, evidence of the use of different systemic therapy approaches in R/M SDC is summarized. SGC is a heterogeneous and rare disease, and SDC is only one of many subtypes. This impedes treating physicians' search for the best treatment for their patients as it is hard to get an overview of the known evidence. This review aimed to aid in this search and is, to our best knowledge, the first systematic review presenting all known evidence specifically for systemic therapy approaches in R/M SDC patients through a broad search (although other reviews have summarized evidence for SGC in general, thus including SDC) (65-67).

Due to the rarity of the disease, most studies had small sample sizes and included not only SDC patients, but a broader range of SGC patients. However, recently two relatively large prospective studies have been performed (29, 54). Overall, given the close histologic resemblance of SDC to high grade intraductal and ductal carcinoma of the breast and the analogy with prostate cancer due to the omnipresence of the AR pathway activation, investigated therapeutic strategies closely mimic the treatment of these common cancers (e.g. ADT and HER2 targeted therapy) (1, 68-70).

Comparing systemic treatments

No prospective randomized studies comparing different systemic treatments in SDC patients have been performed. Retrospectively, one study compared first-line ADT versus chemotherapy. Selection between these regimes was mainly based on the availability of AR testing at the time of patient encounter, possibly introducing bias in the comparison (59). Median overall survival was comparable between first-line ADT and first-line chemotherapy, but as most patients received other lines of therapy after progression on the first-line of ADT/chemotherapy effect size of both interventions is hard to establish. An ongoing phase II study is currently evaluating the efficacy of chemotherapy versus ADT in patients with advanced, AR-positive SGCs (EORTC-1206, NCT01969578).

Several studies evaluated the efficacy of ADT. Only in the study of Boon *et al.* the majority of patients received bicalutamide monotherapy. The results of this study are less favourable as compared to studies where the majority of patients received a LHRH analogue with or without bicalutamide (CAB). In prostate cancer, where the role of ADT has been examined extensively, LHRH analogues are recommended as standard of care when hormonal therapy is advised (71). The role of bicalutamide is less prominent, although a large study showed a small survival advantage when bicalutamide is added to LHRH analogues (72). This suggests, for male patients, that treatment with a LHRH analogue with or without bicalutamide is the reasonable choice over bicalutamide monotherapy (although bicalutamide monotherapy has the advantage of libido and sexual potency retention) (73).

The registration of novel AR-targeted drugs even widens the options for AR-targeted therapy in SDC (74). For female patients, bicalutamide monotherapy might still be considered due to lower physiological testosterone levels, but pre-treatment measurement of testosterone would be advisable. Additional ADT treatment optimization might arise from research on the mutational landscape of SDC, which reveals several potential resistance mechanisms to androgen receptor blockade, including AR-V7 splice variants and *FOXA1* mutations (16).

HER2 targeted therapy has previously shown positive results in breast cancer and has subsequently been explored for other malignancies with HER2 expression, including SDC (69, 75). In SDC the combination of trastuzumab and chemotherapy showed impressive response rates in a phase II clinical trial, which makes this combination a good choice in the palliative treatment of HER2-positive SDC patients (54). In breast cancer, the addition of pertuzumab to trastuzumab plus docetaxel has also shown favourable results. In a phase III study, PFS was 18.5 months (OS 56.5 months) for the group with addition of pertuzumab, compared to 12.4 months (OS 40.8 months) for the control group (placebo, trastuzumab, docetaxel) (76, 77). These results provide a rationale for addition of pertuzumab to trastuzumab plus docetaxel in SDC patients, although the high response rates for this combination in SDC makes it more difficult to study the potential additive value of pertuzumab as it already is a very effective regime.

Treatment recommendations

To guide treatment decisions it is recommended to test for AR and HER2 in all SDC patients. Especially in R/M disease this is a prerequisite, although ADT might also be important in the adjuvant setting (78). AR should be assessed by immunohistochemistry (IHC), ideally AR positivity should be scored similarly to the scoring method of the prospective trial of Fushimi et al. (29), where AR positivity was evaluated just like the estrogen and progesterone receptors in accordance with the American Society of Clinical Oncology/College of American Pathologists guidelines for the evaluation of breast cancer predictive factors (79). However, preliminary results indicate that functional AR-pathway activation measurements on mRNA level might better predict response to ADT (80). HER2 status should be assessed by both IHC and *in situ* hybridization (ISH) and interpreted following the guidelines for HER2 assessment in breast cancer (81). Remarkably, the response rate of HER2 targeted therapy in HER2+ SDC patients is high (70%), even compared to the response rate in HER2+ breast cancer patients (41.3%) (54, 82). Whether lower HER2 expression in SDC patients could potentially be sufficient for clinical response to HER2 targeted therapy needs further investigation.

In case of both AR and HER2 positivity (approximately 30% of the patients), AR targeted strategies or HER2 targeted therapy have not yet been compared head-to-head. Indirect comparison of these treatments in different (prospective and retrospective) studies hints toward superior response rates and OS for HER2 targeted therapy combined with taxane

chemotherapy as compared to ADT therapy (29, 54). Especially in SDC patients with visceral metastases, extensive or rapidly progressive disease, a HER2-targeted agent in combination with a taxane is recommended over ADT as first-line therapy (83). In addition, expected side effects might steer the choice between these therapies. ADT-related side effects include bone loss, metabolic changes, gynecomastia, muscle loss and hot flashes (84). Upon HER2 targeted therapies (combined with taxane chemotherapy) haematological toxicity can be expected, as well as various other symptoms such as anorexia and fatigue (54, 69). Furthermore, cardiotoxicity is an important side effect that can occur during HER2 targeted treatment.

As first-line HER2-targeted therapy, trastuzumab combined with chemotherapy is the preferred choice based on the available evidence (54). Evidence supporting the addition of pertuzumab to the first-line treatment is limited, further research is warranted (23, 50, 53, 58). Recently, preliminary results from a phase II basket study in SGC patients were reported on ASCO. In total 10 HER2+ SGC patients received trastuzumab emtansine (TDM-1) treatment (85). Nine patients showed objective responses (90%), including 5 complete responses after prior trastuzumab, pertuzumab and ADT. Based on these promising results, TDM-1 should be considered for patients who progressed after trastuzumab +/- pertuzumab. TDM-1 might even be considered as first-line treatment since the response rates of this study seem higher (90%) than the response rates of trastuzumab (60-70%), although trastuzumab has been studied in larger patient populations as compared to TDM-1.

Recently, another HER2-targeted therapy, the antibody-drug conjugate trastuzumab deruxtecan (T-DXd), led to clinical responses in HER2+ SDC patients. In this phase I study in various solid malignancies 8 SGC patients were included. Two patients with SDC had PR upon treatment with T-DXd and two SDC patients had SD (other responses were not reported) (86).

When initiating ADT, the combination of a LHRH analogue with bicalutamide is preferred over monotherapy in terms of response rate and PFS, although CAB has higher rates of (mild) adverse events. When considering chemotherapy in HER2 negative SDC, although still limited, most evidence exists for the efficacy of carboplatin in combination with taxane chemotherapy in SDC, but CAP chemotherapy may be considered as an alternative. Practical guidelines and dosage recommendations are listed in table 4.

Table 4: Dosage recommendations

Tumour characteristics SDC	Treatment	Drug(s)	Dosage	Duration	Rationale
AR+	ADT	leuprorelin + bicalutamide	- Leuprorelin acetate s.c. 3.75 mg every 4 weeks or 11.25mg every 12 weeks - Bicalutamide 80mg OD	Until PD	Dosage from prospective clinical trial of Fushimi et al.(29)
HER2+	HER2-targeted + chemotherapy	trastuzumab + docetaxel	Every 3 weeks: - Trastuzumab i.v.: loading dose 8 mg/kg, followed by 6 mg/kg - Docetaxel: 70 mg/m^2 Patients ≥ 70 years of age: reduce docetaxel to 55 mg/m^2	6 cycles Consider continuation of combination or trastuzumab monotherapy	Dosage from prospective clinical trial of Takahashi et al.(54)
n.a.*	Chemotherapy	paclitaxel + carboplatin	Every 3 weeks: - carboplatin: AUC = 6 - paclitaxel: 200mg/m^2	The median number cycles was five (range: 2–12)	Dosage from retrospective trial of Nakano et al.(46)
n.a.*	Chemotherapy	docetaxel + carboplatin	Every 3 weeks: - carboplatin: AUC = 5 - docetaxel: 70 mg/m^2	6 cycles	Dosage from retrospective trial of Okada et al.(49)

Abbreviations: AR: androgen receptor, ADT: androgen deprivation therapy, HER2: human epidermal growth factor receptor 2, OD: once daily, n.a: not applicable, i.v.: intravenously, PD: progressive disease, SDC: salivary duct carcinoma, s.c.: subcutaneously.

*These treatments could be considered for HER2 negative patients, in HER2+ patients, chemotherapy should be combined with HER2-targeted agents.

In addition, patients might benefit from other targeted therapies as positive results have been described in small numbers of patients. This requires testing for genetic alterations (e.g. for patients not (longer) eligible for ADT or HER2 targeted strategies) known to be targetable with targeted agents. Whole exome or targeted sequencing of 31 SDC tumours showed potentially druggable targets in 61% of these tumours (87). Although this also includes amplification of *ERBB2* (35%), which would likely have been identified through alternative tests: IHC and FISH, other relevant mutations such as *PIK3CA* (23%), *HRAS* (23%) or *BRAF* V600E mutations were identified in a substantial amount. Patients with PIK3CA or HRAS mutations might benefit from PI3K inhibitors and/or tipifarnib and patients with *BRAF* mutations might benefit from treatment with vemurafenib, since these therapies showed antitumour effects in patients with other malignancies (32, 88, 89). Preferably, such treatments should be examined in a clinical trial setting. This also applies for immunotherapy; more research is required to establish the effect of immunotherapy in SDC patients. A recent study provided more insight in the immune microenvironment of SDC, which might aid in the selection of precision immunotherapy. Through transcriptomic analyses it is indicated that the SDC does not escape immune responses by excluding T-cells, in fact SDC has relatively high levels of immune cell infiltration. The immune evasive capacity appears to rely on high expression of T-cell checkpoints and high levels of T cell dysfunction (90).

Strengths and limitations

In this systematic review all known evidence for systemic treatment strategies in R/M SDC is identified and summarized through a thorough and broad literature search following systematic review guidelines. However, the strength of our treatment recommendations is severely impacted by the low quality of the overall evidence synthesis. Included and described studies are mostly limited to retrospective data, consisting of case series and case reports. This bears the risk of substantial publication bias and limits drawing firm conclusions.

CONCLUSION

This systematic review exposes an overall paucity in well-performed studies on the efficacy of treatment strategies in R/M SDC, although there is an urgent unmet clinical need in this patient category with a dismal prognosis. The evidence that is present is of low quality and the vast majority of cases is retrospectively analysed, although recently relatively large prospective studies (>30 SDC patients) have been published (2018 and 2019) (29, 54). The available knowledge points towards a strategy in which testing for activation of the AR and HER2 pathway is a prerequisite in choosing the right treatment option. In AR-positive patients, ADT should be the first-line whereas in HER2+ patients HER2 based treatment with trastuzumab, combined with a taxane is the reasonable first choice. Upon progression in HER2+ SDC trastuzumab-emtansine is another promising strategy. Treatment decisions in

patients co-expressing AR and HER2 should be guided by clinical factors, but we advocate the use of HER2-targeted treatment in most patients. When chemotherapy is considered in HER2-negative patients, the combination of carboplatin with a taxane should be considered. Eligibility of patients for treatment with specific targeted therapies depends on the presence of mutations targetable with currently registered drugs or drugs under investigation in basket trials. Testing for these targets is therefore recommended.

REFERENCES

1. Kleinsasser O, Klein HJ, Hubner G. [Salivary duct carcinoma. A group of salivary gland tumors analogous to mammary duct carcinoma]. Arch Klin Exp Ohren Nasen Kehlkopfheilkd. 1968;192(1):100-5.
2. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.
3. Gilbert MR, Sharma A, Schmitt NC, Johnson JT, Ferris RL, Duvvuri U, et al. A 20-Year Review of 75 Cases of Salivary Duct Carcinoma. JAMA otolaryngology-- head & neck surgery. 2016;142(5):489-95.
4. El-Naggar AK CJ, Grandis JR, Takata T, Slootweg PJ, eds. WHO classification of head and neck tumours, 4th edn.: World Health Organization (IARC); 2017.
5. Bjorndal K, Krogdahl A, Therkildsen MH, Overgaard J, Johansen J, Kristensen CA, et al. Salivary gland carcinoma in Denmark 1990-2005: a national study of incidence, site and histology. Results of the Danish Head and Neck Cancer Group (DAHANCA). Oral oncology. 2011;47(7):677-82.
6. McHugh JB, Visscher DW, Barnes EL. Update on selected salivary gland neoplasms. Arch Pathol Lab Med. 2009;133(11):1763-74.
7. Luukkaa H, Klemi P, Leivo I, Koivunen P, Laranne J, Mäkitie A, et al. Salivary gland cancer in Finland 1991–96: an evaluation of 237 cases. Acta Otolaryngol. 2005;125(2):207-14.
8. Roh JL, Lee JI, Choi SH, Nam SY, Kim SO, Cho KJ, et al. Prognostic factors and oncologic outcomes of 56 salivary duct carcinoma patients in a single institution: high rate of systemic failure warrants targeted therapy. Oral oncology. 2014;50(11):e64-6.
9. Jayaprakash V, Merzianu M, Warren GW, Arshad H, Hicks WL, Jr., Rigual NR, et al. Survival rates and prognostic factors for infiltrating salivary duct carcinoma: Analysis of 228 cases from the Surveillance, Epidemiology, and End Results database. Head & neck. 2014;36(5):694-701.
10. Villepelet A, Lefevre M, Verillaud B, Janot F, Garrel R, Vergez S, et al. Salivary duct carcinoma: Prospective multicenter study of 61 cases of the Reseau d'Expertise Francais des Cancers ORL Rares. Head & neck. 2019;41(3):584-91.
11. Johnston ML, Huang SH, Waldron JN, Atenafu EG, Chan K, Cummings BJ, et al. Salivary duct carcinoma: Treatment, outcomes, and patterns of failure. Head & neck. 2016;38 Suppl 1:E820-6.
12. Boon E, van Boxtel W, Buter J, Baatenburg de Jong RJ, van Es RJJ, Bel M, et al. Androgen deprivation therapy for androgen receptor-positive advanced salivary duct carcinoma: A nationwide case series of 35 patients in The Netherlands. Head & neck. 2018;40(3):605-13.
13. Schmitt NC, Kang H, Sharma A. Salivary duct carcinoma: An aggressive salivary gland malignancy with opportunities for targeted therapy. Oral oncology. 2017;74:40-8.
14. Takase S, Kano S, Tada Y, Kawakita D, Shimura T, Hirai H, et al. Biomarker immunoprofile in salivary duct carcinomas: clinicopathological and prognostic implications with evaluation of the revised classification. Oncotarget. 2017;8(35):59023-35.
15. Shimura T, Tada Y, Hirai H, Kawakita D, Kano S, Tsukahara K, et al. Prognostic and histogenetic roles of gene alteration and the expression of key potentially actionable targets in salivary duct carcinomas. Oncotarget. 2017;9(2).

16. Dalin MG, Desrichard A, Katabi N, Makarov V, Walsh LA, Lee KW, et al. Comprehensive Molecular Characterization of Salivary Duct Carcinoma Reveals Actionable Targets and Similarity to Apocrine Breast Cancer. Clinical cancer research : an official journal of the American Association for Cancer Research. 2016;22(18):4623-33.
17. Vital D, Ikenberg K, Moch H, Rossle M, Huber GF. The expression of PD-L1 in salivary gland carcinomas. Sci Rep. 2019;9(1):12724.
18. Xu B, Jungbluth AA, Frosina D, Alzumaili B, Aleynick N, Slodkowska E, et al. The immune microenvironment and expression of PD-L1, PD-1, PRAME and MHC I in salivary duct carcinoma. Histopathology. 2019;75(5):672-82.
19. Pai M, Iorio A, Meerpohl J, Taruscio D, Laricchiuta P, Mincarone P, et al. Developing methodology for the creation of clinical practice guidelines for rare diseases: A report from RARE-Bestpractices. Rare Dis. 2015;3(1):e1058463.
20. Balshem H, Helfand M, Schunemann HJ, Oxman AD, Kunz R, Brozek J, et al. GRADE guidelines: 3. Rating the quality of evidence. Journal of clinical epidemiology. 2011;64(4):401-6.
21. Agulnik M, Cohen EW, Cohen RB, Chen EX, Vokes EE, Hotte SJ, et al. Phase II study of lapatinib in recurrent or metastatic epidermal growth factor receptor and/or erbB2 expressing adenoid cystic carcinoma and non adenoid cystic carcinoma malignant tumors of the salivary glands. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2007;25(25):3978-84.
22. Almquist D, Umakanthan JM, Ganti AK. Sequential HER2-Targeted Therapy in Salivary Ductal Carcinoma With HER2/neu Overexpression and a Concomitant ERBB2 Mutation. JCO Precision Oncology. 2018(2):1-5.
23. Correa TS, Matos GDR, Segura M, Dos Anjos CH. Second-Line Treatment of HER2-Positive Salivary Gland Tumor: Ado-Trastuzumab Emtansine (T-DM1) after Progression on Trastuzumab. Case reports in oncology. 2018;11(2):252-7.
24. De Block K, Vander Poorten V, Dormaar T, Nuyts S, Hauben E, Floris G, et al. Metastatic HER-2-positive salivary gland carcinoma treated with trastuzumab and a taxane: a series of six patients. Acta clinica Belgica. 2016;71(6):383-8.
25. de Cecio R, Cantile M, Fulciniti F, Collina F, Scognamiglio G, Longo F, et al. Metastatic salivary ductal carcinoma androgen receptor-positive with V600E BRAF gene mutation. International Journal of Clinical and Experimental Medicine. 2016;9(11):22463-9.
26. Elkrief A, Saleh R. Androgen deprivation therapy for metastatic salivary gland cancer. Cmaj. 2018;190(33):E985-E7.
27. Falchook GS, Lippman SM, Bastida CC, Kurzrock R. Human epidermal receptor 2-amplified salivary duct carcinoma: regression with dual human epidermal receptor 2 inhibition and anti-vascular endothelial growth factor combination treatment. Head & neck. 2014;36(3):E25-7.
28. Fiedler W, Stoeger H, Perotti A, Gastl G, Weidmann J, Dietrich B, et al. Phase i study of TrasGEX, a glyco-optimised anti-HER2 monoclonal antibody, in patients with HER2-positive solid tumours. ESMO Open. 2018;3 (4) (no pagination) (e000381).
29. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.
30. Gibo T, Sekiguchi N, Gomi D, Noguchi T, Fukushima T, Kobayashi T, et al. Targeted therapy with trastuzumab for epidermal growth factor

receptor 2 (HER2)-positive advanced salivary duct carcinoma: A case report. Molecular and clinical oncology. 2019;11(2):111-5.

31. Graham LJ, Meininger LJ. Salivary ductal adenocarcinoma with complete response to androgen blockade. Journal of Community and Supportive Oncology. 2018;16(4):e200-e1.
32. Hyman DM, Puzanov I, Subbiah V, Faris JE, Chau I, Blay JY, et al. Vemurafenib in multiple nonmelanoma cancers with BRAF V600 mutations. New England Journal of Medicine. 2015;373(8):726-36.
33. Jakob JA, Kies MS, Glisson BS, Kupferman ME, Liu DD, Lee JJ, et al. Phase II study of gefitinib in patients with advanced salivary gland cancers. Head & neck. 2015;37(5):644-9.
34. Kadowaki S, Yatabe Y, Hirakawa H, Komori A, Kondoh C, Hasegawa Y, et al. Complete Response to Trastuzumab-Based Chemotherapy in a Patient with Human Epidermal Growth Factor Receptor-2-Positive Metastatic Salivary Duct Carcinoma ex Pleomorphic Adenoma. Case reports in oncology. 2013;6(3):450-5.
35. Kaidar-Person O, Billan S, Kuten A. Targeted therapy with trastuzumab for advanced salivary ductal carcinoma: case report and literature review. Medical oncology (Northwood, London, England). 2012;29(2):704-6.
36. Kawahara K, Hiraki A, Yoshida R, Arita H, Matsuoka Y, Yamashita T, et al. Salivary duct carcinoma treated with cetuximab-based targeted therapy: A case report. Molecular and clinical oncology. 2017;6(6):886-92.
37. Kim Y, Lee SJ, Lee JY, Lee SH, Sun JM, Park K, et al. Clinical trial of nintedanib in patients with recurrent or metastatic salivary gland cancer of the head and neck: A multicenter phase 2 study (Korean Cancer Study Group HN14-01). Cancer. 2017;123(11):1958-64.
38. Krishnamurthy J, Krishnamurty DM, Baker JJ, Zhen W, Lydiatt D, Ganti AK. Salivary duct carcinoma responding to trastuzumab-based therapy: case report and review of the literature. Head & neck. 2013;35(12):E372-5.
39. Laurie SA, Siu LL, Winquist E, Maksymiuk A, Harnett EL, Walsh W, et al. A phase 2 study of platinum and gemcitabine in patients with advanced salivary gland cancer: a trial of the NCIC Clinical Trials Group. Cancer. 2010;116(2):362-8.
40. Lee JS, Kwon OJ, Park JJ, Seo JH. Salivary duct carcinoma of the parotid gland: Is adjuvant HER-2-targeted therapy required? Journal of Oral and Maxillofacial Surgery. 2014;72(5):1023-31.
41. Licitra L, Cavina R, Grandi C, Palma SD, Guzzo M, Demicheli R, et al. Cisplatin, doxorubicin and cyclophosphamide in advanced salivary gland carcinoma. A phase II trial of 22 patients. Annals of oncology : official journal of the European Society for Medical Oncology. 1996;7(6):640-2.
42. Limaye SA, Posner MR, Krane JF, Fonfria M, Lorch JH, Dillon DA, et al. Trastuzumab for the treatment of salivary duct carcinoma. The oncologist. 2013;18(3):294-300.
43. Lin VTG, Nabell LM, Spencer SA, Carroll WR, Harada S, Yang ES. First-Line Treatment of Widely Metastatic BRAF-Mutated Salivary Duct Carcinoma With Combined BRAF and MEK Inhibition. Journal of the National Comprehensive Cancer Network : JNCCN. 2018;16(10):1166-70.
44. Locati LD, Perrone F, Cortelazzi B, Bergamini C, Bossi P, Civelli E, et al. A phase II study of sorafenib in recurrent and/or metastatic salivary gland carcinomas: Translational analyses and clinical impact. European Journal of Cancer. 2016;69:158-65.
45. Locati LD, Perrone F, Cortelazzi B, Lo Vullo S, Bossi P, Dagrada G, et al. Clinical activity of androgen deprivation therapy in patients with metastatic/

relapsed androgen receptor-positive salivary gland cancers. Head & neck. 2016;38(5):724-31.

46. Nakano K, Sato Y, Sasaki T, Shimbashi W, Fukushima H, Yonekawa H, et al. Combination chemotherapy of carboplatin and paclitaxel for advanced/metastatic salivary gland carcinoma patients: differences in responses by different pathological diagnoses. Acta oto-laryngologica. 2016;136(9):948-51.

47. Nardi V, Sadow PM, Juric D, Zhao D, Cosper AK, Bergethon K, et al. Detection of novel actionable genetic changes in salivary duct carcinoma helps direct patient treatment. Clinical cancer research : an official journal of the American Association for Cancer Research. 2013;19(2):480-90.

48. Nashed M, Casasola RJ. Biological therapy of salivary duct carcinoma. The Journal of laryngology and otology. 2009;123(2):250-2.

49. Okada T, Saotome T, Nagao T, Masubuchi T, Fushimi C, Matsuki T, et al. Carboplatin and docetaxel in patients with salivary gland carcinoma: A retrospective study. In Vivo. 2019;33(3):843-53.

50. Park JC, Ma TM, Rooper L, Hembrough T, Foss RD, Schmitt NC, et al. Exceptional responses to pertuzumab, trastuzumab, and docetaxel in human epidermal growth factor receptor-2 high expressing salivary duct carcinomas. Head & neck. 2018;40(12):E100-e6.

51. Piha-Paul SA, Cohen PR, Kurzrock R. Salivary duct carcinoma: targeting the phosphatidylinositol 3-kinase pathway by blocking mammalian target of rapamycin with temsirolimus. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2011;29(26):e727-30.

52. Prat A, Parera M, Reyes V, Peralta S, Cedres S, Andreu J, et al. Successful treatment of pulmonary metastatic salivary ductal carcinoma with trastuzumab-based therapy. Head & neck. 2008;30(5):680-3.

53. Swed BL, Cohen RB, Aggarwal C. Targeting HER2/neu Oncogene Overexpression With Ado-Trastuzumab Emtansine in the Treatment of Metastatic Salivary Gland Neoplasms: A Single-Institution Experience. JCO precision oncology. 2019;3.

54. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.

55. Thorpe LM, Schrock AB, Erlich RL, Miller VA, Knost J, Le-Lindqwister N, et al. Significant and durable clinical benefit from trastuzumab in 2 patients with HER2-amplified salivary gland cancer and a review of the literature. Head & neck. 2017;39(3):E40-e4.

56. Ueki Y, Tada Y, Togashi T, Kawakita D, Nagao T, Sato Y. Pathological response of salivary duct carcinoma to trastuzumab and docetaxel therapy. International cancer conference journal. 2016;5(3):150-3.

57. Urban D, Rischin D, Angel C, D'Costa I, Solomon B. Abiraterone in metastatic salivary duct carcinoma. Journal of the National Comprehensive Cancer Network : JNCCN. 2015;13(3):288-90.

58. van Boxtel W, Boon E, Weijs WLJ, van den Hoogen FJA, Flucke UE, van Herpen CML. Combination of docetaxel, trastuzumab and pertuzumab or treatment with trastuzumab-emtansine for metastatic salivary duct carcinoma. Oral oncology. 2017;72:198-200.

59. Viscuse PV, Price KA, Garcia JJ, Schembri-Wismayer DJ, Chintakuntlawar AV. First Line Androgen Deprivation Therapy vs. Chemotherapy for Patients With Androgen Receptor Positive Recurrent or Metastatic

Salivary Gland Carcinoma-A Retrospective Study. Frontiers in oncology. 2019;9:701.

60. Wang K, Russell JS, McDermott JD, Elvin JA, Khaira D, Johnson A, et al. Profiling of 149 Salivary Duct Carcinomas, Carcinoma Ex Pleomorphic Adenomas, and Adenocarcinomas, Not Otherwise Specified Reveals Actionable Genomic Alterations. Clinical cancer research : an official journal of the American Association for Cancer Research. 2016;22(24):6061-8.

61. Yamamoto H, Uryu H, Segawa Y, Tsuneyoshi M. Aggressive invasive micropapillary salivary duct carcinoma of the parotid gland. Pathology international. 2008;58(5):322-6.

62. Yamamoto N, Minami S, Fujii M. Clinicopathologic study of salivary duct carcinoma and the efficacy of androgen deprivation therapy. American journal of otolaryngology. 2014;35(6):731-5.

63. Kuroda H, Sakurai T, Yamada M, Uemura N, Ono M, Abe T, et al. [Effective treatment by both anti-androgen therapy and chemotherapy for a patient with advanced salivary duct carcinoma]. Gan To Kagaku Ryoho. 2011;38(4):627-30.

64. Iguchi F, Taniguchi Z, Kusano J, Takahashi Y, Murai N. [A case of metastatic salivary duct carcinoma successfully treated with trastuzumab-based targeted therapy]. Nihon Jibiinkoka Gakkai Kaiho. 2014;117(8):1108-14.

65. Alfieri S, Granata R, Bergamini C, Resteghini C, Bossi P, Licitra LF, et al. Systemic therapy in metastatic salivary gland carcinomas: A pathology-driven paradigm? Oral oncology. 2017;66:58-63.

66. Lagha A, Chraiet N, Ayadi M, Krimi S, Allani B, Rifi H, et al. Systemic therapy in the management of metastatic or advanced salivary gland cancers. Head Neck Oncol. 2012;4:19-.

67. Laurie SA, Licitra L. Systemic therapy in the palliative management of advanced salivary gland cancers. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2006;24(17):2673-8.

68. Udager AM, Chiosea SI. Salivary Duct Carcinoma: An Update on Morphologic Mimics and Diagnostic Use of Androgen Receptor Immunohistochemistry. Head and neck pathology. 2017;11(3):288-94.

69. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. The New England journal of medicine. 2001;344(11):783-92.

70. Sharifi N, Gulley JL, Dahut WL. Androgen deprivation therapy for prostate cancer. Jama. 2005;294(2):238-44.

71. Cornford P, Bellmunt J, Bolla M, Briers E, De Santis M, Gross T, et al. EAU-ESTRO-SIOG Guidelines on Prostate Cancer. Part II: Treatment of Relapsing, Metastatic, and Castration-Resistant Prostate Cancer. European urology. 2017;71(4):630-42.

72. Akaza H, Hinotsu S, Usami M, Arai Y, Kanetake H, Naito S, et al. Combined androgen blockade with bicalutamide for advanced prostate cancer: long-term follow-up of a phase 3, double-blind, randomized study for survival. Cancer. 2009;115(15):3437-45.

73. Kolvenbag GJ, Blackledge GR, Gotting-Smith K. Bicalutamide (Casodex) in the treatment of prostate cancer: history of clinical development. The Prostate. 1998;34(1):61-72.

74. Feng Q, He B. Androgen Receptor Signaling in the Development of Castration-Resistant Prostate Cancer. Frontiers in oncology. 2019;9:858.

75. Piccart-Gebhart MJ, Procter M, Leyland-Jones B, Goldhirsch A, Untch M, Smith I, et al. Trastuzumab after Adjuvant Chemotherapy in HER2-Positive Breast Cancer. New England Journal of Medicine. 2005;353(16):1659-72.

76. Baselga J, Cortes J, Kim SB, Im SA, Hegg R, Im YH, et al. Pertuzumab plus trastuzumab plus docetaxel for metastatic breast cancer. The New England journal of medicine. 2012;366(2):109-19.

77. Swain SM, Baselga J, Kim SB, Ro J, Semiglazov V, Campone M, et al. Pertuzumab, trastuzumab, and docetaxel in HER2-positive metastatic breast cancer. The New England journal of medicine. 2015;372(8):724-34.

78. van Boxtel W, Locati LD, van Engen-van Grunsven ACH, Bergamini C, Jonker MA, Fiets E, et al. Adjuvant androgen deprivation therapy for poor-risk, androgen receptor-positive salivary duct carcinoma. European journal of cancer (Oxford, England : 1990). 2019;110:62-70.

79. Hammond ME, Hayes DF, Dowsett M, Allred DC, Hagerty KL, Badve S, et al. American Society of Clinical Oncology/College of American Pathologists guideline recommendations for immunohistochemical testing of estrogen and progesterone receptors in breast cancer (unabridged version). Arch Pathol Lab Med. 2010;134(7):e48-72.

80. van Boxtel W, Verhaegh GW, van Engen-van Grunsven IA, van Strijp D, Kroeze LI, Ligtenberg MJ, et al. Prediction of clinical benefit from androgen deprivation therapy in salivary duct carcinoma patients. International journal of cancer. 2019.

81. Wolff AC, Hammond MEH, Allison KH, Harvey BE, Mangu PB, Bartlett JMS, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Clinical Practice Guideline Focused Update. Journal of Clinical Oncology. 2018;36(20):2105-22.

82. Balduzzi S, Mantarro S, Guarneri V, Tagliabue L, Pistotti V, Moja L, et al. Trastuzumab-containing regimens for metastatic breast cancer. Cochrane Database Syst Rev. 2014;2014(6):Cd006242.

83. Lassche G, van Boxtel W, Ligtenberg MJL, van Engen-van Grunsven ACH, van Herpen CML. Advances and challenges in precision medicine in salivary gland cancer. Cancer Treat Rev. 2019;80:101906.

84. Nguyen PL, Alibhai SMH, Basaria S, D'Amico AV, Kantoff PW, Keating NL, et al. Adverse Effects of Androgen Deprivation Therapy and Strategies to Mitigate Them. European urology. 2015;67(5):825-36.

85. Li BT, Shen R, Offin M, Buonocore DJ, Myers ML, Venkatesh A, et al. Ado-trastuzumab emtansine in patients with HER2 amplified salivary gland cancers (SGCs): Results from a phase II basket trial. Journal of Clinical Oncology. 2019;37(15_suppl):6001-.

86. Tsurutani J, Iwata H, Krop I, Janne PA, Doi T, Takahashi S, et al. Targeting HER2 with Trastuzumab Deruxtecan: A Dose-Expansion, Phase I Study in Multiple Advanced Solid Tumors. Cancer discovery. 2020;10(5):688-701.

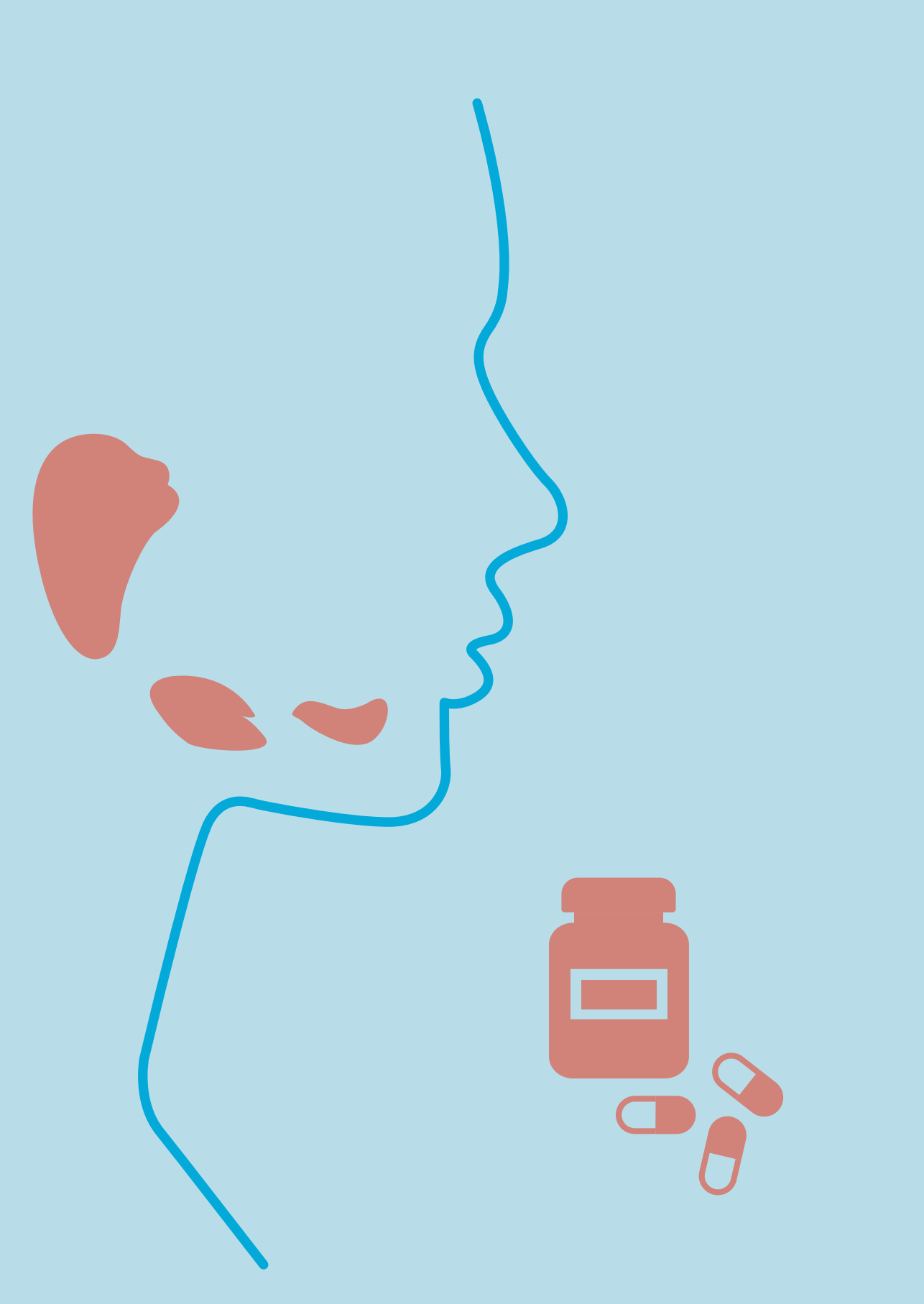
87. Dalin MG, Desrichard A, Katabi N, Makarov V, Walsh LA, Lee K-W, et al. Comprehensive Molecular Characterization of Salivary Duct Carcinoma Reveals Actionable Targets and Similarity to Apocrine Breast Cancer. Clinical Cancer Research. 2016.

88. Ho AL, Hanna GJ, Scholz CR, Gualberto A, Park SH. Preliminary activity of tipifarnib in tumors of the head and neck, salivary gland and urothelial tract with HRAS mutations. J Clin Oncol 38: 2020 (suppl; abstr 6504).

89. André F, Ciruelos E, Rubovszky G, Campone M, Loibl S, Rugo HS, et al. Alpelisib for PIK3CA-Mutated, Hormone Receptor-Positive Advanced Breast Cancer. The New England journal of medicine. 2019;380(20):1929-40.

90. Morris LG, Linxweiler M, Kuo F, Katabi N, Nadeem Z, Dalin MG, et al. The immune microenvironment and neoantigen landscape of aggressive salivary gland carcinomas differ by subtype. Clinical cancer

research : an official journal of the American Association for Cancer Research. 2020.



Chapter 3

Excessive toxicity of cabozantinib in a phase II study in patients with recurrent and/or metastatic salivary gland cancer.

Wim van Boxtel*, Maike J.M. Uijen*, Stefanie D. Krens, Tim Dijkema, Stefan M. Willems, Marianne A. Jonker, Sjoert A.H. Pegge, Adriana C.H. van Engen-van Grunsven, Carla M.L. van Herpen

* These authors contributed equally

European Journal of Cancer, 2022. ***161****:128-137.*

ABSTRACT

Aim

Because the tyrosine kinases c-MET and vascular endothelial growth factor receptors (VEGFR) are often overexpressed in salivary gland cancer (SGC), this study evaluated the efficacy and safety of cabozantinib in recurrent/metastatic (R/M) SGC patients.

Patients and Methods

A single-centre phase II study was conducted. Patients with immunohistochemical c-MET-positive R/M SGC were included in three cohorts: adenoid cystic carcinoma (ACC); salivary duct carcinoma (SDC; and other miscellaneous SGCs. No prior systemic treatments were required. Patients started cabozantinib 60 mg once daily. The primary outcome was the objective response rate (ORR). Secondary outcomes included survival, safety and quality of life. Per Simon-two-stage design, depending on efficacy, a maximum of 43 patients would be included.

Results

In total, 25 patients were included until premature closure owing to severe toxicity. Six patients (24%) had grade 3-5 wound complications, occurring at a median of 7.1 months on cabozantinib treatment (range 2.1-12.6). Remarkably, four of these six patients developed this complication in the area prior exposed to high-dose radiotherapy. Other grade ≥3 adverse events in >1 patient were hypertension (20%), diarrhoea (8%) and dehydration (8%).

Twenty-one patients were evaluable for response; 1/15 ACC (ORR: 7%), 1/4 SDC, and 0/2 patients with other miscellaneous SGC responded. Median progression-free survival was 9.4 months (95% confidence interval [CI] 7.4–11.4 months), 7.2 months (95%CI 0.0–15.1) and 6.9 months (95%CI 0.0–15.1), respectively.

Conclusion

This study showed too many severe cabozantinib-associated wound complications in patients with SGC, especially in prior irradiated areas. Therefore, the study closed prematurely. The efficacy in the limited number of evaluable patients was low to moderate.

INTRODUCTION

Salivary gland cancer (SGC) is a rare cancer with an annual incidence of 0.5-2 cases per 100.000 persons. Twenty-two different subtypes are recognised with their own clinical behaviour and prognosis (1, 2). Primary treatment consists of a tumour resection, frequently combined with a neck dissection and postoperative radiotherapy. However, rates of locoregional recurrence and distant metastases (R/M) are high in certain subtypes; especially in adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC) (3). In patients with R/M ACC, chemotherapy remains the cornerstone of treatment, with a response rate of 25% of cyclophosphamide plus doxorubicin plus cisplatin (4). Recently, several phase II trials explored the efficacy of anti-angiogenic tyrosine kinase inhibitors (TKIs) in patients with ACC. Lenvatinib showed a response rate of 16% in 32 ACC patients (5) and apatinib showed the most promising results, with a response rate of 47% in 59 ACC patients (6).

For patients with R/M SDC, androgen deprivation therapy and human epidermal growth factor receptor 2 (HER2)-targeted therapies are well-established for patients with androgen receptor-positive (78-96%) and HER2-positive (29-46%) SDC, with response rates of 42% and 70%, respectively (7-9). Although these treatment options altered the prognosis in subgroups of patients with SGC, new treatment options are needed to substantially improve the prognosis of patients with R/M SGC.

Cabozantinib is a TKI that targets among others c-MET and vascular endothelial growth factor receptor 2 (VEGFR2) and is registered for patients with R/M medullary thyroid carcinoma, renal cell carcinoma and hepatocellular carcinoma (10-12). c-MET expression has been shown in approximately 53-67% of ACC tumours and 40-50% of SDC tumours (13-16). Furthermore, VEGFR expression is seen in 76% of ACC tumours (17), and trials with angiogenesis inhibitors in patients with ACC showed promising results (as listed previously). The aim of this phase II trial was to evaluate the efficacy and safety of cabozantinib in patients with R/M SGC with c-MET expression.

PATIENTS AND METHODS

Patients, treatment and assessments

Patients with locally advanced, recurrent and/or metastatic SGC were included in 3 cohorts: ACC; SDC and other miscellaneous SGCs. Main inclusion criteria included immunohistochemical c-MET expression (H-score ≥10/300) and measurable disease per Response Evaluation Criteria in Solid Tumours (RECIST) version 1.1 (18). There was no limit on the number of prior anticancer treatments. Objective growth or complaints owing to the disease were required before

inclusion in the ACC and the other miscellaneous SGC cohort but not for the SDC cohort because of its aggressive natural behaviour. Additional criteria are listed in the Supplementary.

Participants were treated with a starting dose of cabozantinib tablets 60 mg once daily (OD). In case of grade ≥3 (hypertension excepted) or intolerable grade 2 adverse events, treatment was temporally interrupted. Subsequently, the dosage was reduced to 40 mg OD or 20mg OD (minimal dose). Cabozantinib treatment was discontinued at disease progression or in case of unacceptable toxicity.

Patients were monitored regularly (Supplementary table 1). Tumour imaging consisted of magnetic resonance (MR) scanning of the primary tumour (in case of local recurrence) and computed tomography (CT) scan of the neck, chest and abdomen. Tumour imaging was performed every 8 weeks during the first year of treatment and thereafter every 12 weeks Tumour response was assessed as per RECIST version 1.1. Toxicity was scored as per the National Cancer Institute's Common Terminology Criteria for Adverse Events, version 5.0. Furthermore, quality of life was measured as per validated questionnaires (e.g. core quality of life questionnaire [QLQ-C30]) and cabozantinib trough concentration levels were measured, details and results of these outcomes are listed in the Supplementary (Supplementary table 3/Supplementary figure 2). This study was approved by the local medical ethics committee.

Study endpoints and statistical analysis

The primary endpoint consisted of the objective response rate (ORR), defined as the proportion of patients with a complete (CR) or partial response (PR) as the best response. Secondary endpoints included progression-free survival (PFS), overall survival (OS), clinical benefit rate (CR + PR + stable disease [SD] ≥6 months) and safety. Only patients with a treatment duration of ≥8 weeks were considered evaluable for response. All patients who started cabozantinib treatment were included in the toxicity analysis, PFS and OS.

A Simon two-stage design was used for the ACC and SDC cohort, with a null hypothesis of at most 5% response rate and an alternative hypothesis of at least 25% response rate (α: 0.05, power: 80%). The first stage consisted of nine patients per cohort. In case of at least one response in the first stage, the cohort would be expanded to 17 patients. If >2 of 17 patients had a response, the null hypothesis would be rejected. A maximum of nine patients would be included in the other SGC cohort.

Kaplan-Meier methods were used for the assessment of the OS and PFS. Statistical analyses were performed using IBM SPSS Statistics version 25.0 (IBM Corp, Armonk, New York).

RESULTS

The study started in September 2018 and was closed prematurely owing to toxicity on 6th of November 2019. In total, 32 patients were screened for eligibility and 25 patients enrolled in the study. Reasons for ineligibility were rapid clinical deterioration (*n*=3), comorbidity (*n*=2), secondary malignancy (*n*=1) and abnormal liver function (*n*=1).

Patient characteristics

In total, 17 patients with ACC, 5 patients with SDC and 3 patients with other miscellaneous SGC subtypes (carcinoma ex pleomorphic adenoma *n*=1, acinic cell carcinoma *n*=1 and mucoepidermoid carcinoma *n*=1) were included in the study. Most patients (*n*=17) were treated for distant metastatic disease (68%), three patients (12%) were treated for local recurrent disease only and five patients (20%) had both local recurrent disease and distant metastases. Baseline patient characteristics per cohort are listed in table 1.

Table 1: Baseline patient characteristics

	ACC (*n*=17) No. of pts (%)*	SDC (*n*=5) No. of pts (%)*	Other SGC (*n*=3) No. of pts (%)*
Gender			
Male	8 (47)	3 (60)	1 (33)
Female	9 (53)	2 (40)	2 (67)
Age, median (range)	56 (49-71)	54 (51-71)	65 (64-72)
ECOG PS			
0	9 (53)	2 (40)	0 (0)
1	8 (47)	3 (60)	3 (100)
Primary site			
Parotid gland	4 (24)	5 (100)	3 (100)
Submandibular gland	3 (18)	0 (0)	0 (0)
Sublingual gland	0 (0)	0 (0)	0 (0)
Minor salivary gland	4 (24)	0 (0)	0 (0)
Other†	6 (35)	0 (0)	0 (0)
Disease distribution			
Locoregional disease	3 (18)	0 (0)	0 (0)
Locoregional and metastatic disease	3 (18)	2 (40)	0 (0)
Metastatic disease	11 (65)	3 (60)	3 (100)
Sites of metastatic disease			
Lung	12 (71)	3 (60)	2 (67)
Pleural	7 (41)	0 (0)	0 (0)
Liver	5 (29)	0 (0)	0 (0)
Bone	5 (29)	1 (20)	2 (67)
Distant lymph nodes	5 (29)	1 (20)	3 (100)
Other	6 (35)	1 (20)	2 (67)

Table 1: Continued

Prior treatments			
Surgery			
Tumour resection	14 (82)	2 (40)	2 (67)
Lymph node neck dissection	5 (29)	3 (60)	1 (33)
Radiotherapy			
Postoperative	11 (65)	3 (60)	2 (67)
Primary treatment‡	3 (18)	1 (20)	1 (33)
Palliative	6 (35)	2 (40)	3 (100)
Systemic therapy			
Adjuvant	0 (0)	2 (40)	0 (0)
ADT	-	2 (40)	-
Palliative	4 (24)	3 (60)	1 (33)
Median number of prior lines (range)	1 (1-3)	4 (2-5)	1 (-)
ADT	0 (0)	2 (40)	0 (0)
Anti-HER-2§	0 (0)	3 (100)	0 (0)
Chemotherapy	4 (24)	1 (20)	0 (0)
other	2 (12)	0 (0)	1 (33)
c-MET expression, median H-score (range)	110 (20-300)	60 (25-120)	15 (10-180)

Abbreviations: ACC: adenoid cystic carcinoma, ADT: androgen deprivation therapy, ECOG PS: Eastern Cooperative Oncology Group performance status, HER-2: Human epidermal growth factor receptor 2, pts: patients, SDC: salivary duct carcinoma, SGC: salivary gland cancer.

* Values are numbers and percentages, unless indicated otherwise.

† Other include: nasal cavity (*n*=1), breast (*n*=1), trachea (*n*=1), sphenoid sinus (*n*=1), pterygopalatine fossa (*n*=1), nasopharynx (*n*=1).

‡ Patients whom received radiotherapy as primary treatment (when the primary tumour was inoperable). A complete radiotherapy overview is presented in Supplementary table 3.

§ All 3 SDC patients received multiple lines of HER2 targeted therapy: first-line trastuzumab +/- pertuzumab combined with chemotherapy (most often docetaxel), followed by second-line: ado-trastuzumab emtansine (T-DM1).

Safety

In total, six patients developed grade ≥3 wound complications. Four of these patients developed this complication in the area previously exposed to high-dose radiotherapy (dose ≥66 Gy in most patients); of these, one patient developed a tracheoesophageal fistula which resulted in death. Another patient with a pre-existing small fistula in the neck developed severe ulcerating wounds that covered a large part of the neck. After cessation of cabozantinib, the wound slowly healed over a time course of one year, see figure 1. One patient developed anal fistula with abscesses which required surgical drainage (after high-dose tailbone radiation), and in one patient with a sore throat, imaging showed pharyngeal ulceration. Details can be found in table 2. The time between radiotherapy and the start of cabozantinib ranged from 10.5 to 93.1 months.

In addition, two patients developed wound complications without prior radiotherapy; in one patient a small pre-existing salivary gland fistula increased in size, and one patient required surgery for perforated appendicitis.

These complications occurred at a median of 7.1 months on cabozantinib treatment (range 2.1-12.6). Two of these patients had a pre-existing fistula at the site of the wound complication, see table 2. These severe wound complications were the reason for the premature closing of the study.

Other frequently observed adverse events included: fatigue; elevated liver enzymes; hand-foot syndrome; diarrhoea and anorexia; further details can be found in table 3 and supplementary table 2.

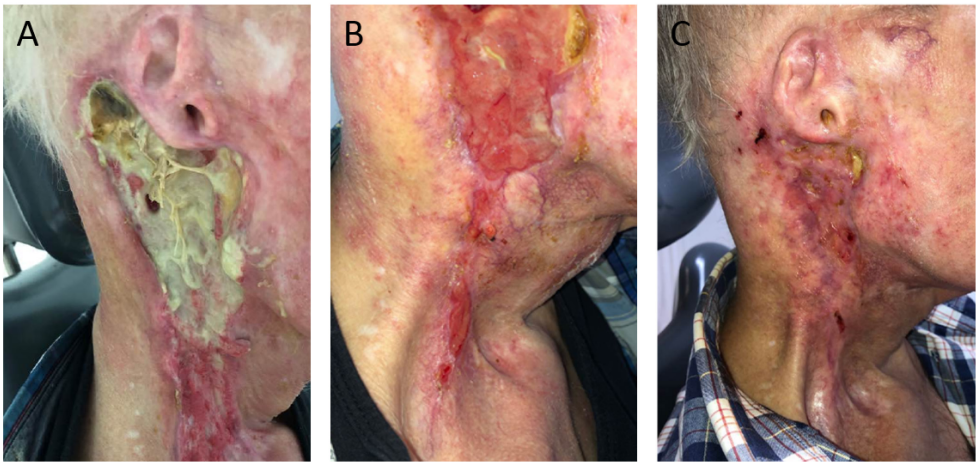


Figure 1: Ulcerative wound in the neck during cabozantinib treatment; (A) wound at 24 weeks on cabozantinib treatment, (B) 3 months after treatment cessation, (C) 1 year after treatment cessation. Shown with permission from the patient.

Table 2: Wound complications grade ≥3 related to cabozantinib treatment

Adverse event	CTCAE Grade	Pre-existing complications	Recurrent or metastatic tumour at site of AE	Time between start of cabozantinib treatment and occurrence of AE	Relevant prior radiotherapy (dose)	Time between radiotherapy and start of cabozantinib treatment	Prior systemic therapies
Tracheoesophageal fistula	Grade 5	-	No	7.8 months	Local recurrence (70 Gy)	51.1 months	CAP (cyclophosphamide + doxorubicin + cisplatin)
Ulcerating wound	Grade 4	Pre-existing small fistula in the neck area	Yes	5.3 months	Postoperative radiotherapy (66 Gy)	89.0 months	Goserelin + bicalutamide
Anal fistula and abscess	Grade 3	-	No	2.1 months	Bone metastasis tailbone (39 Gy) (because of oligometastases)	10.5 months	-
Pharyngeal ulceration	Grade 3	-	Yes	12.6 months	Primary tumour (70 Gy)	93.1 months	Cisplatin
Salivary gland fistula (cutaneous)	Grade 3	Pre-existing small salivary gland fistula	Yes	7.8 months	-	-	-
Perforated appendicitis	Grade 3	-	No	6.4 months	-	-	Paclitaxel with bevacizumab

Abbreviations: AE: adverse event, CTCAE: Common Terminology Criteria for Adverse Event

Table 3: Adverse events probably related to cabozantinib treatment

Adverse event	Any Grade No. of pts (%)	Grade 3 No. of pts (%)	Grade 4 No. of pts (%)	Grade 5 No. of pts (%)
Fatigue	22 (88)	0	0	0
ALAT increased	17 (68)	1 (4)	0	0
Hand-foot syndrome	16 (64)	1 (4)	0	0
ASAT increased	15 (60)	0	0	0
Diarrhoea	15 (60)	2 (8)	0	0
Dysgeusia	15 (60)	0	0	0
ALP increased	12 (48)	0	0	0
Anorexia	12 (48)	1 (4)	0	0
Hypophosphatemia	12 (48)	0	0	0
Mucositis oral	12 (48)	0	0	0
Weight loss	11 (44)	1 (4)	0	0
Hypertension	10 (40)	5 (20)	0	0
Nausea	10 (40)	1 (4)	0	0
Platelet count decreased	10 (40)	1 (4)	0	0
Dry mouth	9 (36)	0	0	0
Dry skin	9 (36)	0	0	0
Dyspepsia	9 (36)	0	0	0
Hoarseness	9 (36)	0	0	0
Alopecia	8 (32)	0	0	0
Dyspnoea	8 (32)	0	0	0
Headache	8 (32)	0	0	0
Constipation	7 (28)	0	0	0
Muscle cramp	6 (24)	0	0	0
Vomiting	6 (24)	1 (4)	0	0
Anaemia	5 (20)	0	0	0
Blood bilirubin increased	5 (20)	0	0	0
Hair colour changes	5 (20)	0	0	0
Oral pain	5 (20)	0	0	0
GGT increased	3 (12)	1 (4)	0	0
Skin ulceration	3 (12)	0	1 (4)	0
Hypokalaemia	3 (12)	1 (4)	0	0
Dehydration	2 (8)	2 (8)	0	0
Pharyngeal mucositis	2 (8)	1 (4)	0	0
Anal fistula	1 (4)	1 (4)	0	0
Appendicitis perforated	1 (4)	1 (4)	0	0
Lung infection	1 (4)	1 (4)	0	0
Myositis	1 (4)	1 (4)	0	0
Salivary gland fistula	1 (4)	1 (4)	0	0
Tracheal fistula	1 (4)	0	0	1 (4)

Table lists treatment-related adverse events that occurred in 20% or more of the patients (any grade) and any grade 3, 4 or 5 events reported in a patient, regardless of frequency.

Adverse events were graded according to National Cancer Institute Common Terminology Criteria for Adverse Events (version 5.0). Patients were counted once at the highest grade for each adverse event.

Abbreviations: ALAT: alanine aminotransferase, ALP: alkaline phosphatase, ASAT: aspartate aminotransferase, GGT, Gamma-glutamyltransferase, pts: patients

Efficacy

In total, 15/17 patients with ACC , 4/5 patients with SDC and 2/3 patients with other miscellaneous SGC were eligible for response assessment (treatment duration of ≥8 weeks). The ORR was 7% (1/15 patients) in the ACC cohort; furthermore 1/4 patients with SDCs and 0/2 patients with other miscellaneous SGC responded. One ACC patient achieved a PR after 25 weeks on cabozantinib, and the duration of response was 32 weeks. One SDC patient achieved a PR after 9 weeks on cabozantinib with progressive disease at 40 weeks on treatment. All other assessable patients achieved SD as the best response (table 4). Figure 2 shows the maximum percentage change in tumour size from baseline, most evaluable patients showed a decrease in target lesion diameter on cabozantinib treatment. Of the four non-evaluable patients, one patient ended treatment <8 weeks owing to side-effects of cabozantinib, and three patients were on treatment <8 weeks and had to stop owing to the closing of the study. Details on the duration of treatment and treatment modifications are listed in table 4.

The median follow-up was 14.2 months. The median PFS for the ACC, SDC and other miscellaneous SGC cohorts were 9.4 months (95% CI 7.4-11.4 months), 7.2 months (95% CI 0.0-15.1 months) and 6.9 months (95% CI 0.0-15.1), respectively. The median OS for the ACC, SDC and other miscellaneous SGC cohorts were 27.5 months (95% CI 15.7-39.4), 14.2 months (95% CI 0.0-28.5) and 15.1 (insufficient events for 95% CI), respectively. Survival plots can be found in figure 2.

Table 4: Cabozantinib treatment and efficacy

	ACC (*n*=17) No. of patients (%)*	SDC (*n*=5) No. of patients (%)*	Other SGC (*n*=3) No. of patients (%)*
Treatment			
Median duration on treatment, months (range)	5.7 (0.8-12.8)	5.7 (1.1-8.5)	6.6 (0.7-7.6)
Median time to first treatment modification†, months (range)	0.9 (0.5-2.0)	1.0 (0.3-5.3)	0.7 (0.5-0.7)
Reasons for first treatment modification†			
Side-effects	15 (88)	3 (60)	2 (67)
Other	0	1 (20)	1 (33)
Premature closing study	2 (12)	1 (20)	0
Efficacy			
Evaluable patients‡	15 (88)	4 (80)	2 (67)
CR	0	0	0
PR	1 (7)	1 (25)	0
SD	14 (93)	3 (75)	2 (100)
≥ 6 months§	10 (67)	2 (50)	2 (100)
PD	0	0	0
ORR	7%	25%	0%
CBR	73%	75%	100%
Median PFS, months (95% CI)	9.4 (7.4-11.4)	7.2 (0.0-15.1)	6.9 (0.0-15.1)
Median OS, months (95% CI)	27.5 (15.7-39.4)	14.2 (0.0-28.5)	15.1¶

Abbreviations: ACC: adenoid cystic carcinoma, CBR: clinical benefit rate (CR+PR+SD ≥6 months), CI: confidence interval, CR: complete response, ORR: objective response rate (CR+PR) PD: progressive disease, PFS: progression free survival, PR: partial response, SD: stable disease, SDC: salivary duct carcinoma, SGC: salivary gland cancer

* Values are numbers and percentages, unless indicated otherwise.
† Treatment modification is defined as: dose reduction, treatment interruption or discontinuation of treatment.
‡ Only patients who were on treatment ≥ 8 weeks were considered evaluable.
§ These numbers are affected by the premature closure of the study.
¶ Insufficient events for 95% CI.

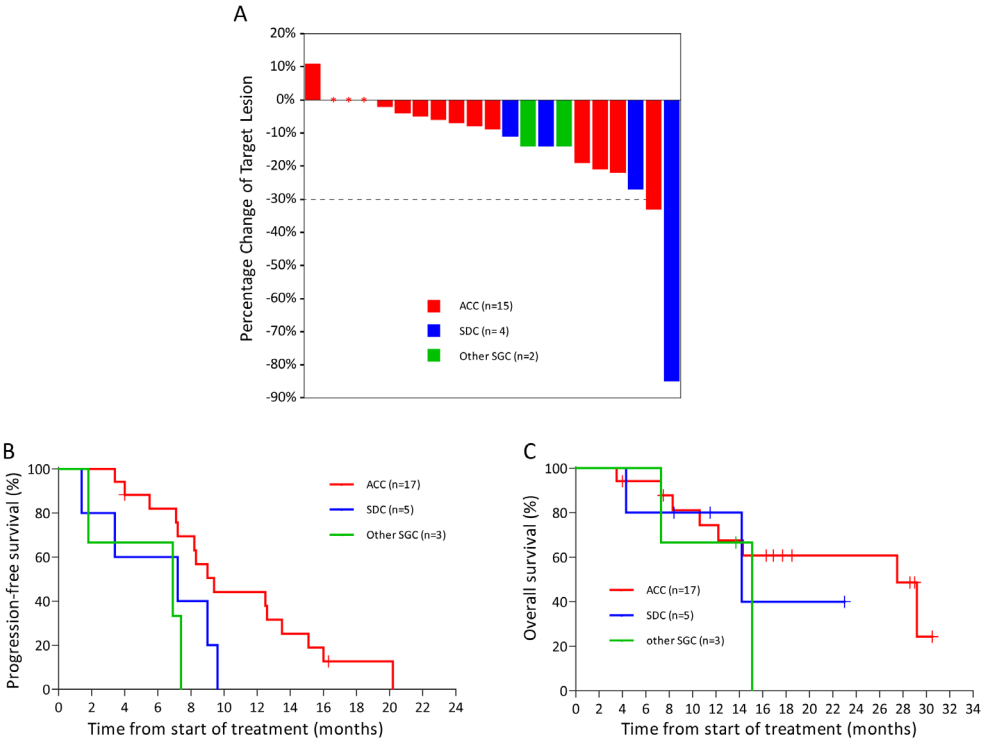


Figure 2: Treatment efficacy; (A) waterfall plot of evaluable patients, (B) Kaplan-Meier plots of progression-free survival, (C) overall survival. Only evaluable patients (treatment duration of ≥ 8 weeks) are presented in the waterfall plot. All included patients are included in the PFS and OS Kaplan-Meier plots. The plus signs on the Kaplan-Meter plots indicates censored data.

Correlation c-MET expression and treatment response

There was no significant relationship between these variables (Spearman's rho correlation coefficient: 0.119, p=0.6). A scatter plot of these variables is presented in supplementary figure 1.

DISCUSSION

This study showed that cabozantinib led to considerable toxicity in patients with SGC. Apart from expected adverse events such as hand-foot syndrome and gastrointestinal side effects, a remarkable high number of severe wound complications were observed. In total, six patients (24%) had grade ≥3 wound complications. This included a tracheoesophageal fistula which resulted in death and a life-threatening ulcerating wound in the neck (figure 1). These severe wound complications are possibly the result of prior tissue damage owing to several previous treatments (e.g. surgery and radiotherapy, often combined as primary treatment, and in some cases, prior systemic therapy) in combination with the antiangiogenic effects of cabozantinib.

Especially prior radiotherapy was considered a major risk factor for the occurrence of wound complications in this study, because four of these six patients developed these complications in previously irradiated areas. These wound complications occurred even when the radiotherapy was given several years before the start of cabozantinib. Comparable toxicity was observed in a phase II study of cabozantinib in patients with Merkel cell carcinoma, which was closed prematurely owing to the toxicity and lack of responses (19). Of eight included patients, two (25%) developed non-healing ulcers and tumour skin fistula. The report did not associate these adverse to previous radiotherapy. Remarkably, in the phase III studies of cabozantinib (10-12, 20, 21) wound complications do not seem to be a major issue: they were not described in the prostate cancer and renal cell carcinoma studies (20-22), the study in patients with hepatocellular carcinoma only reported on one grade 5 bronchoesophageal fistula (10), and the study in medullary thyroid carcinoma reported on wound complications, gastrointestinal fistula and other fistula in 1.9%, 0.9% and 3.7% of the patients, respectively (12). According to cabozantinib drug registration reports, fistula/perforations occurred in 1-4% of patients treated with cabozantinib, and wound complications occurred in 2% (23, 24). A possible explanation for this discrepancy is that all patients with SGC in this study were exposed to radiotherapy, often in high-dose (see supplementary table 4 for details). This is not the case for most patients with renal cell, hepatocellular, and prostate cancer, in these cancers, radiotherapy is given less frequently, and if used, it will often be administered at a lower dose. A review on adverse events of anti-VEGF drugs stated that especially in head and neck cancer, prior radiotherapy is a risk factor for fistula formation (25), which is likely owing to the high dose of radiotherapy.

In studies with other VEGFR TKIs in patients with SGC, lower rates of wound complications were observed. One of 32 patients treated with lenvatinib had an oral cutaneous fistula and one tracheal fistula occurred in 14 patients treated with sunitinib (5, 26). Both reports mentioned that the fistula arose in previously irradiated areas. In addition one of 32 patients treated with sorafenib had a grade 4 skin ulceration (27). Other studies in patients with SGC with VEGFR inhibitors, such as axitinib or sorafenib, did not mention the development of wounds or fistulas (28-32). Thus, wound complications also occurred in other VEGFR-TKI studies in patients with SG, but seemingly at a lower rate than in this study. Because the other VEGFR-TKI SGC studies consisted of patient populations with similar disease distributions (e.g. locoregional recurrence and/or metastatic disease) and comparable rates of prior radiotherapy exposure, these factors are unlikely to account for the difference in wound complications. We consider the difference in the other tyrosine kinase targets of the different VEGFR-TKIs as the most plausible explanation. Although sorafenib, lenvatinib, sunitinib, and cabozantinib all inhibit VEGFRs, each TKI has its own target profile. Cabozantinib distinguishes itself from the other TKIs through inhibition of c-MET and AXL. Both MET and AXL are tyrosine kinases that are involved in wound healing (33, 34). Inhibition of these targets

by cabozantinib, especially in areas with prior tissue atrophy, fibrosis and vascular damage as a result of previous radiotherapy and/or surgery, might be the most plausible hypothesis for the high toxicity observed in this study.

Because the study was closed prematurely owing to toxicity, the efficacy of cabozantinib could only be determined based on the 25 included patients of which 21 patients were considered evaluable (cabozantinib ≥8 weeks). ORRs were 7%, 25% and 0% for the ACC, SDC and other miscellaneous SGC subtype cohorts, respectively.

Based on preclinical data, cabozantinib can inhibit both c-MET-positive tumours, as well as c-MET-negative tumours, by inhibiting other cancer-specific targets, such as AXL, RET and KIT. In mouse models, improved cabozantinib efficacy was correlated with c-MET expression (35). Therefore, we assumed that tumours with high c-MET expression might respond better to cabozantinib treatment. However, we did not find a significant correlation between c-MET expression and treatment response. Prior studies in patients with renal cell carcinoma, breast carcinoma and cholangiocarcinoma also did not find a correlation between c-MET levels and treatment response (22, 36, 37).

Limitations of this study include the small sample size and the single-arm design. We describe a possible relation between cabozantinib treatment and prior radiotherapy for patients who developed severe wound complications, however statistics could not be performed to support or reject this relation owing to the small sample size.

CONCLUSION

This phase II study showed limited efficacy in patients with R/M SGC and was ended prematurely owing to severe wound complications, especially in prior irradiated areas. Therefore, cabozantinib is not recommended in SGC and caution is suggested when prescribing cabozantinib to patients previously exposed to high-dose radiotherapy.

ACKNOWLEDGMENTS

The authors would like to thank the patients and their families for participating in this study.

SUPPLEMENTARY

Inclusion and exclusion criteria

In order to be eligible to participate in this study, a patient must meet all of the following criteria:

Inclusion criteria:

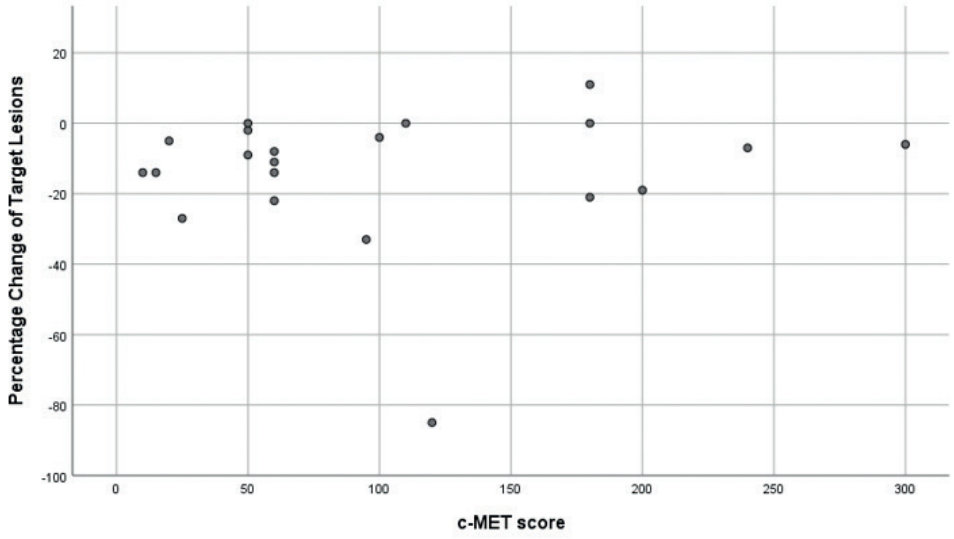
- Disease specific
  - locally advanced, recurrent, and/or metastatic SGC (excluding sarcomas and mesenchymal tumours)
  - c-MET-positive disease*
  - Measurable disease per RECIST version 1.1
  - Cohort-specific criteria:
    - SDC cohort:
      - Direct inclusion (no objective tumour growth prior to inclusion needed)
    - ACC cohort:
      - Inclusion after objective growth in the last three months or complaints due to the disease
    - Other SGC's
      - Inclusion after objective growth in the last three months or complaints due to the disease
- General conditions
  - Age ≥18 years
  - Eastern Cooperative Oncology Group performance status of 0 or 1.
  - Normal number of neutrophils and thrombocytes
  - Liver function:
    - ALT and AST < 2.5 x upper limit of normal (ULN)
    - Total bilirubin ≤ 1.5 x ULN (except for Gilbert's syndrome)
    - Serum albumin ≥28 g/L
  - Renal function:
    - Creatinine < 1.5 x ULN or calculated creatinine clearance ≥ 40 ml/min
    - Urine protein/creatinine ratio ≤113.1 mg/mmol (≤1 mg/mg) or 24-hour urine protein <1 g
  - Haemoglobin A1c (HbA1c) ≤ 8% or a fasting serum glucose ≤ 9 mmol/l

(exclusion criteria on next page)

Exclusion criteria:

- General conditions
  - A known allergy for cabozantinib or its components
  - Long QT-syndrome
  - Pregnancy or lactation
  - Patients (M/F) with reproductive potential not implementing adequate contraceptives measures
  - Known brain metastases or cranial epidural disease unless adequately treated with radiotherapy and/or surgery and stable for at least 3 months before inclusion
  - Major surgery within 3 months before randomization. Complete wound healing from major surgery must have occurred 1 month before inclusion and from minor surgery at least 10 days before inclusion
  - Uncontrolled illness including, but not limited to
    - Cardiovascular disorders including symptomatic congestive heart failure, unstable angina pectoris, or serious cardiac arrhythmias
    - Uncontrolled hypertension defined as sustained systolic BP > 150 mm Hg, or diastolic BP > 100 mm Hg
    - Stroke (including TIA), myocardial infarction, or other ischemic event within 6 months before inclusion
    - Serious active infections
  - Concomitant treatments
- Concomitant (or within 4 weeks before inclusion) administration of any other experimental drug under investigation.
  - Concurrent treatment with any other anti-cancer therapy.
  - Concomitant anticoagulation.
    - Low dose aspirin for cardio protection and low dose LMWH are permitted.
  - Radiation therapy within the last 4 weeks before inclusion.

* c-MET expression was analysed by immunohistochemical staining. A rabbit monoclonal IgG antibody targeting the C-terminus of human MET (D1C2; Cell Signalling) was used. Only membranous staining was taken into account. The stained slides were scored for intensity of the staining (0, 1+, 2+, 3+) and percentage (0-100%). For each slide, the H-score was calculated according to the following formula: 3 x percentage of strongly staining cells + 2 x percentage of moderately staining cells + percentage of weakly staining cells, resulting in a range of 0 to 300. Minimal c-MET expression (H-score ≥10) was sufficient to be included in the study.



Supplementary figure 1: Correlation of c-MET score and percentage change of target lesions

c-MET expression was evaluated by immunohistochemical staining. The staining was scored using the H-score according to the following formula: 3 x percentage of strongly staining cells + 2 x percentage of moderately staining cells + percentage of weakly staining cells, resulting in a range of 0 to 300.

There was no significant relation between these variables (Spearman's rho correlation coefficient: 0.119, p=0.6).

Supplementary table 1: Study assessments

	History / physical exam	Blood analysis	CtDNA	Urine	ECG	Imaging	Questionarres	Toxicity	Any other
	1	2	3	4	5	6	7	8	9
Week 0	X	X	X	X	X	X	X		X
Week 2	X	X	X	X	X			X	X
Week 4	X	X	X					X	X
Week 6	X	X						X	X
Week 8	X	X	X			X	X	X	X
Week 12	X	X						X	X
Week 16	X	X	X			X	X	X	X
Week 20	X	X						X	X
Week 24	X	X	X			X	X	X	X
Week 32	X	X	X			X		X	X
Week 40	X	X	X			X	X	X	X
Week 48	X	X	X			X		X	X
Week 56	X	X	X			X	X	X	X
Week 68	X	X	X			X		X	X
Week 80	X	X	X			X		X	X
Week 92	X	X	X			X		X	X
Week 104	X	X	X			X		X	X
At PD	X	X	X			X	X	X	X

1. History / physical exam
    - Standard history and physical examination
2. Blood analysis
    - Haematology: haemoglobin, blood cell count (RBC, WBC count and differential, platelets)
    - Biochemistry: AST, ALT, AP, GGT, LD, bilirubin, creatinine, urea, Na, K, Ca, P, Mg, albumin.
    - TSH and free T4 in week 0.
3. ctDNA
    - blood sample for ctDNA analysis
4. Urine
    - protein/creatinine ratio
5. ECG
    - Standard ECG
6. Imaging
    - CT- or MR-scanning of primary tumour area and regional lymph nodes
    - CT chest and abdomen
7. Questionnaires:
    - EORTC QLQ-C30
    - EORTC QLQ-H&N35
    - VAS
8. Toxicity
    - Toxicity profile according to NCI CTC common criteria v 5.0
9. Any other
    - Any other diagnostic procedure that the investigator deems necessary

Supplementary table 2: changes in quality of life between baseline and at 16 weeks on treatment

	Mean change from baseline (95% CI)	P value*
EORTC QLQ-C30		
Global health status	-4.2 (-15.6 to 7.3)	0.553
Functional scales		
Physical functioning	-8.6 (-20 to 3.5)	0.109
Role functioning	-6.0 (-21.8 to 9.9)	0.481
Emotional functioning	11.9 (4.4 to 19.4)	**0.009**
Cognitive functioning	-8.3 (-20.7 to 4.4)	0.161
Social functioning	-7.1 (-28.7 to 14.4)	0.811
Symptom scales		
Fatigue	4.0 (-11.0 to 9.0)	0.562
Nausea & vomiting	1.2 (-3.4 to 5.8)	0.276
Pain	6.0 (-8.5 to 20.4)	0.343
Dyspnoea	0.0 (-16.9 to 16.9)	0.607
Insomnia	0.0 (-15.1 to 15.1)	0.785
Appetite loss	23.8 (-2.8 to 50.4)	0.072
Constipation	11.9 (-0.3 to 24.1)	0.059
Diarrhoea	31.0 (10.3 to 51.6)	**0.010**
Financial difficulties	4.8 (-5.5 to 15.0)	0.257
EORTC QLQ-H&N35		
Symptom scales		
Pain	17.9 (4.8 to 30.9)	**0.005**
Swallowing	7.1 (-2.8 to 17.1)	0.196
Sense problems	22.6 (7.7 to 37.5)	**0.013**
Speech problems	9.5 (-2.5 to 21.6)	0.115
Trouble with social eating	23.8 (13.2 To 34.5)	**0.003**
Trouble with social contact	6.2 (2.4 to 10.0)	**0.011**
Less sexuality	22.6 (3.2 to 42.1)	**0.024**
Teeth	16.7 (-4.3 to 37.7)	0.107
Opening mouth	14.3 (-8.0 to 36.6)	0.140
Dry mouth	2.4 (-13.6 to 18.3)	0.739
Sticky saliva	0.0 (-23.9 to 23.9)	0.916
Coughing	-7.1 (-25.9 to 11.6)	0.347
Felt ill	2.4 (-9.5 to 14.2)	0.783
Pain killers†	NA	0.500
Nutritional supplements†	NA	0.250
Feeding tube†	NA	1.000
Weight loss†	NA	0.125
Weight gain†	NA	0.500
Pain scores		
Average pain	3.7 (-10.5 to 17.9)	0.442
Worst pain	9.2 (-9.4 to 27.9)	0.235

Only patients whom completed the questionnaires at both baseline and 16 weeks on treatment are included in this analysis (N=14).

Abbreviations: CI: confidence interval, NA: not applicable

*For each item differences from baseline were analysed by the Wilcoxon Signed-Rank Test. Items where the mean showed a statistically significant difference are highlighted in bold.

†Because these items are dichotomous, McNemar's test was performed.

Interpretation of quality of life before and during treatment (at 16 weeks)

Fourteen out of 16 patients who were on treatment for 16 weeks completed all questionnaires. Two of these patients (14%) showed a more than ten point improvement and 4 patients (29%) showed a more than ten point decline in global health status compared to baseline, but overall mean scores for global health status were not significantly different (p=0.6).

Regarding the functional scales of the QLQc-30, only emotional functioning showed a statistically significant improvement (p=0.009). Of the symptom scales of both the QLQc-30 and H&N-35, statistically significant differences were observed for the following items: diarrhoea, pain, sense problems, trouble with social eating, troubles with social contact, and less sexuality. All these symptoms increased at 16 weeks on cabozantinib treatment, compared to baseline. The VAS pain scores, both average and worst pain, did not change significantly during treatment (p=0.4 and p=0.2).

Supplementary table 3: Prior radiotherapy overview

Pt No.	Subtype	Postoperative radiotherapy (Gy)	Palliative radiotherapy (Gy)
1	ACC	Tumour (63)	NA
2	ACC	Tumour + neck level II (70/46)*	NA
3	SDC	Tumour + neck level V (57)	NA
4	ACC	Tumour/operation area (70/46)	NA
5	ACC	Tumour/operation area (66/59.4)	NA
6	ACC	Tumour/operation area + neck level I (66/51.2)	Bone metastasis Th12 (8)
7	SDC	Tumour/operation area (66/54)	Brain metastases (24)
8	ACC	Tumour/operation area (63/51)	Lung metastases (32)
9	ACC	NA	Bone metastasis hip (8)
10	ACC	NA	Local recurrence (unknown)
11	ACC	NA	Local recurrence (70) Liver metastases (7.5)
12	Other SGC	Tumour/operation area (66/54)	Bone metastasis rib (8) Bone metastasis hand (8) Bone metastasis pelvis (20)
13	SDC	NA	Tumour (50)
14	Other SGC	Neck (66)	Bone metastasis sacrum (39)
15	Other SGC	Tumour/neck level Ib-V (66/51)*	Muscle metastasis arm (20) Recurrence muscle metastasis arm (8)
16	ACC	Tumour/operation area (66/51)	NA
17	SDC	Tumour + positive lymph nodes/operation area (including neck) (66/51.2)	NA
18	ACC	Tumour (66)	Bone metastasis coccyx (39) Bone metastases Th11-L2 (30)
19	ACC	Tumour (70)*	NA
20	ACC	Tumour*† (unknown)	NA
21	ACC	Tumour (66)	NA
22	ACC	Tumour (unknown)	NA
23	SDC	Tumour + positive lymph nodes + neck level Ib to V (70/56)*	NA
24	ACC	Tumour/operation area and neck level I (66/52)	NA
25	ACC	Tumour (66)	NA

Abbreviations: ACC: adenoid cystic carcinoma, NA= not applicable, Pt: patient, SDC: salivary duct carcinoma, SGC: salivary gland carcinoma

*These patients received radiotherapy as primary treatment, e.g. when primary tumour was inoperable.
† Patient 20 received carbon ion therapy in Germany (considered to have received a dose >50Gy).

REFERENCES

1. Cancer incidence in five continents. Volume VIII. IARC Sci Publ. 2002(155):1-781.
2. El-Naggar AK CJ, Grandis JR, Takata T, Slootweg PJ. WHO Classification of Head and Neck Tumours. 4th ed: WHO; 2017.
3. Chen AM, Garcia J, Granchi PJ, Johnson J, Eisele DW. Late recurrence from salivary gland cancer: when does "cure" mean cure? Cancer. 2008;112(2):340-4.
4. Laurie SA, Ho AL, Fury MG, Sherman E, Pfister DG. Systemic therapy in the management of metastatic or locally recurrent adenoid cystic carcinoma of the salivary glands: a systematic review. The Lancet Oncology. 2011;12(8):815-24.
5. Tchekmedyian V, Sherman EJ, Dunn L, Tran C, Baxi S, Katabi N, et al. Phase II Study of Lenvatinib in Patients With Progressive, Recurrent or Metastatic Adenoid Cystic Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(18):1529-37.
6. Zhu G, Zhang L, Li R, Dou S, Yang W, Zhang C. Phase II trial of apatinib in patients with recurrent and/or metastatic adenoid cystic carcinoma of the head and neck: Updated analysis. Journal of Clinical Oncology. 2018;36(15_suppl):6026-.
7. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.
8. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.
9. Uijen MJM, Lassche G, van Engen-van Grunsven ACH, Tada Y, Verhaegh GW, Schalken JA, et al. Systemic therapy in the management of recurrent or metastatic salivary duct carcinoma: A systematic review. Cancer Treat Rev. 2020;89:102069.
10. Abou-Alfa GK, Meyer T, Cheng AL, El-Khoueiry AB, Rimassa L, Ryoo BY, et al. Cabozantinib in Patients with Advanced and Progressing Hepatocellular Carcinoma. The New England journal of medicine. 2018;379(1):54-63.
11. Choueiri TK, Escudier B, Powles T, Mainwaring PN, Rini BI, Donskov F, et al. Cabozantinib versus Everolimus in Advanced Renal-Cell Carcinoma. The New England journal of medicine. 2015;373(19):1814-23.
12. Elisei R, Schlumberger MJ, Muller SP, Schoffski P, Brose MS, Shah MH, et al. Cabozantinib in progressive medullary thyroid cancer. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2013;31(29):3639-46.
13. Bell D, Ferrarotto R, Fox MD, Roberts D, Hanna EY, Weber RS, et al. Analysis and significance of c-MET expression in adenoid cystic carcinoma of the salivary gland. Cancer Biol Ther. 2015;16(6):834-8.
14. Herpen CML, Boxtel W, Flucke UE, Bloemena E, Boon E, Verhaegh G, et al. 1046PDOverexpression of the c-MET proto-oncogene in salivary duct carcinoma patients. Annals of Oncology. 2017;28(suppl_5).
15. Ryu HJ, Koh YW, Yoon SO. The implications of TrkA and MET aberrations in de novo salivary duct carcinoma. Human pathology. 2018;81:18-25.

16. Suzuki K, Cheng J, Watanabe Y. Hepatocyte growth factor and c-Met (HGF/c-Met) in adenoid cystic carcinoma of the human salivary gland. J Oral Pathol Med. 2003;32(2):84-9.
17. Zhang J, Peng B, Chen X. Expressions of nuclear factor kappaB, inducible nitric oxide synthase, and vascular endothelial growth factor in adenoid cystic carcinoma of salivary glands: correlations with the angiogenesis and clinical outcome. Clinical cancer research : an official journal of the American Association for Cancer Research. 2005;11(20):7334-43.
18. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). European journal of cancer (Oxford, England : 1990). 2009;45(2):228-47.
19. Rabinowits G, Lezcano C, Catalano PJ, McHugh P, Becker H, Reilly MM, et al. Cabozantinib in Patients with Advanced Merkel Cell Carcinoma. The oncologist. 2018;23(7):814-21.
20. Basch EM, Scholz M, de Bono JS, Vogelzang N, de Souza P, Marx G, et al. Cabozantinib Versus Mitoxantrone-prednisone in Symptomatic Metastatic Castration-resistant Prostate Cancer: A Randomized Phase 3 Trial with a Primary Pain Endpoint. European urology. 2019;75(6):929-37.
21. Smith M, Bono JD, Sternberg C, Moulec SL, Oudard S, Giorgi UD, et al. Phase III Study of Cabozantinib in Previously Treated Metastatic Castration-Resistant Prostate Cancer: COMET-1. Journal of Clinical Oncology. 2016;34(25):3005-13.
22. Choueiri TK, Escudier B, Powles T, Tannir NM, Mainwaring PN, Rini BI, et al. Cabozantinib versus everolimus in advanced renal cell carcinoma (METEOR): final results from a randomised, open-label, phase 3 trial. The Lancet Oncology. 2016;17(7):917-27.
23. FDA. Cabometyx prescribing information. 2020. https://www.accessdata.fda.gov/drugsatfda_docs/label/2020/208692s008lbl.pdf. Accessed 20 Oct 2020.
24. FDA. Cometriq prescribing information. 2020. https://www.accessdata.fda.gov/drugsatfda_docs/label/2020/203756s008lbl.pdf. Accessed 20 Oct 2020.
25. Chen HX, Cleck JN. Adverse effects of anticancer agents that target the VEGF pathway. Nat Rev Clin Oncol. 2009;6(8):465-77.
26. Chau NG, Hotte SJ, Chen EX, Chin SF, Turner S, Wang L, et al. A phase II study of sunitinib in recurrent and/or metastatic adenoid cystic carcinoma (ACC) of the salivary glands: current progress and challenges in evaluating molecularly targeted agents in ACC. Annals of oncology : official journal of the European Society for Medical Oncology. 2012;23(6):1562-70.
27. Locati LD, Perrone F, Cortelazzi B, Bergamini C, Bossi P, Civelli E, et al. A phase II study of sorafenib in recurrent and/or metastatic salivary gland carcinomas: Translational analyses and clinical impact. European journal of cancer (Oxford, England : 1990). 2016;69:158-65.
28. Ho AL, Dunn L, Sherman EJ, Fury MG, Baxi SS, Chandramohan R, et al. A phase II study of axitinib (AG-013736) in patients with incurable adenoid cystic carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2016;27(10):1902-8.
29. Kim Y, Lee SJ, Lee JY, Lee SH, Sun JM, Park K, et al. Clinical trial of nintedanib in patients with recurrent or metastatic salivary gland cancer of the head and neck: A multicenter phase 2 study (Korean Cancer Study Group HN14-01). Cancer. 2017;123(11):1958-64.
30. Locati LD, Cavalieri S, Bergamini C, Resteghini C, Alfieri S, Calareso G, et al. Phase II trial with

axitinib in recurrent and/or metastatic salivary gland cancers of the upper aerodigestive tract. Head & neck. 2019;41(10):3670-6.

31. Thomson DJ, Silva P, Denton K, Bonington S, Mak SK, Swindell R, et al. Phase II trial of sorafenib in advanced salivary adenoid cystic carcinoma of the head and neck. Head & neck. 2015;37(2):182-7.

32. Wong SJ, Karrison T, Hayes DN, Kies MS, Cullen KJ, Tanvetyanon T, et al. Phase II trial of dasatinib for recurrent or metastatic c-KIT expressing adenoid cystic carcinoma and for nonadenoid cystic malignant salivary tumors. Annals of oncology : official journal of the European Society for Medical Oncology. 2016;27(2):318-23.

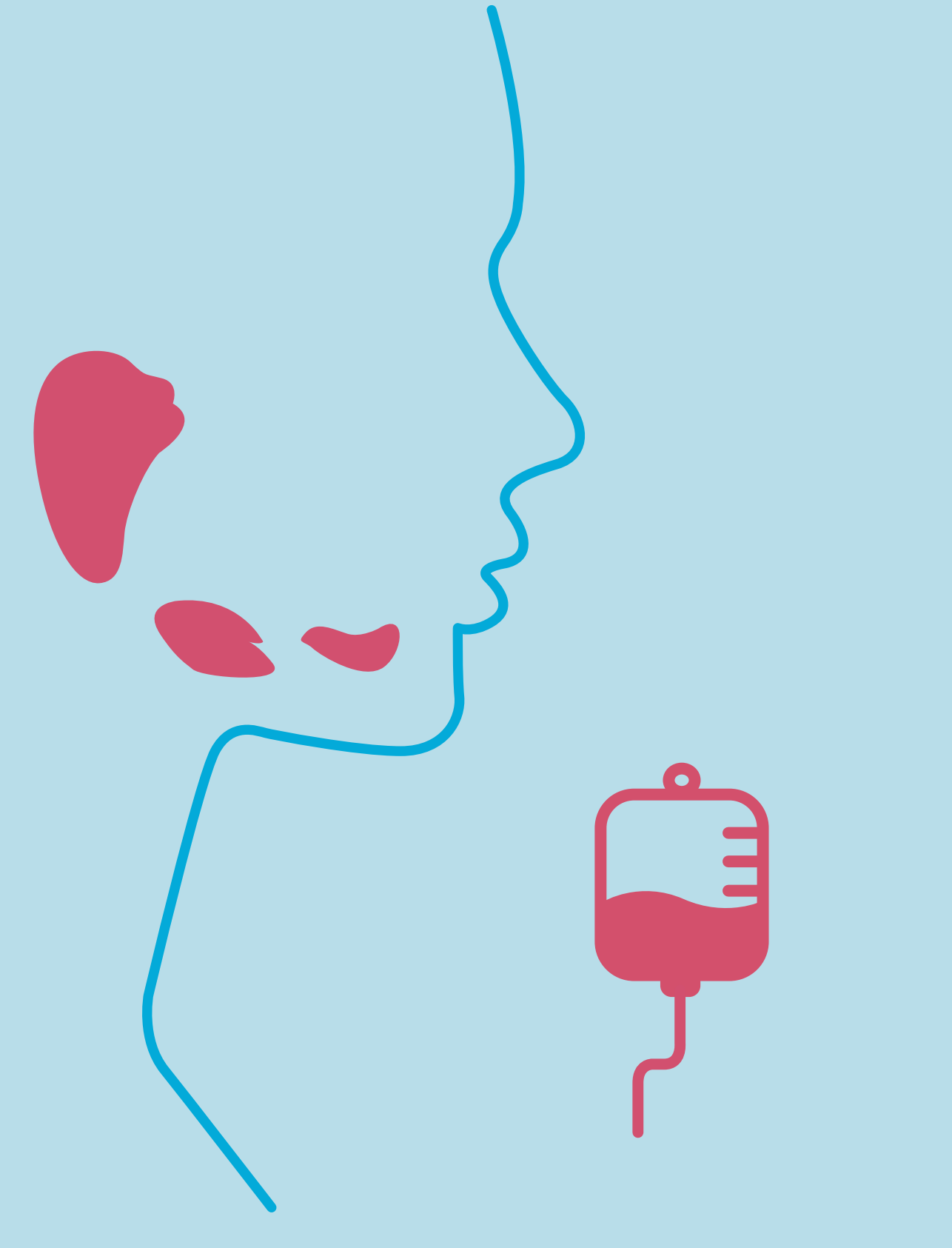
33. Ruan GX, Kazlauskas A. Lactate engages receptor tyrosine kinases Axl, Tie2, and vascular endothelial growth factor receptor 2 to activate phosphoinositide 3-kinase/Akt and promote angiogenesis. J Biol Chem. 2013;288(29):21161-72.

34. Chmielowiec J, Borowiak M, Morkel M, Stradal T, Munz B, Werner S, et al. c-Met is essential for wound healing in the skin. J Cell Biol. 2007;177(1):151-62.

35. Xiang Q, Chen W, Ren M, Wang J, Zhang H, Deng DYB, et al. Cabozantinib Suppresses Tumor Growth and Metastasis in Hepatocellular Carcinoma by a Dual Blockade of VEGFR2 and MET. Clinical Cancer Research. 2014;20(11):2959-70.

36. Goyal L, Zheng H, Yurgelun MB, Abrams TA, Allen JN, Cleary JM, et al. A phase 2 and biomarker study of cabozantinib in patients with advanced cholangiocarcinoma. Cancer. 2017;123(11):1979-88.

37. Tolaney SM, Ziehr DR, Guo H, Ng MR, Barry WT, Higgins MJ, et al. Phase II and Biomarker Study of Cabozantinib in Metastatic Triple-Negative Breast Cancer Patients. The oncologist. 2017;22(1):25-32.



Chapter 4

Case series of docetaxel, trastuzumab, and pertuzumab (DTP) as first-line anti-HER2 therapy and ado-trastuzumab emtansine (T-DM1) as second-line for recurrent or metastatic HER2-positive salivary duct carcinoma.

Maike J.M. Uijen*, Gerben Lassche*, Adriana C.H. van Engen-van Grunsven, Chantal M.L. Driessen, Carla M.L. van Herpen

* These authors contributed equally

Oral Oncology, 2022. ***125****: 105703.*

ABSTRACT

Objective

Salivary duct carcinoma (SDC) overexpresses Human Epidermal growth factor Receptor 2 (HER2) in 29-46% of cases, favouring anti-HER2 therapy. Here, we present the results of patients with recurrent or metastatic HER2-positive SDC treated with docetaxel, trastuzumab, and pertuzumab (DTP) as first-line anti-HER2 therapy and subsequently ado-trastuzumab emtansine (T-DM1) in second-line. Furthermore, we searched for potential biomarkers.

Methods

Retrospective case series from a tertiary hospital. First-line anti-HER2 treatment consisted of DTP, after progression T-DM1 was considered for patients with an adequate performance status. Objective response rate (ORR), progression-free survival (PFS) and overall survival (OS) were assessed and related to mRNA-based PI3K and MAPK signalling pathway activity scores.

Results

Thirteen SDC HER2+ patients received DTP. In twelve evaluable patients, one complete response (CR) and six partial responses (PR) were observed (ORR 58%), with a median PFS of 6.9 months (95%-CI 5.3-8.5). Seven patients received subsequent T-DM1 in second-line, resulting in four PR (ORR 57%), with a median PFS of 4.4 months (95%-CI 0-18.8). Median OS after start of DTP was 42.0 months (95%-CI 13.8-70.1). Grade ≥3 toxicity on DTP was seen in 39% of patients, and 14% on T-DM1. Highest combined PI3K and MAPK signalling was seen in the patient with CR and lowest in the patient with progressive disease on DTP.

Conclusion

In R/M HER2-positive SDC patients DTP followed by T-DM1 upon progression are promising treatments, leading to responses in the majority (58%) of the patients at an acceptable toxicity profile.

INTRODUCTION

Salivary duct carcinoma (SDC) is an aggressive subtype of salivary gland cancer. It was first described by Kleinsasser et al. in 1968 as histologically highly similar to ductal carcinoma of the breast, and recently the resemblance of SDC to apocrine breast cancer has also been recognized regarding genetic background (1, 2). SDC most often occurs in the parotid gland, but it can also originate in other salivary glands in the head and neck region. Primary treatment consists of surgery, which is often combined with a lymph node neck dissection due to the high rates (49-72% of patients) of extensive lymph node involvement. Surgery is regularly followed by postoperative radiotherapy, and in a retrospective case control study with androgen receptor (AR) positive SDC (67-97% expresses AR) adjuvant androgen deprivation therapy (ADT) showed to be possibly effective, but it is not standard, yet (3). The prognosis of SDC is negatively affected by the high rates of distant metastases, and approximately half of the patients diagnosed with SDC are faced with metastatic disease during their disease course, with a median time of 16 months until the occurrence of distant metastases (4). This results in poor survival rates, with a median overall survival (OS) ranging between 48 to 79 months from diagnosis (4-6), and only 5 months in recurrent or metastatic disease when best supportive care is given (7).

In the case of recurrent and/or metastatic (R/M) disease, both androgen deprivation therapy and chemotherapy have previously shown clinical activity, with objective responses ranging from 18-53% and 10-50%, respectively (7-13). Additionally, 29-46% of the SDC tumours overexpress the Human Epidermal growth factor Receptor 2 (HER2), which could serve as a key to targeted therapy (4, 14, 15).

The transmembrane protein HER2 is a member of the epidermal growth factor receptor family and its overexpression is widely recognized to play a critical role in the initiation and maintenance of several malignancies, including SDC. HER2 dimerization leads to the activation of a complex interplay of several signal transduction cascades, which include the important PI3K/AKT/mTOR and RAS/RAF/MEK/ERK (MAPK) cascades (16-18). Currently, a wide palette of therapies aiming to interrupt this signalling exists, targeting at several levels. Small molecule tyrosine kinase inhibitors targeting the receptor pathway (e.g. lapatinib) are available as are agents inhibiting more downstream signalling. Most used are however monoclonal antibodies that upon binding to HER2 uncouple or block dimerization and thereby disrupt initiation of the signalling cascades, besides triggering antibody-dependent cell-mediated cytotoxicity. These include trastuzumab and pertuzumab, which both bind to different epitopes on HER2 and could therefore have synergistic effects (17, 18).

The efficiency of these HER2-targeted therapies has been investigated extensively in HER2-positive breast cancer, following the favourable results of chemotherapy and trastuzumab,

and the addition of pertuzumab has shown even better outcomes. In a phase III study, progression-free survival (PFS) was 18.5 months (OS: 56.5 months) for the group with the addition of pertuzumab, compared to 12.4 months (OS: 40.8 months) for the control group (docetaxel, trastuzumab, placebo) (19, 20). In breast cancer, second-line HER2 targeted therapy with ado-trastuzumab emtansine (T-DM1) is available for patients who progressed on first-line HER2 targeted therapy, following the results of a phase III study in which objective responses were observed in 43.6% of the cases (21).

In SDC, agents targeting the HER2 pathway have shown impressive response rates (4, 14, 15). In the prospective phase II study of Takahashi *et al.,* 57 HER2-positive R/M SDC patients were treated with docetaxel and trastuzumab, which resulted in objective responses in 70% of the patients and a median PFS and OS of 8.9 and 39.7 months (22), respectively. However, in SDC patients, literature is scarce on the effects of the possible synergistic combination of docetaxel, trastuzumab, and pertuzumab (DTP) and subsequent T-DM1, that is used in breast cancer patients. Existing literature on these treatments mainly consist of case reports (23-28).

In this paper we therefore describe the results of SDC patients treated with DTP as first-line anti-HER2 therapy and T-DM1 in second-line in our tertiary referral hospital specialized in salivary gland cancer in the Netherlands. Besides this, we provide a preliminary analysis of possible biomarkers predicting response to these HER2 targeting agents, based on quantification of PI3K and MAPK pathway activities.

PATIENTS AND METHODS

Patients

All HER2-positive SDC patients that were treated with DTP as first-line anti-HER2 therapy (and followed with T-DM1 in second-line in part of patients) at the Radboud University Medical Centre (a tertiary centre for recurrent and metastatic salivary gland cancer in the Netherlands), were retrospectively identified for this retrospective case-series. According to Dutch law, a review by a medical ethical committee was not required due to the retrospective nature of this research.

HER2 status

HER2 status was assessed a combination of immunohistochemistry (IHC) and in situ hybridization (ISH) and interpreted following the guidelines for HER2 assessment in breast cancer (29, 30). For IHC the HercepTest (Dako Agilent) was used (including a rabbit anti-human HER2 monoclonal antibody). An experienced pathologist scored the staining intensity which ranged from 0 (no immunoreactivity) to 3+ (strong immunoreactivity in >10% of tumour cells). Fluorescence in situ hybridization (FISH) was performed with dual ERBB2 FISH probes

(Z-2077-200; ZytoVision or KB-00007; Leica). An HER2-CEP17 ratio was calculated, tumours with a ratio of >2 were considered to be amplified.

Treatment

DTP consisted of the combination docetaxel, trastuzumab, and pertuzumab, administered every 3 weeks. After 6 dosages of docetaxel (75 mg/m2), patients continued the combination of trastuzumab and pertuzumab until disease progression or intolerable toxicity. In the case of docetaxel-related toxicity, the dose of docetaxel could be reduced. Trastuzumab was given either intravenously (i.v., starting dose 8 mg/kg, with subsequent dosages of 6 mg/kg) or subcutaneously (s.c., 600 mg). The starting dose of pertuzumab consisted of 840 mg with subsequent dosages of 420 mg, intravenously. Termination of treatment was at the discretion of the treating physician. In some cases, the treatment could have been continued despite progression according to Response Evaluation Criteria in Solid Tumours (RECIST) criteria (e.g. in case of the ongoing response of target lesions despite the occurrence of a new metastatic lesion).

After DTP treatment, second-line HER2 targeted therapy consisting of ado-trastuzumab emtansine (T-DM1) was considered for patients with an adequate performance status (Karnofsky ≥70). T-DM1 treatment consisted of 3.6 mg/kg intravenously, every 3 weeks. The dose could be adjusted in case of toxicity. T-DM1 was continued until disease progression or intolerable toxicity.

For both treatments, patients were evaluated approximately every 3 months, consisting of MR-scanning or CT of the head and neck area (in case of local recurrence or brain metastases) and CT-scan of the chest and abdomen. Patients could receive local treatment during or in between DTP or T-DM1 therapy, including stereotactic brain radiotherapy in case of brain metastases.

Outcomes

Treatment evaluation was performed according to the RECIST version 1.1 (31). Objective response rate (ORR) was defined as complete response (CR) or partial response (PR). Other response categories were stable disease (SD) and progressive disease (PD). Progression-free survival (PFS) was defined as the time between the start of HER2 targeted therapy and disease progression or death. Overall survival (OS) was defined as the time between the start of HER2 targeted therapy until the death of any cause or lost follow-up. Time on treatment was defined as the time between the start of HER2 targeted therapy and the last administration of that treatment. Furthermore, treatment-related adverse events were retrospectively identified from medical files. Adverse events were scored according to the Common Terminology Criteria for Adverse Events, version 5.0 (32).

Signalling pathway activity quantification

If residual tumour tissue from regular diagnostics prior to DTP initiation was available, three 10 µm slices of formalin-fixated paraffin-embedded (FFPE) material were collected. These slices were annotated for the presence of tumour material on an adjacent haematoxylin and eosin slide. RNA was extracted using VERSANT Tissue Preparation Reagents kit (Siemens, Munich, Germany) according to the manufacturer's instructions. The Philips pathway activity profiling OncoSignal test (Philips Molecular Pathway Diagnostics, Eindhoven, The Netherlands, model version O4.4) was used to quantify the Phosphoinositide 3-Kinase pathway (PI3K, as the inverse of Forkhead Box-O (FOXO) signalling) and Mitogen-Activated Protein Kinase pathway (MAPK) activities. If enough RNA was available Androgen Receptor pathway (AR), Notch signalling pathway (Notch), Transforming Growth Factor beta signalling pathway (TGFβ), Estrogen Receptor pathway (ER) and Hedgehog signalling pathway (HH) activities were also quantified. For each of these pathways, output of this test is the odds of the transcription complex of this pathway being active vs. not active, expressed on a logarithmic scale and scaled to range from 0-100. This approach has previously been published and is validated in several tissue types for different pathways, which include AR pathway activity in SDC (33-37).

As both the PI3K and MAPK pathway are downstream signalling cascades for activated HER2 receptors, a composite metric of these two activity scores was used, defined as the sum of PI3K and MAPK scores.

Statistical analysis

Descriptive measures were summarized as medians with their respective ranges (minimum and maximum). Survival curves using Kaplan-Meier estimates were constructed, for OS and for PFS on both therapies. Kaplan-Meier curves were made for the entire cohort and after dichotomization was made using the median PI3K/MAPK scores. Using the Kaplan-Meier estimates, median survival with 95% confidence intervals (CI) were estimated. A log-rank test was performed to compare differences in survival. Analyses were performed in SPSS version 25. Graphical work was created using Python version 3.8 with Matplotlib, Pandas, Numpy, Seaborn and Lifelines packages and R Studio version 3.5.3.

RESULTS

Patient characteristics

In total 13 patients received DTP as first-line anti-HER2 therapy and 7 patients T-DM1 as second-line. The first HER2-positive SDC patient started with DTP in 2015. The median age at the start of DTP therapy was 61 years (range 48-75). The majority of patients was male (77%). Most often the SDC tumour occurred in the parotid gland (92%), only in one patient (8%) the primary tumour was located in the submandibular gland. HER2 status was assessed on the primary tumour in 8/13 patients and on metastatic tissue in 5/13 patients. IHC staining intensity ranged from 2+ to 3+. All patients had HER2 amplified SDC tumours assessed with FISH. In addition, the tumours of all 13 patients were AR-positive. Prior systemic therapy included adjuvant ADT (23%), palliative ADT (46%) and chemotherapy (8%) (table 2). Additional information on baseline patient characteristics is listed in table 1. Median duration of follow-up, from start of DTP to current analysis, was 15.4 months (range: 5.5-55.0). Last data for this study was collected on 27 July 2021. Figure 1 graphically summarizes treatment and response information per individual patient.

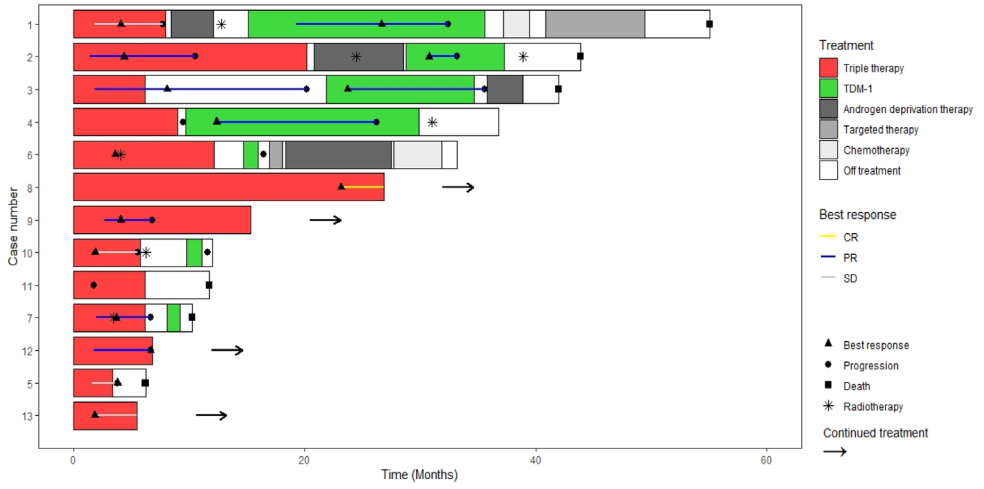


Figure 1: Swimmers plot, graphically summarizing treatment and response information per individual case

Table 1: Baseline patient characteristics before start of HER2 targeted therapy

Patient No.	Age	Gender	Primary tumour	Prior treatments	Disease distribution	Sites of DM	HER2 status assessed on	HER2 IHC	HER2 FISH	AR IHC ‡
1	48	F	Parotid gland	Surgery + PORT	DM	Lung	Primary tumour	3+	amplified	positive
2	64	M	Parotid gland	Surgery + PORT Palliative ADT	DM	Lung, liver, lymph node	Primary tumour	3+	amplified	positive
3	54	M	Parotid gland	Surgery + PORT Palliative ADT	DM	Lung, liver, bone	Bone metastasis	3+	amplified	positive
4	59	M	Parotid gland	Surgery Palliative ADT	DM	Brain, bone, lymph node	Primary tumour	2-3+	amplified	positive
5	54	F	Parotid gland	Palliative ADT	LR + DM	Lung, liver, lymph node	Liver metastasis	2-3+	amplified	positive
6	51	M	Parotid gland	-	LR	-	Primary tumour	2-3+	amplified	positive
7	55	M	Parotid gland	Surgery	DM	Brain, lung, lymph node	Lung metastasis	3+	amplified	positive
8	66	F	Parotid gland	Surgery + PORT Adjuvant ADT	DM	Lung	Primary tumour	3+	amplified	positive
9	75	M	Submandibular gland	Surgery + PORT Adjuvant ADT	DM	Lung, lymph node	Primary tumour	3+	amplified	positive
10	64	M	Parotid gland	Palliative chemo† Palliative ADT	LR + DM	Lymph node	Lymph node metastasis	3+	amplified	positive
11	61	M	Parotid gland	Surgery + PORT Adjuvant ADT	DM	Liver	Liver metastasis	2-3+	amplified	positive
12	62	M	Parotid gland	Surgery + PORT Palliative Rx	LR + DM	Lymph node, brain	Primary tumour	3+	amplified	positive
13	67	M	Parotid gland	Surgery+ PORT Palliative ADT	LR + DM	Lung, pancreas	Primary tumour	3+	amplified	positive

ADT: androgen deprivation therapy, AR: androgen receptor, DM: distant metastases, F: female, FISH: fluorescence in situ hybridization, IHC: immunohistochemistry, LR: locoregional disease, M: male, PORT: Postoperative radiotherapy, Rx: radiotherapy

†Cisplatin, etoposide and ifosfamide

‡ AR status was assessed by an experienced pathologist through immunohistochemistry in routine clinical practice.

DTP therapy

All 13 patients received DTP, with a median duration of treatment of 6.9 months (range 3.4-26.8+). Six patients (46%) required a dose reduction of docetaxel during DTP treatment. Of the 13 patients, two patients consistently received trastuzumab i.v., seven patients consistently received trastuzumab s.c. and four patients have received trastuzumab both i.v. and s.c. during their treatment. Twelve patients were RECIST evaluable (one patient did not have target lesions). The ORR was 58%, 1 CR (8%) and 6 PR (50%). Furthermore, 4 patients (33%) had stable disease (SD) of which only one patient had SD with a response duration of >6 months (table 2). At the time of this report, four patients are still on DTP therapy. Median overall survival (calculated using Kaplan-Meier estimates) was 42.0 (95%-CI 13.8-70.1 months) after start of DTP (figure 2A). Median PFS on DTP was 6.9 months (95%-CI 5.2-8.5) (figure 2B) and median time on treatment 8.0 months (95%-CI 3.8-12.1). Three patients with brain metastases received DTP. All these patients received radiation therapy on the brain metastases prior to the start of DTP. The brain metastases of two patients showed a significant reduction in size over time; after the second treatment evaluation (several months after the start of DTP), the brain metastases decreased further than shown on the first treatment evaluation. In one other patient new brain metastases were detected on the second treatment evaluation scans.

T-DM1 therapy

Seven patients received T-DM1 after DTP therapy. The median time on treatment was 8.5 months (range 1.1-20.4). During T-DM1 treatment, three patients (43%) required a dose reduction. T-DM1 resulted in an ORR of 57%, 4 PR (57%), see table 2. The three other patients (43%) had progressive disease (PD) as best response. In figure 3 the response of a patient with pulmonary metastases on T-DM1 is visualized. Median PFS on T-DM1 after progression on DTP was 4.4 months (95%-CI 0-18.8) (figure 2C). Two patients with brain metastases received second-line T-DM1, one of these patients died before the first treatment evaluation, in the other patient a reduction in tumour size of the brain metastasis was observed (PR).

Table 2: Response to HER2 targeted therapy

Patient No.	First-line HER2 targeted treatment (DTP therapy)	Best response	Best percentage change in target lesions	Duration of response	Second-line HER2 targeted treatment (T-DM1)	Best response	Best percentage change in target lesions	Duration of response
1	Docetaxel + trastuzumab + pertuzumab	SD	-17%	7.7 mo	T-DM1	PR	-78%	17.3 mo
2	Docetaxel + trastuzumab + pertuzumab	PR	-62%	10.6 mo	T-DM1	PR	-32%	4.4 mo
3	Docetaxel + trastuzumab + pertuzumab	PR	-100%	20.2 mo	T-DM1	PR	-42%	13.7 mo
4	Docetaxel + trastuzumab + pertuzumab	IR/SD*	N.A.	9.5 mo	T-DM1	PR	-40%	16.6 mo
5	Docetaxel + trastuzumab + pertuzumab	SD	-27%	3.8 mo	-	-	-	-
6	Docetaxel + trastuzumab + pertuzumab	PR	-33%	Unclear†	T-DM1	PD	+22%	1.8 mo
7	Docetaxel + trastuzumab + pertuzumab	PR	-45%	6.7 mo	T-DM1	PD	Unclear‡	Unclear‡
8	Docetaxel + trastuzumab + pertuzumab	CR	-100%	Ongoing at 26.8 mo	-	-	-	-
9	Docetaxel + trastuzumab + pertuzumab	PR	-78%	6.9 mo¶	-	-	-	-
10	Docetaxel + trastuzumab + pertuzumab	SD	+1%	5.6 mo	T-DM1	PD	+34%	1.8 mo
11	Docetaxel + trastuzumab + pertuzumab	PD	+38%	1.8 mo	-	-	-	-
12	Docetaxel + trastuzumab + pertuzumab	PR	-68%	6.7 mo¶	-	-	-	-
13	Docetaxel + trastuzumab + pertuzumab	SD	-2%	Ongoing at 5.5 mo	-	-	-	-

Abbreviations: CR: complete response, mo: months, PR: partial response, SD: stable disease.

Table 2 legend on next page.

*This patient had no RECIST measurable target lesions, based on non-target lesions the response is defined as: incomplete response/stable disease (IR/SD

†This patient was treated for locally advanced disease, based on the good response on HER2 therapy, the patient received local radiotherapy with a curative intent. Therefore the duration of response could not be assessed.

‡Patient died before first treatment evaluation imaging.

¶ According to RECIST criteria, these patients have progressive disease. However, treatment in these patients is still ongoing; treatment discontinuation is at the discretion of the treating physician, as previously described in methods section.

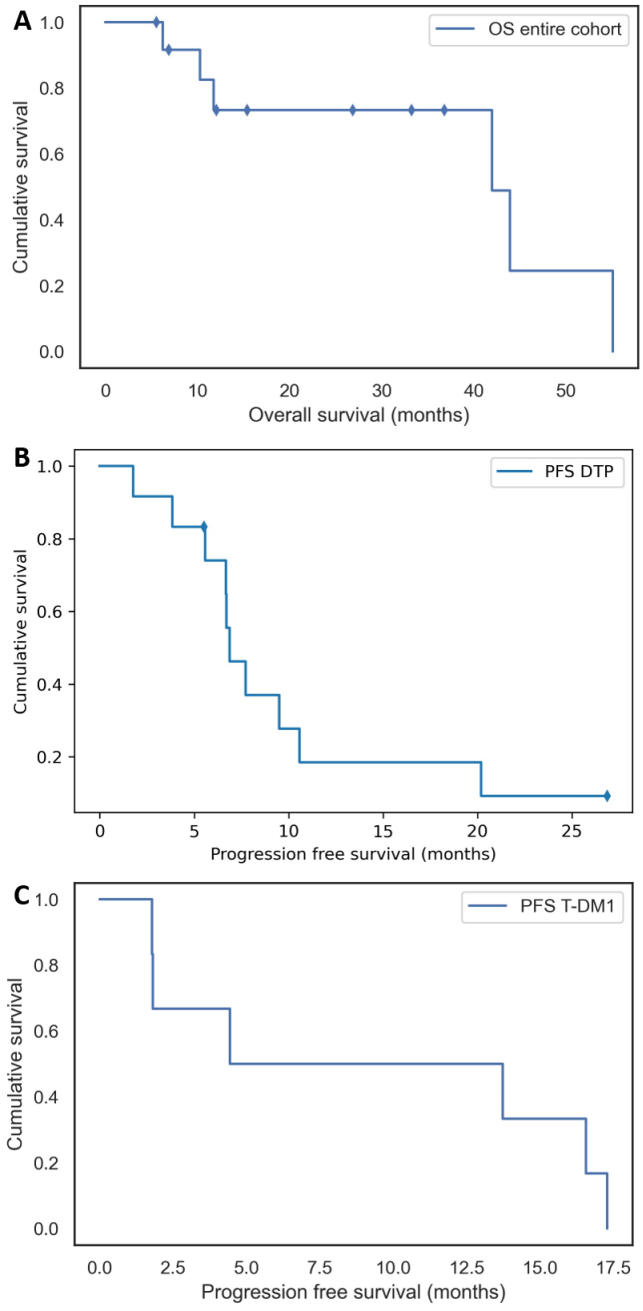


Figure 2: A. Kaplan-Meier curve of overall survival for the entire cohort. **B.** Kaplan-Meier curve of progression free survival on DTP therapy. C. Kaplan-Meier curve of progression free survival on T-DM1.

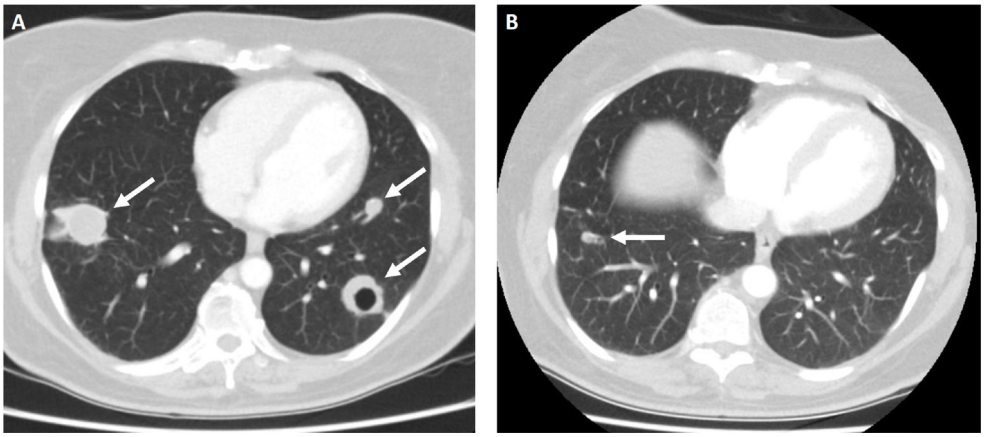


Figure 3: Visualization of response on T-DM1 therapy in SDC patient with pulmonary metastases, A: before start of T-DM1 therapy B: 1 year on treatment. Pulmonary metastases are indicated by white arrows.

Response prediction potential of pathway analysis

Of the 13 patients, material for RNA extraction was available in 11 cases (85%). RNA quantities were sufficient to assess PI3K and MAPK pathway activity scores in these 11 cases. In 10 cases AR, ER, HH, Notch and TGFβ pathway activity scores could also be assessed. The median MAPK pathway activity score was 62 (range 47-69) and PI3K pathway activity score 34 (range 27-48). Composite scores, summing these metrics ranged from 88-107 (median 94). Figure 4A summarizes pathway activity scores of the samples in which all pathways could be determined. In all patients, activity scores followed the same pattern, with a relatively narrow bandwidth of scores within a single pathway, with a range of usually <25 points (except for one outlier in the HH pathway, figure 4A). AR pathway activity scores ranged from 42-60, with a median of 51 (figure 4A).

The patient with the highest combined PI3K/MAPK score was the only patient experiencing CR and the patient with the lowest combined score was the only patient experiencing PD as best response upon DTP therapy (figure 4B). PR and SD scores ranged in between. No relation was observed between the combined PI3K/MAPK score and the response on T-DM1 (supplementary figure 1).

After dichotomization of the cohort on the median PI3K/MAPK score, OS and PFS did not differ significantly between the groups with a low and high score (p=0.15, p=0.59 and p=0.21, regarding OS, PFS on DTP therapy and PFS on T-DM1, respectively, Supplementary figure 2).

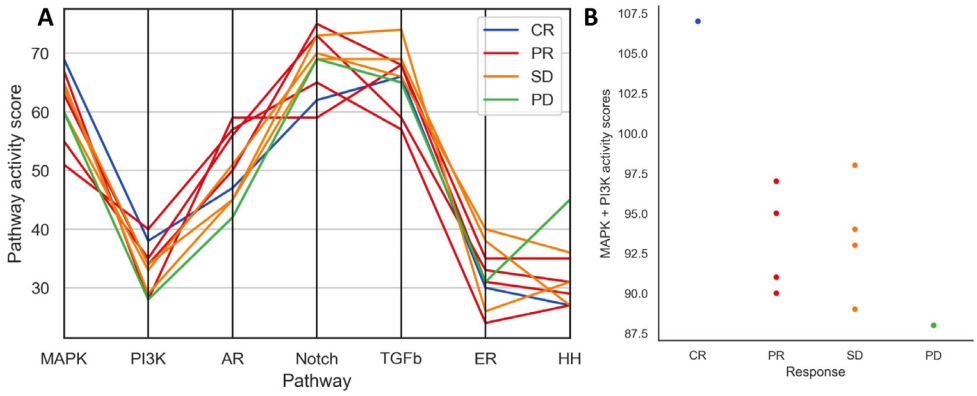


Figure 4: A: parallel coordinates plot of all seven pathway activity scores, color-coded for response on DTP therapy. **B:** Swarm plot of combined MAPK and PI3K score versus response on DTP therapy.

Toxicity

Regarding DTP therapy, most common toxicities were mainly docetaxel related and included haematological toxicity, gastrointestinal toxicity, fatigue, and peripheral sensory neuropathy. Five patients (39%) developed grade ≥3 toxicity on DTP, including infections (n=3), neutropenia (n=1), and heart failure (related to trastuzumab/pertuzumab) (n=1) (table 3, Supplementary table 1). Regarding T-DM1 therapy, often reported toxicities were fatigue, nausea, peripheral sensory neuropathy, and increasing levels of liver enzymes. One patient (14%) developed grade ≥3 hyponatremia during T-DM1 treatment, this was however deemed unrelated to T-DM1 treatment.

Table 3: Grade 3 and 4 treatment-related toxicity

	DTP therapy (N=13)	**T-DM1 therapy (N=7)**
Adverse events	Grade 3-4 N (%)	Grade 3-4 N (%)
Bone marrow toxicity		
White blood count decreased	1 (8)	-
Lymphocyte count decreased	1 (8)	-
Neutrophil count decreased	2 (15)	-
Gastrointestinal toxicity		
Pharyngeal mucositis	1 (8)	-
Infectious toxicity		
Skin infection	2 (15)	-
Enterocolitis infectious	1 (8)	-
Heart failure	1 (8)	-

Toxicity was retrospectively scored per CTCAE version 5.0.

DISCUSSION

In this retrospective case series, the efficacy of two sequential HER2 targeted treatment strategies was evaluated in HER2-positive recurrent or metastatic SDC patients. The majority of the 13 patients treated with DTP responded to this therapy, with an objective response rate of 58%. Also on subsequent T-DM1 therapy, objective responses were observed for the majority of patients (4/7; 57%). Besides this, the PI3K and MAPK signalling pathway activity scores show potential in predicting response to DTP treatment, although numbers of patients in this study are too low to draw firm conclusions, so these findings should be considered as hypothesis generating rather than confirming.

The combination of docetaxel and trastuzumab has previously shown a comparable, even slightly better response rate in a prospective phase II study in SDC patients as compared to our DTP regimen that also includes pertuzumab, ORR 70% with median PFS of 8.9 (95%-CI 7.8-9.9) and OS of 39.7 months (95%-CI not reached) versus ORR of 58% with PFS of 6.9 months (95%-CI 5.2-8.5) and OS of 42.0 months (95%-CI 13.8-70.1), respectively (22). This might be due to selection bias, since generally prospective studies include a different patient population as compared to a routine clinical setting. For instance, brain metastases occurred in 5% of the patients in that study, compared to 23% in our study. This is also supported by other retrospective data, in which an OS for R/M SDC in case of best supportive care as low as 5 months and 17 months upon administration of ADT was seen, although these were not all HER2-positive patients (HER2 is probably not a prognostic factor in SDC) (4, 7). Besides this, the sample size in this study is relatively small, influencing robustness of the found ORR, as one response more or one response less substantially alters the ORR. Regarding efficacy of T-DM1 in salivary gland cancer, preliminary results of a recent phase II basket study in HER2-positive patients also reported high response rates (28). Although unclear how many of SDC subtype, the ORR in this basket study was higher than our results: 90% (9/10 patients) versus 57% (4/7 patients), but both results point towards efficacy of T-DM1 most patients (28). Interestingly, recent research also focusses on the efficacy of HER2 targeted treatments in adjuvant setting of HER2-positive SDC (38). A prospective study on adjuvant T-DM1 is currently recruiting patients (NCT04620187).

Given the histological and molecular similarity between breast cancer and SDC and the frequent overexpression of HER2 in breast cancer, achieved results on anti-HER2 therapy in breast cancer are of interest. The addition of pertuzumab to docetaxel and trastuzumab (DTP) has shown favourable results in HER2-positive breast cancer with a PFS of 18.5 months for DTP versus 12.4 for docetaxel and trastuzumab (19, 20). Also in HER2-positive gastric cancers, slightly better outcomes were seen upon addition of pertuzumab to trastuzumab combined with chemotherapy: PFS 8.5 months (pertuzumab) versus 7.0 months (placebo) (39, 40).

Toxicity was on average bearable, highlighting the potential of this therapy for this aggressive cancer. Overall, the toxicity of docetaxel and trastuzumab seems comparable to DTP. However, in the prospective SDC trial of docetaxel and trastuzumab no grade ≥2 or higher heart failure was observed, yet one patient in our case series developed grade 3 heart failure. Cardiotoxicity is a known side-effect of trastuzumab treatment, but in larger clinical trials the addition of pertuzumab to trastuzumab did not result in higher rates of cardiotoxicity (19, 20, 39). It, therefore, seems unlikely that the case of heart failure in this case series is related to the addition of pertuzumab.

In our cohort, the treatment-related toxicity profile of T-DM1 was more favourable than that of DTP (grade ≥3 toxicity in 0% versus 39%). The relatively favourable toxicity profile of T-DM1 is also observed in phase III studies of HER2-positive breast cancer patients; adverse events are generally of low grade and manageable (41). This raises the question of whether T-DM1 should be considered as first-line treatment (before DTP). To date, no clinical study in metastatic breast cancer compared DTP to T-DM1 in the first-line setting. Yet, results of a retrospective multicentre study in early-relapsing breast cancer patients, suggests the superiority of DTP over T-DM1 (42). Therefore, first-line HER-2 therapy with DTP followed by T-DM1 remains probably the favourable choice.

In this study, PI3K and MAPK pathway signalling cascades were quantified at mRNA level to explore their potential as predictive or prognostic biomarkers based on the hypothesis that the high activity of these cascades is a result of HER2 activation and that tumour cells with high PI3K/MAPK signalling depend on these pathways for survival and proliferation. Inhibition of HER2, preventing downstream PI3K/MAPK signalling, would hit tumours with high scores harder than those with low activity scores.

Quantification of downstream HER2 signalling is however hard, given the complex nature of the involved and intertwined signalling cascades (17, 18). The summation of the MAPK and PI3K pathway activity scores, which are both optimized towards the quantification of the single pathway, might be a too simple representation of this complex biology. It is unknown which of these two pathways most, let alone in which amount, contributes to the pro-tumorigenic effect in SDC. It is however promising to see that despite these limitations, the combined PI3K/MAPK scores could still be of predictive value as the one patient in our small series experiencing PD on DTP had the lowest score and the only patient experiencing CR the highest score. For T-DM1 response prediction PI3K/MAPK pathway activity scores (Supplementary figure 1, 2C) might not be optimal biomarkers, as HER2 is mainly used as a target to deliver the cytotoxic emtansine rather than to specifically inhibit downstream signalling.

Recently, the interplay between HER2 and AR signalling cascades is gaining increasing interest, and in models of other cancers reciprocal activation of these pathways has been observed (43, 44). However, the extent of this cross-talk in SDC and its clinical consequences still needs to be investigated. In our case series, all SDC patients were both HER2-positive and AR-positive. If HER2-targeted therapy influenced AR-targeted treatment and vice versa is difficult to deduce from our case series due to the limited patient numbers and heterogenous treatment strategies (e.g. in several patients HER2-directed therapy was administered before AR-directed therapy was given, and in other patients the other way around).

The aggressive nature and rarity of SDC hampers patient accrual in large clinical trials, and learning from advances made in more common cancers is therefore an appealing strategy. Although this study, translating the advances made in breast cancer to SDC patients, has several limitations, such as the retrospective nature and limited sample size, it may still be of great value to SDC patients given the abovementioned difficulties in clinical studies. The absence of a control cohort impedes the drawing of a robust conclusion about anti-HER2 therapy on PFS and OS, but our results seem promising in this patient group with dismal prognoses. This study leaves the question unanswered which sequence of anti-HER2 treatment strategies for HER2-positive SDC patients is optimal and what the additive effect pertuzumab onto trastuzumab and docetaxel is. As overall toxicity does not seem to be increased upon addition of pertuzumab, and synergy of trastuzumab/pertuzumab in blocking HER2 downstream signalling is to be expected both because of the working mechanisms of these agents and as a result of clinical trials in other cancers, we suggest the addition of pertuzumab to trastuzumab/docetaxel in SDC patients (18), realizing that a formal phase III study comparing both treatment arms could not be performed because of the rarity of the disease.

Furthermore, recent research in breast cancer indicates that Trastuzumab deruxtecan, a new antibody-drug conjugate targeting HER2, improved PFS when compared to T-DM1 as second-line HER2 targeted therapy. Trastuzumab deruxtecan might replace T-DM1 as second-line HER-2 treatment in the future (45, 46). This also encourages future research on Trastuzumab deruxtecan in SDC patients.

CONCLUSION

In R/M HER2-positive SDC patients, DTP followed by T-DM1 upon progression as second-line anti-HER2 therapy are promising treatment strategies, leading to responses in the majority of the patients at an acceptable toxicity profile.

ACKNOWLEDGEMENTS

We would like to thank Wim Verhaegh (Philips Research, Eindhoven, The Netherlands) and Diederick Keizer (Philips molecular pathway diagnostics, Eindhoven, The Netherlands) for performing the pathway analyses and critical revision of the manuscript.

REFERENCES

1. Kleinsasser O, Klein HJ, Hubner G. [Salivary duct carcinoma. A group of salivary gland tumors analogous to mammary duct carcinoma]. Arch Klin Exp Ohren Nasen Kehlkopfheilkd. 1968;192(1):100-5.
2. Dalin MG, Desrichard A, Katabi N, Makarov V, Walsh LA, Lee KW, et al. Comprehensive Molecular Characterization of Salivary Duct Carcinoma Reveals Actionable Targets and Similarity to Apocrine Breast Cancer. Clinical cancer research : an official journal of the American Association for Cancer Research. 2016;22(18):4623-33.
3. van Boxtel W, Locati LD, van Engen-van Grunsven ACH, Bergamini C, Jonker MA, Fiets E, et al. Adjuvant androgen deprivation therapy for poor-risk, androgen receptor-positive salivary duct carcinoma. European journal of cancer (Oxford, England : 1990). 2019;110:62-70.
4. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.
5. Roh JL, Lee JI, Choi SH, Nam SY, Kim SO, Cho KJ, et al. Prognostic factors and oncologic outcomes of 56 salivary duct carcinoma patients in a single institution: high rate of systemic failure warrants targeted therapy. Oral oncology. 2014;50(11):e64-6.
6. Jayaprakash V, Merzianu M, Warren GW, Arshad H, Hicks WL, Jr., Rigual NR, et al. Survival rates and prognostic factors for infiltrating salivary duct carcinoma: Analysis of 228 cases from the Surveillance, Epidemiology, and End Results database. Head & neck. 2014;36(5):694-701.
7. Boon E, van Boxtel W, Buter J, Baatenburg de Jong RJ, van Es RJJ, Bel M, et al. Androgen deprivation therapy for androgen receptor-positive advanced salivary duct carcinoma: A nationwide case series of 35 patients in The Netherlands. Head & neck. 2018;40(3):605-13.
8. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.
9. Viscuse PV, Price KA, Garcia JJ, Schembri-Wismayer DJ, Chintakuntlawar AV. First Line Androgen Deprivation Therapy vs. Chemotherapy for Patients With Androgen Receptor Positive Recurrent or Metastatic Salivary Gland Carcinoma-A Retrospective Study. Frontiers in oncology. 2019;9:701.
10. Locati LD, Perrone F, Cortelazzi B, Lo Vullo S, Bossi P, Dagrada G, et al. Clinical activity of androgen deprivation therapy in patients with metastatic/relapsed androgen receptor-positive salivary gland cancers. Head & neck. 2016;38(5):724-31.
11. Okada T, Saotome T, Nagao T, Masubuchi T, Fushimi C, Matsuki T, et al. Carboplatin and docetaxel in patients with salivary gland carcinoma: A retrospective study. In Vivo. 2019;33(3):843-53.
12. Nakano K, Sato Y, Sasaki T, Shimbashi W, Fukushima H, Yonekawa H, et al. Combination chemotherapy of carboplatin and paclitaxel for advanced/metastatic salivary gland carcinoma patients: differences in responses by different pathological diagnoses. Acta oto-laryngologica. 2016;136(9):948-51.
13. Uijen MJM, Lassche G, van Engen-van Grunsven ACH, Tada Y, Verhaegh GW, Schalken JA, et

al. Systemic therapy in the management of recurrent or metastatic salivary duct carcinoma: A systematic review. Cancer Treat Rev. 2020;89:102069.

14. Takase S, Kano S, Tada Y, Kawakita D, Shimura T, Hirai H, et al. Biomarker immunoprofile in salivary duct carcinomas: clinicopathological and prognostic implications with evaluation of the revised classification. Oncotarget. 2017;8(35):59023-35.
15. Schmitt NC, Kang H, Sharma A. Salivary duct carcinoma: An aggressive salivary gland malignancy with opportunities for targeted therapy. Oral oncology. 2017;74:40-8.
16. Vu T, Claret FX. Trastuzumab: updated mechanisms of action and resistance in breast cancer. Frontiers in oncology. 2012;2:62.
17. Yarden Y, Pines G. The ERBB network: at last, cancer therapy meets systems biology. Nat Rev Cancer. 2012;12(8):553-63.
18. Arteaga CL, Engelman JA. ERBB receptors: from oncogene discovery to basic science to mechanism-based cancer therapeutics. Cancer Cell. 2014;25(3):282-303.
19. Baselga J, Cortes J, Kim SB, Im SA, Hegg R, Im YH, et al. Pertuzumab plus trastuzumab plus docetaxel for metastatic breast cancer. The New England journal of medicine. 2012;366(2):109-19.
20. Swain SM, Baselga J, Kim SB, Ro J, Semiglazov V, Campone M, et al. Pertuzumab, trastuzumab, and docetaxel in HER2-positive metastatic breast cancer. The New England journal of medicine. 2015;372(8):724-34.
21. Verma S, Miles D, Gianni L, Krop IE, Welslau M, Baselga J, et al. Trastuzumab emtansine for HER2-positive advanced breast cancer. The New England journal of medicine. 2012;367(19):1783-91.
22. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.
23. Park JC, Ma TM, Rooper L, Hembrough T, Foss RD, Schmitt NC, et al. Exceptional responses to pertuzumab, trastuzumab, and docetaxel in human epidermal growth factor receptor-2 high expressing salivary duct carcinomas. Head & neck. 2018;40(12):E100-e6.
24. van Boxtel W, Boon E, Weijs WLJ, van den Hoogen FJA, Flucke UE, van Herpen CML. Combination of docetaxel, trastuzumab and pertuzumab or treatment with trastuzumab-emtansine for metastatic salivary duct carcinoma. Oral oncology. 2017;72:198-200.
25. Almquist D, Umakanthan JM, Ganti AK. Sequential HER2-Targeted Therapy in Salivary Ductal Carcinoma With HER2/neu Overexpression and a Concomitant ERBB2 Mutation. JCO Precision Oncology. 2018(2):1-5.
26. Correa TS, Matos GDR, Segura M, Dos Anjos CH. Second-Line Treatment of HER2-Positive Salivary Gland Tumor: Ado-Trastuzumab Emtansine (T-DM1) after Progression on Trastuzumab. Case reports in oncology. 2018;11(2):252-7.
27. Swed BL, Cohen RB, Aggarwal C. Targeting HER2/neu Oncogene Overexpression With Ado-Trastuzumab Emtansine in the Treatment of Metastatic Salivary Gland Neoplasms: A Single-Institution Experience. JCO precision oncology. 2019;3.
28. Li BT, Shen R, Offin M, Buonocore DJ, Myers ML, Venkatesh A, et al. Ado-trastuzumab emtansine in patients with HER2 amplified salivary gland cancers (SGCs): Results from a phase II basket trial. Journal of Clinical Oncology. 2019;37(15_suppl):6001-.

29. Wolff AC, Hammond MEH, Allison KH, Harvey BE, Mangu PB, Bartlett JMS, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Clinical Practice Guideline Focused Update. Archives of pathology & laboratory medicine. 2018;142(11):1364-82.
30. Wolff AC, Hammond MEH, Hicks DG, Dowsett M, McShane LM, Allison KH, et al. Recommendations for Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Clinical Practice Guideline Update. Journal of Clinical Oncology. 2013;31(31):3997-4013.
31. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). European journal of cancer (Oxford, England : 1990). 2009;45(2):228-47.
32. Sood S, McGurk M, Vaz F. Management of Salivary Gland Tumours: United Kingdom National Multidisciplinary Guidelines. The Journal of laryngology and otology. 2016;130(S2):S142-s9.
33. Lassche G, Tada Y, van Herpen CML, Jonker MA, Nagao T, Saotome T, et al. Predictive and Prognostic Biomarker Identification in a Large Cohort of Androgen Receptor-Positive Salivary Duct Carcinoma Patients Scheduled for Combined Androgen Blockade. Cancers (Basel). 2021;13(14).
34. van Boxtel W, Verhaegh GW, van Engen-van Grunsven IA, van Strijp D, Kroeze LI, Ligtenberg MJ, et al. Prediction of clinical benefit from androgen deprivation therapy in salivary duct carcinoma patients. International journal of cancer. 2020;146(11):3196-206.
35. van de Stolpe A. Quantitative Measurement of Functional Activity of the PI3K Signaling Pathway in Cancer. Cancers (Basel). 2019;11(3).
36. van de Stolpe A, Holtzer L, van Ooijen H, Inda MA, Verhaegh W. Enabling precision medicine by unravelling disease pathophysiology: quantifying signal transduction pathway activity across cell and tissue types. Sci Rep. 2019;9(1):1603.
37. van de Stolpe A, Verhaegh W, Blay JY, Ma CX, Pauwels P, Pegram M, et al. RNA Based Approaches to Profile Oncogenic Pathways From Low Quantity Samples to Drive Precision Oncology Strategies. Front Genet. 2020;11:598118.
38. Hanna GJ, Bae JE, Lorch JH, Haddad RI, Jo VY, Schoenfeld JD, et al. The Benefits of Adjuvant Trastuzumab for HER-2-Positive Salivary Gland Cancers. The oncologist. 2020;25(7):598-608.
39. Tabernero J, Hoff PM, Shen L, Ohtsu A, Shah MA, Cheng K, et al. Pertuzumab plus trastuzumab and chemotherapy for HER2-positive metastatic gastric or gastro-oesophageal junction cancer (JACOB): final analysis of a double-blind, randomised, placebo-controlled phase 3 study. The Lancet Oncology. 2018;19(10):1372-84.
40. Tabernero J, Hoff PM, Shen L, Ohtsu A, Shah MA, Siddiqui A, et al. 1423MO End-of-study analysis from JACOB: A phase III study of pertuzumab (P) + trastuzumab (H) and chemotherapy (CT) in HER2-positive metastatic gastric or gastro-esophageal junction cancer (mGC/GEJC). Annals of Oncology. 2020;31:S900-S1.
41. Kowalczyk L, Bartsch R, Singer CF, Farr A. Adverse Events of Trastuzumab Emtansine (T-DM1) in the Treatment of HER2-Positive Breast Cancer Patients. Breast Care (Basel). 2017;12(6):401-8.
42. Schettini F, Conte B, Buono G, De Placido P, Parola S, Griguolo G, et al. T-DM1 versus pertuzumab, trastuzumab and a taxane as first-line therapy of early-relapsed HER2-positive metastatic breast cancer: an Italian multicenter observational study. ESMO Open. 2021;6(2):100099.
43. Locati LD, Cavalieri S, Bergamini C, Resteghini C, Colombo E, Calareso G, et al. Abiraterone

Acetate in Patients With Castration-Resistant, Androgen Receptor–Expressing Salivary Gland Cancer: A Phase II Trial. Journal of Clinical Oncology.0(0):JCO.21.00468.

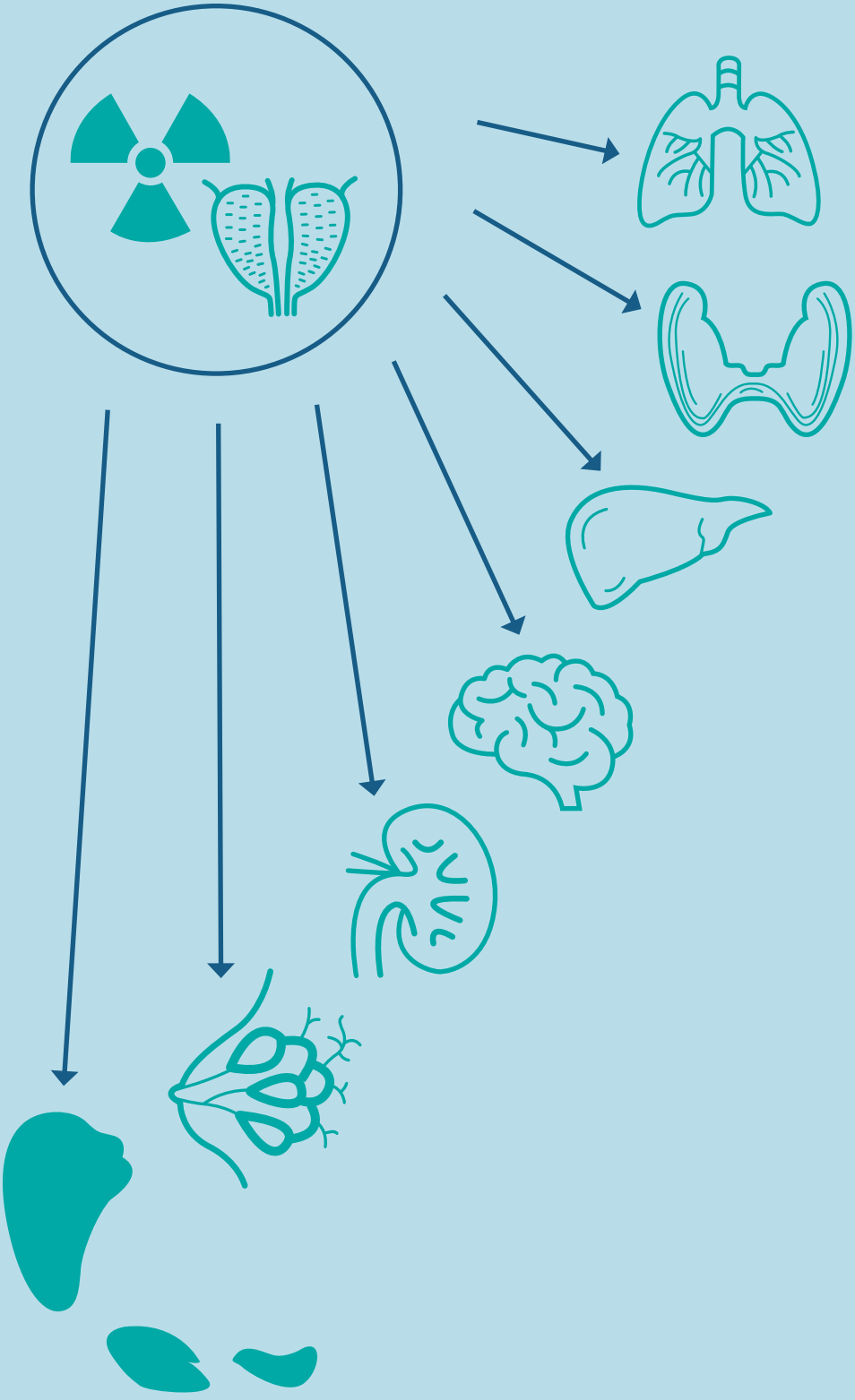
44. Ho AL. Androgen Receptor Pathway in Salivary Gland Cancer. Journal of Clinical Oncology.0(0):JCO.21.01983.

45. J. Cortés SK, W. Chung, S. Im, Y.H. Park, R. Hegg, M.H. Kim, L. Tseng, V. Petry, C. Chung, H. Iwata, E. Hamilton, G. Curigliano, B. Xu, C. Lee, Y. Liu, J. Cathcart, E. Bako, S. Verma, S.A. Hurvitz. LBA1 - Trastuzumab deruxtecan (T-DXd) vs trastuzumab emtansine (T-DM1) in patients (Pts) with HER2+ metastatic breast cancer (mBC): Results of the randomized phase III DESTINY-Breast03 study. Annals of Oncology (2021) 32 (suppl_5): S1283-S1346. 10.1016/annonc/annonc741; 2021.

46. Perez J, Garrigós L, Gion M, Jänne PA, Shitara K, Siena S, et al. Trastuzumab deruxtecan in HER2-positive metastatic breast cancer and beyond. Expert Opin Biol Ther. 2021;21(7):811-24.

Part 2

PSMA radioligand therapy for Salivary Gland Cancer



Chapter 5

PSMA radioligand therapy for solid tumours: background, opportunities, challenges and first clinical reports.

Maike J.M. Uijen*, Yonne H.W. Derks*, Robin I.J. Merkx, Melline G.M. Schilham, Joey Roosen, Bastiaan M. Privé, Sanne A.M. van Lith, Carla M.L. van Herpen, Martin Gotthardt, Sandra Heskamp, Willemijn A.M. van Gemert§, James Nagarajah§

* These authors contributed equally
§ These authors contributed equally

European Journal of Nuclear Medicine and Molecular Imaging, 2021. ***48****: 4350–4368.*

ABSTRACT

In the past decade, a growing body of literature has reported promising results for prostate-specific membrane antigen (PSMA) targeted radionuclide imaging and therapy in prostate cancer. First clinical studies evaluating the efficacy of [^{177}Lu]Lu-PSMA radioligand therapy (PSMA-RLT) demonstrated favourable results in prostate cancer patients. [^{177}Lu]Lu-PSMA is generally well tolerated due to its limited side-effects. While PSMA is highly overexpressed in prostate cancer cells, varying degrees of PSMA expression have been reported in other malignancies as well, particularly in the tumour-associated neovasculature. Hence, it is anticipated that PSMA-RLT could be explored for other solid cancers.

Here, we describe the current knowledge of PSMA expression in other solid cancers and define a perspective towards broader clinical implementation of PSMA-RLT. This review focusses specifically on: salivary gland cancer, glioblastoma, thyroid cancer, renal cell carcinoma, hepatocellular carcinoma, lung cancer and breast cancer. An overview of the (pre)clinical data on PSMA immunohistochemistry and PSMA PET/CT imaging is provided and summarized. Furthermore, the first clinical reports of non-prostate cancer patients treated with PSMA-RLT are described.

INTRODUCTION

Prostate-specific membrane antigen (PSMA) is a transmembrane protein that is encoded by the *FOLH1* (folate hydrolase 1) gene and was first discovered in prostate cancer cells (1). Contrary to what the name suggests, PSMA is not selective to prostate cancer cells only (2, 3) but also expressed by neovascular endothelial cells of various cancers, including glioblastoma, kidney cancer, lung cancer and breast cancer (4-6). Preclinical data suggests that PSMA might be involved in cancer-related angiogenesis by degrading the extracellular matrix and participating in integrin signal transduction (7, 8).

To date, most clinical research on PSMA focusses on prostate cancer due to its exceptional high level of PSMA expression by tumour cells. Clinical studies evaluated the potential of PSMA imaging using radiolabelled PSMA antibodies (ProstaScint®, J591) and ligands (namely [^{68}Ga] Ga-PSMA-11 and [^{18}F]F-PSMA-1007), mainly by positron emission tomography (PET), revealing higher tumour detection rates and higher tumour-to-background ratios when compared to conventional imaging modalities (9-12). Subsequently, PSMA targeting antibodies (J591) or ligands (PSMA-617 or PSMA-I&T) were labelled with therapeutic radionuclides such as lutetium-177 (^{177}Lu) or actinium-225 (^{225}Ac) respectively (13-19). Driven by the favourable binding features and pharmacokinetics of ligands as compared to currently available antibodies (low bone marrow toxicity due to faster clearance), PSMA ligands are currently the main focus for PSMA therapy in prostate cancer patients (13, 19). Yet, comparative studies are still lacking. [^{177}Lu]Lu-PSMA-617 has demonstrated promising results in prostate cancer patients one prospective study, as well as several retrospective studies and compassionate-use programs world-wide (14-16). A phase II trial on Lu-PSMA in heavily pre-treated progressive prostate cancer showed efficacy, i.e. a PSA decline >50%, in 57% of patients and a progression free survival of 7.6 months (13). Moreover, a phase III registration trial (VISION) in advanced prostate cancer patients completed recruiting and final results are awaited end of 2021 (NCT03511664).

Since PSMA radioligand therapy (PSMA-RLT) demonstrated remarkable therapeutic efficacy in prostate cancer patients, the question arises whether PSMA-RLT could also achieve beneficial effects in other cancers expressing PSMA on the tumours cells themselves, or in the tumour-associated neovasculature.

The aim of this review is to assess which other solid cancers could potentially benefit from PSMA-RLT, based on PSMA expression levels and PSMA imaging data. Potential challenges and differences when compared to prostate cancer are discussed. Additionally, the results of the first clinical reports of PSMA-RLT in solid tumours other than prostate cancer are presented.

METHODS

Search strategy

The selection of cancer types for this review was based on a combination of PSMA expression analysis and electronic library searches. First, the *FOLH1* gene expression levels (this gene encodes PSMA) of all cancers included in the TCG PanCancer Atlas were obtained from cBioPortal (figure 1) (20, 21). Second, literature was searched by universal PubMed searches (see Supplementary 1) for the fifteen cancers with the highest PSMA expression level on the PanCancer Atlas. Solely cancers with a substantial (>20) amount of PubMed results were included in this review.

This resulted in the inclusion of: glioblastoma, thyroid cancer, renal cell carcinoma, hepatocellular carcinoma, lung cancer and breast cancer. Additionally, we included salivary gland cancer. Although this rare tumour entity is not included in the TCG PanCancer Atlas, several relevant PSMA-related studies were conducted for this cancer type.

For each type of cancer, the PubMed results were screened for papers or case reports which investigated PSMA expression levels through immunohistochemistry, PSMA imaging (e.g. PET/CT scans) or reports on PSMA-RLT. Both preclinical and clinical studies were included. The last search was performed on the 23rd of October 2020.

PSMA immunohistochemistry

All articles reporting on PSMA immunohistochemistry (IHC) of the above mentioned seven solid cancer types were included. No selection was made based on the type of antibody used for IHC staining, since there is no golden standard for PSMA IHC. Antibodies targeting the intracellular- and extracellular domain of PSMA were included. A distinction was made between IHC staining on the tumour cells or the neovasculature. For each tumour type, the percentage of tumours which are PSMA-positive on the IHC staining were described.

PSMA PET/CT imaging

Although, mRNA and PSMA IHC data provide relevant information on PSMA expression levels, in clinical practice, eligibility for PSMA-RLT in prostate cancer is based on *in vivo* tracer uptake revealed by PSMA PET/CT, semi-quantitatively expressed as standardized uptake values (SUV). According to the European Association of Nuclear Medicine (EANM) guideline based on the phase II trial on [^{177}Lu]Lu-PSMA-617, the required maximum SUV (SUVmax) at dominant sites of tumour involvement should be at least 1.5-fold higher than the baseline mean SUV (SUVmean) of the liver on PET/CT (using renally excreted ligands such as [^{68}Ga]Ga-PSMA-11) to qualify for therapy (13, 22). Therefore, we looked at the tumour/liver ratio, if this was not reported, we used a SUVmean of 4-8 for liver as reported the in literature (23, 24). This suggests that a tumour uptake (SUVmax) of at least 12 might be considered sufficient to explore PSMA-RLT.

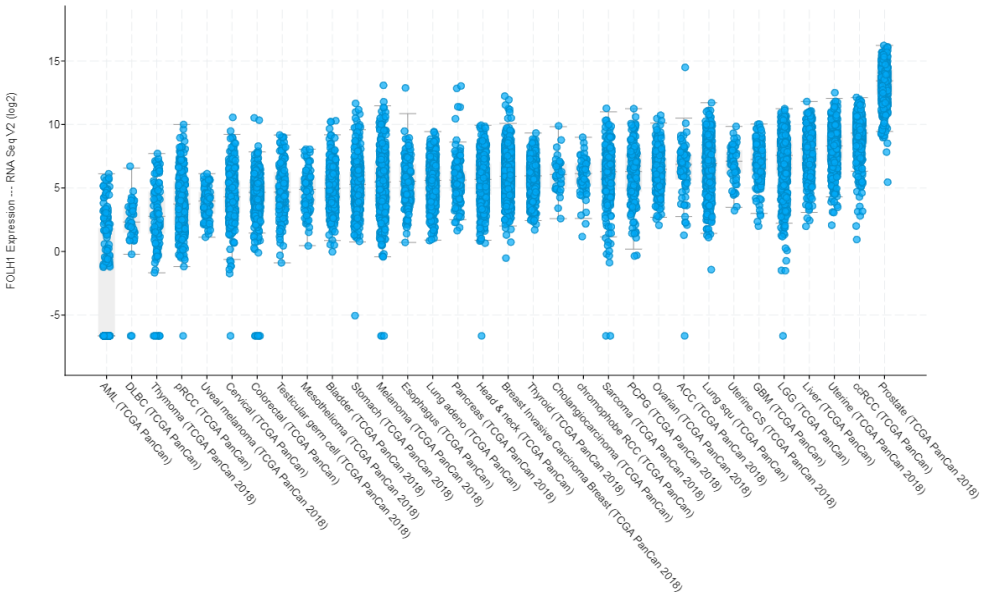


Figure 1: *FOLH1* expression levels of cancers included in TCGA PAN-CAN Atlas studies
This figure is was adapted from cBioportal.org.

Cancers are sorted based on median. Negative values are the result of the log(2)scale, where expression of 0 up to 1 in log(2) scaling results in negative values.
Overall expression of mRNA in other cancers is considerably lower (log scale) than in prostate cancer. All cancers show a large variation in *FOLH1* expression levels.

Abbreviations: ACC: adrenocortical carcinoma, AML: Acute Myloid Leukemia, DLBC: Diffuse Large B-Cell Lymphoma, pRCC: papillary renal cell carcinoma, RCC: renal cell carcinoma, PCPG: Pheochromocytoma and Paraganglioma, Uterine CS: Uterine Carcinosarcoma, GBM: Glioblastoma Multiforme, LGG: Lower Grade Glioma, ccRCC: clear cell renal cell carcinoma.

RESULTS

Salivary gland cancer

Salivary gland cancer (SGC) is a rare and complex disease with an annual incidence of 2 per 100.000, consisting of 22 subtypes each with different clinical behaviour and prognosis (25, 26). PSMA related research has solely been conducted for adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC).
Healthy salivary glands show high physiological tracer uptake on PSMA PET scans (9). Interestingly, unlike prostate cancer cells, the uptake of PSMA ligands by the salivary glands does not seem to be completely mediated by PSMA; at least part of the uptake is aspecific (27, 28).

PSMA expression has been examined using IHC for both ACC and SDC in primary tumour material as well as metastases (details can be found in table 1). The majority of ACC express

PSMA on the tumour cells (91% - 145/159 patients), while none of the tumours showed PSMA expression in the vasculature. In contrast, in SDC the majority of the vessels (90% - 9/10), and some of the tumour cells express PSMA (40% - 4/10) (29-33).

PSMA tracer uptake in ACC, as visualized with PSMA PET/CT, was first described in several case reports. Some patients demonstrated high PSMA uptake (SUVmax: 23.3) in metastatic lesions, as compared to other patients who only showed low to modest tracer uptake in the tumour cells (SUVmax: 1.2) (31, 32, 34, 35). This variation in uptake was confirmed in larger studies (van Boxtel *et al.* also included SDC patients), also describing a large variation of PSMA tracer uptake between patients (figure 2) (29, 30). Even within a patient, a relatively large inter-metastatic variation in tracer uptake was detected (29). Van Boxtel *et al.* reported a tumour/parotid ratio, which was <1 in the vast majority of cases (29). Therefore, PSMA PET/CT imaging might be of limited value for detecting primary tumours or local recurrences, but could be useful for detecting lymph node- or distant metastases. Overall, SUVmax values ranged from 1.1 to 30.2 in ACC patients and from 0.3 to 25.9 in SDC patients. This suggests that PSMA-RLT might of interest for a subset of salivary gland cancer patients, since some patient showed lesions with a SUVmax >12.

Regarding clinical studies on PSMA-RLT (table 2), one patient with stage IV ACC received a single dose of [^{177}Lu]Lu-PSMA (7.5 GBq) (34). Treatment was well tolerated with no side-effects and some pain relief was reported. Whole-body [^{177}Lu]Lu-PSMA SPECT/CT imaging after therapy showed intense uptake in the metastases. A planned second cycle of [^{177}Lu]Lu-PSMA was cancelled due to malignancy induced hypercalcemia, and the patient deceased soon after. Another study stated that one ACC patient was undergoing [^{177}Lu]Lu-PSMA treatment, but details on the dose, toxicity and therapeutic effect were not reported (30). Currently, a prospective phase II pilot study of [^{177}Lu]Lu-PSMA-I&T for ACC and SDC patients is recruiting (NCT04291300) offering a maximum of four cycles containing 7.4 GBq, every 6 weeks.

Table 1: Summary of PSMA expression and PSMA PET/CT imaging of seven different solid tumours

Cancer type	Subtype	PSMA expression tumour cells IHC	PSMA expression vasculature IHC	PSMA PET imaging	Proportion of patients possibly eligible for future PSMA-RLT studies
Salivary gland cancer	Adenoid cystic carcinoma	*Primary tumour*[29-33] N=135 PSMA+: 93% (125/135) Positive cells: range: <1%-90% *Metastases*[29, 30, 33] N=24 PSMA+: 83% (20/24) Positive cells: range: 5%-100%	*Primary tumour*[29] N=14 PSMA+: 0% (0/14) *Metastases*[29] N=9 PSMA+: 0% (0/9)	*Primary/recurrent tumour*[29-31] N=8 PET tracer uptake: 100% (8/8) SUVmax: range: 1.1 to 30.2* *Metastases*[29, 30, 32, 34, 35] N=26 PET tracer uptake: 100% (26/26) SUVmax: range: 1.1 to 30.2*	93% of adenoid cystic carcinoma patients; tumour/liver-ratio >1 in 13/14 patients.[29] Percentage of patients with tumour/liver-ratio >1.5 not reported.
	Salivary duct carcinoma	*Primary tumour*[29] N=9 PSMA+: 44% (4/9) Positive cells: range: <1%-50% *Metastases*[29] N=1 PSMA+: 0% (0/1)	*Primary tumour*[29] N=9 PSMA+: 89% (8/9) *Metastases*[29] N=1 PSMA+: 100% (1/1)	*Primary/recurrent tumour*[29] N=3 PET tracer uptake: 100% (3/3) SUVmax: range: 0.3 to 25.9* *Metastases*[29] N=9 PET tracer uptake: 100% (3/3) SUVmax: range: 0.3 to 25.9*	40% of salivary duct carcinoma patients; tumour/liver-ratio >1 in 4/10 patients.[29] Percentage of patients with tumour/liver-ratio >1.5 not reported.
Glioblastoma	-	*Primary/recurrent tumour*[38-40] N=8 PSMA+: 0% (0/8)	*Primary/recurrent tumour*[38-44] N=128 PSMA+: 72% (92/128)	*Primary/recurrent tumour*[40, 42, 45-51, 53-55] N=46 PET tracer uptake: 100% (46/46) SUVmax: range: 2.1 to 24.6	13% of glioblastoma patients tumour/liver ratio >1.5 in 2/15 patients.[54]

Table 1: Continued

Thyroid cancer	Differentiated thyroid cancer	*Primary tumour*[58, 59] N=209 PSMA+: 0% (0/209) *Metastases(58)* N=9 PSMA+: 0% (0/9)	*Primary tumour*[58-60, 62] N=258 PSMA+: 74% (192/258) *Metastases*[58] N=9 PSMA+: 100% (9/9)	*Primary/recurrent tumour*[63, 64, 66, 74, 77, 108] N=9 PET tracer uptake: 100% (9/9) SUVmax: range: 1.4 to 13.7 *Metastases*[63, 65, 67, 68, 73-75, 77] N=29 PET tracer uptake: 100% (29/29) SUVmax: range: 0.9 to 101.8	Especially in metastatic disease, high tracer uptake has been reported.[75] Some patients might be eligible for PSMA-RLT.
	Anaplastic thyroid cancer	*Primary tumour*[58, 59] N=15 PSMA+: 0% (0/15)	*Primary tumour*[58-60] N=19 PSMA+: 63% (12/19)	*Primary/recurrent tumour*[72, 73] N=2 PET tracer uptake: 100% (2/2) SUVmax: 6.0‡ *Metastases*[72] N= 1 PET tracer uptake: + SUVmax: NR	Insufficient data.
	Medullary thyroid cancer	*Primary tumour*[59] N=10 PSMA+: 0% (0/10)	*Primary tumour*[59-61] N=126 PSMA+: 83% (104/126)	*Primary/recurrent tumour*[69, 71] N=2 PET tracer uptake: 100% (2/2) SUVmax‡: 4.5 *Metastases*[70] N=1 PET tracer uptake: + SUVmax: 19.7	Insufficient data. Imaging data of one metastatic patient indicate sufficient tracer uptake of metastases.

Renal cell carcinoma	Clear cell	*Primary tumour*[38, 132] N=12 PSMA+: 0% (0/12)	*Primary tumour*[38, 81-83, 132, 133] N=299 PSMA+: 79% (236/299) *Metastases*[134] N=20 PSMA+: 75% (15/20)	*Primary/recurrent tumour*[85, 87, 88, 132, 135-140] N=28 PET tracer uptake: 96% (27/28) SUVmax: range: 1.7 to 39.4 *Metastases*[85-89, 138-145] N=36 PET tracer uptake: 89% (32/36) SUVmax: range: 0.9 to 48	In metastatic patients high tracer uptake has been reported. Some patients might be eligible for PSMA-RLT.
	Papillary	-	*Primary tumour*[81-83, 133] N=59 PSMA+: 27% (16/59)	*Primary/recurrent tumour*[85, 87, 89, 137] N=4 PET tracer uptake: 50% (2/4) SUVmax: range: 3.6 to 5.1 *Metastases*[84] N=3 PET tracer uptake: 67% (2/3) SUVmax: range: 1.8 to 4.1	Available data shows relatively low tracer uptake.
	NS†	-	-	*Primary/recurrent tumour*[85, 137] N=7 PET tracer uptake: 71% (5/7) SUVmax‡: 18.3 *Metastases*[84, 146] N=3 PET tracer uptake: 67% (2/3) SUVmax: range: 0.5 to 6.2	Available data shows relatively low tracer uptake in metastatic patients.

Table 1: Continued

Hepatocellular carcinoma	-	*Primary tumour*[91-94] N=282 PSMA+: 24% (69/282) Positive cells: NR	*Primary tumour*[91-94] N=282 PSMA+: 83% (235/282)	*Primary/recurrent tumour*[92-102] N=117 PET tracer uptake: 96% (112/117) SUVmax: range: 3.7 to 55.4 *Metastases*[94, 95, 99, 102] N=16 PET tracer uptake: 100% (16/16) SUVmax: 2.2-21.3	100% of hepatocellular carcinoma patients; tumour/liver ratio >1.5 in 15/15 patients.[94]
Lung cancer	NSCLC – Adenocarcinoma	*Primary tumour*[104, 105] N=141 PSMA+: 15% (21/141) Positive cells: NR	*Primary tumour*[104, 105] N=141 PSMA+: 45% (63/141)	*Primary tumour*[106, 107, 111] N=3 PET tracer uptake: 100% (3/3) SUVmax: range: 4.8 to 5.6	Available PSMA imaging data indicates relatively low tracer uptake.
	NSCLC – Squamous cell carcinoma	*Primary tumour*[104, 105] N=151 PSMA+: 19% (29/151) Positive cells: NR	*Primary tumour*[104, 105] N=151 PSMA+: 64% (97/151)	-	Insufficient data
	NSCLC – Large cell carcinoma	*Primary tumour*[104, 105] N=70 PSMA+: 20% (14/70) Positive cells: NR	*Primary tumour*[104, 105] N=70 PSMA+: 70% (49/70)	-	Insufficient data
	NSCLC – NS†	*Primary tumour*[38] N=5 PSMA+: 0% (0/5)	*Primary tumour*[4, 38, 109] N=13 PSMA+: 100% (13/13)	*Primary tumour*[108, 109] N=9 PET tracer uptake: 100% (9/9) SUVmax: range: 3.7-7.0 *Metastases*[110] N=1 PET tracer uptake: yes SUVmax: 4.4	Available PSMA imaging data indicates relatively low tracer uptake.

Lung cancer (continued)	Small-cell lung cancer	*Primary tumour*[104] N=30 PSMA+: 0% (0/30)	*Primary tumour*[104] N=30 PSMA+: 70% (21/30)	-	Insufficient data
Breast cancer	Invasive carcinoma of no special type	*Primary tumour*[38, 114] N=56 PSMA+: 46% (26/56) Positive cells: NR	*Primary tumour*[38, 114, 118, 133] N=312 PSMA+: 67% (209/312)	*Primary/recurrent tumour*[147, 148] N=2 PET tracer uptake: 100% (2/2) SUVmax: range: 3.2 to 9.7	Insufficient data. Available PSMA imaging data indicates relatively low tracer uptake.
	Invasive lobular carcinoma	*Primary tumour*[38] N=1 PSMA+: -	*Primary tumour*[38, 118] N=65 PSMA+: 42% (27/65)	-	Insufficient data.
	NS†	*Primary tumour*[114] N=17 PSMA+: 29% (5/17) Positive cells: NR *Metastases*[114] N=12 PSMA+: 75% (9/12) Positive cells: NR	*Primary tumour*[4, 114, 149] N=110 PSMA+: 70% (77/110) *Metastases*[114, 149] N=23 PSMA+: 96% (22/23)	*Primary/recurrent tumour*[116, 150] N=14 PET tracer uptake: 57% (8/14) SUVmax: range: NR Mean SUVmax¶: 2.45 *Metastases*[116, 130, 131, 150-152] N=19 PET tracer uptake: 89% (17/19) SUVmax: range: NR Mean SUVmax¶: 6.86	Available PSMA imaging data indicates relatively low tracer uptake.

Abbreviations: IHC: immunohistochemistry, N: number of patients, NR: not reported, NS: not specified, NSCLC: non-small-cell lung cancer, PSMA: prostate specific membrane antigen, RLT: radioligand therapy, SUVmax: maximum standardized uptake value,

*This study did not report separate SUVmax ranges for local recurrences or distant metastases. In ACC patients SUVmax ranged from 1.1 to 30.2 and in SDC patients SUVmax ranged from 0.3 to 25.9.

†: some studies did not further specify the histology or outcomes were not separately reported for each histology.

‡: some studies did not report SUVmax values, therefore only reported SUVmax values are presented in this table, but no range could be presented.

¶: because SUVmax range could not be reported, the mean SUVmax of the study of *Sathekge et al.* was reported as an alternative indication of SUVmax values.

Glioblastoma

Glioblastoma is the most frequently occurring type of brain cancer, with an annual incidence of 5 per 100.000 and is highly aggressive (36). Glioblastomas are known to be highly vascularized tumours (37).

The first reports on immunohistochemical staining in glioblastoma tumours observed PSMA expression only in the neovasculature and not in the tumour cells (38-40). Therefore, subsequent IHC studies primarily focused on the PSMA expression of the neovasculature (41-44). Overall, 72% (92/128) of the glioblastoma tumours express PSMA in the neovasculature (table 1). Two reports also quantified the vasculature staining by scoring the percentage of PSMA-positive vessels and staining intensity (41, 44). Wernicke *et al.* (41) reported that in 69% of the tumours >50% of the vessels were PSMA-positive, while this was only the case in 32% of the tumours in Mahzouni *et al.* (44).

PSMA ligand uptake by glioblastoma tumours has been observed with different diagnostic radiotracers (40, 42, 45-51). Bertagna *et al.* previously published a systematic review with a focus on the possible diagnostic role of PSMA PET/CT imaging, including most of these studies (52). They concluded that glioblastomas are PSMA-avid tumours and that PSMA PET/CT imaging could be a useful diagnostic tool in glioblastoma. Articles published since then are in line with these conclusions (49-51, 53-55). Regarding the diagnostic value, a major advantage of PSMA PET/CT imaging over [^{18}F]FDG PET/CT imaging is the lower background uptake, since normal brain parenchyma shows physiological [^{18}F]FDG uptake but no physiological PSMA uptake. In glioblastoma, [^{18}F]FLT PET/CT is regularly performed, but no studies comparing this tracer with PSMA PET/CT are known. Overall, SUVmax in glioblastoma ranged between 2.1 and 24.6. Kunikowksa *et al.* reported tumour/liver ratios after [^{68}Ga]Ga-PSMA-11 PET (54); 40% (6/15 patients) of the glioblastoma patients showed a tumour/liver ratio >1, and 13% (2/15) had a tumour/liver ratio >1.5. This suggests that at least part of the glioblastoma patients might have sufficient uptake to be considered for PSMA-RLT.

Kunikowska *et al.* published the first case report of PSMA-RLT in a glioblastoma patient (table 2) (53). This patient had a glioblastoma recurrence after prior treatments of surgery and chemo-radiotherapy. On [^{68}Ga]Ga-PSMA-11 PET the patient had a SUVmax of 10.3 with homogenous tumour PSMA uptake. The patient received a single dose of 8.4 GBq [^{177}Lu]Lu-PSMA. Although the report did not mention clinical outcome, intra-therapeutic serial SPECT imaging showed tracer accumulation in the tumour over time, with a calculated absorbed radiation dose of 14 Gy within the tumour. Recently, Kumar *et al.* reported about a patient who received PSMA-RLT which resulted in tumour shrinkage. This patient was pre-treated with surgery, radiotherapy and temozolomide before receiving 3 cycles of 3.7 GBq (every 8 weeks) of [^{177}Lu]Lu-PSMA-617. Post-therapy MRI showed a partial response with a tumour shrinkage (from 18 to 5.4 mL) and importantly improvement of quality of life (55).

Thyroid cancer

Thyroid cancer is an endocrine malignancy with an annual incidence of 2-6 per 100.000 (56). The most common subtype is differentiated thyroid carcinoma (DTC), which includes papillary thyroid carcinoma (PTC) and follicular thyroid carcinoma (FTC) (57). Other rare subtypes of thyroid cancer are medullary thyroid carcinoma (MTC) and anaplastic thyroid carcinoma (ATC) that have a dismal prognosis.

In thyroid cancer, the available literature did not report any PSMA expression on tumour cells itself in any of the subtypes (58, 59). Immunohistochemical PSMA expression on the neovasculature has been examined for all thyroid carcinoma subtypes, details are shown in table 1 (58-62). Overall, PSMA expression in the neovasculature was observed in PTC (61% - 134/220 patients), FTC (56% - 43/77), MTC (83% - 104/126), and ATC (63% - 12/19). Of the PTC and FTC tumours that became dedifferentiated (so called radio-iodine (RAI)-refractory), neovascular PSMA expression was reported in 63% of tumours (15/24) (59). Interestingly, Sollini *et al.* found that PSMA expression levels in DTC patients contributed to the prediction of tumour aggressiveness and patient outcome (62).

PSMA tracer uptake on PET/CT imaging of thyroid cancer has been described in several case reports and subsequent larger prospective studies (figure 2) (63-75). Overall, in DTC patients PSMA tracer uptake seemed to differ between primary/recurrent lesions and metastatic lesions. SUVmax of primary/recurrent tumours ranged between 1.4 and 13.7, whereas in metastatic lesion the SUVmax range was 0.9 to 101.8. Therefore, especially metastatic DTC patients might have sufficient tracer uptake to be eligible for PSMA-RLT. PSMA uptake in ATC and MTC patients was only described in few patients, with relatively low SUVmax values (primary tumour uptake 4.5-6) (69-73). Of these, Arora *et al.* reported a tumour/liver ratio >2 in some MTC patients, indicating the possible eligibility of these patients for PSMA-RLT (69).

Regarding PSMA-RLT, literature reports on three treated thyroid cancer patients (table 2) (76, 77). Assadi *et al.* treated a progressing metastatic RAI-refractory DTC patient with [^{177}Lu]Lu-PSMA (76). The patient previously received RAI therapy, sorafenib therapy for 6 months and radioligand therapy targeting the somatostatin receptor using [^{177}Lu]Lu-DOTATATE (1 cycle, 7.4 GBq). Thereafter, 1 cycle of 7.4 GBq [^{177}Lu]Lu-PSMA was given. Post-therapy whole-body SPECT imaging revealed higher uptake of [^{177}Lu]Lu-PSMA compared with whole-body SPECT imaging following [^{177}Lu]Lu-DOTATE treatment, PSMA-RLT therapy is therefore more likely to be effective in this patient. Two weeks after [^{177}Lu]Lu-PSMA therapy the patient deceased unexpectedly due to cardiac arrest. In the study of De Vries *et al.,* five patients with RAI-refractory DTC underwent PSMA PET/CT to determine their eligibility for [^{177}Lu]Lu-PSMA therapy (77). Three patients were considered eligible for PSMA-RLT, of whom two were treated with 2 cycles of 6 GBq [^{177}Lu]Lu-PSMA-617. One of the patients did not respond to therapy and showed disease progression on [^{18}F]FDG PET/CT after one month. Interestingly, the second

patient did have a partial response of lung and liver metastases on imaging, and a transient decrease of the tumour marker thyroglobulin from 17 to 9 µg/L. Seven months post treatment disease progression was observed on imaging and the thyroglobulin level increased to 14 µg/L. Both of these DTC case reports did not report on side-effects of PSMA-RLT (76, 77).

Renal cell carcinoma

Renal cell carcinoma (RCC) has an incidence of 4.4 per 100.000 (78). Renal tumours are divergent and their clinical behaviour is highly dependent on the histological subtype (79). Clear cell RCC is the most common subtype and accounts for the majority of kidney cancer related deaths (80). Importantly, pro-angiogenic factors (VEGF, PDGF) are strongly upregulated in clear cell RCC, leading to high vascularized tumours. Other frequently occurring RCC subtypes include papillary RCC and chromophobe RCC.

Regarding the PSMA expression in RCC, most research has been conducted for clear cell RCC and papillary RCC (table 1). PSMA expression of primary renal neoplasms demonstrated an exclusive PSMA expression in the tumour-associated neovasculature (5). This holds true for clear cell RCC, papillary RCC, chromophobe RCC and oncocytoma. Clear cell RCC was found to have the highest percentage of PSMA-positive tumours and also the highest PSMA staining intensity. In contrast, transitional cell and angiomyolipoma showed no PSMA expression (81, 82). Overall, seventy-nine percent (236/299 patients) of the primary clear cell RCC tumours showed positive PSMA staining, in contrast to 27% (16/59) in primary papillary RCC. In addition, in metastatic clear cell RCC 75% (15/20) of the tumours showed PSMA expression in the neovasculature (81-83). Spatz *et al*. presented the largest cohort, with 257 RCC patients (including papillary, clear cell and chromophobe subtypes). Interestingly, this cohort related stronger PSMA expression patterns with high grade and advanced tumours and increased staining intensity was associated with poorer overall survival (81).

The role of PSMA PET/CT imaging in RCC has yet to be defined, but its potential has been investigated in multiple clinical studies. Due to the highest PSMA expression in clear cell RCC, this subtype has gained the most interest for clinical application. This consensus is reinforced by a recent report that showed inconsistent detection of non-clear cell RCC lesions (84). Several explorative studies showed heterogeneity of PSMA-uptake in clear cell RCC lesions; in primary/recurrent tumours SUVmax ranged from 1.7 to 39.4 and in metastatic lesions a range between 0.9 and 48 was reported (85-89). Since some of the SUVmax values described in literature are above 12, a part of the patients might be considered for PSMA-RLT. This is supported by Siva *et al*. who also mentioned that [^{177}Lu]Lu-PSMA treatment might be feasible in a part of the recurrent RCC patients based on the high PSMA tracer uptake (89).

To date, no RCC patients have been treated with PSMA-RLT according to literature.

Hepatocellular carcinoma

Hepatocellular carcinoma (HCC) is the most frequent primary liver cancer with an incidence of 10.1 cases per 100.000 person-years (90).

Immunohistochemical PSMA expression has been examined in 282 primary HCC tissue samples (table 1) (91-94). Overall, PSMA expression was mostly observed in the tumour associated neovasculature (83% - 235/282), and it was associated with poor prognosis in patients with HCC (91). Only one of the studies also identified some PSMA expression by the tumour parenchyma (41% of samples), in a canalicular pattern (93).

PSMA PET/CT imaging of HCC tumours has been investigated in multiple case reports and three clinical imaging studies (figure 2) (92, 93, 95-102). In the study by Kelser *et al.* a correlation was described between the SUVmax on PSMA PET/CT and the hyper-vascularization status of tumours on CT, underlining the hypothesis that PSMA is mainly present on the neo-vascularization (92). Overall, SUVmax of primary lesions ranged between 3.7 and 55.4. Reports on PSMA uptake in metastatic HCC lesions are scarce, a total of 16 lesions were reported (92, 95, 99, 102). Metastatic lesions were all PSMA-positive on PSMA PET/CT. Almost all patients had a tumour-to-liver ratio >1.5, many even had a ratio >2 (92, 94, 95). Therefore, this stimulates further research of PSMA-RLT in HCC patients. Currently, a prospective trial of [^{68}Ga]Ga-PSMA-11 PET/CT imaging in HCC patients is recruiting (NCT04310540). In this study, one of the secondary research goals is to establish PSMA as a theranostic target for [^{177}Lu]Lu-PSMA-RLT.

Regarding PSMA-RLT (table 2), two HCC patients received PSMA-RLT (102). Both patients were treated with one cycle of [^{177}Lu]Lu-PSMA-617 (activity 5.9-6.2 GBq). Although the treatment was well tolerated, intra-therapeutic SPECT/CT based dosimetry revealed disappointing radiation dosages. According to the authors, the PSMA-RLT dose was at least ten-fold lower than typically achieved by one cycle of external beam radiation therapy for HCC. Therefore PSMA-RLT was discontinued after one cycle for both patients.

Lung cancer

The incidence of lung cancers varies largely between countries and differs between sexes. It ranges from <10 to >50 per 100.000 person-years (103). Lung cancer is generally divided into small cell lung cancer (SCLC) or non-small cell lung cancer (NSCLC). NSCLC can be further classified into adenocarcinoma (most common form), squamous cell carcinoma and large cell carcinoma (103).

In lung cancer, presence of PSMA is mainly observed on the neovasculature (table 1), with expression levels in primary tumours ranging from 45% (63/141 patients) in adenocarcinoma to 70% in large cell carcinoma (49/70) and SCLC (21/30) (104, 105). Positive staining of the

tumour cells was shown by Wang *et al*. in all three subtypes of NSCLC, in approximately half of the cases (104). However, this was not observed by Schmidt *et al.* where only a small fraction of the NSCLC cases of were PSMA-positive on the tumour cells (2-12%) (105). PSMA expression on SCLC tumour cells has only been studied by Wang *et al*., who showed no PSMA expression (104).

All literature on PSMA tracer uptake by lung cancer derives from accidental findings of lung lesions on PSMA PET/CT scans in patients who received PSMA-imaging for their prostate cancer (106-111). The reported SUVmax of lung cancer ranged from 4.8 to 5.6 in lung adenocarcinoma, table 1. In 9 patients with NSCLC (without further details on subtype) SUVmax ranged from 3.7 to 7.0. However, these lung cancers are identified due to their PSMA tracer uptake, and most likely PSMA negative lung cancers have not been published. Therefore, it remains unclear which proportion of lung cancer patients show PSMA tracer uptake and to what extent (SUV values).

Case reports on PSMA-RLT for lung cancer patients were not identified.

Breast cancer

Breast cancer is one of the most prevailing types of cancer, with an incidence of 128.5 per 100.000 woman per year (112). The most common histopathological subtype, accounting for 75% of all breast cancers, is invasive carcinoma of no special type (IC-NST), formerly known as invasive ductal carcinoma (113). The second most common type is invasive lobular carcinoma (ILC) (5-10%).

Immunohistochemical PSMA expression has been examined for both primary IC-NST and ILC tumours (table 1). Overall, 67% (209/312 patients) of IC-NST tumours and 42% (27/65) of ILC tumours expressed PSMA in the neovasculature. Interestingly, Kasoha *et al.* also found weak to moderate PSMA expression on the tumour cells in 51% of IC-NST tumours, yet Chang *et al.* did not observe PSMA expression on the tumour cells in breast cancer (38, 114). In metastatic breast cancer, PSMA expression was described in two reports, 96% (22/23) of these samples were positive for PSMA in the neovasculature (114, 115). Remarkably, Wernicke *et al.* described that PSMA expression of all tumour metastases correlated with PSMA-expression intensity of the primary tumour. They also found that both estrogen and progesterone receptor-negative tumours were more likely to have higher PSMA-expression as compared to hormone receptor-positive tumours (115).

Reports on PSMA PET/CT imaging mainly consisted of case reports (figure 2), which provided limited data on SUVmax or tumour/liver ratios. One article reported on PSMA imaging in 19 breast cancer patients (68% with IC-NST) (116). The SUVmean was 2.5 ± 2.6 for primary or local recurrences (n=13) and 3.2 ± 1.8 for involved lymph nodes (n=15). Distant metastases

(*n*=53) showed a significantly higher SUVmean of 6.9 ±5.7 when compared to primary tumour/local recurrence (p=0.04) and lymph node metastases (p=0.011). SUVmean did not show a significant correlation with hormone receptor status, however PSMA uptake increased with tumour grading and was more often seen in IC-NST as compared to other histological subtypes. Based on the limited literature, the SUVmax values that have been reported in breast cancer are generally low (mean SUVmax: 2.5 - 6.9) suggesting limited potential for PSMA-RLT in breast cancer.

Interestingly, we found a preclinical PSMA related study in breast cancer. This study investigated the potential of PSMA-RLT in breast cancer, showing that [^{177}Lu]Lu-PSMA strongly impaired the vitality and angiogenic capacity of endothelial cells cultured in breast cancer conditioned medium (117).

Regarding the clinical application of PSMA-RLT (table 2), a 38-year-old female with an aggressive triple negative breast carcinoma, previously unresponsive to chemotherapy and bevacizumab, received [^{177}Lu]Lu-PSMA-RLT (2 cycles 7.5 GBq) based on intense tumour tracer uptake on PSMA imaging (SUV not reported). Post-therapy SPECT imaging showed uptake in the tumour lesions and the treatment was well tolerated. However, severe disease progression was seen after the second treatment cycle and treatment was terminated (118). No other case reports on PSMA-RLT in breast cancer were found.

Table 2: Clinical reports and studies on PSMA-RLT in seven different solid tumours

Cancer	Subtype	Author, year	Number of patients	Radioligand	Injected activity, number of cycles	Efficacy*	Dosimetry	Comments
Salivary gland cancer	Adenoid cystic carcinoma	Klein Nulent et al. 2017. (30)	N=1	[^{177}Lu]Lu-PSMA-617	NR	NR	NR	Article mentions that one patient was undergoing [^{177}Lu]Lu-PSMA treatment. But further details have not been reported.
	Adenoid cystic carcinoma	Simsek et al. 2019. (34)	N=1	[^{177}Lu]Lu-PSMA	7.5 GBq 1 cycle	Pain reduction	Scan after 24h, showed intense uptake of metastases	PSMA ligand not specified. Second dose was intended but cancelled to malignancy-induced hypercalcemia.
	Adenoid cystic carcinoma and Salivary duct carcinoma	Study protocol: recruiting	Intended: N=10	[^{177}Lu]Lu-PSMA-I&T	7.4 GBq 2-4 cycles	NA	Will be performed after 1h, 24h, 48h, 72h and 7d	Clinical study: NCT04291300. Recruiting.
Glioblastoma	-	Kunikowska et al. 2020. (53)	N=1	[^{177}Lu]Lu-PSMA-617	8.4 GBq 1 cycle	NR	Scans after 3h, 24h, 48h, 7d and 14 d. Calculated tumour absorbed dose: 14.07 Gy	There were no efficacy related outcomes reported.
	-	Kumar et al. 2020. (55)	N=1	[^{177}Lu]Lu-PSMA-617	3.7 GBq 3 cycles	- improvement performance status - symptom improvement - tumour reduction: from 17 mL to 5.4 mL	NR	

Thyroid cancer	Papillary thyroid carcinoma	De Vries et al. 2020. (77)	N=2	[^{177}Lu]Lu-PSMA-617	6 GBq 2 cycles	**Patient 1:** Partial temporary response of lung and liver metastases PFS: 7 months. **Patient 2:** No response	NR	Both patients were heavily pre-treated.
	Radioactive iodine-refractory differentiated thyroid carcinoma	Assadi et al. 2019 (76)	N=1	[^{177}Lu]Lu-PSMA	7.4 GBq 1 cycle	NR	NR	PSMA ligand not specified Patient deceased 2 weeks post therapy of sudden cardiac arrest.
Renal cell carcinoma	-	-	-	-	-	-	-	
Hepatocellular carcinoma	-	Hirmas et al. 2021 (102)	N=2	[^{177}Lu]Lu-PSMA-617	5.9-6.2 GBq 1 cycle	NR	Intra-therapeutic SPECT/CT based dosimetry revealed low tumour radiation dose	Treatment was discontinued in both patients after low radiation doses based on SPECT/CT dosimetry.
Lung cancer	-	-	-	-	-	-	-	
Breast cancer	Unknown, triple negative	Tolkach et al. 2018	N=1	[^{177}Lu]Lu-PSMA	7.5 GBq 2 cycles	No response.	NR	PSMA ligand not specified. Treatment was well tolerated, no side effects. Clinical follow-up showed severe progress after the second cycle, so no further cycles applied.

Abbreviations: NA: not applicable NR: not reported, h: hours, d: days, RLT: radioligand therapy, PFS: progression-free survival, PR: partial response

*Efficacy: this included any of the following: objective or subjective response, progression-free survival, overall survival, quality of life.

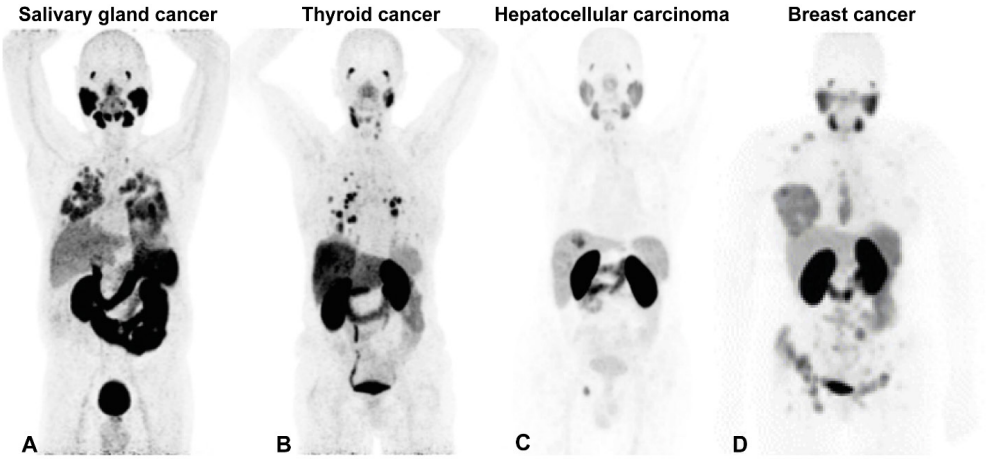


Figure 2: Four example PSMA PET/CT whole body images of patients with salivary gland cancer, thyroid cancer, hepatocellular carcinoma and breast cancer

(A) Patient with adenoid cystic carcinoma (salivary gland cancer) showing PSMA ligand uptake in lung metastases with a mean SUVmax of 10.0 and tumour-to-liver ratio of 2.5. **(B)** Patient with papillary thyroid carcinoma where PSMA PET/CT showed medium-high PSMA uptake in pulmonary metastases (median SUVmax 8.0). Additionally, new hotspots were seen on PSMA PET/CT (compared to [^{18}F]FDG PET) in the left cervical lymph nodes (SUVmax 3.33) and liver (SUVmax 7.2). **(C)** Patient with hepatocellular carcinoma showing focal uptake with an SUVmax of 17.6 and tumour-to-liver ratio of 4.0, as well as a tiny lesion in the cutting line with an SUVmax of 8.4. **(D)** Patient with breast cancer where PSMA PET/CT imaging demonstrated multiple osseous metastasis and a primary right breast cancer.

Patient A was originally published in van Boxtel et al., [68Ga]Ga-PSMA-HBED-CC PET/CT imaging for adenoid cystic carcinoma and salivary duct carcinoma: a phase 2 imaging study, Theranostics 2020, Ivyspring International Publisher© (29). Patient B was originally published in de Vries et al., [68Ga]Ga-PSMA PET/CT in radioactive iodine-refractory differentiated thyroid cancer and first treatment results with [177Lu]Lu-PSMA-617. EJNMMI Research 2020, Springer Nature© (77). Patient C was originally published in Kunikowska et al., [68Ga]Ga-Prostate-Specific Membrane Antigen PET/CT: a novel method for imaging patients with hepatocellular carcinoma. Eur J Nucl Med Mol Imaging, Copyright 2020, Spinger Nature© (94). Patient D was originally published in Sathekge et al., [68Ga]Ga-PSMA-HBED-CC PET imaging in breast carcinoma patients. Eur J Nucl Med Mol Imaging, 2017, Springer Nature© (116). These PSMA PET/CT images of four example patients were reprinted from open access articles distributed under the terms of the Creative Commons Attribution 4.0 International License (http://creativecommons.org/licenses/by/4.0/).

DISCUSSION

Increasing evidence shows that PSMA-RLT is an effective treatment for prostate cancer patients with a favourable toxicity profile (13, 119). Currently PSMA-RLT is investigated in a phase III trial (VISION; NCT03511664). These promising results in prostate cancer in combination with literature showing PSMA expression and PSMA tracer uptake in other malignancies, encouraged us to assess the potential role of PSMA-RLT for other solid cancer types (120). We focused on PSMA expression, PSMA PET/CT tracer uptake, and results of clinical attempts of PSMA-RLT in seven different solid cancers.

Regarding PSMA immunohistochemistry, in the majority of the solid cancers included in this review >70% of the primary tumours showed PSMA expression on the tumour associated neovasculature. Of all included primary tumours, medullary thyroid carcinomas and hepatocellular carcinomas most often expressed PSMA in the neovasculature. In contrast, in adenoid cystic carcinoma (a subtype of salivary gland cancer) and papillary renal cell carcinoma, only few of the tumours showed PSMA-positive staining on the neovasculature. Interestingly, although most of the solid cancers did not express PSMA on the tumour cells, it was still observed in salivary gland tumours (especially in adenoid cystic carcinoma), and to a minimal extent on hepatocellular-, lung- and breast cancer tissue.

On PSMA PET/CT imaging, PSMA tracer uptake differed considerably between the solid cancers. In patients with salivary gland cancer, glioblastoma, thyroid cancer, hepatocellular cancer and clear cell renal cell cancer several patients showed relevant tumour tracer accumulation on PET imaging (SUVmax values ≥12). Extremely high SUVmax values up to 101.8 were seen in metastatic medullary thyroid carcinoma. On the other hand, in several types of lung cancer and breast cancer, tracer uptake was low to moderate at best (SUVmax <10). Noteworthy, in solid cancers intra-patient tumour heterogeneity was observed.

PSMA-RLT eligibility in prostate cancer is assessed through PSMA PET/CT imaging, with an eligibility cut-off tumour/liver ratio >1.5 in [^{68}Ga]Ga-PSMA PET/CT according to the EANM guideline (22). As elaborated on in the methods section, for the aim of this review we considered a tumour SUVmax of >12 sufficient to potentially investigate PSMA-RLT. Bearing this in mind, we conclude that salivary gland cancer, glioblastoma, thyroid cancer (differentiated and medullary), hepatocellular carcinoma and renal cell cancer (clear cell) are the most relevant tumours to further explore the potential of PSMA-RLT. In line with this, the first case reports on PSMA-RLT in patients other than prostate cancer included salivary gland cancer, glioblastoma, thyroid cancer and hepatocellular carcinoma (30, 34, 53, 55, 76, 77, 102). These nine heavily pre-treated end-stage patients received 1-2 cycles of 5.9-8.4 GBq [^{177}Lu]Lu-PSMA per cycle in compassionate use programs (table 2). In some of these case reports positive treatment outcomes were reported. In one salivary gland cancer patient, pain reduction was

observed (34). One thyroid cancer patient showed a partial response that lasted 7 months (77). In a glioblastoma patient, tumour volume decreased upon PSMA-RLT (55). Importantly, the treatment was generally well tolerated, with no or low-grade adverse events.

Despite the use of the same PSMA PET/CT based eligibility criteria as in prostate cancer patients to assess possible PSMA-RLT application, there are essential differences between prostate cancer and the solid cancers included in this review.

First, in prostate cancer, PSMA is expressed on the tumour cells compared with the mainly neovascular expression in most of the other solid cancers. Still, even if PSMA is solely expressed on the neovasculature of well-perfused tumours, PSMA-RLT could hypothetically induce a tumoricidal local radiation dose to the tumour cells due to the tissue range (2 mm) of beta particles emitted by radionuclides such as ^{177}Lu [120]. Furthermore, radiation dosages to the neovasculature and tumour micro-environment might also lead to a harmful effect and induce secondary immune responses. It has been speculated that PSMA expression solely on the neovasculature could result in a shortened tracer washout (53, 94), meaning that PSMA-RLT is not retained in the tumour for a longer time, resulting in a lower radiation dose to the tumour and less effective treatment. Yet, a case report on a glioblastoma patient treated with [^{177}Lu]Lu-PSMA showed good tumour tracer retention on post-therapy imaging, resulting in a substantial tumour absorbed dose (53) and in another glioblastoma patient [^{177}Lu]Lu-PSMA treatment resulted in a decrease in tumour volume (55). This suggests that a sufficient radiation dose might still be reached while PSMA expression is limited to the tumour vasculature. Nonetheless, dedicated studies including dosimetry are required to prove this.

Second, SUVmax values in other solid cancers are generally lower than the SUVmax (>15-40) values in prostate cancer (121, 122). This suggests that lower radiation doses in the tumour could be reached, likely leading to a lower fraction of patients responding to PSMA-RLT as compared to prostate cancer patients.

Third, more intra-patient tumour heterogeneity in terms of PSMA expression is seen in other solid tumours when compared to prostate cancer (123). Supposedly, this is a result of the neovasculature versus tumour cell PSMA expression, as described above. To illustrate this, metastases with high neo-vascularization would have higher PSMA uptake than metastases with low neo-vascularization within a patient. However, even in patients with heterogenous PSMA tracer uptake, the bystander and abscopal effect are known in radiation-oncology, which might lead to tumour responses in PSMA negative tumours (124, 125).

Fourth, it has been long known that cancers vary in radiosensitivity (126). Prostate cancer is generally radiosensitive and external beam radiotherapy is effective in early-stage disease,

which provided a good rationale for PSMA-RLT in metastatic prostate cancer (127). In contrast, hepatocellular carcinoma, for example, is considered less radiosensitive (128, 129). Therefore, some of the other solid cancers might require higher PSMA-RLT radiation doses, as compared to prostate cancer, to achieve a clinically relevant response.

Based on this review, PSMA-RLT could potentially be investigated for certain solid cancers (e.g. salivary gland cancer, glioblastoma, thyroid cancer, liver cancer and clear cell renal cell cancer). This has also been proposed for these cancers by other authors (29, 30, 46, 54, 76, 89, 95). Sufficient PSMA uptake on PSMA PET/CT is a crucial parameter to consider therapy, and since a significant fraction of prostate cancer patients with high PSMA PET/CT tracer uptake does not respond to PSMA-RLT, other parameters, which are not clearly identified yet, obviously play also a relevant role. Hence, a good pre-selection of patients is crucial to apply this therapy to these patients in the future.

The current literature on PSMA uptake in cancers other than prostate cancer is scarce and prospective studies are rare. Therefore, it is not possible to draw firm, generalized conclusions. Furthermore, many of the papers included in this review are case reports. It is likely that patients with high PSMA uptake are reported, while negative results are less likely to be published, known as publication bias. Therefore, it may appear as if more tumours are PSMA avid or have higher SUV values than is actually the case. In addition, relevant data such as PSMA tracer uptake (SUVmax) was frequently not reported (40, 49, 50, 64, 67, 69, 107, 130, 131).

Future prospective

Currently, prospective PSMA PET/CT imaging studies in end-stage non-prostate cancer patients are lacking, consequently reliable information on PSMA uptake is missing. In our opinion, prospective imaging studies are the key way towards exploring PSMA-RLT for non-prostate cancers, especially imaging studies in patients with advanced disease, as PSMA-RLT is likely to be explored in end-stage disease with limited other treatment options. This will provide essential information on PSMA uptake and enables estimating which proportion of patients could be eligible for PSMA-RLT.

Furthermore, when evaluating the potential of PSMA-RLT in other cancers, preclinical studies on the therapeutic effects of PSMA-RLT are currently lacking and would be advisable. These could provide relevant insight into fundamental questions such as PSMA tracer retention in tumours where PSMA is limited to the neovasculature. Preclinical studies could also provide information on the sensitivity of non-prostate cancers to PSMA-RLT. In a clinical setting, prospective therapeutic studies should be performed instead of single patient reports to prevent trial-and-error-based science. As a different PSMA localization (tumour cell surface in prostate cancer versus neovasculature in other solid cancers) might lead to different PSMA tracer kinetics, preferably these prospective studies should include

multi-timepoint post-therapy imaging (dosimetry), to provide more information on PSMA tracer kinetics. Currently, a prospective therapeutic study in salivary gland cancer patients is recruiting (NCT04291300). Furthermore, pending the outcome of the pivotal trial for [^{177}Lu]Lu-PSMA (VISION trial; NCT03511664) in prostate cancer patients, it is anticipated that with positive results the translation to other solid cancers may be accelerated.

CONCLUSION

In summary, PSMA expression in solid cancers other than prostate cancer is primarily observed in the tumour neovasculature, except for adenoid cystic carcinoma (subtype salivary gland cancer), where PSMA is expressed on the tumour cells. Although there is heterogeneity in PSMA expression and tracer uptake, a subset of patients with advanced salivary gland cancer, glioblastoma, thyroid cancer, hepatocellular carcinoma and clear cell renal carcinoma show sufficient PSMA PET/CT tracer uptake in the tumour. These patients might potentially benefit from PSMA-RLT, so future research in this setting is encouraged. To date, ten patients with non-prostate solid cancers (salivary gland cancer, glioblastoma, thyroid cancer, hepatocellular carcinoma and breast cancer) have been treated with PSMA-RLT and some beneficial effects were seen, making this an interesting topic for further exploration.

SUPPLEMENTARY

Final universal PubMed search strategy (23-10-2020)†

	Search strategy PubMed
Pubmed	
#1	((((((((((((PSMA[tiab]) OR prostate specific membrane antigen[tiab]) OR FOLH1[tiab]) OR FOLH[tiab]) OR folate hydrolase[tiab]) OR "FOLH1 protein, human" [Supplementary Concept])) OR Glutamaat carboxypeptidase II[tiab]) OR GCPII[tiab]) OR GCP2[tiab]) OR NAAG peptidase[tiab]) OR NAALADase I[tiab]) OR NAALDase1[tiab])
#2	((***type of cancer***) AND (carcinoma*[tiab] OR cancer*[tiab] OR malignanc*[tiab] OR neoplasm*[tiab] OR tumor*[tiab] OR tumour*[tiab]))
#3	#1 AND #2

† In addition to the articles identified through the elaborate search strategy, we included a recent article (published after de date of the final search on 23-10-2020). This article showed important PSMA imaging in hepatocellular carcinoma and reports on 2 patients treated with PSMA-radioligand therapy.

REFERENCES

1. Evans JC, Malhotra M, Cryan JF, O'Driscoll CM. The therapeutic and diagnostic potential of the prostate specific membrane antigen/glutamate carboxypeptidase II (PSMA/GCPII) in cancer and neurological disease. Br J Pharmacol. 2016;173(21):3041-79.
2. Israeli RS, Powell CT, Fair WR, Heston WD. Molecular cloning of a complementary DNA encoding a prostate-specific membrane antigen. Cancer Res. 1993;53(2):227-30.
3. Horoszewicz JS, Kawinski E, Murphy GP. Monoclonal antibodies to a new antigenic marker in epithelial prostatic cells and serum of prostatic cancer patients. Anticancer Res. 1987;7(5b):927-35.
4. Liu H, Moy P, Kim S, Xia Y, Rajasekaran A, Navarro V, et al. Monoclonal antibodies to the extracellular domain of prostate-specific membrane antigen also react with tumor vascular endothelium. Cancer Res. 1997;57(17):3629-34.
5. Chang SS, O'Keefe DS, Bacich DJ, Reuter VE, Heston WD, Gaudin PB. Prostate-specific membrane antigen is produced in tumor-associated neovasculature. Clin Cancer Res. 1999;5(10):2674-81.
6. Silver DA, Pellicer I, Fair WR, Heston WD, Cordon-Cardo C. Prostate-specific membrane antigen expression in normal and malignant human tissues. Clinical cancer research : an official journal of the American Association for Cancer Research. 1997;3(1):81-5.
7. Conway RE, Petrovic N, Li Z, Heston W, Wu D, Shapiro LH. Prostate-specific membrane antigen regulates angiogenesis by modulating integrin signal transduction. Mol Cell Biol. 2006;26(14):5310-24.
8. Conway RE, Rojas C, Alt J, Novakova Z, Richardson SM, Rodrick TC, et al. Prostate-specific membrane antigen (PSMA)-mediated laminin proteolysis generates a pro-angiogenic peptide. Angiogenesis. 2016;19(4):487-500.
9. Afshar-Oromieh A, Malcher A, Eder M, Eisenhut M, Linhart HG, Hadaschik BA, et al. PET imaging with a [68Ga]gallium-labelled PSMA ligand for the diagnosis of prostate cancer: biodistribution in humans and first evaluation of tumour lesions. Eur J Nucl Med Mol Imaging. 2013;40(4):486-95.
10. Afshar-Oromieh A, Holland-Letz T, Giesel FL, Kratochwil C, Mier W, Haufe S, et al. Diagnostic performance of (68)Ga-PSMA-11 (HBED-CC) PET/CT in patients with recurrent prostate cancer: evaluation in 1007 patients. Eur J Nucl Med Mol Imaging. 2017;44(8):1258-68.
11. Hofman MS, Lawrentschuk N, Francis RJ, Tang C, Vela I, Thomas P, et al. Prostate-specific membrane antigen PET-CT in patients with high-risk prostate cancer before curative-intent surgery or radiotherapy (proPSMA): a prospective, randomised, multicentre study. Lancet. 2020;395(10231):1208-16.
12. Taneja SS. ProstaScint(R) Scan: Contemporary Use in Clinical Practice. Rev Urol. 2004;6 Suppl 10(Suppl 10):S19-28.
13. Hofman MS, Violet J, Hicks RJ, Ferdinandus J, Thang SP, Akhurst T, et al. [(177)Lu]-PSMA-617 radionuclide treatment in patients with metastatic castration-resistant prostate cancer (LuPSMA trial): a single-centre, single-arm, phase 2 study. Lancet Oncol. 2018;19(6):825-33.
14. Ahmadzadehfar H, Rahbar K, Kurpig S, Bogemann M, Claesener M, Eppard E, et al. Early side effects and first results of radioligand therapy with (177)Lu-DKFZ-617 PSMA of castrate-resistant metastatic prostate cancer: a two-centre study. EJNMMI Res. 2015;5(1):114.

15. Rahbar K, Ahmadzadehfar H, Kratochwil C, Haberkorn U, Schafers M, Essler M, et al. German Multicenter Study Investigating 177Lu-PSMA-617 Radioligand Therapy in Advanced Prostate Cancer Patients. J Nucl Med. 2017;58(1):85-90.
16. Kratochwil C, Giesel FL, Stefanova M, Benesova M, Bronzel M, Afshar-Oromieh A, et al. PSMA-Targeted Radionuclide Therapy of Metastatic Castration-Resistant Prostate Cancer with 177Lu-Labeled PSMA-617. J Nucl Med. 2016;57(8):1170-6.
17. Heck MM, Tauber R, Schwaiger S, Retz M, D'Alessandria C, Maurer T, et al. Treatment Outcome, Toxicity, and Predictive Factors for Radioligand Therapy with (177)Lu-PSMA-I&T in Metastatic Castration-resistant Prostate Cancer. Eur Urol. 2019;75(6):920-6.
18. Kratochwil C, Bruchertseifer F, Giesel FL, Weis M, Verburg FA, Mottaghy F, et al. 225Ac-PSMA-617 for PSMA-Targeted alpha-Radiation Therapy of Metastatic Castration-Resistant Prostate Cancer. J Nucl Med. 2016;57(12):1941-4.
19. Tagawa ST, Milowsky MI, Morris M, Vallabhajosula S, Christos P, Akhtar NH, et al. Phase II study of Lutetium-177-labeled anti-prostate-specific membrane antigen monoclonal antibody J591 for metastatic castration-resistant prostate cancer. Clin Cancer Res. 2013;19(18):5182-91.
20. Cerami E, Gao J, Dogrusoz U, Gross BE, Sumer SO, Aksoy BA, et al. The cBio Cancer Genomics Portal: An Open Platform for Exploring Multidimensional Cancer Genomics Data. Cancer Discovery. 2012;2(5):401-4.
21. Gao J, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, Sumer SO, et al. Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. Sci Signal. 2013;6(269):pl1.
22. Kratochwil C, Fendler WP, Eiber M, Baum R, Bozkurt MF, Czernin J, et al. EANM procedure guidelines for radionuclide therapy with 177Lu-labelled PSMA-ligands (177Lu-PSMA-RLT). European Journal of Nuclear Medicine and Molecular Imaging. 2019;46(12):2536-44.
23. Jansen BHE, Kramer GM, Cysouw MCF, Yaqub MM, de Keizer B, Lavalaye J, et al. Healthy Tissue Uptake of (68)Ga-Prostate-Specific Membrane Antigen, (18)F-DCFPyL, (18)F-Fluoromethylcholine, and (18)F-Dihydrotestosterone. J Nucl Med. 2019;60(8):1111-7.
24. Demirci E, Sahin OE, Ocak M, Akovali B, Nematyazar J, Kabasakal L. Normal distribution pattern and physiological variants of 68Ga-PSMA-11 PET/CT imaging. Nucl Med Commun. 2016;37(11):1169-79.
25. El-Naggar AK CJ GJ, Takata T, Slootweg PJ, eds. WHO classification of head and neck tumours, 4th edn. World Health Organization (IARC), 2017.
26. Boukheris H, Curtis RE, Land CE, Dores GM. Incidence of carcinoma of the major salivary glands according to the WHO classification, 1992 to 2006: a population-based study in the United States. Cancer Epidemiol Biomarkers Prev. 2009;18(11):2899-906.
27. Rupp NJ, Umbricht CA, Pizzuto DA, Lenggenhager D, Topfer A, Muller J, et al. First Clinicopathologic Evidence of a Non-PSMA-Related Uptake Mechanism for (68)Ga-PSMA-11 in Salivary Glands. J Nucl Med. 2019;60(9):1270-6.
28. Tonnesmann R, Meyer PT, Eder M, Baranski AC. [(177)Lu]Lu-PSMA-617 Salivary Gland Uptake Characterized by Quantitative In Vitro Autoradiography. Pharmaceuticals (Basel). 2019;12(1).
29. van Boxtel W, Lutje S, van Engen-van Grunsven ICH, Verhaegh GW, Schalken JA, Jonker MA, et al. (68)Ga-PSMA-HBED-CC PET/CT imaging for adenoid cystic carcinoma and salivary duct carcinoma: a phase 2 imaging study. Theranostics. 2020;10(5):2273-83.

30. Klein Nulent TJW, van Es RJJ, Krijger GC, de Bree R, Willems SM, de Keizer B. Prostate-specific membrane antigen PET imaging and immunohistochemistry in adenoid cystic carcinoma-a preliminary analysis. Eur J Nucl Med Mol Imaging. 2017;44(10):1614-21.
31. Konig L, Hauswald H, Flechtenmacher C, Heller M, Debus J, Haberkorn U, et al. Uptake of Prostate-Specific Membrane Antigen (PSMA) in adenoid cystic carcinoma - Is PSMA-PET-CT a helpful tool in radiation oncology? Clin Transl Radiat Oncol. 2017;7:79-82.
32. de Keizer B, Krijger GC, Ververs FT, van Es RJJ, de Bree R, Willems S. (68)Ga-PSMA PET-CT Imaging of Metastatic Adenoid Cystic Carcinoma. Nucl Med Mol Imaging. 2017;51(4):360-1.
33. Klein Nulent TJW, Valstar MH, Smit LA, Smeele LE, Zuithoff NPA, de Keizer B, et al. Prostate-specific membrane antigen (PSMA) expression in adenoid cystic carcinoma of the head and neck. BMC Cancer. 2020;20(1):519.
34. Has Simsek D, Kuyumcu S, Agaoglu FY, Unal SN. Radionuclide Therapy With 177Lu-PSMA in a Case of Metastatic Adenoid Cystic Carcinoma of the Parotid. Clin Nucl Med. 2019;44(9):764-6.
35. Lutje S, Sauerwein W, Lauenstein T, Bockisch A, Poeppel TD. In Vivo Visualization of Prostate-Specific Membrane Antigen in Adenoid Cystic Carcinoma of the Salivary Gland. Clin Nucl Med. 2016;41(6):476-7.
36. Omuro A, DeAngelis LM. Glioblastoma and other malignant gliomas: a clinical review. Jama. 2013;310(17):1842-50.
37. Das S, Marsden PA. Angiogenesis in glioblastoma. N Engl J Med. 2013;369(16):1561-3.
38. Chang SS, Reuter VE, Heston WD, Bander NH, Grauer LS, Gaudin PB. Five different anti-prostate-specific membrane antigen (PSMA) antibodies confirm PSMA expression in tumor-associated neovasculature. Cancer Res. 1999;59(13):3192-8.
39. Nomura N, Pastorino S, Jiang P, Lambert G, Crawford JR, Gymnopoulos M, et al. Prostate specific membrane antigen (PSMA) expression in primary gliomas and breast cancer brain metastases. Cancer Cell Int. 2014;14(1):26.
40. Salas Fragomeni RA, Menke JR, Holdhoff M, Ferrigno C, Laterra JJ, Solnes LB, et al. Prostate-Specific Membrane Antigen-Targeted Imaging With [18F]DCFPyL in High-Grade Gliomas. Clin Nucl Med. 2017;42(10):e433-e5.
41. A Gabriella Wernicke, Mark A. Edgar, Ehud Lavi, He Liu, Pamela Salerno, Neil H. Bander, et al. Prostate-Specific Membrane Antigen as a Potential Novel Vascular Target for Treatment of Glioblastoma Multiforme. Archives of Pathology & Laboratory Medicine. 2011;135(11):1486-9.
42. Schwenck J, Tabatabai G, Skardelly M, Reischl G, Beschorner R, Pichler B, et al. In vivo visualization of prostate-specific membrane antigen in glioblastoma. Eur J Nucl Med Mol Imaging. 2015;42(1):170-1.
43. Saffar H, Noohi M, Tavangar SM, Saffar H, Azimi S. Expression of Prostate-Specific Membrane Antigen (PSMA) in Brain Glioma and its Correlation with Tumor Grade. Iran J Pathol. 2018;13(1):45-53.
44. Mahzouni P, Shavakhi M. Prostate-Specific Membrane Antigen Expression in Neovasculature of Glioblastoma Multiforme. Adv Biomed Res. 2019;8:18.
45. Sasikumar A, Joy A, Pillai MR, Nanabala R, Anees KM, Jayaprakash PG, et al. Diagnostic Value of 68Ga PSMA-11 PET/CT Imaging of Brain Tumors-Preliminary Analysis. Clin Nucl Med. 2017;42(1):e41-e8.
46. Kunikowska J, Bartosz K, Leszek K. Glioblastoma multiforme: another potential application for (68) Ga-PSMA PET/CT as a guide for targeted therapy. Eur J Nucl Med Mol Imaging. 2018;45(5):886-7.

47. Sasikumar A, Kashyap R, Joy A, Charan Patro K, Bhattacharya P, Reddy Pilaka VK, et al. Utility of 68Ga-PSMA-11 PET/CT in Imaging of Glioma-A Pilot Study. Clin Nucl Med. 2018;43(9):e304-e9.
48. Verma P, Malhotra G, Goel A, Rakshit S, Chandak A, Chedda R, et al. Differential Uptake of 68Ga-PSMA-HBED-CC (PSMA-11) in Low-Grade Versus High-Grade Gliomas in Treatment-Naive Patients. Clin Nucl Med. 2019;44(5):e318-e22.
49. Gupta M, Choudhury PS, Premsagar IC, Gairola M, Ahlawat P. Role of 68Ga-Prostate-Specific Membrane Antigen PET/CT in Disease Assessment in Glioblastoma Within 48 Hours of Surgery. Clin Nucl Med. 2020;45(3):204-5.
50. Marafi F, Sasikumar A, Fathallah W, Esmail A. 18F-PSMA 1007 Brain PET/CT Imaging in Glioma Recurrence. Clin Nucl Med. 2020;45(1):e61-e2.
51. Pilati E, Nicolotti DG, Ceci F, Finessi M, Cerio I, Dionisi B, et al. 68Ga-Prostate-Specific Membrane Antigen 11 PET/CT Detects Residual Glioblastoma After Radical Surgery in a Patient With Synchronous Recurrent Prostate Cancer: A Case Report. Clin Nucl Med. 2020;45(3):e151-e3.
52. Bertagna F, Albano D, Cerudelli E, Gazzilli M, Giubbini R, Treglia G. Potential of radiolabelled PSMA PET/CT or PET/MRI diagnostic procedures in gliomas/glioblastomas. Curr Radiopharm. 2019.
53. Kunikowska J, Charzynska I, Kulinski R, Pawlak D, Maurin M, Krolicki L. Tumor uptake in glioblastoma multiforme after IV injection of [(177)Lu]Lu-PSMA-617. Eur J Nucl Med Mol Imaging. 2020.
54. Kunikowska J, Kulinski R, Muylle K, Koziara H, Krolicki L. 68Ga-Prostate-Specific Membrane Antigen-11 PET/CT: A New Imaging Option for Recurrent Glioblastoma Multiforme? Clin Nucl Med. 2020;45(1):11-8.
55. Kumar A, Ballal S, Yadav MP, ArunRaj ST, Haresh KP, Gupta S, et al. 177Lu-/68Ga-PSMA Theranostics in Recurrent Glioblastoma Multiforme: Proof of Concept. Clin Nucl Med. 2020.
56. La Vecchia C, Malvezzi M, Bosetti C, Garavello W, Bertuccio P, Levi F, et al. Thyroid cancer mortality and incidence: a global overview. Int J Cancer. 2015;136(9):2187-95.
57. Cabanillas ME, McFadden DG, Durante C. Thyroid cancer. Lancet. 2016;388(10061):2783-95.
58. Moore M, Panjwani S, Mathew R, Crowley M, Liu YF, Aronova A, et al. Well-Differentiated Thyroid Cancer Neovasculature Expresses Prostate-Specific Membrane Antigen-a Possible Novel Therapeutic Target. Endocr Pathol. 2017;28(4):339-44.
59. Bychkov A, Vutrapongwatana U, Tepmongkol S, Keelawat S. PSMA expression by microvasculature of thyroid tumors - Potential implications for PSMA theranostics. Sci Rep. 2017;7(1):5202.
60. Heitkotter B, Steinestel K, Trautmann M, Grunewald I, Barth P, Gevensleben H, et al. Neovascular PSMA expression is a common feature in malignant neoplasms of the thyroid. Oncotarget. 2018;9(11):9867-74.
61. Lodewijk L, Willems SM, Dreijerink KMA, de Keizer B, van Diest PJ, Schepers A, et al. The theranostic target prostate-specific membrane antigen is expressed in medullary thyroid cancer. Hum Pathol. 2018;81:245-54.
62. Sollini M, di Tommaso L, Kirienko M, Piombo C, Erreni M, Lania AG, et al. PSMA expression level predicts differentiated thyroid cancer aggressiveness and patient outcome. EJNMMI Res. 2019;9(1):93.
63. Lengana T, Lawal IO, Mokoala K, Vorster M, Sathekge MM. 68Ga-PSMA: a One-stop Shop in Radioactive Iodine Refractory Thyroid Cancer? Nucl Med Mol Imaging. 2019;53(6):442-5.
64. Sager S, Vatankulu B, Uslu L, Sonmezoglu K. Incidental Detection of Follicular Thyroid

Carcinoma in 68Ga-PSMA PET/CT Imaging. J Nucl Med Technol. 2016;44(3):199-200.

65. Sasikumar A, Joy A, Pillai MRA, Oommen KE, Jayakumar R. Rare Case of Intratracheal Metastasis Detected on 68Ga-Prostate-Specific Membrane Antigen PET/CT Scan in a Case of Thyroglobulin Elevated Negative Iodine Scan Syndrome. Clin Nucl Med. 2018;43(4):282-3.

66. Singh D, Horneman R, Nagra NK. More than the prostate: Intrapancreatic accessory spleen and papillary thyroid cancer detected with (18) F-PSMA PET/CT. Hell J Nucl Med. 2018;21(2):145-7.

67. Taywade SK, Damle NA, Bal C. PSMA Expression in Papillary Thyroid Carcinoma: Opening a New Horizon in Management of Thyroid Cancer? Clin Nucl Med. 2016;41(5):e263-5.

68. Verburg FA, Krohn T, Heinzel A, Mottaghy FM, Behrendt FF. First evidence of PSMA expression in differentiated thyroid cancer using [(6)(8)Ga] PSMA-HBED-CC PET/CT. Eur J Nucl Med Mol Imaging. 2015;42(10):1622-3.

69. Arora S, Damle NA, Parida GK, Singhal A, Nalli H, Dattagupta S, et al. Recurrent Medullary Thyroid Carcinoma on 68Ga-Prostate-Specific Membrane Antigen PET/CT: Exploring New Theranostic Avenues. Clin Nucl Med. 2018;43(5):359-60.

70. Arora S, Prabhu M, Damle NA, Bal C, Kumar P, Nalla H, et al. Prostate-specific Membrane Antigen Imaging in Recurrent Medullary Thyroid Cancer: A New Theranostic Tracer in the Offing? Indian J Nucl Med. 2018;33(3):261-3.

71. Ciappuccini R, Edet-Sanson A, Saguet-Rysanek V, Gauthe M, Bardet S. Thyroid Incidentaloma on 18F-fluorocholine PET/CT and 68Ga-PSMA PET/CT Revealing a Medullary Thyroid Carcinoma. Clin Nucl Med. 2019;44(8):663-5.

72. Damle NA, Bal C, Singh TP, Gupta R, Reddy S, Kumar R, et al. Anaplastic thyroid carcinoma on 68 Ga-PSMA PET/CT: opening new frontiers. Eur J Nucl Med Mol Imaging. 2018;45(4):667-8.

73. Lawhn-Heath C, Yom SS, Liu C, Villanueva-Meyer JE, Aslam M, Smith R, et al. Gallium-68 prostate-specific membrane antigen ([(68)Ga] Ga-PSMA-11) PET for imaging of thyroid cancer: a feasibility study. EJNMMI Res. 2020;10(1):128.

74. Lutje S, Gomez B, Cohnen J, Umutlu L, Gotthardt M, Poeppel TD, et al. Imaging of Prostate-Specific Membrane Antigen Expression in Metastatic Differentiated Thyroid Cancer Using 68Ga-HBED-CC-PSMA PET/CT. Clin Nucl Med. 2017;42(1):20-5.

75. Verma P, Malhotra G, Agrawal R, Sonavane S, Meshram V, Asopa RV. Evidence of Prostate-Specific Membrane Antigen Expression in Metastatic Differentiated Thyroid Cancer Using 68Ga-PSMA-HBED-CC PET/CT. Clin Nucl Med. 2018;43(8):e265-e8.

76. Assadi M, Ahmadzadehfar H. (177)Lu-DOTATATE and (177)Lu-prostate-specific membrane antigen therapy in a patient with advanced metastatic radioiodine-refractory differentiated thyroid cancer after failure of tyrosine kinase inhibitors treatment. World J Nucl Med. 2019;18(4):406-8.

77. de Vries LH, Lodewijk L, Braat A, Krijger GC, Valk GD, Lam M, et al. (68)Ga-PSMA PET/CT in radioactive iodine-refractory differentiated thyroid cancer and first treatment results with (177)Lu-PSMA-617. EJNMMI Res. 2020;10(1):18.

78. Capitanio U, Bensalah K, Bex A, Boorjian SA, Bray F, Coleman J, et al. Epidemiology of Renal Cell Carcinoma. European urology. 2019;75(1):74-84.

79. Keegan KA, Schupp CW, Chamie K, Hellenthal NJ, Evans CP, Koppie TM. Histopathology of surgically treated renal cell carcinoma: survival differences by subtype and stage. J Urol. 2012;188(2):391-7.

80. Saad AM, Gad MM, Al-Husseini MJ, Ruhban IA, Sonbol MB, Ho TH. Trends in Renal-Cell Carcinoma Incidence and Mortality in the United

States in the Last 2 Decades: A SEER-Based Study. Clin Genitourin Cancer. 2019;17(1):46-57.e5.

81. Spatz S, Tolkach Y, Jung K, Stephan C, Busch J, Ralla B, et al. Comprehensive Evaluation of Prostate Specific Membrane Antigen Expression in the Vasculature of Renal Tumors: Implications for Imaging Studies and Prognostic Role. J Urol. 2018;199(2):370-7.

82. Baccala A, Sercia L, Li J, Heston W, Zhou M. Expression of prostate-specific membrane antigen in tumor-associated neovasculature of renal neoplasms. Urology. 2007;70(2):385-90.

83. Al-Ahmadie HA, Olgac S, Gregor PD, Tickoo SK, Fine SW, Kondagunta GV, et al. Expression of prostate-specific membrane antigen in renal cortical tumors. Mod Pathol. 2008;21(6):727-32.

84. Yin Y, Campbell SP, Markowski MC, Pierorazio PM, Pomper MG, Allaf ME, et al. Inconsistent Detection of Sites of Metastatic Non-Clear Cell Renal Cell Carcinoma with PSMA-Targeted [(18)F]DCFPyL PET/CT. Mol Imaging Biol. 2019;21(3):567-73.

85. Rhee H, Blazak J, Tham CM, Ng KL, Shepherd B, Lawson M, et al. Pilot study: use of gallium-68 PSMA PET for detection of metastatic lesions in patients with renal tumour. EJNMMI Res. 2016;6(1):76.

86. Rowe SP, Gorin MA, Hammers HJ, Som Javadi M, Hawasli H, Szabo Z, et al. Imaging of metastatic clear cell renal cell carcinoma with PSMA-targeted (1)(8)F-DCFPyL PET/CT. Ann Nucl Med. 2015;29(10):877-82.

87. Sawicki LM, Buchbender C, Boos J, Giessing M, Ermert J, Antke C, et al. Diagnostic potential of PET/CT using a (68)Ga-labelled prostate-specific membrane antigen ligand in whole-body staging of renal cell carcinoma: initial experience. Eur J Nucl Med Mol Imaging. 2017;44(1):102-7.

88. Meyer AR, Carducci MA, Denmeade SR, Markowski MC, Pomper MG, Pierorazio PM, et al. Improved identification of patients with oligometastatic clear cell renal cell carcinoma with PSMA-targeted (18)F-DCFPyL PET/CT. Ann Nucl Med. 2019;33(8):617-23.

89. Siva S, Callahan J, Pryor D, Martin J, Lawrentschuk N, Hofman MS. Utility of (68) Ga prostate specific membrane antigen - positron emission tomography in diagnosis and response assessment of recurrent renal cell carcinoma. J Med Imaging Radiat Oncol. 2017;61(3):372-8.

90. Forner A, Reig M, Bruix J. Hepatocellular carcinoma. Lancet. 2018;391(10127):1301-14.

91. Jiao D, Li Y, Yang F, Han D, Wu J, Shi S, et al. Expression of Prostate-Specific Membrane Antigen in Tumor-Associated Vasculature Predicts Poor Prognosis in Hepatocellular Carcinoma. Clin Transl Gastroenterol. 2019;10(5):1-7.

92. Kesler M, Levine C, Hershkovitz D, Mishani E, Menachem Y, Lerman H, et al. (68)Ga-PSMA is a novel PET-CT tracer for imaging of hepatocellular carcinoma: A prospective pilot study. J Nucl Med. 2018.

93. Tolkach Y, Goltz D, Kremer A, Ahmadzadehfar H, Bergheim D, Essler M, et al. Prostate-specific membrane antigen expression in hepatocellular carcinoma: potential use for prognosis and diagnostic imaging. Oncotarget. 2019;10(41):4149-60.

94. Kunikowska J, Cieślak B, Gierej B, Patkowski W, Kraj L, Kotulski M, et al. [(68) Ga]Ga-Prostate-Specific Membrane Antigen PET/CT: a novel method for imaging patients with hepatocellular carcinoma. Eur J Nucl Med Mol Imaging. 2020.

95. Kuyumcu S, Has-Simsek D, Iliaz R, Sanli Y, Buyukkaya F, Akyuz F, et al. Evidence of Prostate-Specific Membrane Antigen Expression in Hepatocellular Carcinoma Using 68Ga-PSMA PET/CT. Clin Nucl Med. 2019;44(9):702-6.

96. Alipour R, Gupta S, Trethewey S. Ga-68-PSMA Uptake in Combined Hepatocellular

Cholangiocarcinoma With Skeletal Metastases. Clinical Nuclear Medicine. 2017;42(10):E452-E3.

97. Das J, Ray S, Tapadia R, Midha D, Mallick I. Prostate-specific membrane antigen-expressing hepatic lesion: Metastatic or hepatocellular carcinoma. Indian Journal of Nuclear Medicine. 2020;35(1):58-60.

98. Erhamamci S, Aslan N. Primary Hepatocellular Carcinoma With Intense Ga-68-PSMA Uptake But Slight F-18-FDG Uptake on PET/CT Imaging. Clinical Nuclear Medicine. 2020;45(3):E176-E7.

99. Patel D, Loh H, Le K, Stevanovic A, Mansberg R. Incidental Detection of Hepatocellular Carcinoma on Ga-68-Labeled Prostate-Specific Membrane Antigen PET/CT. Clinical Nuclear Medicine. 2017;42(11):881-4.

100. Perez PM, Flavell RR, Kelley RK, Umetsu S, Behr SC. Heterogeneous Uptake of F-18-FDG and Ga-68-PSMA-11 in Hepatocellular Carcinoma. Clinical Nuclear Medicine. 2019;44(3):e133-e5.

101. Taneja S, Taneja R, Kashyap V, Jha A, Jena A. Ga-68-PSMA Uptake in Hepatocellular Carcinoma. Clinical Nuclear Medicine. 2017;42(1):E69-E70.

102. Hirmas N, Leyh C, Sraieb M, Barbato F, Schaarschmidt BM, Umutlu L, et al. [(68)Ga]Ga-PSMA-11 PET/CT improves tumor detection and impacts management in patients with hepatocellular carcinoma (HCC). J Nucl Med. 2021.

103. Janssen-Heijnen MLG, Coebergh J-WW. Trends in incidence and prognosis of the histological subtypes of lung cancer in North America, Australia, New Zealand and Europe. Lung Cancer. 2001;31(2):123-37.

104. Wang HL, Wang SS, Song WH, Pan Y, Yu HP, Si TG, et al. Expression of prostate-specific membrane antigen in lung cancer cells and tumor neovasculature endothelial cells and its clinical significance. PLoS One. 2015;10(5):e0125924.

105. Schmidt LH, Heitkötter B, Schulze AB, Schliemann C, Steinestel K, Trautmann M, et al. Prostate specific membrane antigen (PSMA) expression in non-small cell lung cancer. PLoS One. 2017;12(10):e0186280.

106. Chia JY, Loi HY, Khor LK, Joe Lee KC, Seow YH. Primary Lung Adenocarcinoma With (68) Gallium Prostate-Specific Membrane Antigen-PET/CT Scan Avidity in a Patient on Surveillance After Prostatectomy. Clin Genitourin Cancer. 2018;16(3):e525-e7.

107. Jochumsen MR, Gormsen LC, Nielsen GL. 68Ga-PSMA Avid Primary Adenocarcinoma of the Lung With Complementary Low 18F-FDG Uptake. Clin Nucl Med. 2018;43(2):117-9.

108. Osman MM, Iravani A, Hicks RJ, Hofman MS. Detection of Synchronous Primary Malignancies with (68)Ga-Labeled Prostate-Specific Membrane Antigen PET/CT in Patients with Prostate Cancer: Frequency in 764 Patients. J Nucl Med. 2017;58(12):1938-42.

109. Pyka T, Weirich G, Einspieler I, Maurer T, Theisen J, Hatzichristodoulou G, et al. 68Ga-PSMA-HBED-CC PET for Differential Diagnosis of Suggestive Lung Lesions in Patients with Prostate Cancer. J Nucl Med. 2016;57(3):367-71.

110. Shetty D, Loh H, Bui C, Mansberg R, Stevanovic A. Elevated 68Ga Prostate-Specific Membrane Antigen Activity in Metastatic Non-Small Cell Lung Cancer. Clin Nucl Med. 2016;41(5):414-6.

111. Usmani S, Sadeq A, Marafi F, Esmail A, Al Kandari F, Ahmed N. 68Ga-PMSA Uptake in the Lung: Metastatic Versus Primary Lung Tumor. Clin Nucl Med. 2020;45(2):e80-e2.

112. SEER database. Cancer Stat Facts: Female Breast Cancer. https://seer.cancer.gov/statfacts/html/breast.html. Accessed in 2020. .

113. Kim J, Kim JY, Lee H-B, Lee YJ, Seong M-K, Paik N, et al. Characteristics and prognosis of 17 special histologic subtypes of invasive breast

cancers according to World Health Organization classification: comparative analysis to invasive carcinoma of no special type. Breast Cancer Research and Treatment. 2020;184(2):527-42.

114. Kasoha M, Unger C, Solomayer E-F, Bohle RM, Zaharia C, Khreich F, et al. Prostate-specific membrane antigen (PSMA) expression in breast cancer and its metastases. Clinical & experimental metastasis. 2017;34(8):479-90.

115. Wernicke AG, Varma S, Greenwood EA, Christos PJ, Chao KS, Liu H, et al. Prostate-specific membrane antigen expression in tumor-associated vasculature of breast cancers. Apmis. 2014;122(6):482-9.

116. Sathekge M, Lengana T, Modiselle M, Vorster M, Zeevaart J, Maes A, et al. 68Ga-PSMA-HBED-CC PET imaging in breast carcinoma patients2017 2017-Apr-. 689-94 p.

117. Morgenroth A, Tinkir E, Vogg ATJ, Sankaranarayanan RA, Baazaoui F, Mottaghy FM. Targeting of prostate-specific membrane antigen for radio-ligand therapy of triple-negative breast cancer. Breast Cancer Res. 2019;21(1):116.

118. Tolkach Y, Gevensleben H, Bundschuh R, Koyun A, Huber D, Kehrer C, et al. Prostate-specific membrane antigen in breast cancer: a comprehensive evaluation of expression and a case report of radionuclide therapy. Breast cancer research and treatment. 2018;169(3):447-55.

119. Hofman MS, Emmett L, Sandhu S, Iravani A, Joshua AM, Goh JC, et al. [(177)Lu]Lu-PSMA-617 versus cabazitaxel in patients with metastatic castration-resistant prostate cancer (TheraP): a randomised, open-label, phase 2 trial. Lancet. 2021;397(10276):797-804.

120. Herrmann K, Schwaiger M, Lewis JS, Solomon SB, McNeil BJ, Baumann M, et al. Radiotheranostics: a roadmap for future development. Lancet Oncol. 2020;21(3):e146-e56.

121. Paymani Z, Rohringer T, Vali R, Loidl W, Alemohammad N, Geinitz H, et al. Diagnostic Performance of [(18)F]Fluorocholine and [(68)Ga]Ga-PSMA PET/CT in Prostate Cancer: A Comparative Study. J Clin Med. 2020;9(7).

122. Scarpa L, Buxbaum S, Kendler D, Fink K, Bektic J, Gruber L, et al. The (68)Ga/(177)Lu theragnostic concept in PSMA targeting of castration-resistant prostate cancer: correlation of SUV(max) values and absorbed dose estimates. Eur J Nucl Med Mol Imaging. 2017;44(5):788-800.

123. Tsourlakis MC, Klein F, Kluth M, Quaas A, Graefen M, Haese A, et al. PSMA expression is highly homogenous in primary prostate cancer. Appl Immunohistochem Mol Morphol. 2015;23(6):449-55.

124. Brady D, O'Sullivan J, Prise K. What is the Role of the Bystander Response in Radionuclide Therapies? Frontiers in Oncology. 2013;3(215).

125. Demaria S, Ng B, Devitt ML, Babb JS, Kawashima N, Liebes L, et al. Ionizing radiation inhibition of distant untreated tumors (abscopal effect) is immune mediated. Int J Radiat Oncol Biol Phys. 2004;58(3):862-70.

126. RADIOSENSITIVITY OF TUMORS. Journal of the American Medical Association. 1934;102(8):619-20.

127. Kratochwil C, Haberkorn U, Giesel FL. Radionuclide Therapy of Metastatic Prostate Cancer. Seminars in Nuclear Medicine. 2019;49(4):313-25.

128. Molvar C, Lewandowski R. Yttrium-90 Radioembolization of Hepatocellular Carcinoma-Performance, Technical Advances, and Future Concepts. Semin Intervent Radiol. 2015;32(4):388-97.

129. Wang EA, Stein JP, Bellavia RJ, Broadwell SR. Treatment options for unresectable HCC with a focus on SIRT with Yttrium-90 resin microspheres. Int J Clin Pract. 2017;71(11).

130. Malik D, Kumar R, Mittal BR, Singh H, Bhattacharya A, Singh SK. 68Ga-Labeled PSMA Uptake in Nonprostatic Malignancies: Has the Time Come to Remove "PS" From PSMA? ; 2018 2018-Jul-. Report No.: 1536-0229 Contract No.: 7.

131. Passah A, Arora S, Damle NA, Tripathi M, Bal C, Subudhi TK, et al. 68Ga-Prostate-Specific Membrane Antigen PET/CT in Triple-Negative Breast Cancer. 2018 2018-Jun-. Report No.: 1536-0229 Contract No.: 6.

132. Rhee H, Ng KL, Tse BW, Yeh MC, Russell PJ, Nelson C, et al. Using prostate specific membrane antigen (PSMA) expression in clear cell renal cell carcinoma for imaging advanced disease. Pathology. 2016;48(6):613-6.

133. Kinoshita Y, Kuratsukuri K, Landas S, Imaida K, Rovito PM, Jr., Wang CY, et al. Expression of prostate-specific membrane antigen in normal and malignant human tissues. World J Surg. 2006;30(4):628-36.

134. Chang SS, Reuter VE, Heston WD, Gaudin PB. Metastatic renal cell carcinoma neovasculature expresses prostate-specific membrane antigen. Urology. 2001;57(4):801-5.

135. Joshi A, Nicholson C, Rhee H, Gustafson S, Miles K, Vela I. Incidental Malignancies Identified During Staging for Prostate Cancer With (68)Ga Prostate-specific Membrane Antigen HBED-CC Positron Emission Tomography Imaging. Urology. 2017;104:e3-e4.

136. Gao J, Xu Q, Fu Y, He K, Zhang C, Zhang Q, et al. Comprehensive evaluation of (68)Ga-PSMA-11 PET/CT parameters for discriminating pathological characteristics in primary clear-cell renal cell carcinoma. Eur J Nucl Med Mol Imaging. 2020.

137. Raveenthiran S, Esler R, Yaxley J, Kyle S. The use of (68)Ga-PET/CT PSMA in the staging of primary and suspected recurrent renal cell carcinoma. Eur J Nucl Med Mol Imaging. 2019;46(11):2280-8.

138. Mittlmeier LM, Unterrainer M, Todica A, Cyran CC, Rodler S, Bartenstein P, et al. PSMA PET/CT for tyrosine-kinase inhibitor monitoring in metastatic renal cell carcinoma. Eur J Nucl Med Mol Imaging. 2020;47(9):2216-7.

139. Marafi F, Sasikumar A, Al-Terki A, Alfeeli M. 18F-PSMA 1007 in Suspected Renal Cell Carcinoma. Clin Nucl Med. 2020;45(5):377-8.

140. Chen EJ, Tan TH, Chew MT, Chye PC. 68Ga-PSMA PET/CT and 18F-FDG PET/CT in Renal Cell Carcinoma. Clin Nucl Med. 2020;45(7):e317-e9.

141. Demirci E, Ocak M, Kabasakal L, Decristoforo C, Talat Z, Halac M, et al. (68)Ga-PSMA PET/CT imaging of metastatic clear cell renal cell carcinoma. Eur J Nucl Med Mol Imaging. 2014;41(7):1461-2.

142. Rowe SP, Gorin MA, Hammers HJ, Pomper MG, Allaf ME, Javadi MS. Detection of 18F-FDG PET/CT Occult Lesions With 18F-DCFPyL PET/CT in a Patient With Metastatic Renal Cell Carcinoma. Clin Nucl Med. 2016;41(1):83-5.

143. Einspieler I, Tauber R, Maurer T, Schwaiger M, Eiber M. 68Ga Prostate-Specific Membrane Antigen Uptake in Renal Cell Cancer Lymph Node Metastases. Clin Nucl Med. 2016;41(5):e261-2.

144. Sasikumar A, Joy A, Nanabala R, Unni M, Tk P. Complimentary Pattern of Uptake in 18F-FDG PET/CT and 68Ga-Prostate-Specific Membrane Antigen PET/CT in a Case of Metastatic Clear Cell Renal Carcinoma. Clin Nucl Med. 2016;41(12):e517-e9.

145. Zacho HD, Nielsen JB, Dettmann K, Haberkorn U, Petersen LJ. Incidental Detection of Thyroid Metastases From Renal Cell Carcinoma Using 68Ga-PSMA PET/CT to Assess Prostate Cancer Recurrence. Clin Nucl Med. 2017;42(3):221-2.

146. Mittlmeier LM, Unterrainer M, Todica A, Cyran CC, Rodler S, Bartenstein P, et al. PSMA PET/CT for tyrosine-kinase inhibitor monitoring in

metastatic renal cell carcinoma. Eur J Nucl Med Mol Imaging. 2020.

147. Kumar R, Mittal BR, Bhattacharya A, Singh H, Singh SK. Synchronous Detection of Male Breast Cancer and Prostatic Cancer in a Patient With Suspected Prostatic Carcinoma on 68Ga-PSMA PET/CT Imaging. 2018 2018-Jun-. Report No.: 1536-0229 Contract No.: 6.

148. Polverari G, Ceci F, Calderoni L, Cervati V, Farolfi A, Castellucci P, et al. Male Breast Cancer Detected by 68Ga-PSMA-11 PET/CT in a Patient With Prostate Cancer With Pelvic Lymph Node Metastasis. 2019 2019-04-. Report No.: 1938-0682 Contract No.: 2.

149. Wernicke AG, Varma S, Greenwood EA, Christos PJ, Chao KSC, Liu H, et al. Prostate-specific membrane antigen expression in tumor-associated vasculature of breast cancers. APMIS : acta pathologica, microbiologica, et immunologica Scandinavica. 2014;122(6):482-9.

150. Marafi F, Sasikumar A, Alfeeli M, Thuruthel S. 18F-PSMA 1007 Uptake in a Man With Metastatic Breast Cancer. Clin Nucl Med. 2020;45(6):e276-e8.

151. Sathekge M, Modiselle M, Vorster M, Mokgoro N, Nyakale N, Mokaleng B, et al. ^{68}Ga-PSMA imaging of metastatic breast cancer. 2015 2015-Aug-. Report No.: 1619-7089 Contract No.: 9.

152. Marafi F, Sasikumar A, Alfeeli M, Fathallah W. 18F-PSMA 1007 Uptake in Brain Metastases From Breast Cancer. 2020 2020-Feb-. Report No.: 1536-0229 Contract No.: 2.

Chapter 6

^{177}Lu-PSMA radioligand therapy for recurrent/metastatic salivary gland cancer patients, a phase II pilot study.

Maike J.M. Uijen, Wim van Boxtel, Bastiaan M. Privé, Chantal M.L. Driessen, Martin Gotthardt, James Nagarajah, Carla M.L. van Herpen

Work in progress: description of preliminary results 2022.

ABSTRACT

Background

Systemic treatment options for salivary gland cancer (SGC) patients are scarce. Prostate-specific membrane antigen (PSMA) targeted radionuclide therapy is highly effective in prostate cancer. PSMA imaging results in SGC patients, especially in the adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC) subtypes, indicated that PSMA radionuclide therapy may be a useful option in SGC patients. Therefore, this study evaluates the safety and efficacy of PSMA targeted radionuclide therapy in ACC and SDC patients.

Methods

This is an ongoing single-centre, single-arm, phase II pilot study. Incurable recurrent or metastatic ACC and SDC patients with sufficient PSMA tracer uptake on positron emission tomography (PET) imaging are included. Treatment consists of 7.4 GBq (±10%) [^{177}Lu]Lu-PSMA-I&T, administrated intravenously, with an interval of 6±1 weeks between subsequent cycles and with a maximum of 4 cycles. The primary endpoint is safety. Secondary endpoints include the objective response rate (ORR), progression-free survival (PFS), and overall survival (OS).

Results

This study started in May 2020 and is still ongoing. This report presents the preliminary results of this study. The cut-off date for this analysis was 1 December 2021. Until that date, 17 patients (12 ACC and 5 SDC) were screened for eligibility and 10 patients (8 ACC and 2 SDC) met the inclusion criteria and received at least one cycle of [^{177}Lu]Lu-PSMA-I&T. Due to the limited number of SDC patients, this chapter only describes the preliminary results of the 8 ACC patients who received [^{177}Lu]Lu-PSMA-I&T treatment. The median follow-up of these 8 ACC patients at the cut-off date for this analysis was 7.8 months (range: 2.5-14.1 months).

Trial registration

This trial is registered on ClinicalTrials.gov: NCT04291300.

Update: At the time of submission of this thesis (April 2022) the inclusion of the ACC cohort was completed (10/10 patients). The SDC cohort is still open for inclusion; 2/5 patients are included.

Based on the limited number of SDC patients (*n*=2), as mentioned above, this chapter only includes the preliminary results of the first 8 ACC patients.

INTRODUCTION

Salivary gland cancer (SGC) is a rare cancer and consists of many different subtypes. Local recurrences and/or distant metastases occur most often in patients with adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC) subtypes (1, 2). Approximately 60% of all ACC patients and 50% of all SDC patients experience a local recurrence and/or distant metastases over time (1-3). Therefore, ACC and SDC patients often require palliative systemic therapy. ACC tumors often show an indolent growth pattern, however some subgroups of patients may show a more aggressive disease course (e.g. NOTCH1 mutated ACC tumours (4)).

For ACC patients with metastases the median overall survival is approximately 3 years (5). Responses are difficult to achieve in ACC disease (5). Only a few treatments have shown clinically relevant response rates and the most common given therapies (e.g. combined chemotherapy regimens and tyrosine kinase inhibitors) based on single-arm phase II studies often cause side effects that can reduce the quality of life.

For SDC, one of the most aggressive subtypes of SGC, two recent phase II studies showed promising results. For androgen receptor-positive SDC tumors (78-96%), androgen deprivation therapy resulted in a response rate of 42% (6). For Human epidermal growth factor receptor 2 (HER2) -positive SDC tumors (≈30%), the combination of docetaxel and trastuzumab showed a response rate of 70% (7), and case series provided a rationale for second-line HER-2 treatment with ado-trastuzumab emtansine (8). But overall, unfortunately, there are still limited systemic treatment options for ACC and SDC patients, warranting further research (9, 10).

Together, the results of prostate-specific membrane antigen (PSMA) targeted therapy in prostate cancer and PSMA imaging results in SGC, makes PSMA an appealing therapeutic target for SGC patients. PSMA is a transmembrane protein, firstly discovered in prostate cancer (11), and is an example of a theranostic target, where a single target can be imaged with diagnostics and can also be targeted with therapy (12). In prostate cancer, PSMA Positron emission tomography (PET) imaging, using radiotracers that specifically bind to PSMA, showed high tumor detection rates and excellent tumor-to-background ratios (13, 14). Subsequently, research focussed on PSMA-targeted radioligand therapy. By coupling a PSMA ligand with a radionuclide (for example Lutetium-177; ^{177}Lu) a cytotoxic radiation load can be directed to PSMA expressing tumor cells (15, 16). This therapy,^{177}Lu-PSMA, improved both the median progression-free survival (8.7 vs 3.4 months) and median overall survival (15.3 vs 11.3 months) compared to standard care alone in a phase III study of advanced PSMA-positive metastatic castration-resistant prostate cancer patients (17). Furthermore, ^{177}Lu-PSMA treatment also showed a relatively favourable toxicity profile (17, 18). In a randomized study in prostate cancer patients, ^{177}Lu-PSMA showed a higher response rate (66% vs 37%) and a more favourable toxicity profile than cabazitaxel, due to fewer grade 3-4 adverse events (33% vs 53%) (18).

PSMA PET imaging in SGC patients showed that 93% of ACC patients and 40% of SDC have sufficient tracer uptake, which encourages to explore the potential of PSMA targeted therapy in these patients (19). Hence, the aim of this ongoing study is to investigate the safety and efficacy of ^{177}Lu-PSMA therapy for ACC and SDC patients.

METHODS

Study design

This is an ongoing single-centre, single-arm, phase II pilot study of the Radboudumc (an SGC expertise centre in the Netherlands). The study consists of two cohorts. In total, 10 incurable recurrent and/or metastatic ACC patients will be included in the ACC cohort and 5 incurable recurrent and/or metastatic SDC patients will be included in the SDC cohort. Study treatment, as detailed below, is similar for both cohorts. This study was approved by the local medical ethical committee. A Data Safety Monitoring Board (DSMB), consisting of an oncologist with expertise in head and neck cancer, a nuclear medicine physician and a biostatistician, conducts ongoing safety oversight through quarterly study updates.

Study population

Eligible participants, ACC or SDC patients with incurable local or regional recurrent or metastatic disease, are required to be ≥ 18 years of age, to have adequate bone marrow function, renal, and liver function, and an Eastern Cooperative Oncology Group (ECOG) performance status of 0–2. There are no limitations regarding previous cancer treatments. Based on the general indolent tumor growth of ACC and recommendations for clinical trial design in ACC patients (5), ACC patients can only participate in case of objective growth in the last three months or complaints due to their disease. Furthermore, patients need to have measurable disease as per Response Evaluation Criteria in Solid Tumours (RECIST) version 1.1 (20), which is assessed with baseline tumor imaging consisting of a CT scan of the chest and abdomen and depending on the primary tumor location, a CT- or MR scan of the neck. All patients underwent gallium-68 [^{68}Ga]Ga-PSMA-11 and 2-deoxy-2-[^{18}F]fluoro-D-glucose (FDG) PET/CT scans, which we will further refer to as ^{68}Ga-PSMA PET and ^{18}FDG PET in this report. Patients need to have a positive ^{68}Ga-PSMA PET scan, defined by at least one lesion ≥ 1.5 cm with a tracer uptake above the liver level. In the case of clinically relevant discrepancies between tumor detected on ^{68}Ga-PSMA PET and ^{18}FDG PET, the PSMA treatment was at the discretion of the treating physicians.

Exclusion criteria include pregnancy, inadequate contraceptive measurements for patients with reproductive potential, brain or intracardial metastases, cranial epidural disease, concurrent serious conditions, urinary tract obstruction, and an interval of less than 4 weeks since the last myelosuppressive therapy or other radionuclide therapy.

^{177}Lu-PSMA-I&T Treatment

In case of eligibility, patients start with [^{177}Lu]Lu-PSMA-I&T treatment. Each cycle consists of 7.4 GBq (±10%) [^{177}Lu]Lu-PSMA-I&T which is administrated intravenously, with an interval of 6±1 weeks between subsequent cycles and with a maximum of 4 cycles.

To prevent or reduce nausea, patients are offered ondansetron 8 mg orally approximately 1 hour before the treatment administration. Furthermore, patients are advised to drink enough fluids (approximately 2 liters) on the day of treatment administration and the following days.

Single photon emission CT (SPECT-CT) scans of the head-and-neck region and chest-abdomen region as well as whole-body planar scans are performed 24 hours following each [^{177}Lu]Lu-PSMA-I&T treatment, further referred to as ^{177}Lu-PSMA, for safety analysis.

A mid-treatment imaging evaluation is performed approximately 4 weeks after the 2nd ^{177}Lu-PSMA treatment, where baseline tumor imaging is repeated (^{68}Ga-PSMA PET, ^{18}FDG PET, CT scans and in some cases MR scan). In case of disease progression per RECIST 1.1, the treatment is discontinued. In case of a response or stable disease, the patient continues the treatment until reaching the maximum of 4 cycles. Other reasons for early discontinuation of the treatment or dosage modification are at the discretion of the principal investigator (CvH).

Study assessments

During study treatment, participants are monitored every 2 weeks after the 1st and 2nd treatment cycles and every 3 weeks after the 3rd and 4th treatment cycles, this includes blood analysis (routine haematology and biochemistry). A final safety assessment is done at 6 weeks after the 4th treatment cycle, and follow-up continues every 12 weeks thereafter. Tumor imaging is performed at baseline, after the 2nd treatment cycle and every 3 months from the 4th treatment cycle. Furthermore, patient-reported outcome measures include validated quality of life questionnaires (e.g. EORTC's QLQ-C30 (21), EORTC QLQ-HN43 (22)) and visual analogue scale (VAS) pain scores. These are assessed at baseline, before each treatment cycle, 3 months, and 6 months after the 4th cycle. An overview of all study assessments and timepoints can be found in the Supplementary (study assessments table).

Specifically for dosimetry analyses (calculating the delivered doses to the tumors and organs at risks), SPECT-CT and planar scans are made at 1, 24, 48, 72 hours, and 7 days after the first ^{177}Lu-PSMA injection. For predictive and translational purposes circulating tumor DNA (ctDNA) is obtained at predefined timepoints, see assessments table.

The results of patient-reported outcome measures, dosimetry, and ctDNA analyses are not included in this preliminary report of this ongoing study.

Study endpoints and statistical analysis

The primary endpoint of this study is to evaluate the safety of ^{177}Lu-PSMA in ACC and SDC patients. Other endpoints include the objective response rate (ORR), progression-free survival (PFS), and overall survival (OS).

The safety is studied by analysis of the adverse events according to National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE) version 5.0. The ORR is defined as the proportion of patients with a complete response (CR) or a partial response (PR) according to RECIST version 1.1. Kaplan-Meier analyses are used for the assessment of the PFS and OS. To summarize the study population, descriptive statistics are performed. Statistical analyses are performed using IBM SPSS Statistics version 25.0 (IBM Corp, Armonk, New York). Since the primary endpoint of this explorative study consists of safety, no sample size calculation was performed. Considering the rarity of ACC and SDC disease, our research team considered 10 ACC and 5 SDC patients a feasible number of patients for this study, which would provide sufficient information regarding safety.

RESULTS

Here, we present the preliminary results of this ongoing study. This study opened for inclusion on 26 May 2020. The cut-off date for this analysis was 1-12-2021. At this cut-off date, in total 17 patients (12 ACC and 5 SDC) had been screened for study eligibility and 10 patients (8 ACC and 2 SDC) were eligible and received at least one dose of ^{177}Lu-PSMA treatment.

Reasons for ineligibility of 4/12 ACC patients were: insufficient tracer uptake on ^{68}Ga-PSMA PET (*n*=2), brain lesions detected on ^{68}Ga-PSMA PET suspicious for brain metastases which were confirmed by MR imaging (*n*=1), and epidural disease (*n*=1). Reasons for ineligibility of 3/5 SDC patients were: insufficient tracer uptake on ^{68}Ga-PSMA PET (*n*=2), brain lesions detected on ^{68}Ga-PSMA PET suspicious for brain metastases which were confirmed by MR imaging (*n*=1).

Based on the limited number of SDC patients (*n*=2) who were treated with ^{177}Lu-PSMA treatment thus far, this chapter only reports on the preliminary results of the first 8 ACC patients.

Patient characteristics

At the time of this analysis, 8 ACC patients started ^{177}Lu-PSMA treatment. These patients received the first treatment cycle of ^{177}Lu-PSMA within 4 weeks after baseline tumor imaging. Figure 1 illustrates the ^{68}Ga-PSMA PET tracer uptake of three representative patients. Baseline patient characteristics are summarised in table 1. Only one ACC patient was treated for incurable locoregional disease only (figure 1C), all other patients had distant metastatic disease with or without locoregional disease. Four patients (40%) received prior systemic therapy.

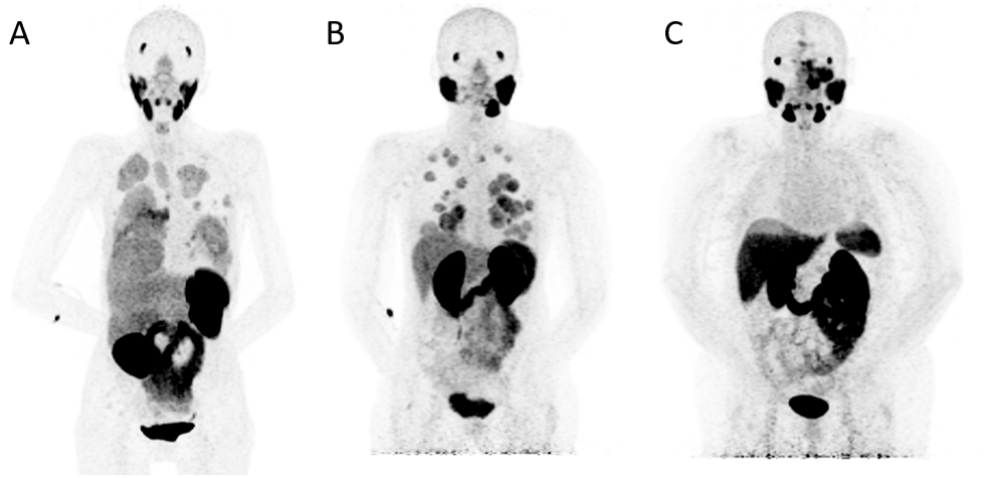


Figure 1: PSMA-PET maximum intensity projections (MIP) of 3 ACC patients before ^{177}Lu-PSMA-I&T treatment

A: ACC patient, primary tumor arose from Bartholin's gland (status post-surgery), with lung, pleural and lymph node metastases, **B**: ACC patient, primary tumor arose from right submandibular gland (status post-surgery), with lung and liver metastases, **C**: ACC patient, primary tumor arose from left lacrimal gland, with an incurable local tumor and lymph node metastasis (near left submandibular gland).

The same scaling was used for all patients.

Table 1: Baseline patient characteristics

	ACC (*n*=8) No. of pts (%)*
Gender	
Male	2 (25)
Female	6 (75)
Age, median (range)	64 (53-73)
ECOG PS	
0	2 (25)
1	5 (62.5)
2	1 (12.5)
Primary site	
Parotid gland	0
Submandibular gland	1 (12.5)
Sublingual gland	0
Minor salivary gland	4 (50)
Lacrimal gland	1 (12.5)
Bartholin's gland	2 (25)
Disease distribution	
LR	1 (12.5)
LR + DM	1 (12.5)
DM	6 (75)
Sites of DM	
Lung	7 (87.5)
Pleural	3 (37.5)
Liver	3 (37.5)
Bone	2 (25)
Distant lymph nodes	2 (25)
Other	1 (12.5)
Prior treatments	
Primary treatment	
Tumor resection	6 (75)
Lymph node neck dissection	1 (12.5)
Radiotherapy	8 (100)
Adjuvant systemic therapy	0
Palliative treatments	
Systemic therapy	2 (25)
Median number of prior lines (range)	1 (-)
Chemotherapy	1 (25)
Other§	1 (25)
Palliative surgery	1 (12.5)
Palliative radiotherapy	2 (25)

* Values are numbers and percentages, unless indicated otherwise.

§ Other includes: ACC patients: cabozantinib (*n*=1)

Abbreviations: ACC: adenoid cystic carcinoma, DM: distant metastases, ECOG PS: Eastern Cooperative Oncology Group performance status, LR: locoregional disease, pts: patients

^{177}Lu-PSMA-I&T Treatment overview

At the time of this analysis, 3/8 patients (37.5%) received all four cycles of ^{177}Lu-PSMA and thus completed the full treatment. In 1/8 patients (12.5%) the treatment is still ongoing, this patient currently received three cycles of ^{177}Lu-PSMA and the fourth cycle is scheduled.

Treatment was discontinued after two cycles of ^{177}Lu-PSMA in 3/8 patients (37.5%), due to progressive disease at the predefined mid-treatment evaluation. Furthermore, one patient (12.5%) received only one cycle of ^{177}Lu-PSMA, due to signs of clinical progression before the second cycle ^{177}Lu-PSMA. Additional imaging was performed in this patient which confirmed the disease progression and no further treatments were given. The median follow-up of these 8 ACC patients at the cut-off date for this analysis was 7.8 months (range: 2.5-14.1 months).

Safety

Thus far, there were no abnormalities in the administered radioactivity per treatment cycle or the interval between two cycles. All injections consisted of 7.4 GBq (±10%) ^{177}Lu-PSMA and were given with an interval of 6 ±1 weeks. Administration of ^{177}Lu-PSMA was well tolerated, with no immediate adverse reactions after the injection.

The most common observed adverse events included: nausea (87.5%), anaemia (87.5%), dry mouth (75%), and fatigue (50%), all of them limited to grade 1 or 2. Two patients (25%) developed grade 3 toxicity. In one patient the lymphocyte count decreased (grade 3) after one cycle of ^{177}Lu-PSMA. Another patient developed a grade 3 hyponatremia after the second cycle of ^{177}Lu-PSMA. No grade 4 or grade 5 toxicity was observed. Table 2 provides an overview of the adverse events observed in the study until the cut-off date.

Noteworthy, one patient developed a grade 2 chronic kidney disease. The estimated glomerular filtration rate (eGFR) decreased four months after the fourth cycle of ^{177}Lu-PSMA and has remained stable (until the last-follow up at the time of this analysis). Nephrology colleagues were consulted and this decrease in kidney function was considered to be a tubulointerstitial nephritis based on other medication the patient was using or potentially a late effect of the ^{177}Lu-PSMA treatment.

Table 2: Adverse events probably related to ^{177}Lu-PSMA-I&T treatment

Adverse event	Any Grade No. of pts (%)	Grade 1 No. of pts (%)	Grade 2 No. of pts (%)	Grade 3 No. of pts (%)	Grade ≥4 No. of pts (%)
Nausea	7 (87.5)	6 (75)	1 (12.5)	0	0
Anaemia	7 (87.5)	7 (87.5)	0	0	0
Dry mouth	6 (75)	5 (62.5)	1 (12.5)	0	0
Fatigue	4 (50)	2 (25)	2 (25)	0	0
Lymphocyte count decreased	4 (50)	1 (12.5)	2 (25)	1 (12.5)	0
White blood cell decreased	3 (37.5)	2 (25)	1 (12.5)	0	0
Platelet count decreased	2 (25)	2 (25)	0	0	0
Vomiting	2 (25)	2 (25)	0	0	0
Anorexia	2 (25)	0	2 (25)	0	0
Constipation	2 (25)	1 (12.5)	1 (12.5)	0	0
Diarrhoea	2 (25)	2 (25)	0	0	0
Neutrophil count decreased	2 (25)	1 (12.5)	1 (12.5)	0	0
Bone pain	1 (12.5)	1 (12.5)	0	0	0
Chronic kidney disease	1 (12.5)	0	1 (12.5)	0	0
Dysgeusia	1 (12.5)	1 (12.5)	0	0	0
Dyspepsia	1 (12.5)	0	1 (12.5)	0	0
Gastrointestinal pain	1 (12.5)	1 (12.5)	0	0	0
Hypokalaemia	1 (12.5)	1 (12.5)	0	0	0
Hyponatremia	1 (12.5)	0	0	1 (12.5)	0

Table lists all treatment-related adverse events, sorted based on frequency of occurrence.

Adverse events were graded according to National Cancer Institute Common Terminology Criteria for Adverse Events (version 5.0). Patients were counted once at the highest grade for each adverse event.

Abbreviations: pts: patients

Efficacy

During this preliminary analysis, no objective responses were observed thus far in the first 8 ACC patients of this ongoing clinical trial. Four ACC patients showed stable disease during the predefined mid-treatment evaluation and two of these patients remained stable for ≥ 6 months. Table 3 shows the responses per cohort. Figure 2 illustrates the tumor growth of target lesions pre- and post- ^{177}Lu-PSMA treatment. In this figure, patients 1 and 6 are the patients with stable disease ≥ 6 months.

Table 3: ^{177}Lu-PSMA-I&T treatment efficacy

Efficacy	ACC (*n*=8) No. of patients (%)
CR	0
PR	0
SD	4 (50)
SD ≥ 6 months*	2 (25)
PD	4 (50)
ORR	0 (0)

* These numbers are affected by the cut-off date; these could change after a longer follow-up.

Abbreviations: ACC: adenoid cystic carcinoma, CR: complete response, ORR: objective response rate (CR+PR), PD: progressive disease, PR: partial response, SD: stable disease.

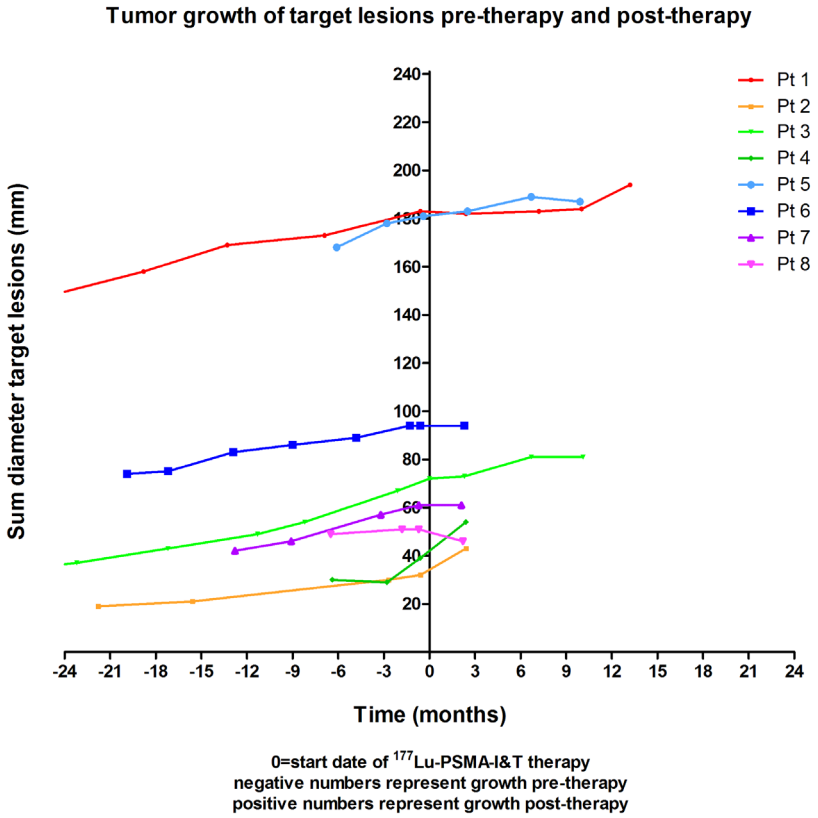


Figure 2: Tumor growth of target lesions pre- and post- ^{177}Lu-PSMA treatment

Pre-therapy imaging: to visualise the natural tumor growth before the start of ^{177}Lu-PSMA therapy, only imaging timepoints were where patients were not on other anticancer therapy are shown.

Post-therapy imaging: all imaging of patients is visualised until lost to follow-up or start of other anticancer therapy.

Abbreviations: ACC: adenoid cystic carcinoma, pt: patient

Regarding survival, for the ACC cohort (8 patients), the median PFS was 2.6 months (95% CI: 0.0-7.7 months) and the median OS is not yet reached. The survival plots are displayed in figure 3.

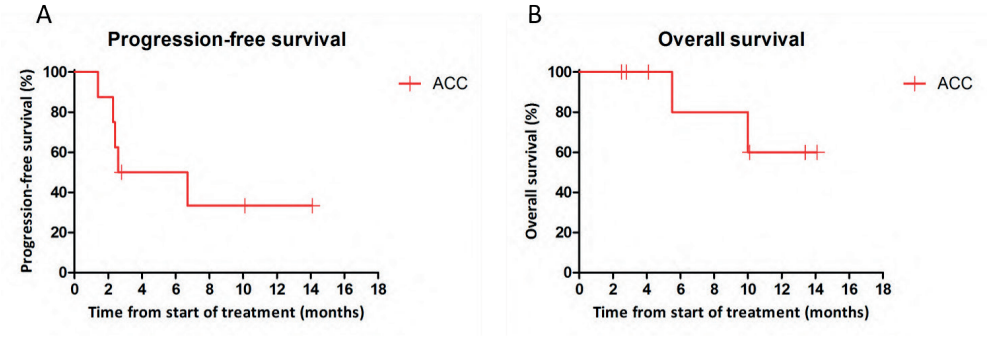


Figure 3: Kaplan-Meier plots of progression-free survival (A), and overall survival (B) of the 8 ACC patients
The plus signs in the graphs indicate censored data.

DISCUSSION

In this report we presented the preliminary results of the first 8 ACC patients included in a clinical study of PSMA-targeted radionuclide therapy with ^{177}Lu-PSMA-I&T for patients with SGC. To our knowledge, this is the first prospective study to evaluate PSMA-targeted radionuclide therapy for a cancer other than prostate cancer.

Regarding the safety, the main endpoint, ^{177}Lu-PSMA treatment in ACC patients was generally well tolerated, with only 2 patients developing a grade 3 adverse event. Furthermore there were was no grade ≥4 toxicity. One of the main differences of toxicity in our ACC population compared to previous studies in prostate cancer is the observed haematological toxicity (16-18). In end-stage prostate cancer patients, grade 3-4 haematological toxicity, e.g. anaemia (13%), thrombocytopenia (8%) and leukopenia (3%) were observed more often than in our population (ACC patients) with only one case of grade 3 haematological toxicity (lymphocytopenia). The most probable explanation for this, is the difference in bone marrow reserve between the populations. In prostate cancer, especially in end-stage disease, patients have extensive bone metastases and subsequently less healthy bone marrow tissue. In contrast, bone metastases occur less often and less extensively in ACC patients. In addition, the prostate cancer patients in ^{177}Lu-PSMA trials already had multiple lines of systemic treatment, which could cause bone marrow suppression, whilst in our study only two patients (25%) received prior systemic therapy.

Another main difference is the proportion of patients with a dry mouth (xerostomia), which was higher in our ACC population compared to prostate cancer studies. In total we observed xerostomia in 75% of our patients, this was limited to grade 1 (62.5%) or grade 2 (12.5%). While a dry mouth was only observed in approximately 40% of prostate cancer patients in the phase III study (all cases were also limited to grade 1 or 2) (17). The healthy salivary gland tissue reserve might be the most rational explanation for this difference, since ACC patients will likely have less healthy salivary gland tissue reserve than prostate cancer patients. Several ACC patients received prior surgery and/or radiotherapy in the head-and-neck region. Usually, xerostomia was reversible over time as is often the case in prostate cancer patients. Yet, in one patient with a primary tumor in the submandibular gland, who received prior surgery and radiotherapy, this side effect was still present at the last follow-up (9 months post-^{177}Lu-PSMA therapy). Although the submandibular gland is smaller than the parotid gland, it produces 2.5 times as much saliva at rest compared to the parotid gland (23). Therefore, patients who received prior surgery or radiotherapy at one of the submandibular glands may hypothetically be more susceptible to prolonged xerostomia after ^{177}Lu-PSMA treatment.

Regarding the efficacy, our preliminary results did not show any objective responses in the first 8 ACC patients treated with ^{177}Lu-PSMA thus far. Two ACC patients did show stable disease >6 months (and still ongoing). Recently, a retrospective study reported the efficacy of ^{177}Lu-PSMA in six SGC patients, including four ACC patients (24). In this study, patients were treated with 6.0–7.4 GBq ^{177}Lu-PSMA-617 every 6–8 weeks, which is relatively comparable to our treatment schedule. In this study, one ACC patient showed stable disease and another ACC patient had a partial response based on ^{68}Ga-PSMA PET (defined by the authors as an SUV decrease ≥ 30% on ^{68}Ga-PSMA PET), whilst the radiological evaluation in this patient showed stable disease. Interestingly, the study states that four patients reported subjective response by clear relief of tumour symptoms within the first weeks after the first cycle. Based on our preliminary study results in ACC patients and the retrospective study in SGC patients, it could be cautiously said that there may be some biological effectiveness of ^{177}Lu-PSMA treatment in SGC patients, however the effectiveness is much less pronounced than in prostate cancer patients. In prostate cancer studies, objective responses were observed in 49-82% of the RECIST evaluable patients (16-18). This difference in effectiveness could be due to the generally lower tracer uptake, measured in SUVmax values, in SGC tumors (SUVmax: 3.5-30.2) (19, 24) compared to SUVmax values in prostate cancer (SUVmax often >15-40, and up to 122) (25, 26), leading to lower radiation doses in SGC patients and therefore less treatment effect. In the future, the dosimetry results of our study will provide more information regarding the achieved radiation doses in SGC tumors and can be compared to dosimetry results in prostate cancer studies (27-29). Depending on these dosimetry results, in case the achieved radiation doses in SGC tumors would be limited, the use of more potent radiation emitters like Actinium-225 (^{225}Ac) may be considered. Furthermore, the difference in effectiveness could also be due to a difference in radiation sensitivity between SGC tumors and prostate cancer tumors.

In summary, the first results of ^{177}Lu-PSMA in SGC patients indicate that this treatment is well-tolerated but the efficacy thus far seems limited to stable disease, which in a proportion of patients lasts >6 months (and is still ongoing at this analysis). Final results are awaited.

ACKNOWLEDGEMENTS

We would like to thank Dr. Esther van Meerten (Erasmus MC, the Netherlands), Dr. Wouter Vogel (NKI-AVL, the Netherlands), and Prof. dr. Kit Roes (Radboudumc, the Netherlands) for their participation and advice in the Data Safety Monitoring Board.

REFERENCES

1. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.
2. Spiro RH. Distant metastasis in adenoid cystic carcinoma of salivary origin. Am J Surg. 1997;174(5):495-8.
3. Johnston ML, Huang SH, Waldron JN, Atenafu EG, Chan K, Cummings BJ, et al. Salivary duct carcinoma: Treatment, outcomes, and patterns of failure. Head & neck. 2016;38 Suppl 1:E820-6.
4. Ferrarotto R, Mitani Y, Diao L, Guijarro I, Wang J, Zweidler-McKay P, et al. Activating NOTCH1 Mutations Define a Distinct Subgroup of Patients With Adenoid Cystic Carcinoma Who Have Poor Prognosis, Propensity to Bone and Liver Metastasis, and Potential Responsiveness to Notch1 Inhibitors. Journal of Clinical Oncology. 2017;35(3):352-60.
5. Laurie SA, Ho AL, Fury MG, Sherman E, Pfister DG. Systemic therapy in the management of metastatic or locally recurrent adenoid cystic carcinoma of the salivary glands: a systematic review. The Lancet Oncology. 2011;12(8):815-24.
6. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.
7. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.
8. Uijen MJM, Lassche G, van Engen-van Grunsven ACH, Driessen CML, van Herpen CML. Case series of docetaxel, trastuzumab, and pertuzumab (DTP) as first line anti-HER2 therapy and ado-trastuzumab emtansine (T-DM1) as second line for recurrent or metastatic HER2-positive salivary duct carcinoma. Oral oncology. 2022;125:105703.
9. Lassche G, van Boxtel W, Ligtenberg MJL, van Engen-van Grunsven ACH, van Herpen CML. Advances and challenges in precision medicine in salivary gland cancer. Cancer Treat Rev. 2019;80:101906.
10. Alfieri S, Granata R, Bergamini C, Resteghini C, Bossi P, Licitra LF, et al. Systemic therapy in metastatic salivary gland carcinomas: A pathology-driven paradigm? Oral oncology. 2017;66:58-63.
11. Evans JC, Malhotra M, Cryan JF, O'Driscoll CM. The therapeutic and diagnostic potential of the prostate specific membrane antigen/glutamate carboxypeptidase II (PSMA/GCPII) in cancer and neurological disease. Br J Pharmacol. 2016;173(21):3041-79.
12. Herrmann K, Schwaiger M, Lewis JS, Solomon SB, McNeil BJ, Baumann M, et al. Radiotheranostics: a roadmap for future development. The Lancet Oncology. 2020;21(3):e146-e56.
13. Lütje S, Heskamp S, Cornelissen AS, Poeppel TD, van den Broek SA, Rosenbaum-Krumme S, et al. PSMA Ligands for Radionuclide Imaging and Therapy of Prostate Cancer: Clinical Status. Theranostics. 2015;5(12):1388-401.
14. Hofman MS, Lawrentschuk N, Francis RJ, Tang C, Vela I, Thomas P, et al. Prostate-specific membrane antigen PET-CT in patients with

high-risk prostate cancer before curative-intent surgery or radiotherapy (proPSMA): a prospective, randomised, multicentre study. Lancet. 2020;395(10231):1208-16.

15. Rahbar K, Ahmadzadehfar H, Kratochwil C, Haberkorn U, Schafers M, Essler M, et al. German Multicenter Study Investigating 177Lu-PSMA-617 Radioligand Therapy in Advanced Prostate Cancer Patients. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2017;58(1):85-90.
16. Hofman MS, Violet J, Hicks RJ, Ferdinandus J, Thang SP, Akhurst T, et al. [(177)Lu]-PSMA-617 radionuclide treatment in patients with metastatic castration-resistant prostate cancer (LuPSMA trial): a single-centre, single-arm, phase 2 study. The Lancet Oncology. 2018;19(6):825-33.
17. Sartor O, de Bono J, Chi KN, Fizazi K, Herrmann K, Rahbar K, et al. Lutetium-177–PSMA-617 for Metastatic Castration-Resistant Prostate Cancer. New England Journal of Medicine. 2021.
18. Hofman MS, Emmett L, Sandhu S, Iravani A, Joshua AM, Goh JC, et al. [(177)Lu]Lu-PSMA-617 versus cabazitaxel in patients with metastatic castration-resistant prostate cancer (TheraP): a randomised, open-label, phase 2 trial. Lancet. 2021;397(10276):797-804.
19. van Boxtel W, Lütje S, van Engen-van Grunsven ICH, Verhaegh GW, Schalken JA, Jonker MA, et al. ⁶⁸Ga-PSMA-HBED-CC PET/CT imaging for adenoid cystic carcinoma and salivary duct carcinoma: a phase 2 imaging study. Theranostics. 2020;10(5):2273-83.
20. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). European journal of cancer (Oxford, England : 1990). 2009;45(2):228-47.
21. Aaronson NK, Ahmedzai S, Bergman B, Bullinger M, Cull A, Duez NJ, et al. The European Organization for Research and Treatment of Cancer QLQ-C30: a quality-of-life instrument for use in international clinical trials in oncology. Journal of the National Cancer Institute. 1993;85(5):365-76.
22. Singer S, Amdal CD, Hammerlid E, Tomaszewska IM, Castro Silva J, Mehanna H, et al. International validation of the revised European Organisation for Research and Treatment of Cancer Head and Neck Cancer Module, the EORTC QLQ-HN43: Phase IV. Head & neck. 2019;41(6):1725-37.
23. Nanci A. Ten Cate's Oral Histology. 9th edition ed: Elsevier; 2018.
24. Klein Nulent TJW, van Es RJJ, Willems SM, Braat A, Devriese LA, de Bree R, et al. First experiences with (177)Lu-PSMA-617 therapy for recurrent or metastatic salivary gland cancer. EJNMMI research. 2021;11(1):126.
25. Paymani Z, Rohringer T, Vali R, Loidl W, Alemohammad N, Geinitz H, et al. Diagnostic Performance of [(18)F]Fluorocholine and [(68)Ga]Ga-PSMA PET/CT in Prostate Cancer: A Comparative Study. J Clin Med. 2020;9(7).
26. Scarpa L, Buxbaum S, Kendler D, Fink K, Bektic J, Gruber L, et al. The (68)Ga/(177)Lu theragnostic concept in PSMA targeting of castration-resistant prostate cancer: correlation of SUV(max) values and absorbed dose estimates. European journal of nuclear medicine and molecular imaging. 2017;44(5):788-800.
27. Violet J, Jackson P, Ferdinandus J, Sandhu S, Akhurst T, Iravani A, et al. Dosimetry of (177)Lu-PSMA-617 in Metastatic Castration-Resistant Prostate Cancer: Correlations Between Pretherapeutic Imaging and Whole-Body Tumor Dosimetry with Treatment Outcomes. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2019;60(4):517-23.
28. Peters SMB, Privé BM, de Bakker M, de Lange F, Jentzen W, Eek A, et al. Intra-therapeutic dosimetry of [(177)Lu]Lu-PSMA-617 in low-

volume hormone-sensitive metastatic prostate cancer patients and correlation with treatment outcome. European journal of nuclear medicine and molecular imaging. 2021.

29. Okamoto S, Thieme A, Allmann J, D'Alessandria C, Maurer T, Retz M, et al. Radiation Dosimetry for ¹⁷⁷Lu-PSMA I&T in Metastatic Castration-Resistant Prostate Cancer: Absorbed Dose in Normal Organs and Tumor Lesions. Journal of Nuclear Medicine. 2017;58(3):445-50.

SUPPLEMENTARY

Study assessments table

Week	Blood analysis	ECG	ctDNA	OPD visit	^{68}Ga-PSMA PET/CT*	^{18}FDG PET/CT	^{177}Lu-PSMA Treatment	SPECT/CT	CT*	Questionnaires	Optional Biopsy†
Screening	X	X	X	X	X	X					
Baseline										X	X
0	X						X	X			
1	X							X			
2	X	X	X	X							
4	X		X	X							
6	X			X			X	X		X	
8	X			X							
10	X		X	X	X	X					X
12	X			X			X	X		X	
15	X			X							
18	X			X			X	X		X	
21	X			X							
24	X		X	X							
Assessments after completion of therapy (in months)											
+3	X		X	X	X					X	
+6	X		X	X					X	X	
At PD	X		X	X					X	X	X

*Contrast enhanced neck-, chest- and abdominal CT

† Biopsies are an optional part of this clinical study, participation is not required.

Visits after 6 months after therapy consists of passive follow-up every 3 months. This consist of OPD visits, with related assessments.

Abbreviations: ^{18}F: Fluor-18, ^{68}Ga: Gallium-68, ^{177}Lu: Lutetium-177, ECG: electrocardiogram, CT: computed tomography, ctDNA: circulating tumor DNA, PET: positron emission tomography, PSMA: prostate-specific membrane antigen, SPECT: single-photon emission computerized tomography, PD: progressive disease

Chapter 7

Effect of androgen deprivation therapy on PSMA-ligand uptake in patients with recurrent or metastatic salivary duct carcinoma: an imaging study protocol.

Maike J.M. Uijen, Wim van Boxtel, Chantal M.L. Driessen, Martin Gotthardt, James Nagarajah, Carla M.L. van Herpen

Study protocol from 2019.

ABSTRACT

Background

For recurrent or metastatic (R/M) salivary duct carcinoma (SDC) patients relatively limited systemic treatment options exist. As the majority of SDC tumors are androgen receptor-positive (AR+), androgen deprivation therapy (ADT) is given as first-line treatment. Prostate-specific membrane antigen (PSMA) is target for treatment in prostate cancer. The presence of PSMA can be visualized ^{68}Ga-PSMA-11 PET/CT. A prior ^{68}Ga-PSMA-11 PET/CT imaging study indicated that approximately 40% of SDC patients also have sufficient PSMA expression in the tumors for potential PSMA-targeted therapy. Since research in prostate cancer indicated that ADT treatment can increase PSMA-ligand uptake on PSMA PET/CT, this imaging study will investigate if ADT can also increase PSMA-ligand uptake in SDC patients.

Methods

This single-centre imaging study in R/M AR+ SDC patients consists of repeated ^{68}Ga-PSMA PET/CT and ^{18}FDG PET/CT scans. Patients who intend to start with ADT (as standard treatment) are eligible for this study. ^{68}Ga-PSMA PET/CT and ^{18}FDG PET/CT scans will be performed before the start of ADT and 3 weeks (± 1 week) after the start of ADT. The primary outcome will be the percentage of patients with an ADT induced increase in PSMA-ligand uptake on ^{68}Ga-PSMA PET/CT. Other outcomes include the number of lesions detected by ^{68}Ga-PSMA PET/CT pre- and post ADT, the change in metabolic (^{18}FDG) uptake of tumor lesions pre- and post ADT, and to explore the metabolic and PSMA uptake patterns of SDC disease.

Discussion

This prospective imaging study will assess the effect of ADT on ^{68}Ga-PSMA-ligand uptake in R/M AR+ SDC patients. In case of an increased ^{68}Ga-PSMA-ligand uptake, as previously demonstrated in prostate cancer, this might have therapeutical consequences for SDC patients.

Trial registration

This study is registered at Clinicaltrial.gov January 2, 2020 (NCT04214353).

BACKGROUND

Salivary duct carcinoma (SDC) is an aggressive histological subtype of salivary gland cancer, representing about 4-10% of all cases (1, 2). Approximately half of all SDC patients will develop a recurrence and/or metastases (R/M) over time (3). Previous research indicated that at this stage the prognosis without systemic therapy is poor, with a median survival of only 5 months with the best supportive care (4). Fortunately, much progress has been made in the field of systemic treatment options for R/M SDC patients in recent decades. Systemic therapy in R/M SDC disease is driven by the presence of molecular targets such as the androgen receptor (AR) and Human Epidermal growth factor Receptor 2 (HER2) on the tumors. Since the vast majority (>90%) of SDC tumors are AR+, androgen deprivation therapy (ADT) is often given as first-line treatment. ADT can consist of either monotherapy with bicalutamide tablets 150 mg once daily, or combined therapy with a LHRH analogue given as subcutaneous injections and bicalutamide tablets (in the combined setting often dosed as 50mg or 80mg once daily). Approximately 40% of R/M SDC patients respond to combined ADT (5). Treatment with bicalutamide monotherapy has shown less favourable results in terms of efficacy (response rate approximately 18%) (4), but may have the advantage of libido and sexual potency retention (6). Furthermore, because 21-44% of SDC tumors are HER2+, the effect of HER2-targeted therapy has been explored and also showed high efficacy (7, 8). The combination of trastuzumab and docetaxel resulted in a response rate of 70% (8). Despite these treatment options, survival in R/M SDC patients remains limited. Therefore new treatment strategies are urgently needed.

Prostate-Specific Membrane Antigen (PSMA)

PSMA is a transmembrane protein expressed by certain healthy tissues (e.g. prostate, duodenum, and proximal renal tubules), and malignant tissue (e.g. prostate cancer tumor cells and the neovasculature of many other solid malignancies) (9). Results on PSMA expression of healthy salivary gland tissue are conflicting (9-12).

In recent years, PSMA has evolved as an interesting target for both diagnosis and therapy in prostate cancer (13). PSMA-ligands can be labelled with radionuclides such as Gallium-68 (^{68}Ga), which is used to visualize PSMA expression in vivo by a ^{68}Ga-PSMA PET/CT scan. For therapeutic purposes, PSMA targeting molecules can be coupled with radionuclides such as Lutetium-177 (^{177}Lu; beta-emitter) or Actinium-225 (^{225}Ac; alfa-emitter), by which a cytotoxic load can be guided specifically to the tumors. In metastatic castration-resistant prostate cancer (mCRPC), there are multiple reports where patients have been effectively treated with PSMA-targeted therapy (14-19).

PSMA a Therapeutic Target for SDC?

In the ^{68}Ga-PSMA PET/CT scans of mCRPC patients, healthy salivary glands also show high PSMA-ligand uptake. This raised the hypothesis that PSMA might also be present in salivary

gland cancer and could potentially pose as a therapeutic target. Therefore, we conducted a prospective imaging study to systematically investigate ^{68}Ga-PSMA-uptake in salivary gland cancer patients. Four of the 10 SDC patients (40%) showed relevant PSMA-ligand uptake (defined as tumor/liver ratio of >1) of the tumors (unpublished own results).

Additionally, ^{68}Ga-PSMA-uptake was correlated to immunohistochemical PSMA expression of resected or biopsied primary tumors and metastases. Interestingly, immunohistochemical PSMA expression did not reliably predict ^{68}Ga-PSMA-uptake in SDC patients, as some patients without immunohistochemical PSMA expression showed good ^{68}Ga-PSMA-ligand uptake. Together with the conflicting immunohistochemical results of healthy salivary glands, this suggests that at least part of the ^{68}Ga-PSMA-uptake in the salivary glands and salivary gland cancers might be non-specific uptake.

Upregulation of PSMA by ADT in Prostate Cancer

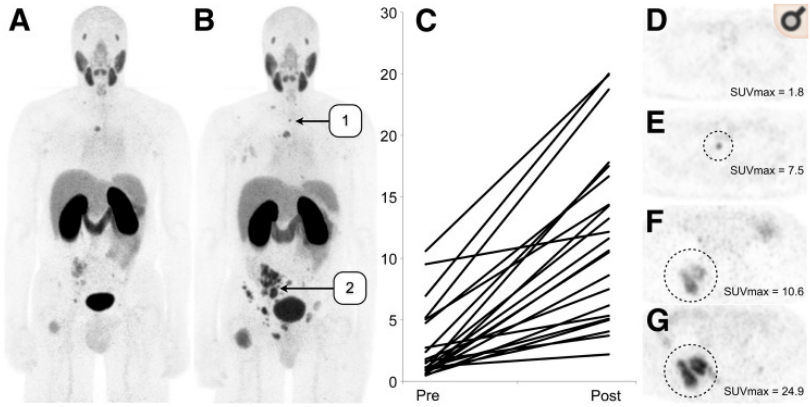
As described above only 40% of the SDC patients have sufficient PSMA-ligand uptake to explore PSMA-targeted therapy. Interestingly, in prostate cancer preclinical and clinical studies indicated that ADT can increase PSMA expression and PSMA-ligand uptake (20-24).

The first clinical result was published by Hope *et al*., an mCRPC patient who showed an increase in ^{68}Ga-PSMA-ligand uptake 4 weeks after the start of ADT (leuprolide acetate and bicalutamide), compared to baseline, see Figure 1 (21).

Furthermore, a clinical pilot study imaged PSMA expression using ^{99m}Tc-MIP-1404 (^{99m}Tc-labeled PSMA inhibitor). Four mCRPC patients underwent baseline SPECT scans prior to ADT (abiraterone or enzalutamide). The SPECT scans were repeated 2 and 12 weeks after the start of ADT. Two weeks after initiation of ADT the PSMA scans showed the same or more lesions with higher intensity compared to baseline. However, after 12 weeks, there was a considerable reduction of the PSMA-ligand uptake in the lesions. This finding is consistent with current literature; there seems to be consensus on increased PSMA expression due to short-term ADT, while the effect of long-term ADT on PSMA-ligand uptake on imaging is debated (24).

Objective

The aim of this imaging study is to explore if ADT (a standard-of-care therapy in R/M SDC) induces an increase of ^{68}Ga-PSMA-ligand uptake on ^{68}Ga-PSMA PET/CT, as has previously been shown in prostate cancer patients. If this is the case, this may have consequences for the proportion of SDC patients that could qualify for PSMA-targeted therapy and furthermore also in patients with baseline high ligand uptake, even higher PSMA-ligand uptake could potentially improve the efficacy of PSMA-targeted therapies.



(A and B) Coronal maximum-intensity projections of patient with castration-sensitive metastatic prostate cancer imaged using ^{68}Ga-PSMA-11 before ADT (A) and after ADT (B) demonstrate marked increase in uptake in lesions. (C) Each visualized lesion demonstrated increased uptake, averaging more than 7 times the initial uptake. (D and E) Numerous lesions (13 of 22) were visualized only on posttreatment imaging, as exemplified by the upper thoracic osseous metastasis seen on these axial PET images. (F and G) Other lesions increased in size and had increased uptake on posttreatment imaging, as exemplified by the lesion seen on these axial PET images.

Figure 1: Example of increased ^{68}Ga-PSMA-ligand uptake after ADT in a prostate cancer patient.

This research was originally published in *JNM*. Hope et al. 68Ga-PSMA-11 PET Imaging of Response to Androgen Receptor Inhibition: First Human Experience. 2017;58(1):81-4. © SNMMI.

Abbreviations: ^{68}Ga: Gallium-68, ADT: androgen deprivation therapy, PET: Positron emission tomography, PSMA: Prostate-Specific Membrane Antigen, SUV: standardized uptake value.

METHODS

Study design

This is an imaging study in R/M AR+ SDC patients who will start ADT (as standard treatment). This study is a performed at a single centre (Radboud university medical centre, Nijmegen, the Netherlands). Imaging consists of repeated ^{68}Ga-PSMA PET/CT and ^{18}FDG PET/CT. Baseline imaging (pre-ADT) will be performed before the start of ADT. After the start of ADT, post-ADT imaging will be performed after 3 weeks (± 1 week).

ADT can consist of either monotherapy with bicalutamide (tablets 150 mg, once daily) or a combination of a luteinizing hormone-releasing hormone (LHRH) analogue (e.g. goserelin 10.8 mg subcutaneously, every 12 weeks) with bicalutamide (50 mg, once daily) (4). The type of ADT treatment is at the discretion of the patient, after careful counselling by the treating physician.

The study protocol was approved by the Medical Review Ethics Committee Arnhem-Nijmegen, The Netherlands and is registered at Clinicaltrial.gov (NCT04214353).

Participants eligibility

Inclusion criteria: patients with locally advanced, recurrent or metastatic AR+ SDC, who intend to start ADT, after this has been recommended by the treating physician as standard-of-care treatment. Patients must have at least one lesion with a diameter of ≥ 1.5 cm. Furthermore, patients must have the ability to provide written informed consent and be ≥ 18 years of age.

Exclusion criteria: patients with a contra-indication for PET imaging, or impaired renal function (MDRD <30 ml/min/1,73m^2) or impaired liver function (AST and ALT ≥ 2.5 x ULN or ≥5 x ULN for patients with liver metastases).

Participants recruitment

SDC patients from the population of salivary gland cancer patients at the Department of Medical Oncology of the Radboudumc who meet the inclusion criteria will be asked to participate in this study. Furthermore, the study will be brought to the attention of the Dutch Working Group for Head and Neck Tumors (NWHHT), a national network of head-and-neck cancer centres in the Netherlands.

Study procedures

- Screening visit: during the screening visit participants will be asked for written informed consent and background data, such as birth date, past medical history, current medication use and allergies.
- Review of electronic patient file: participants will be asked for their consent to review their patient file in order to obtain specific background data, such as type of surgery performed and date of surgery.
- Laboratory tests: if not performed within the last 14 days before the ^{68}Ga-PSMA PET/CT, AST, ALT and creatinine levels will be tested.
- ^{68}Ga-PSMA PET/CT imaging: the participants will be injected with ^{68}Ga-PSMA with a dose of about 2 MBq/kg body weight and PET/CT scanning (from groin to skull top, 3 minutes per bed position, 4-5 bed positions) will be performed 45-60 minutes post-injection.
- ^{18}FDG PET/CT imaging: the participants will be injected with ^{18}FDG with a dose of about 2.1 MBq/kg body weight and PET/CT scanning (from groin to skull top, 3 minutes per bed position, 4-5 bed positions) will be performed 45-60 minutes post-injection.

An overview of the study procedures is presented in Figure 2.

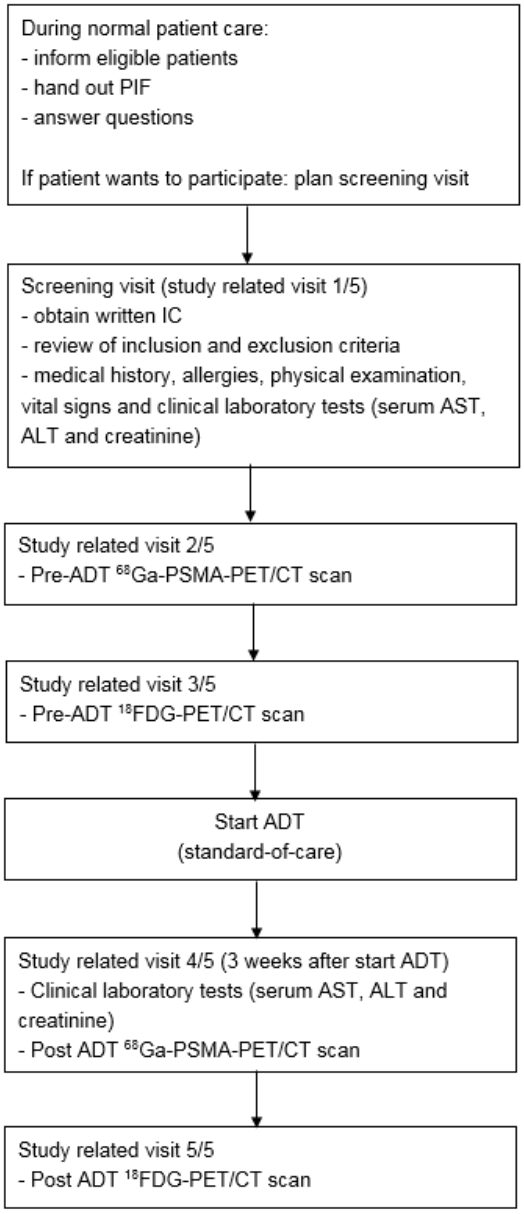


Figure 2: Study flowchart

Abbreviations: ^{68}Ga: Gallium-68, ^{18}F: Fluor-18, ADT: androgen deprivation therapy, ALT: Alanine aminotransferase, AST: Aspartate aminotransferase, CT: Computed tomography, IC: informed consent, PET: Positron emission tomography, PIF: patient information folder, PSMA: Prostate-Specific Membrane Antigen

Follow-up

The follow-up will consist of 12 months. During this period, patients will receive regular diagnostic CT-scans every 2-4 months (as this is standard of care). These CT-scans serve as a monitor for ADT treatment response in daily practice. Thus, during this follow-up period, there will be no additional study-related visits or assessments. The rationale for the follow-up is to evaluate if findings on FDG-PET/CT correspond with CT-findings during follow-up (this is an additional explorative endpoint as described below).

Primary Outcome

The percentage of patients with an ADT induced increase in PSMA-ligand uptake on ^{68}Ga-PSMA-PET/CT is defined as the primary outcome. PSMA-ligand uptake will be expressed in standardized uptake value (SUV). SUV will be calculated according to the following formula: measured activity concentration [MBq/mL] * body weight [g]) / injected activity [MBq].

Secondary Outcomes

- Comparison of SUV in ^{68}Ga-PSMA PET/CT between lesions before and after the initiation of ADT.
- Comparison of SUV in ^{18}FDG-PET/CT between lesions before and after the initiation of ADT.
- To establish whether new metastatic lesions are detected by ^{68}Ga-PSMA PET/CT imaging after initiation of ADT.
- To explore the metabolic and PSMA-ligand uptake patterns of SDC disease.
- To establish the diagnostic added value of ^{68}Ga-PSMA-PET and ^{18}FDG-PET compared to standard imaging.
- To correlate the SUV in ^{68}Ga-PSMA PET/CT to the degree of immunohistochemical PSMA expression.
- To correlate findings on FDG PET/CT scans to CT-findings during follow-up.

Power calculation

Because this research is explorative, no sample size calculations can be performed. We decided to include 14 patients in this study based on the following reasoning:
The sample size is chosen such that for a minimum clinically relevant fraction of patients with an increased PSMA-ligand uptake of 20%, the probability that at least one of the patients in the sample has an increased uptake, is at least 0.95.
If the sample size is equal to 14:
Prob(at least one patient with increased uptake) =
= 1 – Prob (no patient with an increased uptake) =
= 1 – Prob (all patients have no increased uptake) =
= 1 – Prob (a patient has no increased uptake)^14 =
= 1 – 0.80^14 = 0.96.
For 13 patients this probability is below 0.95. So, a sample size of 14 patients is sufficient.

Thus, if none of 14 patients would have an ADT mediated uptake of PSMA ligands on ^{68}Ga-PSMA PET/CT, then we would conclude that the effect of ADT on PSMA is not relevant from a clinical point of view.

Statistical analysis

Pairwise difference between lesions before and after the initiation of ADT will be computed. The variation in the differences will be illustrated with a boxplot. Further, the mean of the differences is presented together with a 95% confidence interval based on the assumption of normality of the differences. If the normality assumption is expected not to be true (based on the form of the boxplot and possibly a histogram), the median and the interquartile range will be given.

If the differences are likely to follow a normal distributed, a paired sample t-test will be used to test the null hypothesis of equal means before and after the initiation of ADT. Otherwise we will use a non-parametric test (the Wilcoxon signed-rank test).

DISCUSSION

In summary, SDC is an aggressive subtype of salivary gland cancer. In R/M SDC first-line systemic therapy often consists of ADT. But despite current treatment options, survival in R/M SDC patients is limited. Therefore, new treatment strategies are warranted. ^{177}Lu-PSMA radioligand therapy is a promising new treatment approach that targets PSMA. Several studies have shown positive results for prostate cancer patients. With a previous imaging study, we assessed if SDC patients could be candidates for ^{177}Lu-PSMA radioligand therapy. Unfortunately, 60% of the SDC patients showed a low ^{68}Ga-PSMA-ligand uptake on ^{68}Ga-PSMA PET/CT, subsequently, these patients are unlikely to respond to ^{177}Lu-PSMA radioligand therapy and will therefore not be considered as candidates for ^{177}Lu-PSMA radioligand therapy. Since preclinical and clinical studies in prostate cancer indicated that ADT could increase PSMA expression, this imaging study will investigate if ADT also increases ^{68}Ga-PSMA-ligand uptake in R/M SDC patients. Furthermore, also in SDC patients with relatively high baseline ^{68}Ga-PSMA-ligand uptake (other 40% of SDC patients), even higher uptake could potentially improve the efficacy of PSMA-targeted therapy.

REFERENCES

1. Bjorndal K, Krogdahl A, Therkildsen MH, Overgaard J, Johansen J, Kristensen CA, et al. Salivary gland carcinoma in Denmark 1990-2005: a national study of incidence, site and histology. Results of the Danish Head and Neck Cancer Group (DAHANCA). Oral oncology. 2011;47(7):677-82.
2. Otsuka K, Imanishi Y, Tada Y, Kawakita D, Kano S, Tsukahara K, et al. Clinical Outcomes and Prognostic Factors for Salivary Duct Carcinoma: A Multi-Institutional Analysis of 141 Patients. Annals of surgical oncology. 2016;23(6):2038-45.
3. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.
4. Boon E, van Boxtel W, Buter J, Baatenburg de Jong RJ, van Es RJJ, Bel M, et al. Androgen deprivation therapy for androgen receptor-positive advanced salivary duct carcinoma: A nationwide case series of 35 patients in The Netherlands. Head & neck. 2018;40(3):605-13.
5. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.
6. Kolvenbag GJ, Blackledge GR, Gotting-Smith K. Bicalutamide (Casodex) in the treatment of prostate cancer: history of clinical development. The Prostate. 1998;34(1):61-72.
7. van Boxtel W, Boon E, Weijs WLJ, van den Hoogen FJA, Flucke UE, van Herpen CML. Combination of docetaxel, trastuzumab and pertuzumab or treatment with trastuzumab-emtansine for metastatic salivary duct carcinoma. Oral oncology. 2017;72:198-200.
8. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.
9. Chang SS, Reuter VE, Heston WD, Bander NH, Grauer LS, Gaudin PB. Five different anti-prostate-specific membrane antigen (PSMA) antibodies confirm PSMA expression in tumor-associated neovasculature. Cancer research. 1999;59(13):3192-8.
10. Wolf P, Freudenberg N, Buhler P, Alt K, Schultze-Seemann W, Wetterauer U, et al. Three conformational antibodies specific for different PSMA epitopes are promising diagnostic and therapeutic tools for prostate cancer. The Prostate. 2010;70(5):562-9.
11. Silver DA, Pellicer I, Fair WR, Heston WD, Cordon-Cardo C. Prostate-specific membrane antigen expression in normal and malignant human tissues. Clinical cancer research : an official journal of the American Association for Cancer Research. 1997;3(1):81-5.
12. Troyer JK, Beckett ML, Wright GL, Jr. Detection and characterization of the prostate-specific membrane antigen (PSMA) in tissue extracts and body fluids. International journal of cancer. 1995;62(5):552-8.
13. Eiber M, Maurer T, Souvatzoglou M, Beer AJ, Ruffani A, Haller B, et al. Evaluation of Hybrid (6)(8)Ga-PSMA Ligand PET/CT in 248 Patients with Biochemical Recurrence After Radical

Prostatectomy. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2015;56(5):668-74.

14. Kratochwil C, Bruchertseifer F, Rathke H, Hohenfellner M, Giesel FL, Haberkorn U, et al. Targeted alpha-Therapy of Metastatic Castration-Resistant Prostate Cancer with (225) Ac-PSMA-617: Swimmer-Plot Analysis Suggests Efficacy Regarding Duration of Tumor Control. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2018;59(5):795-802.

15. Hofman MS, Violet J, Hicks RJ, Ferdinandus J, Thang SP, Akhurst T, et al. [(177)Lu]-PSMA-617 radionuclide treatment in patients with metastatic castration-resistant prostate cancer (LuPSMA trial): a single-centre, single-arm, phase 2 study. The Lancet Oncology. 2018;19(6):825-33.

16. Rahbar K, Ahmadzadehfar H, Kratochwil C, Haberkorn U, Schafers M, Essler M, et al. German Multicenter Study Investigating 177Lu-PSMA-617 Radioligand Therapy in Advanced Prostate Cancer Patients. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2017;58(1):85-90.

17. Heck MM, Retz M, D'Alessandria C, Rauscher I, Scheidhauer K, Maurer T, et al. Systemic Radioligand Therapy with (177)Lu Labeled Prostate Specific Membrane Antigen Ligand for Imaging and Therapy in Patients with Metastatic Castration Resistant Prostate Cancer. The Journal of urology. 2016;196(2):382-91.

18. Fendler WP, Reinhardt S, Ilhan H, Delker A, Boning G, Gildehaus FJ, et al. Preliminary experience with dosimetry, response and patient reported outcome after 177Lu-PSMA-617 therapy for metastatic castration-resistant prostate cancer. Oncotarget. 2017;8(2):3581-90.

19. Yadav MP, Ballal S, Tripathi M, Damle NA, Sahoo RK, Seth A, et al. (177)Lu-DKFZ-PSMA-617 therapy in metastatic castration resistant prostate cancer: safety, efficacy, and quality of life assessment. European journal of nuclear medicine and molecular imaging. 2017;44(1):81-91.

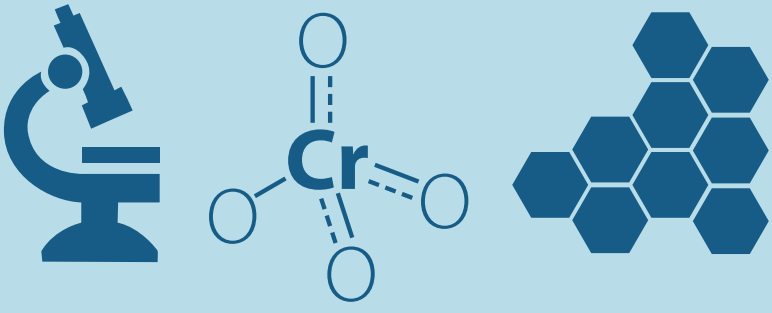
20. Evans MJ, Smith-Jones PM, Wongvipat J, Navarro V, Kim S, Bander NH, et al. Noninvasive measurement of androgen receptor signaling with a positron-emitting radiopharmaceutical that targets prostate-specific membrane antigen. Proceedings of the National Academy of Sciences of the United States of America. 2011;108(23):9578-82.

21. Hope TA, Truillet C, Ehman EC, Afshar-Oromieh A, Aggarwal R, Ryan CJ, et al. 68Ga-PSMA-11 PET Imaging of Response to Androgen Receptor Inhibition: First Human Experience. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2017;58(1):81-4.

22. Meller B, Bremmer F, Sahlmann CO, Hijazi S, Bouter C, Trojan L, et al. Alterations in androgen deprivation enhanced prostate-specific membrane antigen (PSMA) expression in prostate cancer cells as a target for diagnostics and therapy. EJNMMI research. 2015;5(1):66.

23. Miyamoto DT, Lee RJ, Stott SL, Ting DT, Wittner BS, Ulman M, et al. Androgen receptor signaling in circulating tumor cells as a marker of hormonally responsive prostate cancer. Cancer discovery. 2012;2(11):995-1003.

24. Afshar-Oromieh A, Debus N, Uhrig M, Hope TA, Evans MJ, Holland-Letz T, et al. Impact of long-term androgen deprivation therapy on PSMA ligand PET/CT in patients with castration-sensitive prostate cancer. European journal of nuclear medicine and molecular imaging. 2018;45(12):2045-54.


Cr
O
O
O
O

Part 3

Increasing general knowledge of Salivary Gland Cancer

Chapter 8

Results of histopathological revisions of major salivary gland neoplasms in routine clinical practice.

Sam T.H. Reerds*, Maike J.M. Uijen*, Adriana C.H. van Engen-van Grunsven, Henri A.M. Marres, Carla M.L. van Herpen, Jimmie Honings.

* These authors contributed equally

Journal of clinical pathology, 2022. ***0****: 1-5.*

ABSTRACT

Aims

Salivary gland neoplasms are rare and are characterized by overlapping histopathological aspects. Therefore, the assessment of the correct histopathological diagnosis can be challenging. This study evaluated the frequency of pathology consultations and revisions for salivary gland neoplasms during routine clinical practice in the Netherlands. Furthermore, the concordance and discordance rates of these revisions are presented.

Methods

The Dutch Pathology Registry (PALGA) was searched for patients that underwent a resection of a major salivary gland neoplasm between 2006 and 2016. Frequencies of pathology consultations and revisions are presented and, to calculate the rates of concordance and discordance, the results of the initial histopathological review were compared to the results of the revision.

Results

Between 2006 and 2016, 13,441 major salivary gland neoplasms were resected in the Netherlands. 90% (n=12,082) of these tumours were diagnosed as benign and 10% (n=1,359) as malignant. The initial pathologist requested a consultation in 3.3% of resections (n=439). Revision of the histopathological specimen was performed in 2.6% (n=350) of cases. Revisions were discordant in 8.3%; including 5.8% of the initially benign diagnosed lesions reclassified as malignant by the second expert pathologist and 8% of the revised malignant tumours that underwent a subtype change.

Conclusions

The number of discordant histopathological revisions (8.3%) emphasizes the complexity of the histopathological diagnosis of salivary gland neoplasms. An increase in consultations may improve the accuracy of the initial diagnosis and thus treatment in salivary gland tumours while lowering the need for revisions and the number of discordant revisions.

BACKGROUND

Salivary gland tumours are amongst the most histopathologically diverse neoplasms. The WHO classification of 2017 contains 11 benign and 22 malignant salivary gland tumour types (1). Malignant salivary gland tumours are rare. In the Netherlands (approximately 17 million inhabitants), 150-200 patients are diagnosed with salivary gland cancer each year in the last decade (2).Given the wide variety of tumours, the overlap in morphology, and the rarity of specific tumour types, assessment of the correct histopathological diagnosis can be challenging, even for experienced pathologists.

In case of doubt or an inconclusive diagnosis during the initial review, the pathologist can consult a colleague before finalizing the conclusion. These consultations are performed mainly by experienced salivary gland pathologists. On the other hand, a histopathological revision is mostly requested later in the course of the disease by one of the treating physicians. The reason for histopathological revision can be a discrepancy between the initial diagnosis and the clinical course of the disease, or due to referral to another centre in which it might be a standard procedure to perform a reassessment of all performed diagnostics, or to determine if specific targets for targeted therapy are present. A revision might reveal a different tumour subtype or even a different dignity (e.g., benign to malignant or malignant to benign). A summary of the definitions of consultations and revisions is provided in table 1.

Table 1: Definition of consultation and revision

	Requested by	Performed by	Reason for request	Time interval
Consultation	Pathologist	Experienced salivary gland pathologist	- in case of doubt or inconclusive histopathologic diagnosis	Within days/weeks after the initial histopathological assessment
Revision	Physician	Experienced salivary gland pathologist	- discrepancy between initial histopathological diagnosis and tumour's clinical behaviour - after referral to a different hospital (e.g. specialized centre), histopathology reassessment can be a standard procedure.	At a later stage in the course of the disease

Change of dignity can have therapeutic consequences, and in malignant salivary gland tumours, an altered subtype diagnosis might also have therapeutic consequences. Elective neck dissection, for instance, is not necessary for low-grade carcinoma but advocated for high-grade salivary gland cancer, and adjuvant radiotherapy is not routinely considered for benign tumours, whereas it is often performed after the resection of a malignant tumour.

Also, adjuvant hormonal therapy might be considered in patients with androgen receptor-positive salivary duct carcinoma (3). And, in the palliative setting, it is vital to have a correct histopathological diagnosis, as systemic therapeutic options in the palliative setting differ among different subtypes (4). Furthermore, the histopathological diagnosis might have consequences for the patient's prognosis, as it is well known that high-grade histology is a strong negative predictor for nodal metastases, recurrent disease, and distant metastasis (5, 6).

Previous studies have shown that the histopathological reassessment of malignant salivary gland tumours is associated with a change of tumour subtype, tumour origin, or even the change from a benign neoplasm to a malignant neoplasm (7-10). Some of these studies revised benign as well as malignant tumours, while others only revised malignant tumours. Nonetheless, none of these studies reported the results of pathology consultations and revisions in routine clinical practice.

The purpose of this study was to evaluate the frequency of histopathological consultations and the frequency and outcomes of histopathological revisions of salivary gland neoplasms in routine clinical practice.

PATIENTS AND METHODS

Data collection

The database of the nationwide network and registry of histo- and cytopathology in the Netherlands (PALGA) (11) was searched for patients who underwent a salivary gland resection between 1-1-2006 and 31-12-2016. Data regarding patient age at the time of resection, date of resection, side of the tumour, type of centre where the histopathological result was evaluated, and the histopathological result and conclusion itself were anonymously collected from the database.

In the Netherlands, head & neck oncological care is centralized in 14 specialized centres: eight head & neck oncological centres and six affiliated head & neck oncological centres. Therefore, hospitals were categorized as' head & neck oncological centres', 'affiliated head & neck oncological centres', or 'general hospitals'.

As we collected data from 2006-2016, the histopathological diagnoses enlisted in the 2005 WHO classification for salivary gland tumours were included (12). The diagnosis of (mammary analogue) secretory carcinoma was added because this diagnosis was first described in 2010 (13) and has been accepted and used by pathologists thereafter. Metastatic tumours from other sites, lymphomas, and lesions outside of the major salivary glands were excluded. The diagnosis in the initial histopathological report was compared with those from the revisional

report in order to estimate the concordance (agreement) or discordance (conflicting) rates.

Statistical methods

Descriptive measures were summarized as means and their standard deviation. Statistical analyses were performed using SPSS v25.0.

Approval

The current study was approved by the scientific and privacy committee of the PALGA database: "the nationwide network and registry of histo- and cytopathology in the Netherlands" (11). Review by a medical ethical committee was not required by Dutch law due to the anonymous data collection.

RESULTS

Patient and tumour characteristics

The initial search for patients that had undergone a salivary gland resection yielded 24,164 results in the PALGA database. After the exclusion of 10,723 patients, 13,224 patients remained that underwent 13,441 primary resections of at least one of the major salivary glands between 1-1-2006 and 31-12-2016 in the Netherlands (figure 1).

Of all patients, 6,408 were male (48.5%) and 6,816 were female (51.5%). The mean age at resection was 54.6 years (SD: 15.4). Left and right-sided resections were distributed equally (46.3% left, 46.7% right, 7% unknown side). In 217 patients, more than one salivary gland was resected. Overall, major salivary gland tumours were benign in 90% (n=12,082) and malignant in 10% (n=1,359).

Parotid gland resections were most frequently performed (12,364 resections, 92%), followed by 1,049 submandibular (7.8%) and 28 sublingual gland resections (0.2%). The majority of the resected parotid gland tumours were benign (n=11,247, 91%), while 1,117 (9%) were malignant. Submandibular gland resections more frequently yielded malignant neoplasms (n=218, 20.8%), whereas the sublingual gland had the highest incidence of malignant tumours; 24 of 28 lesions (85.7%). The distribution of resected histopathological subtypes throughout the years is shown in the supplementary material.

Benign salivary gland tumours were most often resected in general hospitals (47.8%), while 31.5% were removed in specialized head & neck oncological centres and 20.7% in affiliated head & neck oncological centres. Malignant tumours were most frequently resected in specialized head & neck oncological centres (60.6%), followed by 22.9% in general hospitals and 16.5% in affiliated head & neck oncological centres.

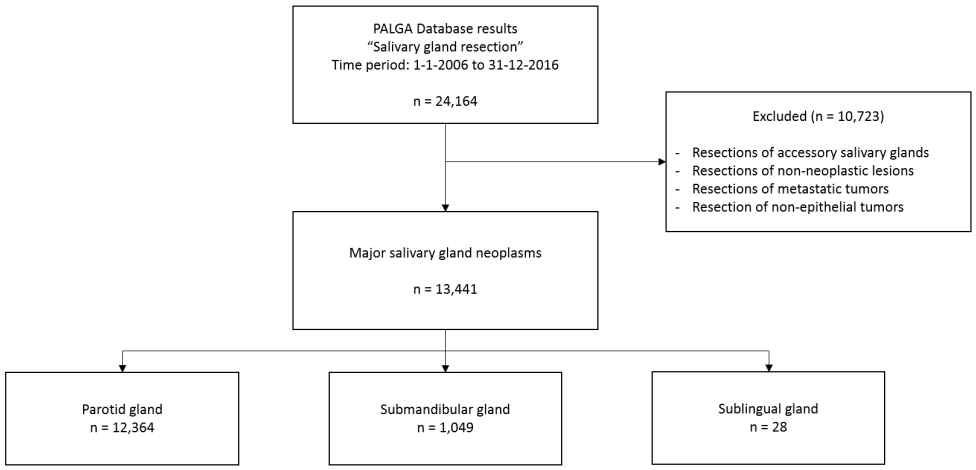


Figure 1: Flow diagram of the search strategy and exclusion process of patients that underwent resection of one of the major salivary glands.

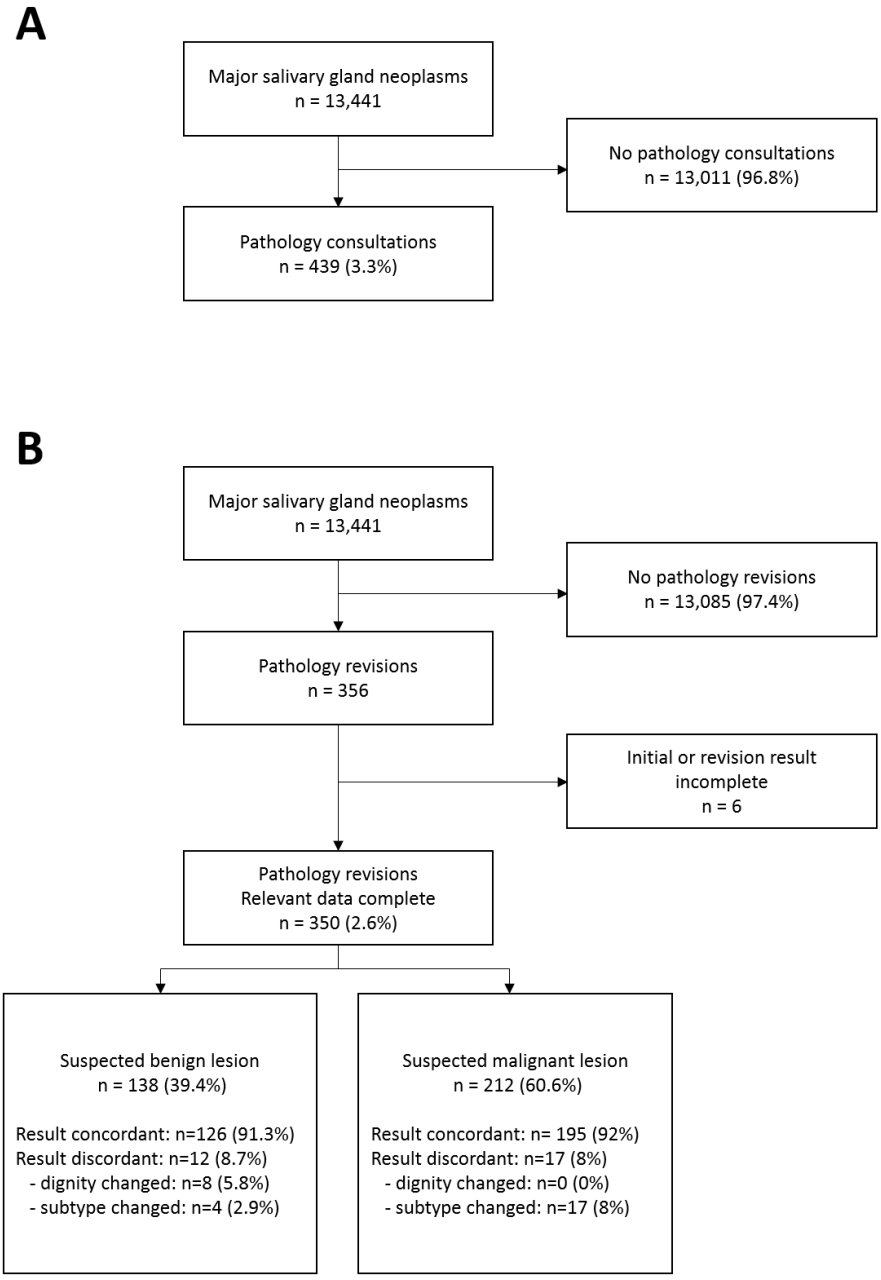


Figure 2: The number of performed consultations (**A**) and specific concordant and discordant rates for revisions (**B**) of benign and malignant salivary gland tumours.

Pathology consultations and revisions

Of all major salivary gland neoplasms evaluated (n=13,441), the initial pathologist requested a consultation in 3.3% of all cases (n=439), figure 2. These consultations were mainly requested by pathologists from general hospitals (54.3%), but also by pathologists from affiliated head & neck oncological centres (22.5%), and head & neck oncological centres (19.0%), and unknown in 4.2% of all consultations.

In 2.6% of all cases, a histopathological revision was asked for by one of the treating clinicians; 321 revisions (91.7%) were concordant, whereas 29 revisions (8.3%) were discordant.

In 39.4% of all revisions performed, the initial histopathological conclusion was a benign salivary gland tumour. The result of the revision was discordant in 8.7%; in 5.8 %, the diagnosis changed from benign to malignant (dignity change), and in 2.9% there was a subtype change.

The benign diagnoses pleomorphic adenoma (n=3) and basal cell adenoma (n=3) were most often associated with a change from benign to malignant. The specific benign diagnoses that changed after revision are shown in table 2.

Table 2: Revisions of benign tumours

Benign tumours that were reclassified as malignant after revision (N=8)		
No	*Initial diagnosis*	*Diagnosis after revision (final diagnosis)*
1	Basal cell adenoma	Adenoid cystic carcinoma
2	Basal cell adenoma	Basal cell adenocarcinoma
3	Basal cell adenoma	Basal cell adenocarcinoma
4	Pleomorphic adenoma	Carcinoma ex pleomorphic adenoma
5	Pleomorphic adenoma	Adenocarcinoma NOS
6	Pleomorphic adenoma	Adenoid cystic carcinoma
7	Sebaceous adenoma	Acinic cell carcinoma
8	Sialadenosis	Salivary duct carcinoma
Benign tumours where the subtype was changed after revision (N=4)		
No	*Initial diagnosis*	*Diagnosis after revision (final diagnosis)*
1	Myoepithelioma	Basal cell adenoma
2	Pleomorphic adenoma	Myoepithelioma
3	Pleomorphic adenoma	Basal cell adenoma
4	Pleomorphic adenoma	Myoepithelioma

Abbreviation: NOS: not otherwise specified

The majority of all revisions were performed for malignant tumours (60.6%). The diagnosis was discordant in 8% of the cases; in all these cases the subtype changed.

The malignant diagnosis that most often led to a subtype change after the revision was adenocarcinoma NOS (n=6), which proved to be a salivary duct carcinoma in three cases. The specific malignant diagnoses that changed are shown in table 3.

Table 3: Revisions of malignant tumours

Malignant tumours that were reclassified as benign after revision (N=0)		
Not applicable		
Malignant tumours where the subtype was changed after revision (N=17)		
No	*Initial diagnosis*	*Diagnosis after revision (final diagnosis)*
1	Acinic cell carcinoma	Cystadenocarcinoma
2	Adenocarcinoma NOS	Salivary duct carcinoma
3	Adenocarcinoma NOS	Mucoepidermoid carcinoma
4	Adenocarcinoma NOS	Basal cell adenocarcinoma
5	Adenocarcinoma NOS	Salivary duct carcinoma
6	Adenocarcinoma NOS	Salivary duct carcinoma
7	Adenocarcinoma NOS	Polymorphous low-grade adenocarcinoma
8	Metastasis of adenocarcinoma	Carcinoma ex pleomorphic adenoma
9	Adenoid cystic carcinoma	Mucoepidermoid carcinoma
10	Carcinoma ex pleomorphic adenoma	Epithelial-myoepithelial carcinoma
11	Carcinoma ex pleomorphic adenoma	Salivary duct carcinoma
12	Carcinosarcoma	Salivary duct carcinoma
13	Large cell carcinoma	Acinic cell carcinoma
14	Large cell carcinoma	Adenocarcinoma NOS
15	Mucoepidermoid carcinoma	Polymorphous low-grade adenocarcinoma
16	Polymorphous low-grade adenocarcinoma	Acinic cell carcinoma
17	Salivary duct carcinoma	Adenocarcinoma NOS

Abbreviation: NOS: not otherwise specified

DISCUSSION

The histopathological evaluation of salivary gland tumours is challenging. This is underlined by the results of our study, in which we evaluated histopathological revisions performed in the Netherlands during routine clinical practice and revealed a relatively high frequency of discordant results of revisions (8.3%).
Two previous studies that revised all malignant salivary gland tumours treated within a specific time frame found even higher rates of discordance of 14% and 25.2% (9, 10). However, these studies were performed in a tertiary care facility and revisions were carried out for every patient that was referred. Therefore, it is difficult to compare these results with ours, as our study evaluated histopathological revisions in routine clinical practice on a nationwide basis. Both these results emphasize the difficulty in assessing the correct histopathological subtype.

One way to increase the rate of initially correct diagnoses and lower the rate of discordant revisions could be to ask for a consultation more frequently. In the current study, only 439 external consultations took place for 13,441 salivary gland resections, which equals a consultation rate of only 3.3%. One of the most recent achievements in pathology is the ability to digitally evaluate a specimen, which might lower practical barriers previously attached to the consultation of a colleague. However, digital evaluation may not always prove sufficient, as the consulting pathologists might need additional material to perform stainings or molecular diagnostics.

Interestingly, pathology revisions in our study frequently led to the reclassification of adenocarcinoma NOS to a different subtype. A recent study observed a decline in the incidence of adenocarcinoma NOS over time and suggested this was due to modern diagnostic classification schemes and new immunohistochemical profiling in salivary gland cancers (14). These authors reassessed tumours previously classified as adenocarcinoma NOS, of which 72% could be reclassified as a more specific entity. Thus, general pathologists should be aware of this and have an even lower threshold to ask for a consultation by a specialized colleague in the case of a suspected adenocarcinoma NOS.

Furthermore, apart from the correct diagnosis, an experienced pathologist might identify clinically relevant tumour characteristics that inexperienced pathologists may not be aware of. An example of this is the growth pattern in adenoid cystic carcinoma (e.g. solid, cribriform, or tubular). This is infrequently mentioned by not specialized pathologists, but a solid growth pattern is a very well-known negative prognostic factor. When identified, it could potentially change the interval of follow-up imaging (15).

Another not necessarily anticipated result of this study was the relatively high proportion of benign salivary gland tumour resections compared to the current literature (16-23). This was the case for each of the individual major salivary glands. Our study showed that 91% of parotid gland tumours were benign, compared with 68-91% of benign parotid gland tumours in previous studies (16-23). Submandibular gland tumours were benign in 79.2% of cases, in contrast to 63-76% in previous studies (17-23). In the group of sublingual gland tumours, 15.7% was benign, compared with 0-14% in literature (17-23). However, these studies are heterogeneous in terms of varying time periods in which the studies were conducted, demographically and ethnically diverse cohorts, and some (16, 18-22) are single-centre cohorts resulting in selection bias. Also, environmental and genetic factors may partly explain the variance in incidence and prevalence of tumours (24). However, when comparing our results to those of a recent nationwide study from Iceland (23), the percentage of benign tumours of the parotid and submandibular glands are highly comparable: 90.8% vs. 91% and 78.4% vs. 76%.

Head and neck cancer care in the Netherlands is centralized in dedicated head & neck oncological centres. This was agreed upon in the late 1980s in an effort to increase the quality of healthcare and reduce costs. Our data, however, shows that 22.9% of the malignant salivary gland tumours were surgically resected in a general hospital. Recent research has confirmed that the centralization of care for rare diseases is beneficial to overall survival (25) and that high-volume centres achieve lower rates of positive surgical margins in the treatment of major salivary gland carcinoma (26). Therefore, we believe it is in the patient's best interest to refer (possible) malignant salivary gland tumours to dedicated head & neck oncological centres.

The study's retrospective nature prevented us from collecting additional data on the neoplasms, such as histopathological tumour grade or information about clinical management and the reason for the revision. Furthermore, in some cases, the PALGA database lacked relevant information about consultations or revisions. For example, six cases that had a revision lacked the conclusion of the initial review or final result, which is why these were excluded from part of the analysis.

Our study reports on the rate of discordance of pathology revisions in routine clinical practice, causing bias because revision is often performed because of a specific (clinical) reason. Therefore, revisions may be more often performed for tumours with clinical behaviour that proves inconsistent with the initial diagnosis or for tumours that have metastasized, as oncologists in tertiary referral centres often perform revisions. It would be of interest to revise a large cohort of malignant and benign salivary gland tumours to analyse the actual rate of discordance. Also, it would have been of interest to investigate how frequently a discordant result had clinical management implications. Unfortunately, we could not assess this due to the absence of clinical information other than the histopathological result.

CONCLUSION

The relatively high number of discordant results of pathology revisions (8.3%) emphasizes the difficulty of the histopathological review of salivary gland neoplasms. Currently, the consultation rate is relatively low. In only 3.3% of all salivary gland neoplasms, a consultation of an experienced salivary gland pathologist is requested. Increasing the use of consultations in salivary gland pathology might improve the accuracy of the initial histopathologic diagnosis in salivary gland tumours.

ACKNOWLEDGEMENTS

We gratefully acknowledge Dr. C.C.H.J. Epskamp-Kuijpers, data advisor at the Dutch Pathology Registry, for her help in acquiring the data.

REFERENCES

1. A.K. El-Naggar JKCC, J.R. Grandis, T. Takata, P.J. Slootweg WHO Classification of Head and Neck Tumours (4th ed.),. ARC, Lyon, France (2017). 2017.
2. IKNL. Nederlandse Kanker Registratie (NKR) / Dutch Cancer Registration [Available from: iknl.nl/nkr-cijfers.
3. van Boxtel W, Locati LD, van Engen-van Grunsven ACH, Bergamini C, Jonker MA, Fiets E, et al. Adjuvant androgen deprivation therapy for poor-risk, androgen receptor-positive salivary duct carcinoma. Eur J Cancer. 2019;110:62-70.
4. Alfieri S, Granata R, Bergamini C, Resteghini C, Bossi P, Licitra LF, et al. Systemic therapy in metastatic salivary gland carcinomas: A pathology-driven paradigm? Oral Oncol. 2017;66:58-63.
5. Haderlein M, Scherl C, Semrau S, Lettmaier S, Uter W, Neukam FW, et al. High-grade histology as predictor of early distant metastases and decreased disease-free survival in salivary gland cancer irrespective of tumour subtype. Head Neck. 2016;38 Suppl 1:E2041-8.
6. Xiao CC, Zhan KY, White-Gilbertson SJ, Day TA. Predictors of Nodal Metastasis in Parotid Malignancies: A National Cancer Data Base Study of 22,653 Patients. Otolaryngol Head Neck Surg. 2016;154(1):121-30.
7. van der Wal JE, Snow GB, van der Waal I. Histological reclassification of 101 intraoral salivary gland tumours (new WHO classification). J Clin Pathol. 1992;45(9):834-5.
8. van der Wal JE, Leverstein H, Snow GB, Kraaijenhagen HA, van der Waal I. Parotid gland tumors: histologic reevaluation and reclassification of 478 cases. Head Neck. 1998;20(3):204-7.
9. Stodulski D, Majewska H, Skalova A, Mikaszewski B, Biernat W, Stankiewicz C. Histological reclassification of parotid gland carcinomas: importance for clinicians. European archives of oto-rhino-laryngology : official journal of the European Federation of Oto-Rhino-Laryngological Societies (EUFOS) : affiliated with the German Society for Oto-Rhino-Laryngology - Head and Neck Surgery. 2016;273(11):3937-42.
10. Bjorndal K, Krogdahl A, Therkildsen MH, Overgaard J, Johansen J, Kristensen CA, et al. Salivary gland carcinoma in Denmark 1990-2005: a national study of incidence, site and histology. Results of the Danish Head and Neck Cancer Group (DAHANCA). Oral oncology. 2011;47(7):677-82.
11. Casparie M, Tiebosch AT, Burger G, Blauwgeers H, van de Pol A, van Krieken JH, et al. Pathology databanking and biobanking in The Netherlands, a central role for PALGA, the nationwide histopathology and cytopathology data network and archive. Cell Oncol. 2007;29(1):19-24.
12. Eveson J, Auclair PL, Gnepp DR, El Naggar AK. . Tumours of the salivary gland. In: Barnes L, Eveson JW, Reichart P, Sidransky D, editors. World Health Organization classification of tumours: pathology and genetics of head and neck tumours. 2005(Lyon; IARC):164.
13. Skalova A, Vanecek T, Sima R, Laco J, Weinreb I, Perez-Ordonez B, et al. Mammary analogue secretory carcinoma of salivary glands, containing the ETV6-NTRK3 fusion gene: a hitherto undescribed salivary gland tumor entity. Am J Surg Pathol. 2010;34(5):599-608.
14. Rooper LM, Mansour M, Yonescu R, Oliai BR, Bishop JA, Westra WH. The Decline of Salivary Adenocarcinoma Not Otherwise Specified as a Tumor Entity: Reclassification Using

Contemporary Immunohistochemical Profiling and Diagnostic Criteria. Am J Surg Pathol. 2020.

15. da Cruz Perez DE, de Abreu Alves F, Nobuko Nishimoto I, de Almeida OP, Kowalski LP. Prognostic factors in head and neck adenoid cystic carcinoma. Oral Oncol. 2006;42(2):139-46.
16. Thackray AC. Salivary gland tumours [abridged]. Proc R Soc Med. 1968;61(11 Pt 1):1089-90.
17. Eveson JW, Cawson RA. Salivary gland tumours. A review of 2410 cases with particular reference to histological types, site, age and sex distribution. J Pathol. 1985;146(1):51-8.
18. Spiro RH. Salivary neoplasms: Overview of a 35-year experience with 2,807 patients. Head & Neck Surgery. 1986;8(3):177-84.
19. Pinkston JA, Cole P. Incidence rates of salivary gland tumors: results from a population-based study. Otolaryngol Head Neck Surg. 1999;120(6):834-40.
20. Bradley PJ, McGurk M. Incidence of salivary gland neoplasms in a defined UK population. British Journal of Oral and Maxillofacial Surgery. 2013;51(5):399-403.
21. Li LJ, Li Y, Wen YM, Liu H, Zhao HW. Clinical analysis of salivary gland tumor cases in West China in past 50 years. Oral oncology. 2008;44(2):187-92.
22. Tian Z, Li L, Wang L, Hu Y, Li J. Salivary gland neoplasms in oral and maxillofacial regions: a 23-year retrospective study of 6982 cases in an eastern Chinese population. International journal of oral and maxillofacial surgery. 2010;39(3):235-42.
23. Aegisdottir AL, Tryggvason G, Jonsdottir AM, Jonasson JG. Salivary gland tumours in Iceland 1986-2015: a nationwide epidemiological analysis over a 30-year time period. Apmis. 2020.
24. Kolonel LN, Altshuler D, Henderson BE. The multiethnic cohort study: exploring genes, lifestyle and cancer risk. Nat Rev Cancer. 2004;4(7):519-27.
25. Weitz J, Koch M, Friess H, Buchler MW. Impact of volume and specialization for cancer surgery. Dig Surg. 2004;21(4):253-61.
26. Bollig CA, Zitsch RP, 3rd. Impact of treating facilities' type and volume in patients with major salivary gland cancer. Laryngoscope. 2019;129(10):2321-7.


O
Cr
O
O
O

Chapter 9

Case report: Two cases of salivary duct carcinoma in workers with a history of chromate exposure.

Imran Seçin, Maike J.M. Uijen, Chantal M.L. Driessen, Carla M.L. van Herpen, Paul T.J. Scheepers

Frontiers in Medicine, *2021*. ***8***: *730403.*

ABSTRACT

Background

Salivary duct carcinoma (SDC), one subtype of the 22 different salivary gland cancers, is a rare malignancy. Risk factors for the development of salivary gland cancer and SDC are largely unknown, although pollution has been described as one of the risk factors. In other cancers, especially in lung cancer, the carcinogenicity of chromium VI (Cr(VI)) is well-known. Here we report on two SDC patients who were occupationally exposed to Cr(VI) and discuss a potential relationship between their Cr(VI) exposure and the occurrence of SDC.

Case presentation

The work history of two SDC patients was analysed for chemical exposures. Both patients had a history of Cr(VI) exposure, with the maintenance of military equipment considered as the source for this exposure. Inhalation of Cr(VI) containing particles from the removal of old paint by mechanical abrasion was identified as a probable source of exposure for both patients, and one of these patients also applied new paint. Both patients reported not to have used any respiratory protection which may have resulted in substantial inhalation of Cr(VI)-containing chromates. Furthermore, in one patient inhalation of fumes from soldering may have resulted in relevant co-exposure.

Conclusion

A causal relation between Cr(VI) exposure and SDC, a rare cancer, cannot be demonstrated on an individual basis but detection in a population-based study is also unlikely because of the extremely low prevalence. Nevertheless, the work history is considered a relevant risk factor in the onset of SDC as occupational exposures to Cr(VI) occurred in the poorly ventilated working environment and without using appropriate respiratory protective equipment.

INTRODUCTION

Salivary duct carcinoma (SDC) is one of the most aggressive subtypes of salivary gland cancer. Salivary gland cancer is a rare cancer with an incidence rate of probably one to two adults in 100.000 each year (1), which makes SDC extremely rare, since <10% of all salivary gland cancer cases comprise of SDC (1-4). Due to its rarity, no studies investigated risk factors specifically for SDC. Some occupational exposures like radioactive substances and nickel compounds/alloys have previously been suggested as potential risk factors for salivary gland cancer (5). Salivary gland cancers are not considered hereditary, although an association between breast cancer and salivary gland cancer has been reported (6).

SDC is classified as an aggressive adenocarcinoma. Common genetic alterations in SDC include the *TP53* gene (53-68%), *PIK3CA* gene (18-26%), and *HRAS* gene (16%) (7). The vast majority (78-96%) of SDC tumours are positive for the androgen receptor (8-10). Furthermore, overexpression of human epidermal growth factor receptor 2 (*HER2neu*), is found in SDC in about 29-46% of the patients (8, 11, 12).

Hexavalent chromium (Cr(VI)) has been identified as a carcinogenic risk factor in humans and is known to increase the risk of lung cancer (13). Workers can become exposed to different chemical species of Cr(VI): chromic acid in chrome plating and the use of chromates in metal coatings, cement and welding fumes. Due to their high chemical reactivity, direct skin contact with chromic acid and Cr(VI) containing chromates leads to irritation and may induce a skin allergy (14). Prolonged inhalation may lead to nasal irritation, and hypertrophy of the nasal turbinates, and can even lead to ulceration and perforation of the nasal septum of chrome platers (15). Most other signs of toxicity are related to different diseases of the respiratory system including lung cancer. When Cr(VI) is inhaled, it can cross cell membranes of the lung and persist in the bifurcation of the lung for years after inhalation exposure. Due to its instability in the human body, Cr(VI) is reduced to the more stable Cr(III) by ascorbate, glutathione and other substances. During this process, free radicals can be generated, which potentially causes structural DNA damage. Accumulation of scarce water-soluble Cr(VI)-containing particles in the bronchial bifurcation causes a sustained high tissue dose, which is believed to increase Cr(VI)-toxicity. The resulting increased cell proliferation may increase the risk of tumorigenesis (15) Cancers of other sites are less frequent and more difficult to study because of the extremely low prevalence among chromium workers (13).

In this report, we present two SDC patients and discuss a potential association with work-related Cr(VI) exposures.

CASE DESCRIPTIONS

Case 1

A 59-year-old male presented to the general practitioner because of painful ribs. Imaging, performed to rule out pulmonary embolism at the Emergency Department, showed no cause for his symptoms, but accidentally revealed enlarged lymph nodes in the neck. Histopathology obtained from a lymph node biopsy indicated a metastasis of an poorly differentiated adenocarcinoma. The node was almost entirely made up of an epithelial tumour which formed tubular structures. The tumour cells had ample eosinophilic cytoplasm and enlarged polymorphic anisochromatic and elucidated nuclei often with a prominent eosinophilic nucleolus. Both mitoses and apoptosis were observed. At the tumour border vascular invasion was present. Additional imaging examinations revealed a large sinonasal process (figure 1). Subsequently, the patient was diagnosed with a primary SDC originating from the sinonasal cavity based on additional clinical information, histomorphology and immunohistochemistry. Due to cervical and mediastinal lymph node metastases (T3N2bM0 disease), the patient was referred to our hospital, a tertiary referral centre for salivary gland cancer, to discuss systemic treatment options. The tumour cells were positive for androgen receptor (AR) see figure 2. Human epidermal growth factor receptor 2 (*HER2neu*) status was assessed in accordance with the

American Society of Clinical Oncology/College of American Pathologists (ASCO/CAP) guidelines for the evaluation of breast cancer (16). Immunohistochemistry (IHC) was inconclusive; IHC score: 2+, therefore fluorescence in situ hybridization (FISH) was performed which indicated no amplification, resulting in a negative HER2neu status. Additional molecular analysis was performed on a lymph node metastasis to detect other potential actionable targets for future systemic treatment options. The assay used for molecular analysis was the TruSight Oncology 500 (Illumina), which is a next-generation sequencing assay that enables comprehensive genomic profiling of 532 genes, furthermore it measures microsatellite instability and tumour mutational burden. The molecular analysis showed no clinically relevant mutations, there was no microsatellite instability, and the total tumour mutational burden was 1.6 mutations per megabase.

At the first outpatient visit, the patient asked if the occurrence of his SDC could be somehow related to direct contact with Cr(VI) in work-related exposures in the past. A detailed occupational history revealed that the patient had been working as inspector of construction cranes and harbour cranes since 1984 up until the occurrence of his SDC disease. Initially this inspection work did not involve any use of chemicals until, in 2012, he was asked to perform inspections at three military air bases in the Netherlands over a period of 2-3 months a year. This liquid penetrant inspection was performed to detect hairline cracks in welds of military equipment. For this task, the coating was completely removed by mechanical grinding. The

topcoat known as chemical agent resistant coating (CARC) contains the toxic hexamethylene diisocyanate (HDI) and is typically adhered to the metal surface with a chromic acid-containing conversion coat and primer coat containing a chromate pigment. Both are Cr(VI)-based chemical components that generate airborne particles when mechanically abrased (17). According to the patient, this procedure resulted in visible dust clouds that caused work clothes to become contaminated. No respiratory protective equipment was used and protective gloves were not always used. During breaks the worker cleaned hands but did not change clothes. When the patient blew his nose after these activities this resulted in a dusty nose secretion matching the colour of the paint, indicating a clear direct exposure of the nasal mucosa to the paint grinded to dust. As of 2015, the use of the destructive method was discontinued and replaced by an alternative non-destructive inspection method.

Apart from Cr(VI) exposure, the patient may have come into contact with other chemical substances during a previous occupation (figure 3). From 1978 till 1984 he worked as a plumber. Furthermore, the patient had a history of smoking and alcohol consumption. The patient started smoking shag from the age of 16 until 58, with an average of 50 gram of tobacco per week which results in a total of 31 pack years. He started to use alcoholic beverages at the age of 16 and consumes approximately 24 units of alcohol per week. The patient has been treated with combined androgen deprivation therapy, because of androgen receptor expression. Previous research indicated that this therapy might alter the prognosis of patients, without therapy patients with advanced disease had a poor overall survival of 5 months, while with androgen deprivation therapy the overall survival was 17 months (18). Therapy consists of the combination of a luteinizing hormone-releasing hormone (LHRH) agonist, goserelin subcutaneous 10.8 mg every 12 weeks, and an androgen receptor antagonist, bicalutamide tablet 50 mg once daily. Evaluation CT scans performed 3 months after start of therapy showed a partial response according to the RECIST 1.1 criteria for tumour response evaluation (19). The most recent evaluation, 9 months after initiation of treatment, showed an ongoing partial response.

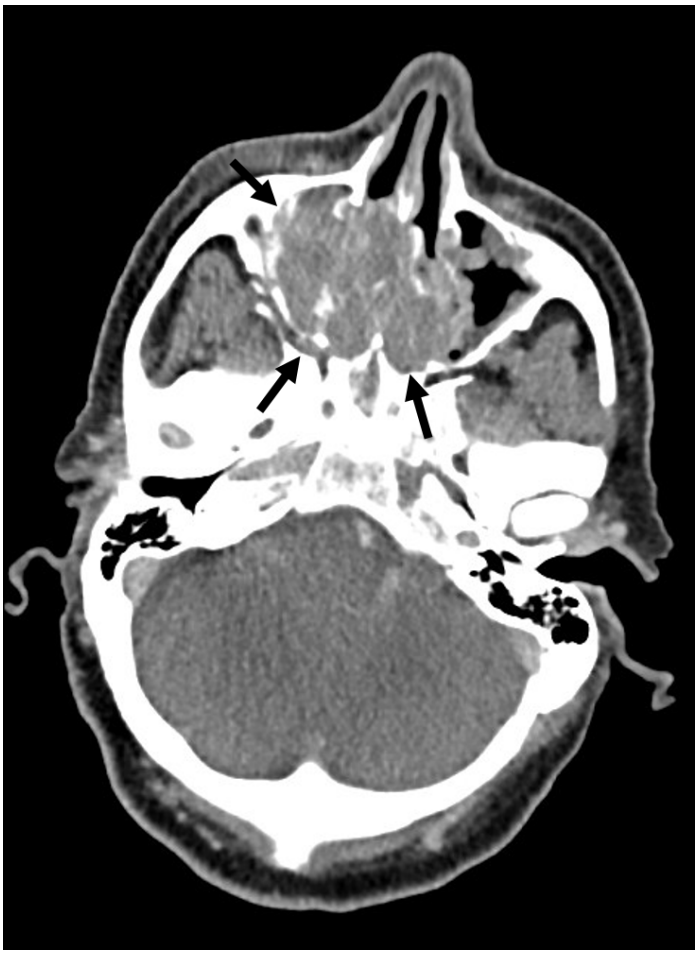


Figure 1: CT image of primary sinonasal SDC tumour (indicated by black arrows)

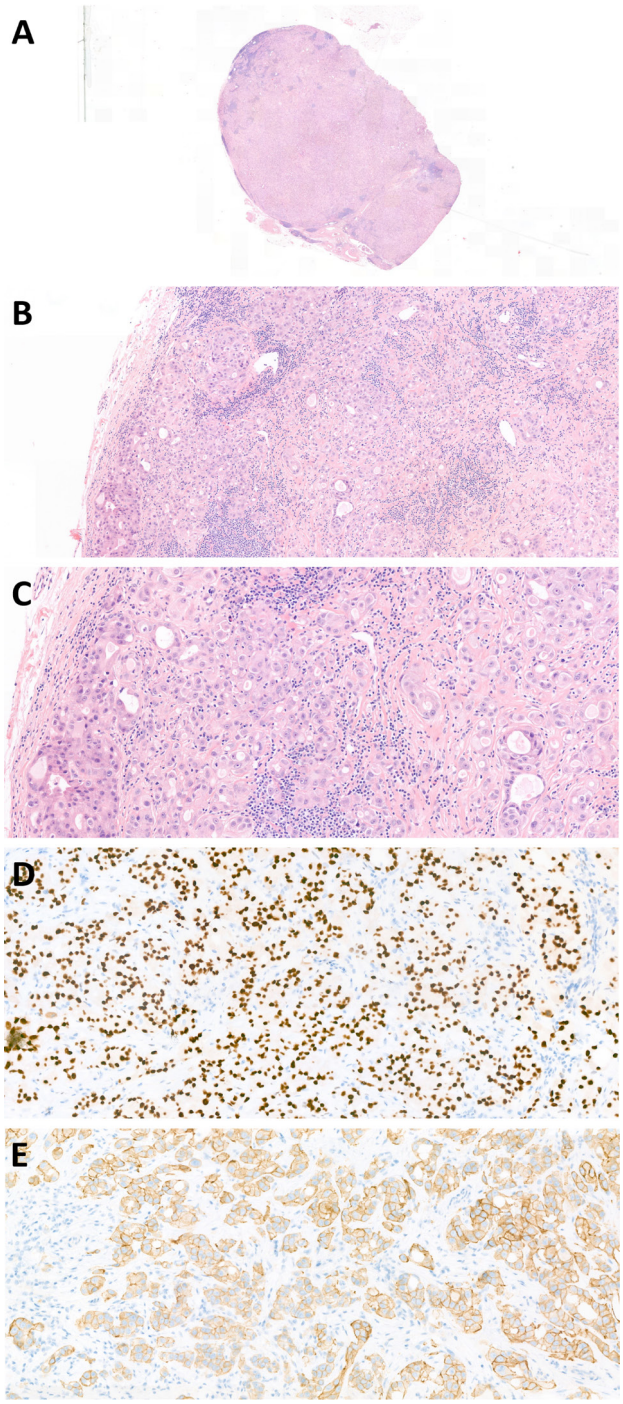


Figure 2: Photomicrographs of case 1: cervical lymph node containing an SDC metastasis.
A: H&E stain 1X, **B**: H&E stain 10X, **C**: H&E stain 21X, **D**: AR stain 20X, AR441 (BioCare Medical),
E: HER2neu stain 21X, HercepTest (Dako Agilent).

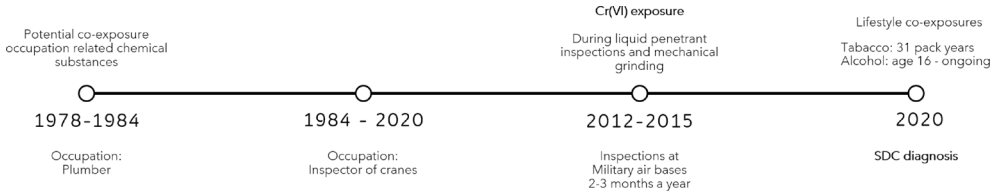


Figure 3: Time-line of case 1

Case 2

A 54-year-old male was referred to a local hospital with a palpable mass in the neck at level 2 and at the right submandibular gland, suspicious for a malignant tumour. PET/CT imaging showed a primary tumour originating from the right submandibular gland (figure 4) and also right lymphadenopathy at level I-V, suspect for lymph node metastases, and a lesion in the 3rd thoracic vertebra. Biopsy of the bone lesion revealed a SDC metastasis (T2N2bM1 disease) (figure 5). The biopsy was largely occupied by a tumour. The tumour was made up of tubes and cribriform tubes of atypical epithelial cells with round-oval nuclei that varied in shape and size and often had a prominent nucleolus and ample amphophilic well delimited cytoplasm. In some of the tubes necrosis was seen. Mitoses were observed. The pathology assessment concluded a bone metastasis from a salivary duct carcinoma. The patient was referred to our hospital for palliative systemic treatment. The tumour cells were positive for AR on IHC and negative for Her2neu (IHC: 2+ staining, but no amplification on FISH). Molecular analysis, using a similar assay as for case 1, presented a mutation in TP53 and CKD12, but overall, no druggable targets. Furthermore, no microsatellite instability was observed and the total tumour mutational burden was 4.7 mutations per megabase. Additionally, the assay showed many fluctuations in number of readings of different chromosomes, suggesting that there were many numerical chromosomal aberrations, which is indicative of genomic instability.

During the first outpatient visit, this patient's history also revealed an occupational exposure to Cr(VI). The patient started working as a paramedic in the army at the age of 18 and has worked there for a total 32 years. In addition to his nursing duties, he was involved in maintenance of 4 x 4 military trucks type YA-314 and YA-328 (approximately 2 hours/day). In the years 1987-1992 old paint was removed by manual abrasion using steel brush and paint scraper. New primer and topcoat were applied by spray and brush painting. In the period 1992-2017 the work continued but painting was mainly done by spot painting. Both tasks were performed indoor in an unventilated large workshop without the use of a spray booth or local exhaust ventilation for approximately 4 hours/month. During spray painting this resulted in substantial exposures to volatile organic compounds from the thinner used. Personal protective equipment such as respirators or gloves were not used and not available. The paint was very sticky and could remain on the hands even when washing hands with water and soap prior to short breaks (for smoking) and longer breaks for a meal in the recreation room.

Contaminated work garment was not changed prior to breaks. Exposure to organic solvents from paint and thinner was indicated by a bad mouth taste. Apart from occasional headaches no acute signs of neurotoxicity were reported on the day of exposure or the day after.
In addition to Cr(VI) exposure, the patient may have come into contact with other chemical substances (figure 6). The patient was sent overseas for military missions for three months at the mission headquarters in Srebrenica, Bosnia that was set up on a former industrial site as part of UNPROFOR in 1994-1995 (20). There were remains of a lead battery factory that was partly destroyed in the conflict. The site was heavily contaminated with lead-containing dust. Lead exposure was confirmed by blood analyses in some of the military staff but did not results in health complaints (21). The patients himself also reported no symptoms of lead exposure. Additionally, he worked as paramedic in Kandahar, Afghanistan (ISAF) in 2006-2007 (22). There, depending on the wind direction the smoke from a nearby burn pit would blow through the compound causing complaints of very bad smell indicating exposure to smoke fumes with unidentified toxic components (23). Lifestyle exposures included; consumption of alcoholic beverages, from the age of 15, approximately 5 units of alcohol per week, and smoking with an average of 20 cigarettes per week during 30 years (30 pack years).

The patient started androgen deprivation therapy, in the form of bicalutamide monotherapy (tablet 150mg once daily) because of androgen receptor positivity. Unfortunately, first treatment evaluation, 3 months after start, showed progressive disease (RECIST v1.1). The patient was subsequently referred for palliative radiotherapy and start of palliative chemotherapy.

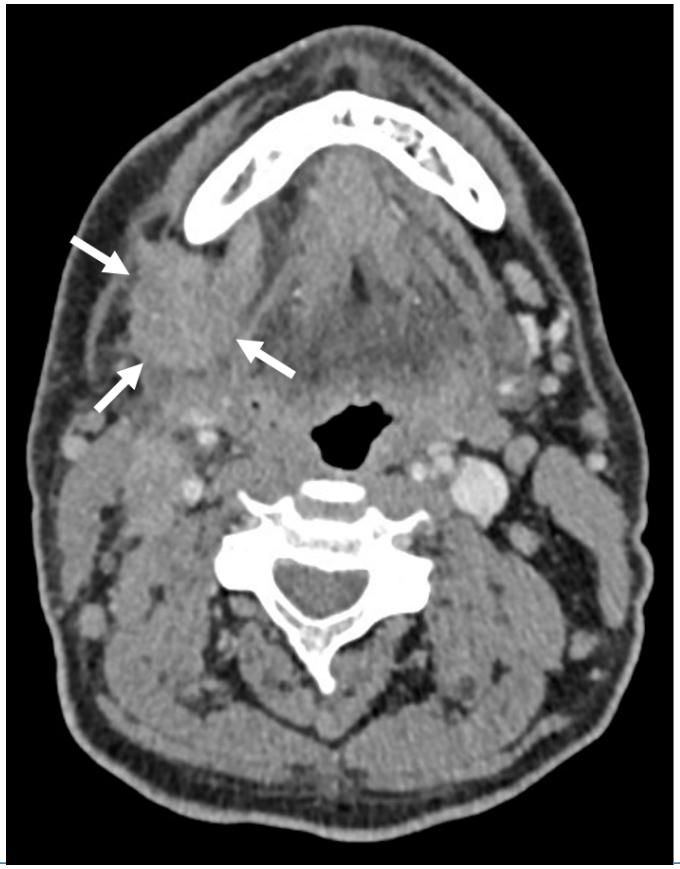


Figure 4: CT image of primary submandibular SDC tumour (indicated by white arrows)

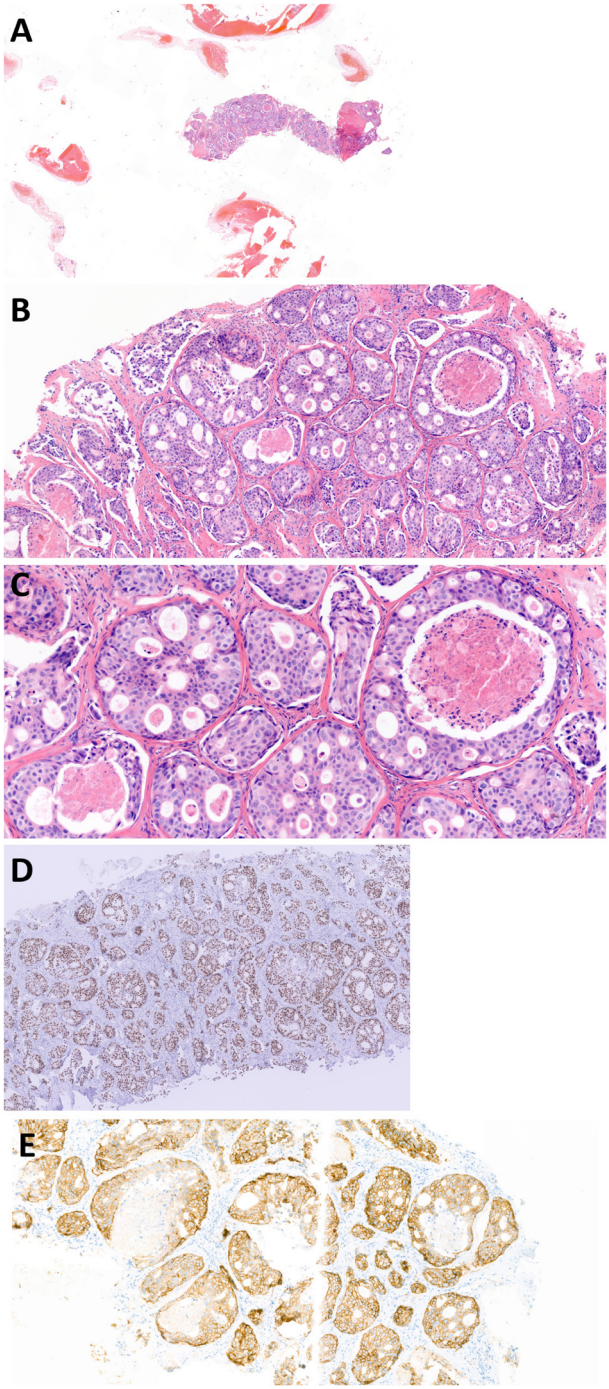


Figure 5: Photomicrographs of case 2: biopsy of SDC bone metastasis in thoracic vertebra 3. **A**: H&E stain 1X, **B**: H&E stain 10X, **C**: H&E stain 20X, **D**: AR stain 5X (photomicrograph from referring centre), **E**: HER2neu stain 10X, HercepTest (Dako Agilent).

Figure 6: Time-line of case 2

DISCUSSION

Here we presented two SDC patients both with a history of Cr(VI) exposure related to mechanical abrasion of chromate containing metal coating in maintenance of military equipment. This triggers the question of a potential relation between the occurrence of SDC and Cr(VI) exposures. To address this question we will first describe known risk factors for SDC and then we will focus on the toxic effects of Cr(VI) and their potential relation to the occurrence of SDC and finally on the prevention of Cr(VI)-exposure.

Little is known about risk factors for SDC specifically, but several studies have investigated potential risk factors for salivary gland cancer. While smoking and alcohol are a clear risk factor for other types of head and neck cancer, especially squamous cell carcinoma (24), most studies showed no or limited association between smoking tobacco or alcohol consumption and salivary gland cancer (25-30). Previously addressed risk factors for developing salivary gland cancer include; prior radiation to the head or neck during radiotherapy or during dental or cervicofacial radiological examinations, occupational exposure to radioactive materials and nickel compounds/alloys, ambient air pollution from waste gas emissions and certain occupations, like plumber, sheet-metal worker and building painter (5, 29, 31). Yet, Cr(VI) exposure is not mentioned specifically in these reports.

Our literature search on Cr(VI) exposure and SDC through PubMed did not yield any relevant articles, also indicating that a possible association has not yet been discussed in literature. Expanding the search to salivary gland cancer resulted in one relevant article (32). In South Korea the incidence of respiratory tract cancers was studied as a function of the distance to Portland cement plants. These plants are known to result in elevated combined emissions of chromium, crystalline silica and polycyclic aromatic hydrocarbons over long distances. The incidence of salivary gland cancer near the cement plants was approximately three times higher in females (standardized incidence ratio: 3.03, 95%CI: 0.98-7.07), using the national incidence in the South-Korean female population as comparator. Evidence for exposure of salivary glands specifically to Cr(VI) was not found but is considered plausible regarding the inhalable size of the cement dust particles.

Regarding the carcinogenic effects of Cr(VI) exposure; Cr(VI) can cause oxidative DNA damage, which in turn could increase the risk of neoplastic formation, especially in the case of reoccurring exposure (33, 34). Cr(VI) containing chromate ions are thought to enter cells by a shunt that is normally used for uptake of sulphate and phosphate and may reach the nucleus (35). When Cr(VI) is reduced reactive oxygen is generated, leading to oxidative and other types of DNA damage confirmed in cell cultures (36). It is still unclear to what extent genotoxicity by itself or combined with epigenetic/inflammatory processes leads to DNA instability, impaired apoptosis and inhibited normal DNA-repair which may all explain tumour induction in animal experiments (37). Interestingly, In both patients molecular analysis has been performed. We did not find remarkable anomalies in case report 1 but In the case report 2, a mutation in TP53 was observed and the assay indicated genomic instability. Mutations in TP53 are common in SDC, present in 53-68% of the tumours (7, 38). The genomic instability can be the result of many possible causes, including TP53 mutations, but has also been previously reported as a result of prior Cr(VI) exposure (39, 40).

In addition to the rarity of SDC itself, in both patients the primary tumour originated at an unusual location. SDC mostly arises from the parotid gland (72% of the cases) (41). In the first patient the SDC originated in the sinonasal tract, which is extremely rare, only four other reports of primary SDC originating in the sinonasal tract have been reported (42). In the second patient the SDC occurred in the submandibular gland, approximately 15% of SDC occur in this salivary gland (41).

The International Agency for Research on Cancer (IARC) classified Cr(VI) as a group 1 human carcinogen based on evidence from population-based studies and experimental animal studies both suggesting an increased risk of lung cancer following occupational exposure to Cr(VI) (13).

In our patient from case report 1 however, cancer arose in a different part of the airway system, namely as mentioned before in the sinonasal tract. A preclinical study which investigated the effect of inhalation of chromium by exposing mice for 12 months to chromic acid mist several mice developed nasal tumours (papillomas) (43). As for sinonasal cancer, the IARC assessed epidemiological evidence for Cr(VI) exposure as the cause for sinonasal cancer and stated that an association was suggestive, however inconclusive (13). Since then, more research regarding this topic has been published. A systematic review and meta-analysis estimated the relative risk of sinonasal cancer to be 18.0 (95%CI: 14.6-22.3) in workers who work with chromium and nickel (44). Another report on Cr(VI) exposure and sinonasal cancer described a maximum relative risk for developing sinonasal cancer among chromate production workers of 15.4 (45). In a recently updated narrative literature review prepared by the Dutch Institute for Public Health and the Environment (RIVM) the available evidence from human observational

combined with experimental animal evidence was evaluated as sufficient to conclude that Cr(VI) exposure can be considered as a risk factor for nose and sinonasal cancer (46).

In the second patient the SDC arose from the submandibular gland. As mentioned before, little has been described in the literature about the association between salivary gland cancer and Cr(VI) exposure. For the submandibular gland tissue there some supporting evidence but not a clear and direct link with occupational Cr(VI) exposure. Oral exposure Cr(VI) is a well-known in oral carcinogenicity (47). Cancer of the oral cavity has been observed in rats who received Cr(VI) in drinking water (48). In workers in metal industry with primarily inhalation exposure dental injury was observed, more specifically a higher incidence of dental caries and mastication deficiency was observed in 100 patients with occupational exposure to chromates in an industrial setting (49).

Apart from Cr(VI) exposure, co-exposure to other carcinogenic substances should always be considered as a contributing factor for the occurrence of cancer. When we considered other potential exposures in our patients, we first evaluated other chemical substances arising from the maintenance of military equipment. Apart from the chromate in the CARC (topcoat of the metal surface), HDI is considered the most toxic ingredient (50). HDI is a known respiratory sensitizer, but has not been classified as a carcinogen and is therefore unlikely to have contributed to the occurrence of SDC (51). Furthermore, our first patient was exposed to fumes from soldering when he worked as a plumber. In these fumes human carcinogens such as nickel oxide and chromates are not often encountered, but aldehydes are (52, 53). Specifically, formaldehyde is a known risk factor of nasal and sinonasal cancer and might have contributed to the occurrence of SDC in this patient (54). Finally, both patients had a history of tobacco and alcohol consumption. The medical file indicated no history of radiation exposure for diagnostic or therapeutic purposes prior to the SDC diagnostic work-up. However, a contribution of this to the occurrence of SDC is considered less likely, given the lack of a clear association with salivary gland cancer as mentioned above.

Because of the deleterious effects of Cr(VI) it is important to minimize or prevent exposure of employees as much as possible. Occupational exposure to Cr(VI) is currently regulated in EU with a binding Occupational Exposure Limit (OEL) expressed as 8-hour time-weighted average (8-h TWA) of 10 $\mu g/m^3$ with the intention to further limit this OEL to 5 $\mu g/m^3$. The Netherlands and France have currently worldwide the strictest 8-h TWA of 1 $\mu g/m^3$ (55, 56). These OELs have been derived based on human lung cancer risk in workers for which there is no evidence for a threshold. So, an exposure below the OEL does not mean that there is no residual cancer risk. In the Netherlands the OEL was derived from a residual accepted risk of 1×10^{-4} per exposure year (corresponding to a risk 4×10^{-3} for a working live exposure of 40 years (57). The first priority is to comply the legally binding OEL. When this is in achieved further reductions of exposure are mandatory in line with international technical state-of-the-

art. For this the occupational hygiene strategy is applied: if the Cr(VI) cannot be eliminated or substituted with an alternative substance with a lower health risk, risk management measures (RMM) are implemented to minimize exposure by segregation of the workers from the source (e.g. by containment of the process or moving the operator at a safe distance from the source), instalment of local air ventilation and additional room ventilation, changing the lay-out of the workroom and/or organizing the work differently, and as a last resort, introduce personal protective equipment such as providing respirators with high-efficient particle filters.

Recently, it was suggested that in addition to direct inhalation exposure also dermal exposure was associated with uptake of Cr(VI) (58). So indirect exposure caused by secondary contamination should also be avoided to prevent exposure by good personal hygiene, e.g. by providing protective gloves and overalls with long sleeves, allowing no consumption of food/beverages or smoking at the workplace and providing standard water sanitation and hygiene facilities and additional facilities for full body decontamination for emergencies. For proper implementation of these measures an occupational hygienist and occupational physician should be involved to perform exposure and health surveillance.

CONCLUSION

In conclusion, our report highlighted two patients with SDC which have both been exposed to Cr(VI). Mechanical abrasion of painted metals of military equipment as part of corrosion control is a known source of inhalation exposure exceeding current standards for protection of worker's health (17). A causal relation between Cr(VI) exposure and SDC is difficult to prove both on individual as well as population basis given the extreme rarity of SDC. Nevertheless, we consider it plausible that Cr(VI) containing chromates might have contributed to the occurrence of the disease as the common factor in both cases as exposures occurred in a poorly ventilated working environment (no local exhaust ventilation) and the work was conducted without appropriate respiratory protective equipment. For case 1 we cannot rule out a contribution from working as a plumber which may also have resulted in exposure to formaldehyde. With this report we aim to increase the awareness of occupational Cr(VI) exposure and occurrence of SDC. Furthermore, we want to emphasize the importance of primary prevention in order to minimize exposure to chemical carcinogens.

ACKNOWLEDGMENTS

The authors would like to thank A.C.H. van Engen-van Grunsven and B. van der Vegt for providing the photomicrographs of the immunohistochemical staining.

REFERENCES

1. de Ridder M, Balm AJ, Smeele LE, Wouters MW, van Dijk BA. An epidemiological evaluation of salivary gland cancer in the Netherlands (1989-2010). Cancer Epidemiol. 2015;39(1):14-20.
2. Luukkaa H, Klemi P, Leivo I, Koivunen P, Laranne J, Mäkitie A, et al. Salivary gland cancer in Finland 1991--96: an evaluation of 237 cases. Acta Otolaryngol. 2005;125(2):207-14.
3. Barnes L, Eveson JW, Reichart P, Sidransky D. World Health Organization Classification of Tumors. Pathology and Genetics of Head and Neck Tumors. IARC Press; Lyon. 2005.
4. Bjørndal K, Krogdahl A, Therkildsen MH, Overgaard J, Johansen J, Kristensen CA, et al. Salivary gland carcinoma in Denmark 1990-2005: a national study of incidence, site and histology. Results of the Danish Head and Neck Cancer Group (DAHANCA). Oral Oncol. 2011;47(7):677-82.
5. Horn-Ross PL, Ljung BM, Morrow M. Environmental factors and the risk of salivary gland cancer. Epidemiology. 1997;8(4):414-9.
6. Shen TK, Teknos TN, Toland AE, Senter L, Nagy R. Salivary Gland Cancer in BRCA-Positive Families : A Retrospective Review. JAMA Otolaryngology–Head & Neck Surgery. 2014;140(12):1213-7.
7. Lassche G, van Boxtel W, Ligtenberg MJL, van Engen-van Grunsven ACH, van Herpen CML. Advances and challenges in precision medicine in salivary gland cancer. Cancer Treatment Reviews. 2019;80:101906.
8. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.
9. Takase S, Kano S, Tada Y, Kawakita D, Shimura T, Hirai H, et al. Biomarker immunoprofile in salivary duct carcinomas: clinicopathological and prognostic implications with evaluation of the revised classification. Oncotarget. 2017;8(35):59023-35.
10. Williams MD, Roberts D, Blumenschein GR, Jr., Temam S, Kies MS, Rosenthal DI, et al. Differential expression of hormonal and growth factor receptors in salivary duct carcinomas: biologic significance and potential role in therapeutic stratification of patients. Am J Surg Pathol. 2007;31(11):1645-52.
11. Locati LD, Perrone F, Losa M, Mela M, Casieri P, Orsenigo M, et al. Treatment relevant target immunophenotyping of 139 salivary gland carcinomas (SGCs). Oral Oncol. 2009;45(11):986-90.
12. Clauditz TS, Reiff M, Gravert L, Gnoss A, Tsourlakis MC, Münscher A, et al. Human epidermal growth factor receptor 2 (HER2) in salivary gland carcinomas. Pathology. 2011;43(5):459-64.
13. IARC. International Agency for Research on Cancer. IARC Monographs on the Evaluation of Carcinogenic Risk to Humans. Arsenic, metals, fibres and dusts. Lyon. 2012;100-C.
14. Hayes RB. Review of occupational epidemiology of chromium chemicals and respiratory cancer. Sci Total Environ. 1988;71(3):331-9.
15. Yatera K, Morimoto Y, Ueno S, Noguchi S, Kawaguchi T, Tanaka F, et al. Cancer Risks of Hexavalent Chromium in the Respiratory Tract. J uoeh. 2018;40(2):157-72.
16. Wolff AC, Hammond MEH, Allison KH, Harvey BE, Mangu PB, Bartlett JMS, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/ College of American Pathologists Clinical

Practice Guideline Focused Update. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2018;36(20):2105-22.

17. Carlton GN. Hexavalent chromium exposures during full-aircraft corrosion control. AIHA J (Fairfax, Va). 2003;64(5):668-72.

18. Boon E, van Boxtel W, Buter J, Baatenburg de Jong RJ, van Es RJJ, Bel M, et al. Androgen deprivation therapy for androgen receptor-positive advanced salivary duct carcinoma: A nationwide case series of 35 patients in The Netherlands. Head Neck. 2018;40(3):605-13.

19. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). Eur J Cancer. 2009;45(2):228-47.

20. Defence. Historische canon Garderegiment Grenadiers en Jagers, Internationale operaties, cited 10 May 2021 2021 [cited 2021 May 10]. Available from: https://www.defensie.nl.

21. Defence. Letter from the Secretary of State of Defense to the President of the House of Representatives of the States General: Leukemie bij uitgezonden militairen. 2001;LEAD RESEARCH (1993/1994) (Srebrenica / Potocari).

22. Defence. International Security Assistance Force (ISAF), Nederlands aandeel in International Security Assistance Force (ISAF), cited 10 May 2021 2021 [Available from: https://www.defensie.nl.

23. Aurell J, Gullett BK, Yamamoto D. Emissions from Open Burning of Simulated Military Waste from Forward Operating Bases. Environmental Science & Technology. 2012;46(20):11004-12.

24. Argiris A, Karamouzis MV, Raben D, Ferris RL. Head and neck cancer. Lancet. 2008;371(9625):1695-709.

25. Alvi S, Chudek D, Limaiem F. Parotid Cancer. In: StatPearls [Internet] Treasure Island (FL): StatPearls 2020.

26. Hayes RB, Bravo-Otero E, Kleinman DV, Brown LM, Fraumeni JF, Harty LC, et al. Tobacco and alcohol use and oral cancer in Puerto Rico. Cancer Causes & Control. 1999;10(1):27-33.

27. Muscat JE, Wynder EL. A case/control study of risk factors for major salivary gland cancer. Otolaryngol Head Neck Surg. 1998;118(2):195-8.

28. Spitz MR, Fueger JJ, Goepfert H, Newell GR. Salivary gland cancer. A case-control investigation of risk factors. Arch Otolaryngol Head Neck Surg. 1990;116(10):1163-6.

29. Radoï L, Barul C, Menvielle G, Carton M, Matrat M, Sanchez M, et al. Risk factors for salivary gland cancers in France: Results from a case-control study, the ICARE study. Oral oncology. 2018;80:56-63.

30. Pan SY, de Groh M, Morrison H. A Case-Control Study of Risk Factors for Salivary Gland Cancer in Canada. Journal of Cancer Epidemiology. 2017;2017:4909214.

31. Cong X. Air pollution from industrial waste gas emissions is associated with cancer incidences in Shanghai, China. Environ Sci Pollut Res Int. 2018;25(13):13067-78.

32. Eom SY, Cho EB, Oh MK, Kweon SS, Nam HS, Kim YD, et al. Increased incidence of respiratory tract cancers in people living near Portland cement plants in Korea. Int Arch Occup Environ Health. 2017;90(8):859-64.

33. Ray RR. Adverse hematological effects of hexavalent chromium: an overview. Interdiscip Toxicol. 2016;9(2):55-65.

34. Wise SS, Aboueissa AE, Martino J, Wise JP, Sr. Hexavalent Chromium-Induced Chromosome Instability Drives Permanent and Heritable Numerical and Structural Changes and a

DNA Repair-Deficient Phenotype. Cancer Res. 2018;78(15):4203-14.

35. de Flora S. Mechanism of chromium (VI) metabolism and genotoxicity. Life Chem Rep. 1989;7:169-244.

36. Ding M, Shi X. Molecular mechanisms of Cr(VI)-induced carcinogenesis. Molecular and Cellular Biochemistry. 2002;234(1):293-300.

37. Chen QY, Murphy A, Sun H, Costa M. Molecular and epigenetic mechanisms of Cr(VI)-induced carcinogenesis. Toxicol Appl Pharmacol. 2019;377:114636-.

38. Ross JS, Gay LM, Wang K, Vergilio JA, Suh J, Ramkissoon S, et al. Comprehensive genomic profiles of metastatic and relapsed salivary gland carcinomas are associated with tumor type and reveal new routes to targeted therapies. Ann Oncol. 2017;28(10):2539-46.

39. Negrini S, Gorgoulis VG, Halazonetis TD. Genomic instability--an evolving hallmark of cancer. Nat Rev Mol Cell Biol. 2010;11(3):220-8.

40. Wise SS, Wise JP, Sr. Chromium and genomic stability. Mutat Res. 2012;733(1-2):78-82.

41. Jayaprakash V, Merzianu M, Warren GW, Arshad H, Hicks WL, Jr., Rigual NR, et al. Survival rates and prognostic factors for infiltrating salivary duct carcinoma: Analysis of 228 cases from the Surveillance, Epidemiology, and End Results database. Head & neck. 2014;36(5):694-701.

42. Agaimy A, Mueller SK, Bishop JA, Chiosea SI. Primary and Secondary/ Metastatic Salivary Duct Carcinoma Presenting within the Sinonasal Tract. Head and Neck Pathology. 2021.

43. Adachi S. [Effects of chromium compounds on the respiratory system. 5. Long term inhalation of chromic acid mist in electroplating by C57BL female mice and recapitulation of our experimental studies]. Sangyo Igaku. 1987;29(1):17-33.

44. Binazzi A, Ferrante P, Marinaccio A. Occupational exposure and sinonasal cancer: a systematic review and meta-analysis. BMC Cancer. 2015;15:49.

45. Comiati V, Scapellato ML, Alexandre E, Volo T, Borsetto D, Carrieri M, et al. Sinonasal cancer in a worker exposed to chromium in an unusual industrial sector. Med Lav. 2017;108(6):477-81.

46. Hessel EVS, den Braver S, Ezendam J, Staal YCM, Piersma AH, Palmen NGM, et al. Nadelige gezondheidseffecten en ziekten veroorzaakt door chroom-6 Actualisatie van de wetenschappelijke literatuur en de risicobeoordeling voor strottenhoofdkanker bij tROM. RIVM-rapport 2020-0019 2020;DOI 10.21945

47. Sun H, Brocato J, Costa M. Oral Chromium Exposure and Toxicity. Curr Environ Health Rep. 2015;2(3):295-303. 48. National Toxicology Program. Toxicology and carcinogenesis studies of sodium dichromate dihydrate (Cas No. 7789-12-0) in F344/N rats and B6C3F1 mice (drinking water studies). Natl Toxicol Program Tech Rep Ser (2008). p. 1–192.

49. Resch CA. Effect of chromates on the teeth, a comparative report. Cleveland Clin J Med. (1950) 17:31–3. doi: 10.3949/ccjm.17.1.31

50. Heringa MB, Bakker J, Hogendoorn E. Evaluatie en prioritering schadelijke stoffen in Chemical Agent Resistant Coating (CARC), gebruikt op de Nederlandse POMS locaties. Evaluating and prioritising harmful substances in Chemical Agent Resistant Coating (CARC), used at POMS sites in the Netherlands: Rijksinstituut voor Volksgezondheid en Milieu RIVM; 2018.

51. Shiotsuka RN, Stuart BP, Charles JM, Simon GS, Malichky P, Mostowy JM. Chronic inhalation exposures of Fischer 344 rats to 1,6-hexamethylene diisocyanate did not reveal a carcinogenic potential. Inhal Toxicol. 2010;22(10):875-87.

52. Smith PA, Son PS, Callaghan PM, Jederberg WW, Kuhlmann K, Still KR. Sampling and analysis of airborne resin acids and solvent-soluble material derived from heated colophony (rosin) flux: a method to quantify exposure to sensitizing compounds liberated during electronics soldering. Toxicology. 1996;111(1-3):225-38.

53. Pengelly MI, Foster RD, Groves JA. An investigation into the composition of solder fume. Annals of Occupational Hygiene. 1994;38(5): 753-63

54. IARC. International Agency for Research on Cancer. Chemical agents and related occupations. Volume 100 F. A review of human carcinogens, Lyon. 2012.

55. Ministère du Travail, de l'Emploi et de l'Insertion. Décret n° 2012-746 du 9 mai 2012 fixant des valeurs limites d'exposition professionnelle contraignantes pour certains agents chimiques (2012).

56. Ministerie voor Sociale Zaken en Werkgelegenheid. Regeling van de Minister van Sociale Zaken en Werkgelegenheid van 18 oktober 2016, 2016-0000222216, tot wijziging van de Arbeidsomstandighedenregeling in verband de wijziging van twee wettelijke grenswaarden in Bijlage XIII (Bisfenol A en Chroom (VI)-verbindingen). The Hague, The Netherlands (2016).

57. Health Council of the Netherlands. Hexavalent chromium compounds. Health-based calculated cancer risk values. The Hague: 57; publication no. 2016/13E (2016).

58. Santonen T, Porras SP, Bocca B, Bousoumah R, Duca RC, Galea KS, et al. HBM4EU chromates study team. HBM4EU chromates study - overall results and recommendations for the biomonitoring of occupational exposure to hexavalent chromium. Environ Res. (2021) 204:111984. doi: 10.1016/j.envres.2021.111984.

Chapter 10

Cutaneous Lymphangitis Carcinomatosa in patients with salivary duct carcinoma resulting in skin lesions of the neck and chest region.

Maike J.M. Uijen*, Jetty A.M. Weijers*, Gerben Lassche, Stefan G. van Ravensteijn, Maartje C. van Rijk, Satish F.K. Lubeek, Adriana C.H. van Engen-van Grunsven, Avital Amir, Chantal M.L. Driessen, Carla van Herpen

* These authors contributed equally

Manuscript in preparation, 2022.

DEAR EDITOR,

Salivary duct carcinoma (SDC) is one of the most aggressive subtypes of salivary gland cancer. It is a very rare cancer (incidence 0.1-0.2 per 100,000 people/year) (1, 2). In recent years, the Radboudumc (Nijmegen, the Netherlands) has evolved to a national tertiary (expert) referral centre for salivary gland cancer patients. Here, we present cutaneous lymphangitis carcinomatosa (CLC) as a manifestation of advanced SDC disease, which is specific for this subtype of salivary gland cancer. To date, little has been reported on this disease manifestation in SDC. Therefore, the aim of this report is to provide an overview of this CLC in SDC patients and to increase the knowledge on this disease manifestation.

A typical characteristic of SDC is the high propensity for lymph node metastases, with a median number of tumour positive lymph nodes of 4 (range: 0–97) (3). The number of lymph node metastases is an important prognostic factor, with a tendency towards worse survival when more lymph nodes are involved (3, 4). Consequently, the primary treatment consists of resecting the affected gland combined with a lymph node neck dissection, followed by post-operative radiotherapy. Furthermore, since SDC tumours are often androgen receptor (AR)-positive (78-96% of cases), androgen deprivation therapy (ADT) can be initiated.
We observed nine SDC patients that developed skin lesions of the neck and chest region, and all these patients had extensive lymph node metastases at diagnosis (table 1). Skin biopsies in these patients showed cutaneous lymphangitis carcinomatosa and cutaneous metastases (figure 1). Pathophysiologically, it is highly plausible that in these patients with extensive lymph node metastases the SDC tumour cells have spread further via the lymphatic vessels, causing cutaneous metastases in the drainage area.

In addition to the overview of all nine SDC patients diagnosed with CLC, we provide a more detailed case description of two representative patients who received several lines of systemic therapy and of whom clinical pictures, diagnostic CT, FDG-PET/CT imaging, and pathology assessments were available. Furthermore, we report on the molecular analysis of these nine patients (supplementary table 1).

Case report 1

A 65-year-old male was diagnosed with an SDC (AR+) arising from the left parotid gland (T3N3bM0). He underwent a parotidectomy and a bilateral neck dissection. He had metastases in 25 lymph nodes (19/19 left and 6/12 right). One week after the initiation of post-operative radiotherapy, two axillary lymph node metastases were discovered and radiotherapy was discontinued. At this stage the patient was referred to our hospital. We initiated combined ADT with goserelin (subcutaneously 10.8 mg 3-monthly) and bicalutamide (tablet 50 mg QD). Two weeks after the start of ADT, the patient noticed skin changes in the neck as well as movement restriction (figure 1A). A CT-scan showed cutaneous thickening with subcutaneous

induration (figure 1B). A skin biopsy revealed cutaneous metastases (supplementary figure 1,2). In addition, tumour cells in the lymphatic vessels (figure 1C, D), and -remarkably- pagetoid tumour growth in the epidermis (supplementary figure 3) were seen. Due to this disease progression, the ADT was discontinued and second-line systemic therapy was initiated with a combination of paclitaxel (175 mg/m^2) and carboplatin (AUC 6). Two weeks after the initiation of this therapy, clinical evaluation indicated stable disease of the affected skin area, potentially even a small reduction. However, thereafter the affected area showed rapid progression. Figure 1E, F, G, H show the affected area approximately 5 months after the initial occurrence. Based on an actionable *PIK3CA* mutation third-line systemic therapy with alpelisib (tablet 300 mg QD) was provided in a clinical study (NCT02925234). This treatment was discontinued after one month, because of limited efficacy and side-effects. Shortly thereafter, the patient died due to respiratory insufficiency caused by rapidly progressive disease.

Case report 2

A 63-year-old male was diagnosed with SDC (AR+) originating from the left submandibular gland with lymph node metastases in the neck, retropharyngeal area and axilla, and also distant metastases in bones (TxN2cM1). He received palliative radiotherapy for the cervical lymph node metastases and bone metastases. Thereafter, he was referred to our hospital for systemic treatment. We initiated ADT with goserelin (subcutaneously 10.8 mg, 3-monthly) and bicalutamide (tablet 50 mg QD). Approximately 3 months after the initial diagnosis, and while still on ADT, the patient noticed skin changes of the neck and thoracic regions (figure 1I), which also caused mechanical skin tension, resulting in movement restriction. Due to the strongly indurated erythematous skin (also observed on CT, figure 1J) with a varying nodular aspect, CLC was suspected. A skin biopsy confirmed the CLC (figure 1K, L and supplementary figure 4,5). Due to this disease progression, ADT was discontinued and chemotherapy was initiated (paclitaxel [175 mg/m^2] and carboplatin [AUC 6]). After two weeks, both the swelling of the neck and the size of the erythematous skin lesions decreased. This clinical response was still present after six cycles of chemotherapy. Also radiologically, the patient had stable disease with a slight reduction of tumour lesions. Because the treatment was well tolerated, two additional cycles were given. Unfortunately, shortly thereafter, the disease progressed. Then, based on limited HER2+ positivity (no amplification on fluorescence in situ hybridization but 2+ HER2 immunostaining) he received third-line systemic therapy in a clinical study (NCT04235101; SYD985 [antibody-drug conjugate targeting HER2], IV, 1.2 mg/m^2, 3-weekly, and niraparib tablet, 200 mg QD). After 3 weeks the patient noticed a positive effect on the skin lesions. Unfortunately, both clinical and radiological progression were observed after 3 months. Figure 1M, N, O, P show the extensiveness of the CLC at this stage, which is approximately 10 months after his CLC presentation. Then, he briefly received fourth-line treatment in a clinical study (NCT04291300). However, due to rapid clinical deterioration, the patient died shortly thereafter.

Table 1: Patient characteristics of SDC patients at time of diagnosis of cutaneous lymphangitis carcinomatosa (CLC)

Patient No.	Age at diagnosis	Sex	Primary tumour	N-stage‡	Number of LN metastases	Prior treatments	Year of onset of CLC	Location of CLC	CLC diagnosed based on	Complaints due to CLC	Sites of metastases
1*	65	M	Parotid gland	N3b	25	Surgery, RT†, palliative ADT	Year 1	Neck and Chest region	Clinical presentation, Imaging, Pathology	Neck stiffness, movement restriction	Lymph nodes
2**	63	M	Submandibular gland	N2c	6¶	RT, palliative ADT	Year 1	Neck and Chest region	Clinical presentation, Imaging, Pathology	Mechanical skin tension, movement restriction	Lymph nodes, muscle, Bone
3	52	M	Submandibular gland	N3b	6¶	RT	Year 1	Head and Neck region	Clinical presentation, Imaging, Pathology	-	Lymph nodes
4	60	M	Parotid gland	N3b	NR	-	Year 1	Neck and Chest region	Pathology§	Pain, ulceration	Lymph nodes
5	54	F	Parotid gland	N2b	41	Surgery, RT, chemotherapy, ADT	Year 4	Head and Neck region	Clinical presentation, Imaging, Pathology	Oedema	Lymph nodes, Bone, Liver
6	76	F	Parotid gland	N3b	45	Surgery, RT, adjuvant ADT	Year 1	Head and Neck region	Clinical presentation, Pathology	Pain	-
7	79	F	Parotid gland	N2b	NR	RT, ADT	Year 4	Axilla	Clinical presentation, Pathology	Pain	Lymph nodes
8	73	M	Parotid gland	N2b	8	Surgery, RT, palliative ADT	Year 4	Neck and Chest region	Clinical presentation, Pathology	Itchiness	Lymph nodes, Lung
9	68	M	Parotid gland	N2b	22	Surgery, RT	Year 1	Neck and Chest region	Clinical presentation, Imaging, Pathology	Mechanical skin tension	Lymph nodes

Abbreviations: ADT: androgen deprivation therapy, CLC: cutaneous lymphangitis carcinomatosa, F: female, LN: lymph node, M: male, NR: not reported , RT: radiotherapy.

* This is the patient presented in the manuscript as case report 1.

** This is the patient presented in the manuscript as case report 2.

† Because axillary lymph node metastases were diagnosed shortly after the start of post-operative radiotherapy, this patient discontinued radiotherapy after 1 week.

§ The primary diagnosis was unknown at time of pathology assessment, the CLC was the first clinical sign of his SDC disease.

‡ Presence and extend of regional lymph node metastases.

¶ In these patients a lymph node neck dissection was not performed, but the number of positive lymph nodes was based on imaging in combination with lymph node biopsies.

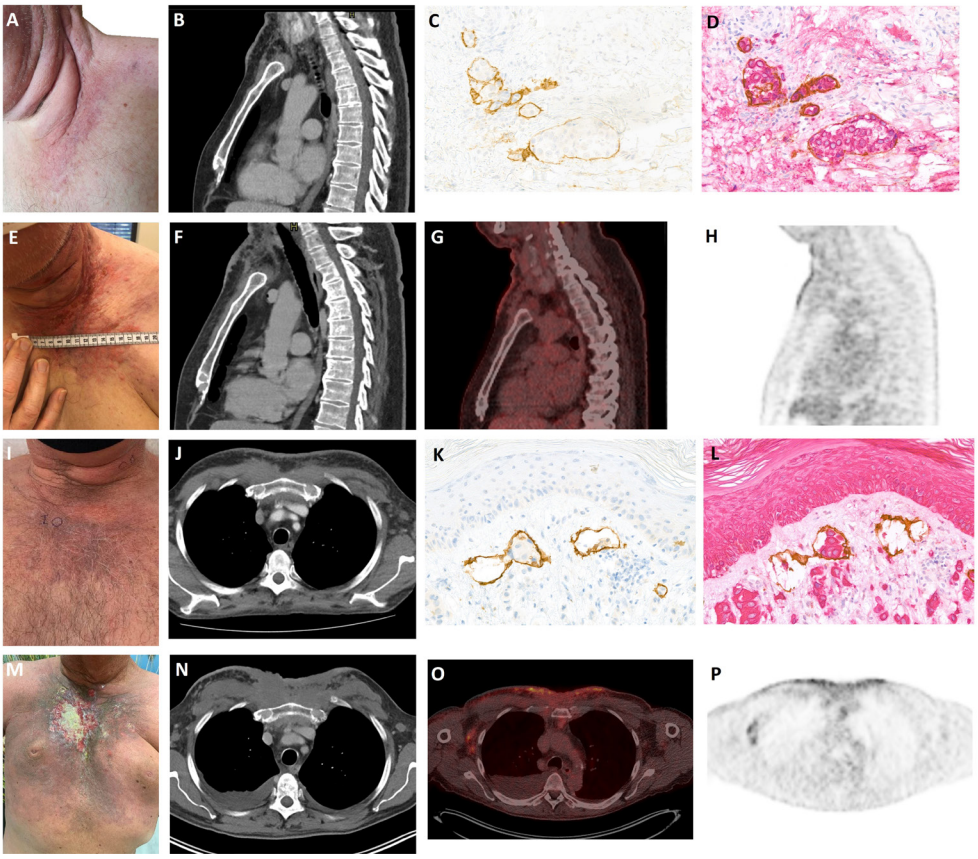


Figure 1: Clinical, radiological, and pathology images of case 1 (A-H) and case 2 (I-P)

Case 1: Figure **A**: clinical picture of case 1 at the first presentation of CLC, showing an erythematous plaque. Figure **B**: sagittal CT image of the case 1 at the first CLC presentation. This CT image shows thickened cutis and subcutaneous induration. Pathological evaluation of the skin biopsy revealed tumour cells within the lumina of dermal lymphatic vessels and infiltration into the dermis and hypodermis (Figure **C:** D2-40 stainings 330X [staining for lymphatic vessels]; Figure **D**: Pan-Cytokeratin staining CK AE 1/3 [red] + D2-40 [brown] stainings 330X). Figure **E:** clinical picture of case 1, approximately 5 months after presentation, with corresponding sagittal CT image (**F**), sagittal fused FDG-PET/CT image (**G**), and sagittal FDG-PET image (**H**). The CT image (F) showed further expansion of the thickened cutis and increase of subcutaneous induration. The PET images showed an increase metabolic activity at the site of the CLC.

Case 2: Figure **I:** clinical picture of case 2 at the first presentation of CLC showing skin changes in the chest region. Figure **J:** axial CT image of case 2, at the first presentation of CLC. On this CT image, a thickened cutis and subcutaneous induration can be observed. Figure **M**: clinical picture of case 2, approximately 10 months after presentation, with corresponding axial CT images (**N**), axial fused FDG-PET/CT image (**O**), and axial FDG-PET image (**P**). At this stage (figure M), ulceration of the skin had occurred. Also in this patient, the radiological evaluation showed further expansion of the thickened cutis and increase of subcutaneous induration. The PET images showed an increase metabolic activity at the site of the CLC. Pathological assessment of the skin biopsy revealed tumour cells within the lumina of dermal lymphatic vessels and infiltration into the dermis and hypodermis (Figure **K:** D2-40 stainings 330X [staining for lymphatic vessels]; Figure **L**: Pan-Cytokeratin staining CK AE 1/3 [red] + D2-40 [brown] stainings 330X).

DISCUSSION

While cutaneous metastases usually present as nodules, CLC manifests itself differently. The skin lesions of the neck and chest region in our SDC patients with CLC varied from an erythematous patch or plaque, with a varying degree of (clinical) oedema, induration, nodules, and/or ulceration. Due to this varying clinical presentation of CLC, the clinical differential diagnosis could be broad and also includes several other (dermatological) conditions, including but not limited to: (chronic) post-radiation dermatitis, erysipelas/cellulitis, herpes zoster, eczema/dermatitis, (fixed) drug eruption, angio-oedema/urticaria, lymphangitis, and dermatomyositis. A representative skin biopsy is recommended to establish the correct diagnosis.

In several cases we observed rapid progression of these skin lesions, such as significant enlargement within weeks to months, which is in line with the general aggressiveness of SDC disease. This rapid progression can limit the utility of local radiotherapy when the radiation area becomes too large. Furthermore, we experienced that CLC did not respond to systemic therapy or only for a very short period of time.

Additionally, we identified four other reports of CLC in SDC patients, each reporting on only one or two patients (5-8). In our experience CLC seems to have a relative high incidence in this rare cancer as compared to other more common cancers where CLC can also be observed, such as breast cancer (9-12), squamous cell carcinoma (10, 13), lung cancer (14), and melanoma (15).

With this report, we want to increase the awareness and the knowledge of CLC as a disease manifestation in SDC patients. Physicians should consider CLC when SDC patients, especially with a history of cervical lymph node metastases, present with skin lesions of the neck and chest regions.

REFERENCES

1. Boukheris H, Curtis RE, Land CE, Dores GM. Incidence of carcinoma of the major salivary glands according to the WHO classification, 1992 to 2006: a population-based study in the United States. Cancer Epidemiol Biomarkers Prev. 2009;18(11):2899-906.
2. McHugh JB, Visscher DW, Barnes EL. Update on Selected Salivary Gland Neoplasms. Archives of Pathology & Laboratory Medicine. 2009;133(11):1763-74.
3. Boon E, Bel M, van Boxtel W, van der Graaf WTA, van Es RJJ, Eerenstein SEJ, et al. A clinicopathological study and prognostic factor analysis of 177 salivary duct carcinoma patients from The Netherlands. International journal of cancer. 2018;143(4):758-66.
4. Kusafuka K, Sato Y, Nakatani E, Baba S, Maeda M, Yamanegi K, et al. The implicated clinical factors for outcomes in 304 patients with salivary duct carcinoma: Multi-institutional retrospective analysis in Japan. Head & neck. 2022.
5. Salivary duct carcinoma metastatic to the skin. Journal of the American Academy of Dermatology. 2016;74(5, Supplement 1):AB71.
6. Takazawa M, Demitsu T, Miyata T, Kakurai M, Tanaka A, Umemoto N. Cutaneous carcinomatous lymphangiosis mimicking angiosarcoma of the face possibly caused by salivary duct carcinoma. The Journal of Dermatology. 2020;47(11):e414-e5.
7. Shin JY, Eun DH, Lee JY, Yoon TY, Kim MK. A Case of Cutaneous Metastases of Salivary Duct Carcinoma Mimicking Radiation Recall Dermatitis. Ann Dermatol. 2020;32(5):436-8.
8. Cohen PR, Prieto VG, Piha-Paul SA, Kurzrock R. The "shield sign" in two men with metastatic salivary duct carcinoma to the skin: cutaneous metastases presenting as carcinoma hemorrhagiectoides. J Clin Aesthet Dermatol. 2012;5(9):27-36.
9. Lymphangitis carcinomatosa: A study of 110 cases. Journal of the American Academy of Dermatology. 2005;52(3, Supplement):P148.
10. Damstra RJ, Jagtman EA, Steijlen PM. Cancer-related secondary lymphoedema due to cutaneous lymphangitis carcinomatosa: clinical presentations and review of literature. Eur J Cancer Care (Engl). 2010;19(5):669-75.
11. LEVER LR, HOLT PJA. Carcinoma erysipeloides. British Journal of Dermatology. 1991;124(3):279-82.
12. Cox SE, Cruz PD. A spectrum of inflammatory metastasis to skin via lymphatics: Three cases of carcinoma erysipeloides. Journal of the American Academy of Dermatology. 1994;30(2, Part 2):304-7.
13. van Kester MS, Genders RE, Quint KD. Lymphangitis carcinomatosa of a cutaneous squamous cell carcinoma. BMJ Case Rep. 2018;2018.
14. Prat L, Chouaid C, Kettaneh A, Fardet L. Cutaneous lymphangitis carcinomatosa in a patient with lung adenocarcinoma: case report and literature review. Lung Cancer. 2013;79(1):91-3.
15. Arfi A, Clavier A, Honart JF, Ammari S, Merabet Z, Mateus C, et al. Blue Lymphangitis of the Breast after Breast Cancer: Case Report. Breast J. 2017;23(3):348-51.

SUPPLEMENTARY

Supplementary table 1: Molecular analysis of SDC patients with cutaneous lymphangitis carcinomatosa (CLC)

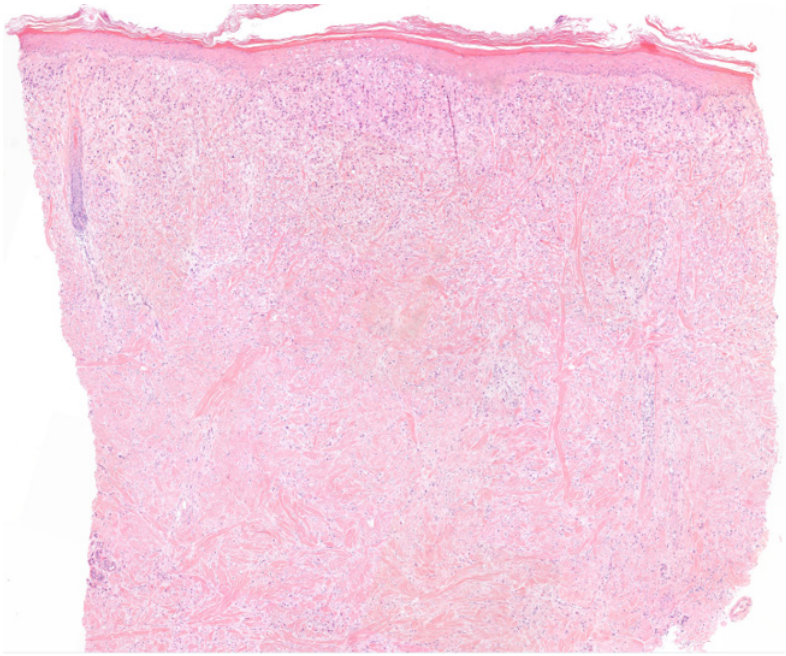
Patient No.	Tissue used for molecular analysis	AR expression (IHC)¶	HER2 status ASCO/CAP§	NGS panel	Identified genetic aberrations	Amplifications	Non-synonymous TMB	Micro Satellite Instability
1	Primary tumour	Positive (% NR)	Negative	TruSight Oncology 500	*PIK3CA** p.(His1047Arg) *HRAS** p.(Gln61Arg) *TP53* p.(Arg209fs)	No	3.2 mut/Mb	No
2	Muscle biopsy	Positive (50%)	Negative	Custom Ampliseq Cancer Hotspot v6 panel	*TP53* p.(Arg175His)	No	N.P.	N.P.
3	Skin biopsy	Positive (60%)	Positive	TruSight Oncology 500	*TP53* p.(Arg248Gln)	*ERBB2* *CDK12*	1.6 mut/Mb	No
4	Primary tumour	Positive (100%)	Positive	TruSight Oncology 500	*PIK3CA*** p.(Glu542Gln) p.(Glu545Lys) *TP53* p.(Asp21fs)	*FGFR1*†	67 mut/Mb	No
5	Skin biopsy‡	Positive (% NR)	Negative	PATH genenpanel version 2.0	*HRAS** p.(Gln61Lys) *PIK3CA** p.(His1047Leu)	No	N.P.	No
6	Primary tumour	Positive (60%)	Negative	TruSight Oncology 500	*MSH6* p.(Glu122fs) *PTEN* p.(Cys136Phe) *TP53* p.(Gln52*)	No	13.8 mut/Mb	No
7	Lymph node	Positive (100%)	Negative	Radboud Cancer Hotspot genenpanel version 2.0	-	No	N.P.	No
8	Primary tumour	Positive (% NR)	N.P.	Radboud Cancer Hotspot genenpanel version 1.0	*AKT1** p.(Glu17Lys) *BRAF** p.(Val600Glu)	No	N.P.	No
9	Primary tumour	Positive (% NR)	Positive	PATH genenpanel version 2.0	*ERBB2** p.(Leu755Ser) *TP53* p.(Gly334Trp)	No	N.P.	No

Abbreviations: AR: androgen receptor, FISH: fluorescence in situ hybridization, IHC: immunohistochemistry, Mb: megabase, N.P.: not performed, N.R.: not reported, NGS: Next generation sequencing, TMB: tumour mutational burden.

Legend continues on next page.

* Druggable target
** Two different mutations found for this gene, positioned on different alleles (trans)
† Amplification of FGFR1, however under validated cut-off. Amplification was not confirmed with other techniques.
§ HER2 status ASCO/CAP according to: Wolff AC *et al*. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology (ASCO)/College of American Pathologists (CAP) Clinical Practice Guideline Focused Update. Arch Pathol Lab Med 2018;142(11):1364–82.
¶ When available, the percentage of AR positive tumour cells is reported below, in case this is not reported in the pathology report this is presented as N.R..
‡ In this patient prior molecular analysis was also performed on the primary tumour. The results of this molecular analysis were similar to the skin biopsy results (e.g. the same mutations identified).

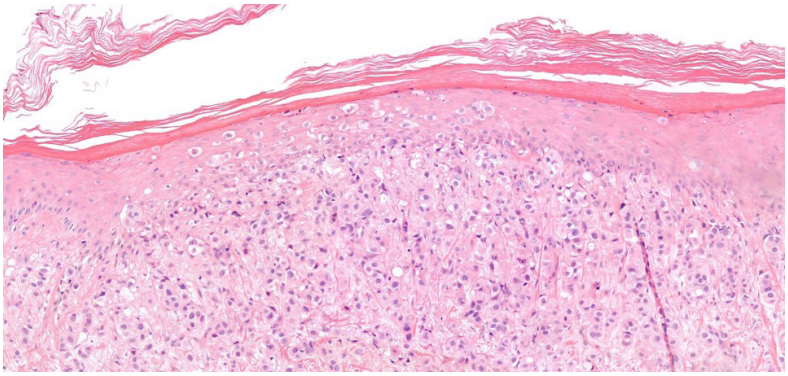
Detailed description of the different NGS panels:
- TSO500 panel; https://www.palga.nl/datasheet/Radboudumc/TSO500.pdf
- CHPv1 panel; https://www.palga.nl/datasheet/Radboudumc/CHPv1.pdf
- CHPv2 panel; https://www.palga.nl/datasheet/Radboudumc/CHPv2.pdf
- Custom CHPv6 panel; https://www.palga.nl/datasheet/LUMC/Pancancer_CHPv6_LUMC.pdf
- PATHv2D panel; https://www.palga.nl/datasheet/Radboudumc/PATHv2D.pdf



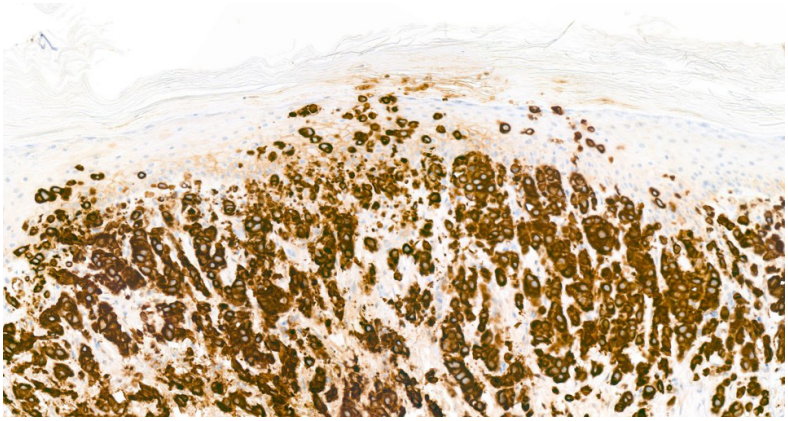
Supplementary Figure 1: H&E stained skin biopsy of case 1 (30X) showing the cutaneous metastases.

Supplementary Figure 2: CK-7 stained skin biopsy of case 1 (30X) showing the cutaneous metastases.

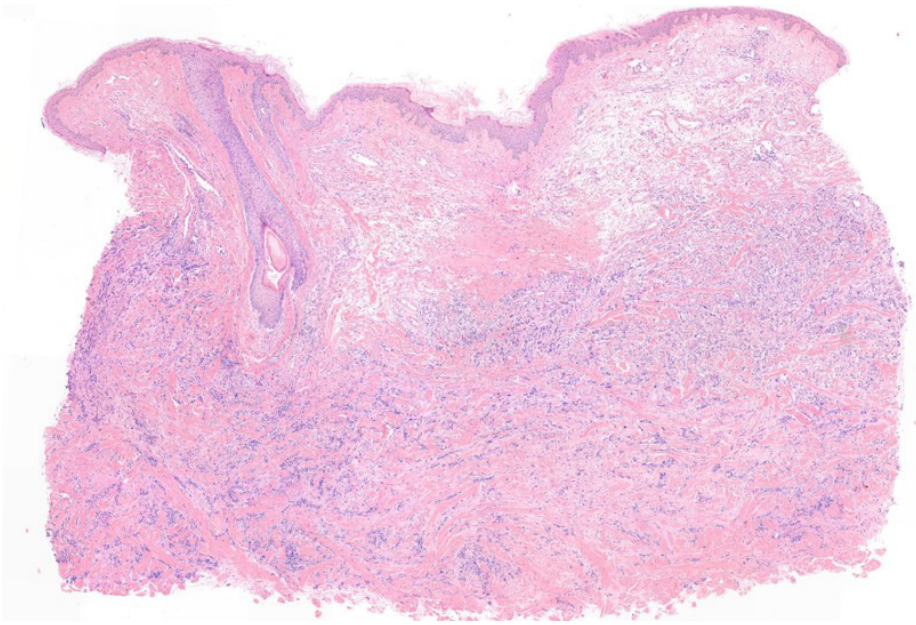
A



B



Supplementary Figure 3: A: H&E stained skin biopsy of case 1 (130X) showing pagetoid spread of tumor cells in the epidermis. **B**: CK-7 stained skin biopsy of case 1 (130X) showing pagetoid spread of tumor cells in the epidermis.



Supplementary Figure 4: H&E stained skin biopsy of case 2 (30X) showing the cutaneous metastases.

Supplementary Figure 5: CK-7 stained skin biopsy of case 2 (30X) showing the cutaneous metastases.

Chapter 11

Summary

SUMMARY

Adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC) are two histological subtypes of salivary gland cancer (SGC) in which local recurrences or distant metastases occur in a large proportion of patients (**chapter 1**). Unfortunately, at that stage, the disease is no longer curable. For these patients, palliative systemic therapy can potentially prevent or treat disease-related symptoms and increase patient survival. Yet, due to the general rarity of SGC and therefore also ACC and SDC, a relatively limited amount of research has focused on palliative systemic therapy for ACC and SDC, as compared to more common cancers.

Therefore, this thesis focusses on palliative systemic therapy for ACC and SDC patients by:
- providing a systematic overview and summary of previously studied palliative systemic therapy for SDC patients (**chapter 2**). A comparable overview for ACC patients had already been published previously;
- exploring the efficacy and safety of new treatments for SGC, i.e. cabozantinib (**chapter 3**) and prostate-specific membrane antigen (PSMA) targeted therapy (**chapter 6**);
- investigating if targeted therapy for well-established therapeutic targets could be optimized, i.e. the addition of pertuzumab to Human epidermal growth factor 2 receptor (HER2) targeted therapy in HER2-positive SDC patients (**chapter 4**).

Furthermore, this thesis also provides more research regarding the PSMA target (**chapter 5 and chapter 7**), insights into the difficulty of histopathological diagnosis of SGC (**chapter 8**), and case reports regarding a potential risk factor for SDC (**chapter 9**).

Part 1: Systemic therapy for Salivary Gland Cancer

In **chapter 2** we conducted a broad systemic literature search on palliative systemic therapy for SDC patients. Through our search, 2014 articles were identified, of which 45 articles contained the relevant and specific information we aimed to find. In total, these 45 articles contained information on 256 SDC patients who received palliative systemic therapy. The results were categorized in the type of systemic therapy: chemotherapy, immunotherapy, hormonal therapy, or targeted therapy. We identified two phase II trials which primarily included SDC patients. One studied the effect of androgen deprivation therapy and the other studied HER2 targeted therapy. Both studies showed promising results. Overall, based on our literature study, we could conclude that androgen deprivation therapy should be considered for androgen receptor-positive SDC patients (response rates 18-53%) and that HER2 therapy is highly effective in HER2-positive SDC patients (response rates 60-70%). Regarding chemotherapy, the combination of carboplatin combined with a taxane was best studied in SDC patients, with response rates of 39-50%. For other targeted agents and immunotherapy, evidence was only anecdotal. This limited treatment recommendations for these agents.

Because prior research showed that the tyrosine kinases c-MET and vascular endothelial growth factor receptors are often overexpressed in SGC, we evaluated the efficacy of cabozantinib (a tyrosine kinase inhibitor) for this cancer. The results of this clinical phase II study on cabozantinib in patients with recurrent or metastatic SGC were described in **chapter 3**. In total 25 SGC patients received cabozantinib treatment. Even though the initial intention was to include more patients (following the sample size calculation), the study was closed prematurely owing to severe toxicity. Six of the 25 patients (24%) had grade ≥3 wound complications. Remarkably, four of these six patients developed this complication in an area that had previously been exposed to (high-dose) radiotherapy. Furthermore, the response rate of cabozantinib was low to moderate. Of the response evaluable patients (cabozantinib treatment ≥8 weeks), 1/15 ACC patients (7%), 1/4 SDC patients (25%), and 0/2 other miscellaneous SGC patients (0%) showed an objective response. Overall, this study showed too many severe cabozantinib-associated wound complications in patients with SGC, especially in prior irradiated areas, and therefore cabozantinib could not be recommended as palliative systemic therapy in SGC patients.

In **chapter 4** we retrospectively evaluated the results of docetaxel, trastuzumab, and pertuzumab as first-line HER2 targeted treatment, and ado-trastuzumab emtansine as second-line HER2 targeted treatment for HER2-positive recurrent or metastatic SDC patients. Between 2015 and 2021, 13 patients received the combination of docetaxel, trastuzumab, and pertuzumab. Of the 12 response evaluable patients, one complete response and six partial responses were observed, resulting in an overall response rate of 58%. The median duration of docetaxel, trastuzumab, and pertuzumab treatment was 6.9 months (range 3.4–26.8+). Seven patients received subsequent ado-trastuzumab emtansine in second-line HER2 therapy, resulting in four partial responses and an overall response rate of 57%. The median time on ado-trastuzumab treatment was 8.5 months (range 1.1–20.4).

The median overall survival of the 13 patients after the start of docetaxel, trastuzumab, and pertuzumab was 42.0 months (95%-confidence interval: 13.8-70.1 months). Grade ≥3 toxicity in 39% of patients on docetaxel, trastuzumab, and pertuzumab, and in 14% of patients on ado-trastuzumab emtansine. We concluded that docetaxel, trastuzumab, and pertuzumab followed by ado-trastuzumab emtansine upon progression are promising treatments for recurrent or metastatic HER2-positive SDC patients, leading to responses in the majority of the patients at an acceptable toxicity profile.

Part 2: PSMA radioligand therapy for Salivary Gland Cancer

Prostate-specific membrane antigen (PSMA) is a transmembrane protein, which was discovered and is highly overexpressed in prostate cancer. PSMA can pose as a target for targeted therapy. Interestingly, PSMA is not solely expressed by prostate cancer cells, but also in other types of cancer. In **chapter 5** we summarized the current knowledge of PSMA

expression in other solid cancers and defined a perspective on broader clinical implementation of PSMA targeted therapy. This review focused on seven different solid cancers, including SGC. Interestingly, PSMA expression in these seven solid cancers was primarily observed on the tumour neovasculature. This contrasts with prostate cancer, where the expression is observed on the tumour cells themselves. Only in ACC patients, PSMA was also expressed on the tumour cells. Furthermore, we found two case reports of ACC patients who received PSMA targeted therapy, but these provided relatively little insight on the treatment effect in these patients. This encouraged us to properly investigate the effect of PSMA targeted treatment with a prospective study in SGC patients.

Chapter 6 described the preliminary results of an ongoing phase II pilot study on PSMA targeted therapy. Targeted treatment consisted of PSMA radioligand therapy with ^{177}Lu-PSMA. By coupling a radionuclide (Lutetium-177 [^{177}Lu, a β-emitter]) to a PSMA targeting molecule (the ligand PSMA-I&T), a cytotoxic radiation load was specifically delivered to the tumours. Only patients with a presence of the PSMA protein in the tumours, assessed by PSMA PET imaging, were eligible for ^{177}Lu-PSMA treatment.
This study started in May 2020 and currently 10/10 ACC patients and 2/5 SDC patients have started treatment. This chapter described the preliminary results of the first 8 ACC patients in this study (with 1 December 2021 as the cut-off date for this analysis). Until that date, 12 ACC patients were screened for eligibility and 8 ACC patients met the inclusion criteria and received at least one cycle of ^{177}Lu-PSMA. Our preliminary results indicate that ^{177}Lu-PSMA is well-tolerated in ACC patients, but the efficacy thus far seems limited only to achieving stable disease. Notably, in 2/8 ACC patients (25%) their stable disease has lasted > 6 months, yet these numbers may change with a longer follow-up.

Chapter 7 reported the study protocol (from 2019) of an ongoing clinical imaging study in patients with recurrent or metastatic SDC. This ongoing study investigates the effect of androgen deprivation therapy on PSMA-ligand uptake, evaluated with repeated PSMA PET scans. Since research in prostate cancer indicated that androgen deprivation therapy can increase PSMA expression/PSMA-ligand uptake, this imaging study explores whether this is also the case in SDC patients.

Part 3: Increasing general knowledge of Salivary Gland Cancer

In **Chapter 8** the results of a project in collaboration with colleagues from the Department of Otorhinolaryngology and Head & Neck Surgery was presented. During this project, we evaluated the frequency and outcomes of pathology consultations and revisions concerning major salivary gland tumours in routine clinical practice through the Nationwide Pathology Network of the Netherlands (PALGA). Between 2006 and 2016, 13,441 major salivary gland neoplasms were resected in the Netherlands. Revision of the histopathological specimen was performed in 2.6% (n=350) of the cases and these were discordant in 8.3% (n=29). This

number (8.3%) emphasizes the complexity of the histopathological diagnosis of salivary gland neoplasms. We proposed that an increase in consultations may improve the accuracy of the initial diagnosis in salivary gland tumours while lowering the need for revisions and the number of discordant revisions.

In **Chapter 9** we described a potential risk factor for the occurrence of SDC in two patients. As mentioned, the risk factors for the development of SGC and SDC are largely unknown, although air pollution has been described as one potential risk factor. In other cancers, especially lung cancer, the carcinogenicity of chromium VI is well-known. In this chapter, we reported on two SDC patients who were occupationally exposed to chromium VI through maintenance of military equipment. However, a causal relation between chromium exposure and the occurrence of SDC cannot be demonstrated on an individual basis, and detection in a population-based study is unlikely because of the extremely low prevalence of SDC. Nevertheless, we considered it plausible that the chromium VI exposure might have contributed to the occurrence of their SDC. With this report we aimed to increase the awareness of occupational chromium exposure and occurrence of SDC

In **Chapter 10** we reported on nine SDC patients who developed cutaneous lymphangitis carcinomatosa (CLC), which resulted in skin lesions of the neck and chest region. All these patients had extensive lymph node metastases at diagnosis. Pathophysiologically, it is highly plausible that in these patients with extensive lymph node metastases the SDC tumour cells have spread further via the lymphatic vessels, causing cutaneous metastases in the drainage area. We observed that CLC manifested itself differently from other cutaneous metastases. While cutaneous metastases usually present as nodules, the skin lesions of the neck and chest region in our SDC patients with CLC varied from an erythematous patch or plaque, with a varying degree of (clinical) oedema, induration, nodules, and/or ulceration. With this report, we wanted to increase the awareness and the knowledge of CLC as a disease manifestation in SDC patients.

Chapter 12

General discussion and future perspectives

DISCUSSION

As described in the general introduction (chapter 1), research on rare cancers, such as salivary gland cancer (SGC), generally faces several challenges compared to more common cancers, e.g. more difficulty in recruiting patients for clinical trials, less research funding, and less publicity.

Nevertheless, the research described in this thesis shows that we overcame several of these obstacles. We received funding for clinical trials for SGC patients due to specific attention for research on rare cancers from research funders (1), and due to international collaboration and collaboration with pharmaceutical companies. Due to the centralization of head and neck cancer in the Netherlands and the development of the Radboudumc towards a tertiary centre for SGC, we were able to include SGC patients in several prospective clinical studies. All this contributed to the research in the framework of this thesis, which investigated new systemic treatment options for SGC patients, with a specific focus on the adenoid cystic carcinoma (ACC) and salivary duct carcinoma (SDC) subtypes, as recurrences and distant metastases often occur in patients with these subtypes.

The unmet need for systemic treatment options for this rare cancer was illustrated by the fact that SGC patients from all over the world contacted us frequently regarding the clinical trials we had reported on international trial registries (clinicaltrials.gov).

Even though research in common cancers is moving faster and with larger patient numbers as compared to rare cancers, we can use these outcomes of common cancers and try to translate these to rare cancers. For example, the beneficial effects of adding pertuzumab to trastuzumab and docetaxel were first demonstrated in HER2-positive breast cancer (common cancer) and therefore we also investigated this addition of pertuzumab in HER2-positive SDC (rare cancer). The fact that PSMA radioligand therapy is not yet registered for prostate cancer (common cancer) and that we are already conducting research into this therapy for SGC, shows that we try to rapidly seize the opportunity of translating new therapeutical innovations to rare cancers.

Here, we will discuss the therapeutical landscape for palliative systemic therapy for ACC and SDC patients. Meanwhile, we describe how our results add to this knowledge and current treatment strategies, and define future perspectives.

PALLIATIVE SYSTEMIC THERAPY FOR RECURRENT OR METASTATIC ACC

Because of the often indolent tumour growth of ACC, early initiation of systemic therapy and its potential benefits must be weighed against its side effects. Recently published international clinical guidelines provide recommendations regarding this important consideration (2).

Chemotherapy

Chemotherapy was one of the first systemic treatment options which showed clinically relevant activity in recurrent or metastatic ACC tumours. Since clinical trials showed the highest response rates (25%) with combinations of cisplatin and anthracycline (such as cyclophosphamide plus doxorubicin plus cisplatin), this proved to be a valid systemic treatment option for ACC patients. However, this regimen is relatively toxic and requires a good baseline performance score of the patient. Therefore, research continues to focus on other treatment options for ACC patients, such as tyrosine kinase inhibitors, which generally show a more favourable toxicity profile as compared to chemotherapy regimens for ACC.

Tyrosine kinase inhibitors (TKIs)

TKIs are drugs that inhibit certain proteins, tyrosine kinases, that are involved in cancer cell growth and survival (3). Analysis of the human genome indicated that there are more than 50 tyrosine kinases, divided into more than 20 subfamilies (4). Tyrosine kinases contribute differently to signalling pathways in each cancer type, and therefore it is a topic of research to identify the most relevant TKI for each cancer type. For ACC, the effect of several TKIs was already investigated before we evaluated the effect of the TKI cabozantinib. Some of these clinical trials on TKIs in ACC showed promising results, e.g. lenvatinib, sorafenib and apatinib, with response rates of 16%, 16% and 47%, respectively (5-7). While other TKIs failed to show objective responses in ACC, e.g. regorafenib, sunitinib (8, 9). Whilst these are all single-arm studies, a first randomized TKI study has also been published recently. In this randomized study, axitinib was compared to observation. Remarkably, axitinib significantly increased the 6-month progression-free survival rate when compared to observation (73% with axitinib vs 23% with observation) (10).

The profile of cabozantinib differs from these previously studied TKIs due to its strong inhibition of the tyrosine kinases c-MET and AXL. Although cabozantinib treatment did lead to an objective response in an ACC patient (objective response rate: 7%), its efficacy seems to be less than lenvatinib, sorafenib, and apatinib in previous studies (5-7). Due to the low efficacy of cabozantinib, we cannot advise the use of this specific TKI in ACC patients. Moreover, the observed toxicity of cabozantinib is an even more important reason not to give this treatment to SGC patients. This toxicity included serious wound complications, which resulted in the premature closure of our clinical trial.

The number of wound complications with cabozantinib was much higher as compared to other TKIs in other ACC studies. Therefore, we hypothesized that this might be due to the specific TKI profile of cabozantinib. Both c-MET and AXL, which are strongly inhibited by cabozantinib, are tyrosine kinases that are involved in wound healing (11, 12). We considered inhibition of these targets by cabozantinib, especially in areas with prior tissue atrophy, fibrosis, and vascular damage due to prior (high-dose) radiotherapy, to be the most plausible explanation for the observed higher number of wound complications in our clinical study.

All in all, several TKIs have shown objective response rates comparable to chemotherapy regimens. Therefore, ACC patients may benefit from TKI treatment such as lenvatinib, sorafenib, apatinib, and axitinib. Interestingly, another phase II study on apatinib in ACC patients in the USA is currently recruiting (NCT04119453), based on the relatively high response rate of the phase II study of apatinib in China (7).

Immunotherapy

To date, only a limited amount of research has focused on the clinical efficacy of immunotherapy in SGC (13, 14).

Generally, tumours with a high tumour mutational burden and high expressions of immune checkpoints are more likely to respond to immunotherapy. However, ACC tumours are considered immunologically 'cold' tumours, since they generally have a low mutational burden (15), are immune-cell deprived (16), and have no or limited expression of immune checkpoints (e.g. PD-1, PD-L1, and CTLA-4) (17), which makes immunotherapy unappealing. Rather than trying to activate the limited number immune cells in ACC tumours with immunotherapy, it may be worthwhile to explore methods that could increase the number of immune cells in these tumours.

Genomic profiling

Despite the fact that previous research indicates that ACC tumours show a relatively low tumour mutational burden, genomic profiling of tumours might still uncover potentially druggable mutations in a low proportion of patients (15). In this case, these patients may benefit from precision medicine and could be included in clinical trials such as DRUP (NCT02925234), TAPUR (NCT02693535) or CAPTUR (NCT03297606). Furthermore, since activating *NOTCH* mutations are found in approximately 20% of ACC patients, an ongoing study is evaluating the effect of a NOTCH inhibitor (NCT0369120).

PALLIATIVE SYSTEMIC THERAPY FOR RECURRENT OR METASTATIC SDC

Androgen deprivation therapy

Androgen deprivation therapy remains an interesting area of research in SDC patients, because the vast majority of SDC tumours express the androgen receptor and because this type of therapy induces relatively limited side effects. As mentioned in our systematic review, approximately half of the SDC patients respond to combined androgen deprivation therapy (LHRH analogue + bicalutamide). Predictive biomarkers may be useful to improve the selection of patients who respond to androgen deprivation therapy. mRNA-based signalling pathway activity scores (OncoSignal test; the same test used in Chapter 4 as a biomarker for HER2 response) showed potential in predicting clinical benefit. In a retrospective cohort of 76 SDC patients, the patients with low androgen-receptor pathway activity scores were less likely to respond to androgen deprivation therapy (18). Interestingly, also 5-alpha reductase type 1 expression (an enzyme responsible for the conversion of testosterone into the more active metabolite dihydrotestosterone) showed predictive value. Patients with a high 5-alpha reductase type 1 expression were more likely to respond to androgen deprivation therapy than patients with low expression (18). SDC tumours with high 5-alpha reductase type 1 expression may potentially be more dependent on androgens for survival. Since 5-alpha reductase type 1 can be inhibited by dutasteride, it may be beneficial to add this (relatively well-tolerated) drug to combined androgen deprivation therapy in the future.

Furthermore, it is not yet clear whether second-line androgen deprivation therapy in SDC tumours will be effective, as is the case in prostate cancer. A prospective study investigating second-line abiraterone therapy in SDC patients showed moderate efficacy (19). Other second-line drugs for prostate cancer (e.g. enzalutamide and apalutamide) have not been properly investigated in a second-line setting in SDC. Enzalutamide (20) and apalutamide (NCT04325828) are investigated, but not in a second-line setting.

Human epidermal growth factor 2 receptor (HER2) targeted therapy

A phase II study that evaluated the effect of docetaxel and trastuzumab in 57 patients with HER2-positive recurrent or metastatic SDC resulted in an objective response rate of 70%. The median progression-free survival and overall survival of this cohort were 8.9 and 39.7 months, respectively (21). In our retrospective study, we aimed to investigate the efficacy of adding pertuzumab to docetaxel and trastuzumab treatment, following the positive results of this combination in clinical studies in HER2-positive breast cancer. Even though our results cannot be compared with the previously mentioned phase II study on docetaxel and trastuzumab, due to the differences in patient population (e.g. 23% of patients with brain metastases in our study compared to 5% in the phase II study) and study design, we provided a rationale to add pertuzumab to the HER2-targeted treatment regime. Since the synergetic effect of

trastuzumab and pertuzumab has been demonstrated in more common cancers and we did not observe an increase in the overall toxicity upon the addition of pertuzumab, we would like to suggest to add pertuzumab to docetaxel and trastuzumab treatment in SDC. Ideally, a randomized study should be performed to compare the efficacy of HER2 treatment with and without pertuzumab, yet this is challenging due to the rarity of SDC.

Furthermore, we reported on the effects of second-line HER2 targeted therapy, with ado-trastuzumab emtansine. We observed a response rate of 57% and an acceptable toxicity profile, advocating the use of this therapy in HER2-positive SDC patients as second-line therapy.

Compellingly, due to new HER2 targeted drugs such as trastuzumab deruxtecan (a new antibody-drug conjugate targeting HER2), the HER2 treatment strategy may be subject to innovations in the future.

Chemotherapy

Although the effect of chemotherapy in SDC patients has mainly been reported in retrospective settings, an ongoing phase II study is currently comparing the effect of chemotherapy versus androgen deprivation therapy as first-line treatment in patients with androgen receptor-positive SGC (NCT01969578). It is likely that most of these SGC patients will be of the SDC subtype, and thus this study will provide more insights in the efficacy and toxicity of chemotherapy in SDC patients in a prospective setting.

Immunotherapy

Our systematic review on systemic therapy in patients with recurrent or metastatic SDC indicated that immunotherapy is the least researched category out of the four different categories of systemic therapy (i.e. chemotherapy, hormonal therapy, targeted therapy, and immunotherapy).

Yet, of all SGC patients, SDC patients may be among the best candidates for response to immunotherapy. This is due to the relatively high tumour mutational burden (14% of SDC tumours has a tumour mutational burden of >10 mutations per megabase (15)) and immunohistochemical expression of immune checkpoints (e.g. PD-L1 positivity [>1%] in 26-60% of cases) (22, 23). As previously mentioned, a high tumour mutational burden and immunohistochemical expression of immune checkpoints are potential biomarkers for response to immunotherapy.

Since the publication of our systematic review (2019), one case report presented a long-term durable response to pembrolizumab (an immune-checkpoint inhibitor) of a patient with metastatic SDC (24).

In contrast, a recent study with nivolumab (another immune-checkpoint inhibitor) in recurrent or metastatic SGC showed low efficacy (25). Of the 24 SGC patients that received nivolumab, SDC was the most common histological subtype; 20 (83%) were SDC patients. The objective response rate in this study was only 4% (1 SDC patient showed a partial response). Better results were obtained by combining two immunotherapy agents. Namely, the combination of nivolumab and ipilimumab (both immune-checkpoint inhibitors, but with a different target) showed promising preliminary results; 25% of SDC patients (3/12) responded (26). Remarkably, these responses was relatively long-lasting, ranging from 15.7 to 29.5 months (26). Thus, to improve the response rate to immunotherapy in SDC patients future research may focus on combination strategies (e.g. combining different immune checkpoint inhibitors). Additional approaches may be to focus on better patient stratification (e.g. focus on immune-infiltrated SDCs) or a different immune checkpoint target (e.g. anti-CTLA-4 (27)).

Genomic profiling

As with ACC tumours, genomic profiling of SDC tumours may show druggable mutations. The probability of druggable mutations and opportunity for precision treatment are likely to be relatively high in this subtype, given the high mutational burden in SDC tumours as compared to other SGC subtypes (15). SDC patients with druggable mutations may benefit from precision treatment in the clinical trials that were mentioned previously in the paragraph on genomic profiling in ACC patients.

PROSTATE-SPECIFIC MEMBRANE ANTIGEN (PSMA) TARGETED RADIOLIGAND THERAPY FOR ACC AND SDC PATIENTS

PSMA targeted radioligand therapy (PSMA-RLT) is a form of systemic therapy in which cytotoxic local radiation is delivered to PSMA expressing tumour cells. This is accomplished by coupling a ligand that binds to PSMA (e.g. PSMA-617 or PSMA-I&T) with a radionuclide that emits radiation (e.g. Lutetium-177 [^{177}Lu; beta-emitter], or Actinium-225 [^{225}Ac; alpha-emitter]).

Multicentre retrospective and small prospective studies showed promising biochemical results for ^{177}Lu-PSMA-RLT in prostate cancer (28-30). More recently, the pivotal phase III study (VISION) showed that ^{177}Lu-PSMA-RLT improved both the median progression-free survival (8.7 vs 3.4 months) and median overall survival (15.3 vs 11.3 months) compared to standard care in advanced PSMA-positive metastatic castration-resistant prostate cancer patients (31). Moreover, all these studies showed a favourable toxicity profile of ^{177}Lu-PSMA-RLT in prostate cancer patients. The most often observed grade 1-2 side effects were: fatigue, dry mouth, and nausea. The most often grade ≥3 side effects was bone marrow toxicity (anaemia, thrombocytopenia, lymphopenia). The combination of positive treatment results and favourable toxicity profile makes ^{177}Lu-PSMA a promising treatment for other PSMA expressing tumours, including SGC.

PSMA expression with immunohistochemistry and PSMA avidity on PSMA PET had previously been demonstrated in ACC and SDC patients (32-36). Based on these findings we are currently conducting a prospective clinical trial of ^{177}Lu-PSMA-RLT in patients with recurrent and/or metastatic ACC and SDC with relevant PSMA avidity on ^{68}Ga-PSMA PET (NCT04291300). The first results in the first ACC patients in this ongoing trial have been presented in chapter 6. Thus far, we did not yet observe an objective response but 25% (2/8) of the patients did show stable disease lasting > 6 months. As this study exploring PSMA targeted-radionuclide therapy is still ongoing, it is too early to draw final conclusions with respect to its efficacy. Importantly, this treatment is generally well tolerated. Overall, the side effects seem comparable to the side effects witnessed in prostate cancer studies, with two exceptions. First, the proportion of patients with a dry mouth (xerostomia) following ^{177}Lu-PSMA-RLT is higher among SGC patients than among prostate cancer patients (75% vs ≈40%). This is probably due to the pre-existing lower salivary gland reserve in patients with SGC, owing to prior localized treatment (i.e. surgery and radiotherapy). Second, bone marrow toxicity is less extensive in SGC patients as compared to prostate cancer patients (grade ≥3 anaemia 0% vs 13%). This is likely due to more bone marrow reserve in SGC patients as compared to prostate cancer, where patients often have extensive bone metastases.

A retrospective case series was recently published on six SGC patients who received ^{177}Lu-PSMA-RLT in a compassionate use program (37). This report also included four ACC patients. Clinical response was observed in one ACC patient: improved facial expression and sensibility, indicating a biological effect of ^{177}Lu-PSMA-RLT. The tumour imaging of this patient showed a partial response on ^{68}Ga-PSMA PET, defined by the researchers as a SUVmax decrease of ≥30% compared to baseline. However, the radiological evaluation of this patient showed stable disease. Based on the retrospective study in SGC patients and our preliminary study results in the first 8 ACC patients, it could be cautiously said that there may be some biological effectiveness of ^{177}Lu-PSMA treatment in SGC patients, however the effectiveness is much less pronounced than in prostate cancer patients. Potential explanations for this lower efficacy of ^{177}Lu-PSMA-RLT in SGC patients could be: a reduced radiation sensitivity, a lower tumour radiation dose (PSMA expression and PSMA ligand uptake in SGC is generally lower than in prostate cancer), or a reduced secondary immune response. Further research, such as dosimetry (calculating the achieved radiation doses) based on post-therapy SPECT imaging, might provide more insights into the tumour radiation doses that are achieved in SGC patients. These dosimetry results can be compared to dosimetry studies in prostate cancer. Furthermore, better outcomes may be obtained by improving the patient selection. This can potentially be accomplished by applying pre-therapeutic PET dosimetry or prolonged in vivo evaluation of PSMA PET tracers with a radionuclide with a longer half-life than Gallium-68 (^{68}Ga), for example Zirconium-89 (^{89}Zr) (38).

CLINICAL IMPLICATIONS

This thesis describes research on various palliative systemic treatments for salivary gland cancer (SGC) patients based on positive results of these treatments in common cancers. Our results show that some of these therapies also lead to positive results in SGC patients, for example ado-trastuzumab emtansin as second-line HER2 targeted therapy in HER2-positive SDC. While on the other hand, some other treatments proved unsuitable for SGC patients, such as cabozantinib therapy.

Additionally, we provide a perspective on where our findings may fit into the broader treatment landscape of systemic therapy for ACC and SDC. With this research, we endeavoured to contribute to the optimization of the treatments for SGC patients.

FUTURE PERSPECTIVES

To further optimize the palliative systemic treatment for SGC patients, future research should focus on the following three areas:

- Research into new therapeutic targets or drug combinations

This applies particularly to ACC patients, as the investigated treatments thus far are unsatisfactory. Despite the fact that several tyrosine kinase inhibitors showed responses in ACC patients, only combined chemotherapy regimens currently remain as alternatives. Therefore, ACC patients would benefit from new therapeutic approaches. However, ACC tumours generally have a low mutational burden, and therefore less targets for potential therapy. Remarkably, *MYB*, *MYBL1* or *NFIB* genes alterations, specifically gene fusions, are observed in the majority of ACC tumours. It is considered likely that these gene fusions increase the oncologic activity of ACC tumours (39). Targeted therapy directed at related pathways may be worth focusing on. Furthermore, as previously described the PSMA receptor is an interesting target in ACC disease, since this target is also present in most of the ACC tumours. In addition to our ongoing study with a PSMA-targeted radionuclide approach, other PSMA-targeted anticancer tactics are being investigated. For example, a method to use this PSMA receptor to induce T-cell enhancement are explored in several solid cancers (NCT04839991). Since ACC tumours are often T-cell deprived, these new methods would be interesting to investigate for ACC in an aim to increase the immunological response.

- Optimization of targeted therapy for well-established therapeutical targets

HER2 is a well-established target for therapy in SDC patients. It would be useful to investigate the optimal anti-HER2 treatment strategy for these patients. Recently, new anti-HER2 drugs have been developed, for example the antibody drug conjugate 'trastuzumab-deruxtecan', which might be even more effective than current anti-HER2 treatments (40). Another example is the optimization of treatment directed against the androgen receptor pathway. As mentioned previously, addition of a 5-alpha reductase inhibitor to existing therapy (LHRH analogue + bicalutamide) might lead to even better results in androgen receptor-positive SDC patients.

- Optimizing patient selection

Since no systemic treatment is without side effects, offering certain therapies only to patients who are likely to benefit from them could also help maintain quality of life for SGC patients. New technologies (e.g. pathway activity quantification) may be able to help optimization of patient selection. Moreover, since impressive long-lasting responses have recently been observed with immunotherapy combinations (nivolumab and ipilimumab) in a subset of SGC patients, it would be very useful, especially for this type of therapy, to be able to select patients that are likely to respond to this treatment.

Apart from the three areas for future research on the palliative systemic treatment for SGC patients, results of treatments in the palliative setting also offers opportunities for treatments earlier in the disease process. For example, androgen-deprivation therapy has demonstrated biological efficacy in the palliative setting (41, 42) and has subsequently also showed to increase the disease-free survival in the adjuvant setting (based on retrospective analysis) (43). Future research, preferably in a prospective manner, could explore the potential of HER2 targeted therapy in the adjuvant, or even neoadjuvant, setting for HER2-positive SDC patients.

REFERENCES

1. Onderzoek naar zeldzame kanker. The Dutch Cancer Foundation (KWF) [Available from: https://www.kwf.nl/kanker/zeldzame-kankers/onderzoek-naar-zeldzame-kanker.] Accessed on 15-12-2021.
2. Geiger JL, Ismaila N, Beadle B, Caudell JJ, Chau N, Deschler D, et al. Management of Salivary Gland Malignancy: ASCO Guideline. Journal of Clinical Oncology. 2021;39(17):1909-41.
3. Pottier C, Fresnais M, Gilon M, Jérusalem G, Longuespée R, Sounni NE. Tyrosine Kinase Inhibitors in Cancer: Breakthrough and Challenges of Targeted Therapy. Cancers (Basel). 2020;12(3).
4. Robinson DR, Wu Y-M, Lin S-F. The protein tyrosine kinase family of the human genome. Oncogene. 2000;19(49):5548-57.
5. Tchekmedyian V, Sherman EJ, Dunn L, Tran C, Baxi S, Katabi N, et al. Phase II Study of Lenvatinib in Patients With Progressive, Recurrent or Metastatic Adenoid Cystic Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(18):1529-37.
6. Locati LD, Perrone F, Cortelazzi B, Bergamini C, Bossi P, Civelli E, et al. A phase II study of sorafenib in recurrent and/or metastatic salivary gland carcinomas: Translational analyses and clinical impact. European journal of cancer (Oxford, England : 1990). 2016;69:158-65.
7. Zhu G, Zhang L, Dou S, Li R, Li J, Ye L, et al. Apatinib in patients with recurrent or metastatic adenoid cystic carcinoma of the head and neck: a single-arm, phase II prospective study. Ther Adv Med Oncol. 2021;13:17588359211013626.
8. Ho AL, Sherman EJ, Baxi SS, Haque S, Ni A, Antonescu CR, et al. Phase II study of regorafenib in progressive, recurrent/metastatic adenoid cystic carcinoma. Journal of Clinical Oncology. 2016;34(15_suppl):6096-.
9. Chau NG, Hotte SJ, Chen EX, Chin SF, Turner S, Wang L, et al. A phase II study of sunitinib in recurrent and/or metastatic adenoid cystic carcinoma (ACC) of the salivary glands: current progress and challenges in evaluating molecularly targeted agents in ACC. Annals of oncology : official journal of the European Society for Medical Oncology. 2012;23(6):1562-70.
10. Kang EJ, Ahn M-J, Ock C-Y, Lee K-W, Kwon JH, Yang Y, et al. Randomized Phase II Study of Axitinib versus Observation in Patients with Recurred or Metastatic Adenoid Cystic Carcinoma. Clinical Cancer Research. 2021;27(19):5272-9.
11. Ruan GX, Kazlauskas A. Lactate engages receptor tyrosine kinases Axl, Tie2, and vascular endothelial growth factor receptor 2 to activate phosphoinositide 3-kinase/Akt and promote angiogenesis. J Biol Chem. 2013;288(29):21161-72.
12. Chmielowiec J, Borowiak M, Morkel M, Stradal T, Munz B, Werner S, et al. c-Met is essential for wound healing in the skin. J Cell Biol. 2007;177(1):151-62.
13. Cohen RB, Delord JP, Doi T, Piha-Paul SA, Liu SV, Gilbert J, et al. Pembrolizumab for the Treatment of Advanced Salivary Gland Carcinoma: Findings of the Phase 1b KEYNOTE-028 Study. American journal of clinical oncology. 2018.
14. Rodriguez CP, Wu Q, Voutsinas J, Fromm JR, Jiang X, Pillarisetty VG, et al. A Phase II Trial of Pembrolizumab and Vorinostat in Recurrent Metastatic Head and Neck Squamous Cell Carcinomas and Salivary Gland Cancer. Clinical Cancer Research. 2020;26(4):837-45.

15. Ross JS, Gay LM, Wang K, Vergilio JA, Suh J, Ramkissoon S, et al. Comprehensive genomic profiles of metastatic and relapsed salivary gland carcinomas are associated with tumor type and reveal new routes to targeted therapies. Annals of oncology : official journal of the European Society for Medical Oncology. 2017;28(10):2539-46.
16. Linxweiler M, Kuo F, Katabi N, Lee M, Nadeem Z, Dalin MG, et al. The Immune Microenvironment and Neoantigen Landscape of Aggressive Salivary Gland Carcinomas Differ by Subtype. Clinical Cancer Research. 2020;26(12):2859-70.
17. Mosconi C, de Arruda JAA, de Farias ACR, Oliveira GAQ, de Paula HM, Fonseca FP, et al. Immune microenvironment and evasion mechanisms in adenoid cystic carcinomas of salivary glands. Oral oncology. 2019;88:95-101.
18. Lassche G, Tada Y, van Herpen CML, Jonker MA, Nagao T, Saotome T, et al. Predictive and Prognostic Biomarker Identification in a Large Cohort of Androgen Receptor-Positive Salivary Duct Carcinoma Patients Scheduled for Combined Androgen Blockade. Cancers (Basel). 2021;13(14).
19. Locati LD, Cavalieri S, Bergamini C, Resteghini C, Colombo E, Calareso G, et al. Abiraterone Acetate in Patients With Castration-Resistant, Androgen Receptor–Expressing Salivary Gland Cancer: A Phase II Trial. Journal of Clinical Oncology.0(0):JCO.21.00468.
20. Ho AL, Foster NR, Zoroufy AJ, Worden FP, Price KAR, Adkins D, et al. Alliance A091404: A phase II study of enzalutamide (NSC# 766085) for patients with androgen receptor-positive salivary cancers. Journal of Clinical Oncology. 2019;37(15_suppl):6020-.
21. Takahashi H, Tada Y, Saotome T, Akazawa K, Ojiri H, Fushimi C, et al. Phase II Trial of Trastuzumab and Docetaxel in Patients With Human Epidermal Growth Factor Receptor 2-Positive Salivary Duct Carcinoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2019;37(2):125-34.
22. Hamza A, Roberts D, Su S, Weber RS, Bell D, Ferrarotto R. PD-L1 expression by immunohistochemistry in salivary duct carcinoma. Ann Diagn Pathol. 2019;40:49-52.
23. Xu B, Jungbluth AA, Frosina D, Alzumaili B, Aleynick N, Slodkowska E, et al. The immune microenvironment and expression of PD-L1, PD-1, PRAME and MHC I in salivary duct carcinoma. Histopathology. 2019;75(5):672-82.
24. Harwood TC, Oliver AS, King S, Shahlapour M, Dhillon WS. Long-Term Durable Response After Treatment of Metastatic Salivary Duct Carcinoma With Pembrolizumab. JCO Oncol Pract. 2020;16(7):397-8.
25. Niwa K, Kawakita D, Nagao T, Takahashi H, Saotome T, Okazaki M, et al. Multicentre, retrospective study of the efficacy and safety of nivolumab for recurrent and metastatic salivary gland carcinoma. Scientific Reports. 2020;10(1):16988.
26. Burman B, Sherman EJ, Dunn L, Fetten JV, Michel LS, Morris LGT, et al. A phase II trial cohort of nivolumab plus ipilimumab in patients (Pts) with recurrent/metastatic salivary gland cancers (R/M SGCs). Journal of Clinical Oncology. 2021;39(15_suppl):6002-.
27. Alame M, Cornillot E, Cacheux V, Tosato G, Four M, De Oliveira L, et al. The molecular landscape and microenvironment of salivary duct carcinoma reveal new therapeutic opportunities. Theranostics. 2020;10(10):4383-94.
28. Hofman MS, Violet J, Hicks RJ, Ferdinandus J, Thang SP, Akhurst T, et al. [(177)Lu]-PSMA-617 radionuclide treatment in patients with metastatic castration-resistant prostate cancer

(LuPSMA trial): a single-centre, single-arm, phase 2 study. The Lancet Oncology. 2018;19(6):825-33.

29. Rahbar K, Ahmadzadehfar H, Kratochwil C, Haberkorn U, Schafers M, Essler M, et al. German Multicenter Study Investigating 177Lu-PSMA-617 Radioligand Therapy in Advanced Prostate Cancer Patients. Journal of nuclear medicine : official publication, Society of Nuclear Medicine. 2017;58(1):85-90.

30. Heck MM, Retz M, D'Alessandria C, Rauscher I, Scheidhauer K, Maurer T, et al. Systemic Radioligand Therapy with (177)Lu Labeled Prostate Specific Membrane Antigen Ligand for Imaging and Therapy in Patients with Metastatic Castration Resistant Prostate Cancer. The Journal of urology. 2016;196(2):382-91.

31. Sartor O, de Bono J, Chi KN, Fizazi K, Herrmann K, Rahbar K, et al. Lutetium-177–PSMA-617 for Metastatic Castration-Resistant Prostate Cancer. New England Journal of Medicine. 2021.

32. van Boxtel W, Lutje S, van Engen-van Grunsven ICH, Verhaegh GW, Schalken JA, Jonker MA, et al. (68)Ga-PSMA-HBED-CC PET/CT imaging for adenoid cystic carcinoma and salivary duct carcinoma: a phase 2 imaging study. Theranostics. 2020;10(5):2273-83.

33. Klein Nulent TJW, van Es RJJ, Krijger GC, de Bree R, Willems SM, de Keizer B. Prostate-specific membrane antigen PET imaging and immunohistochemistry in adenoid cystic carcinoma-a preliminary analysis. European journal of nuclear medicine and molecular imaging. 2017;44(10):1614-21.

34. Konig L, Hauswald H, Flechtenmacher C, Heller M, Debus J, Haberkorn U, et al. Uptake of Prostate-Specific Membrane Antigen (PSMA) in adenoid cystic carcinoma - Is PSMA-PET-CT a helpful tool in radiation oncology? Clin Transl Radiat Oncol. 2017;7:79-82.

35. de Keizer B, Krijger GC, Ververs FT, van Es RJJ, de Bree R, Willems S. (68)Ga-PSMA PET-CT Imaging of Metastatic Adenoid Cystic Carcinoma. Nucl Med Mol Imaging. 2017;51(4):360-1.

36. Klein Nulent TJW, Valstar MH, Smit LA, Smeele LE, Zuithoff NPA, de Keizer B, et al. Prostate-specific membrane antigen (PSMA) expression in adenoid cystic carcinoma of the head and neck. BMC Cancer. 2020;20(1):519.

37. Klein Nulent TJW, van Es RJJ, Willems SM, Braat A, Devriese LA, de Bree R, et al. First experiences with (177)Lu-PSMA-617 therapy for recurrent or metastatic salivary gland cancer. EJNMMI research. 2021;11(1):126.

38. Privé BM, Derks YHW, Rosar F, Franssen GM, Peters SMB, Khreish F, et al. (89)Zr-labeled PSMA ligands for pharmacokinetic PET imaging and dosimetry of PSMA-617 and PSMA-I&T: a preclinical evaluation and first in man. European journal of nuclear medicine and molecular imaging. 2021.

39. Brayer KJ, Frerich CA, Kang H, Ness SA. Recurrent Fusions in MYB and MYBL1 Define a Common, Transcription Factor-Driven Oncogenic Pathway in Salivary Gland Adenoid Cystic Carcinoma. Cancer discovery. 2016;6(2):176-87.

40. Andrikopoulou A, Zografos E, Liontos M, Koutsoukos K, Dimopoulos M-A, Zagouri F. Trastuzumab Deruxtecan (DS-8201a): The Latest Research and Advances in Breast Cancer. Clinical Breast Cancer. 2021;21(3):e212-e9.

41. Fushimi C, Tada Y, Takahashi H, Nagao T, Ojiri H, Masubuchi T, et al. A prospective phase II study of combined androgen blockade in patients with androgen receptor-positive metastatic or locally advanced unresectable salivary gland carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology. 2018;29(4):979-84.

42. Boon E, van Boxtel W, Buter J, Baatenburg de Jong RJ, van Es RJJ, Bel M, et al. Androgen

deprivation therapy for androgen receptor-positive advanced salivary duct carcinoma: A nationwide case series of 35 patients in The Netherlands. Head & neck. 2018;40(3):605-13.

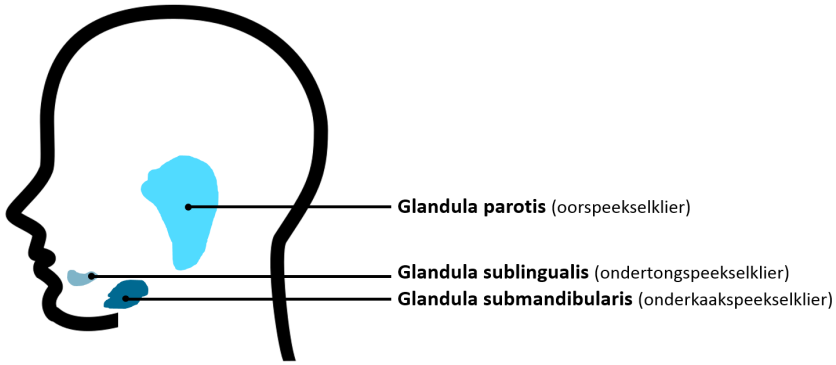
43. van Boxtel W, Locati LD, van Engen-van Grunsven ACH, Bergamini C, Jonker MA, Fiets E, et al. Adjuvant androgen deprivation therapy for poor-risk, androgen receptor-positive salivary duct carcinoma. European journal of cancer (Oxford, England : 1990). 2019;110:62-70.

Chapter 13

Nederlandse samenvatting

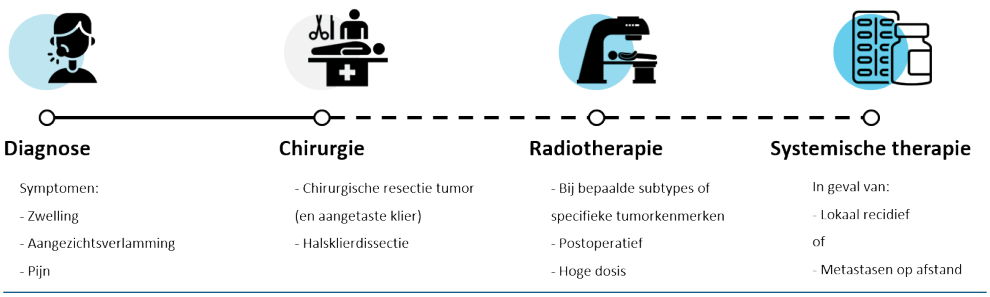
NEDERLANDSE SAMENVATTING

Speekselklierkanker is een zeldzame vorm van kanker die uitgaat van de speekselklieren. De speekselklieren zijn kleine organen rond de mondholte die speeksel produceren. Dit speeksel wordt via de speekselkanalen naar de mondholte getransporteerd, waar het bijdraagt aan het doorslikken van voedsel. Verder speelt speeksel ook een vitale rol bij het spreken en de bescherming van het mondslijmvlies en de tanden. Er bestaan drie grote speekselklieren: de glandula parotis (de oorspeekselklier), de glandula submandibularis (onderkaakspeekselklier) en de glandula sublingualis (ondertongspeekselklier) (figuur 1), die zich alle aan beide zijden van het gezicht bevinden. Daarnaast zijn er verspreid over de mondholte nog 600 tot 1000 kleinere speekselklieren.



Figuur 1: Speekselklieren

Speekselklierkanker is niet alleen een zeldzame vorm van kanker, maar ook een complexe ziekte. Er zijn namelijk 22 verschillende soorten/subtypen, die zich allemaal heel anders gedragen.
Over het algemeen bestaat de behandeling uit een operatie, soms gevolgd door bestraling (figuur 2).



Figuur 2: Overzicht van de behandeling van Speekselklierkanker

Een deel van de patiënten is genezen na een lokale behandeling (operatie +/- bestraling). Helaas komt echter bij een deel van de patiënten de ziekte lokaal terug (lokaal recidief) of ontstaan er uitzaaiingen (metastasen).

Vooral bij de subtypen 'Adenoid Cysteus Carcinoom' (ACC) en 'Salivary Duct Carcinoom' (SDC) keert de ziekte vaak terug. Helaas is de ziekte in dit stadium vaak niet meer te genezen, en komt de nadruk bij de behandeling te liggen op het voorkomen of behandelen van ziektegerelateerde symptomen en het verlengen van het leven, tezamen ook wel palliatieve zorg genoemd. Door de zeldzaamheid van speekselklierkanker is er echter, in vergelijking met meer voorkomende kankersoorten, relatief weinig onderzoek gedaan naar palliatieve systemische therapie. **Daarom richt dit proefschrift zich op de palliatieve systemische behandeling voor speekselklierkankerpatiënten, specifiek voor patiënten die lijden aan ACC of SDC.**

Deel 1: Systemische therapie voor speekselklierkanker

In **hoofdstuk 2** doen we verslag van een uitgebreid literatuuronderzoek dat we gedaan hebben naar palliatieve systemische therapie voor SDC-patiënten. Door onze zoekstrategie werden 2014 artikelen geïdentificeerd, waarvan 45 artikelen de relevante en specifieke informatie bevatten die wij beoogden te vinden. In totaal bevatten deze 45 artikelen informatie over 256 SDC-patiënten die palliatieve systemische therapie hadden gekregen. De resultaten werden gecategoriseerd naar het type systemische therapie: chemotherapie, immunotherapie, hormonale therapie, of doelgerichte therapie.

We hebben twee fase II-studies geïdentificeerd waarin hoofdzakelijk SDC-patiënten waren behandeld. De ene studie bestudeerde het effect van androgeendeprivatietherapie en de andere studie het effect van HER2-gerichte therapie. Beide studies lieten veelbelovende resultaten zien. Op basis van onze literatuurstudie concludeerden wij dat androgeendeprivatietherapie moet worden overwogen voor androgeenreceptor-positieve SDC-patiënten (responspercentages 18-53%) en dat HER2-therapie zeer effectief is bij HER2-positieve SDC-patiënten (responspercentages 60-70%). Wat chemotherapie betreft, is de combinatie van carboplatine met een taxaan (docetaxel/paclitaxel) het best onderzocht bij SDC-patiënten, met responspercentages van 39-50%. Andere doelgerichte middelen en immunotherapie waren nog niet uitgebreid onderzocht in SDC-patiënten.

Omdat eerder onderzoek aantoonde dat de tyrosinekinasen c-MET en vasculaire endotheliale groeifactorreceptoren (VEGFR) vaak tot overexpressie komen in speekselklierkanker, hebben wij de werkzaamheid van cabozantinib (een tyrosinekinaseremmer) in deze vorm van kanker geëvalueerd. De resultaten van deze klinische fase II-studie met cabozantinib in patiënten met gevorderde speekselklierkanker zijn beschreven in **hoofdstuk 3**. In totaal kregen 25 speekselklierkankerpatiënten cabozantinib-behandeling. Hoewel

het aanvankelijk de bedoeling was om meer patiënten te includeren (op basis van de berekening van de steekproefgrootte), werd de studie voortijdig beëindigd vanwege ernstige toxiciteit. Zes van de 25 patiënten (24%) hadden graad ≥3 wondcomplicaties. Opmerkelijk is dat vier van deze zes patiënten deze complicatie ontwikkelden in een gebied dat eerder was blootgesteld aan (hoge dosis) radiotherapie. Verder was de respons op cabozantinib laag tot matig. Van de respons-evalueerbare patiënten (cabozantinib-behandeling ≥8 weken), vertoonden 1/15 ACC-patiënten (7%), 1/4 SDC-patiënten (25%), en 0/2 speekselklierkankerpatiënten met een overig subtype (0%) een objectieve respons. Concluderend kan worden gesteld dat deze studie te veel ernstige met cabozantinib geassocieerde wondcomplicaties toonde bij patiënten met speekselklierkanker, met name in eerder bestraalde gebieden. Daarom is cabozantinib niet aanbevolen als palliatieve systemische therapie bij speekselklierkankerpatiënten.

In **hoofdstuk 4** beschrijven we de retrospectieve evaluatie van de resultaten van docetaxel, trastuzumab en pertuzumab als eerstelijns HER2-gerichte behandeling, en ado-trastuzumab emtansine als tweedelijns HER2-gerichte behandeling voor HER2-positieve SDC-patiënten. Tussen 2015 en 2021 waren 13 SDC-patiënten behandeld met de combinatie van docetaxel, trastuzumab en pertuzumab. Van de 12 respons-evalueerbare patiënten werden één complete respons en zes partiële responsen geobserveerd, wat resulteerde in een totaal responspercentage van 58%. De mediane duur van de behandeling met docetaxel, trastuzumab en pertuzumab was 6,9 maanden (range: 3,4-26,8+). Zeven patiënten kregen vervolgens ado-trastuzumab emtansine als tweedelijns HER2-therapie, wat resulteerde in vier partiële responsen en een totaal responspercentage van 57%. De mediane duur van de behandeling met ado-trastuzumab was 8,5 maanden (range 1,1-20,4).

De mediane overleving van de 13 patiënten na de start van docetaxel, trastuzumab en pertuzumab was 42,0 maanden (95%-betrouwbaarheidsinterval: 13,8-70,1 maanden). Graad ≥3 toxiciteit werd geobserveerd in 39% van de patiënten op docetaxel, trastuzumab en pertuzumab, en in 14% van de patiënten bij ado-trastuzumab emtansine-behandeling. Wij concludeerden dat docetaxel, trastuzumab en pertuzumab-behandeling gevolgd door ado-trastuzumab emtansine bij progressie veelbelovende behandelingen zijn voor HER2-positieve SDC-patiënten. Deze behandelingen hadden immers geleid tot een respons bij de meerderheid van de patiënten en hadden daarbij een aanvaardbaar toxiciteitsprofiel.

Deel 2: PSMA radioligand therapie voor speekselklierkanker

Prostaatspecifiek membraan antigeen (PSMA) is een transmembraaneiwit dat ontdekt werd en sterk tot overexpressie gebracht is in prostaatkanker. PSMA kan een doelwit vormen voor doelgerichte therapie. Inmiddels weten we dat PSMA niet alleen tot expressie komt in prostaatkankercellen, maar ook in andere typen kanker. In **hoofdstuk 5** geven we een samenvatting van de huidige kennis van PSMA-expressie in andere solide kankers en

definiëren we een perspectief voor een bredere klinische implementatie van PSMA-gerichte therapie. Deze review richtte zich op zeven verschillende solide kankersoorten, waaronder speekselklierkanker. Opmerkelijk is dat PSMA-expressie in deze zeven solide kankers voornamelijk werd waargenomen in de neovasculatuur van de tumoren. Dit staat in contrast met prostaatkanker, waar de PSMA-expressie wordt waargenomen op de tumorcellen zelf. Alleen bij ACC-patiënten kwam PSMA ook tot expressie op de tumorcellen. Verder vonden we twee casusbeschrijvingen van ACC-patiënten die PSMA-gerichte therapie hadden gekregen, maar deze gaven relatief weinig inzicht in het effect van de behandeling bij deze patiënten. Dit stimuleerde ons om het effect van PSMA-gerichte behandeling goed te onderzoeken met een prospectieve studie voor speekselklierkankerpatiënten.

In **hoofdstuk 6** beschrijven we de voorlopige resultaten van een lopende fase II-pilotstudie naar PSMA-gerichte therapie. Gerichte behandeling bestond uit PSMA-radioligandtherapie met ^{177}Lu-PSMA. Door koppeling van een radionuclide (Lutetium-177 [^{177}Lu, een β-straler]) aan een PSMA-gerichte molecuul (het ligand PSMA-I&T) werd een cytotoxische stralingslading specifiek aan de tumoren afgegeven. Alleen patiënten bij wie de aanwezigheid van het PSMA-eiwit in de tumoren is vastgesteld middels PSMA PET-beeldvorming, kwamen in aanmerking voor behandeling met ^{177}Lu-PSMA.

Deze studie is gestart in mei 2020 en op dit moment zijn 10/10 ACC-patiënten en 2/5 SDC-patiënten gestart met de behandeling. Dit hoofdstuk beschrijft de voorlopige resultaten van de eerste 8 ACC-patiënten in deze studie (met 1 december 2021 als afkapdatum voor deze analyse). Tot die datum werden 12 ACC-patiënten gescreend voor de studie en voldeden 8 ACC-patiënten aan de inclusiecriteria. Deze 8 ACC-patiënten hadden allen tenminste één cyclus ^{177}Lu-PSMA gekregen. Onze voorlopige resultaten wijzen erop dat ^{177}Lu-PSMA goed wordt verdragen bij ACC-patiënten, maar de werkzaamheid lijkt tot nu toe beperkt tot het bereiken van stabiele ziekte. Opmerkelijk is dat bij 2/8 ACC-patiënten (25%) de stabiele ziekte langer dan 6 maanden aanhoudt, maar deze cijfers kunnen nog veranderen bij een langere follow-up.

In **hoofdstuk 7** rapporteren we over het studieprotocol (uit 2019) van een lopende klinische beeldvormingsstudie bij SDC-patiënten. Deze lopende studie onderzoekt het effect van androgeendeprivatietherapie op PSMA-ligandopname, geëvalueerd middels herhaalde PSMA PET-scans. Aangezien onderzoek bij prostaatkanker heeft aangetoond dat androgeendeprivatietherapie de PSMA-expressie/PSMA-ligandopname kan verhogen, onderzoekt deze beeldvormingsstudie of dit ook het geval is bij SDC-patiënten.

Deel 3: Vergroten van de algemene kennis over speekselklierkanker

In **hoofdstuk 8** worden de resultaten gepresenteerd van een project in samenwerking met collega's van de afdeling KNO en Hoofd-Hals Chirurgie. Tijdens dit project evalueerden we de frequentie en uitkomsten van pathologieconsulten en -revisies van speekselkliertumoren in de routinematige klinische praktijk. De data hiervan werd vergaard via het Pathologisch Anatomisch Landelijk Geautomatiseerd Archief (PALGA). Tussen 2006 en 2016 werden in Nederland 13.441 speekselkliertumoren uitgaande van een van de grote speekselklieren gereseceerd. Revisie van het histopathologisch weefsel werd uitgevoerd in 2,6% (n=350) van de gevallen en deze waren discordant in 8,3% (n=29). Dit aantal (8,3%) benadrukt de complexiteit van de histopathologische diagnose van speekselkliertumoren. Mogelijk zou een toename van het aantal consulten de nauwkeurigheid van de initiële diagnose kunnen verbeteren en tegelijkertijd de noodzaak voor revisies en het aantal discordante revisies kunnen verlagen.

In **hoofdstuk 9** beschrijven wij een mogelijke risicofactor voor het ontstaan van SDC bij twee patiënten. De risicofactoren voor het ontstaan van speekselklierkanker en specifiek het SDC-subtype zijn grotendeels onbekend. In de literatuur wordt luchtvervuiling vaker beschreven als een potentiële risicofactor. Bij andere vormen van kanker, vooral longkanker, is de carcinogeniteit van chroom VI welbekend. In dit hoofdstuk beschrijven wij twee SDC-patiënten die beroepsmatig waren blootgesteld aan chroom VI bij het onderhoud van militaire apparatuur. Alhoewel een oorzakelijk verband tussen blootstelling aan chroom VI en het ontstaan van een SDC niet op individuele basis wordt aangetoond, achten wij het uitvoeren van populatieonderzoek niet haalbaar vanwege de extreem lage prevalentie van SDC. Desondanks achten wij het aannemelijk dat de blootstelling aan chroom VI zou kunnen hebben bijgedragen aan het ontstaan van hun SDC. Met dit hoofdstuk willen wij de bekendheid van beroepsmatige blootstelling aan chroom VI en het optreden van SDC vergroten.

In **hoofdstuk 10** rapporteren wij over negen SDC-patiënten die cutane lymfangitis carcinomatosa (CLC) hadden ontwikkeld, wat zich uitte in huidlaesies in de hals en borstregio. Al deze patiënten hadden uitgebreide lymfekliermetastasen bij diagnose. Pathofysiologisch is het zeer aannemelijk dat bij deze patiënten met uitgebreide lymfekliermetastasen de SDC-tumorcellen zich verder hebben verspreid via de lymfevaten, waardoor cutane metastasen in het drainagegebied (hals en borstregio) zijn ontstaan. Wij observeerden dat CLC zich anders manifesteerde dan gebruikelijke cutane metastasen. Cutane metastasen presenteren zich meestal als noduli. De huidlaesies van de hals en borstregio in onze SDC-patiënten met CLC varieerden van een erythemateuze patch of plaque, met een wisselende graad van (klinisch) oedeem, induratie, noduli, en/of ulceratie. Met dit hoofdstuk beogen wij de bekendheid en de kennis van CLC als ziektemanifestatie bij SDC-patiënten te vergroten.

Appendices

RESEARCH DATA MANAGEMENT

The data described in this thesis were obtained during my PhD research period at the Departments of Medical Oncology and Medical Imaging (Nuclear Medicine) of the Radboud University Medical Centre (Radboudumc) and the Radboud Institute for Health Sciences (RIHS), the Netherlands.

All prospective clinical studies (chapter 3, 6, 7) were approved by the local Medical Ethical Committee (CMO Regio Arnhem-Nijmegen, the Netherlands). These studies were conducted in accordance with the principles of Good Clinical Practice guidelines and the Declaration of Helsinki. Furthermore, this local Medical Ethical Committee also approved for the use of the clinical data and tissue samples from the patients described in the retrospective study (chapter 4). For the research described in chapter 8, a review by a Medical Ethical Committee was not required by Dutch law due to the anonymous data collection. The two patients reported in the case report (chapter 9) both gave written informed consent for the use of their clinical data and medical images. Informed consent was also obtained for patients in chapter 10 in accordance with Dutch law.

The prospective clinical studies (chapter 3, 6, 7) were all registered on ClinicalTrials.gov, a database of privately and publicly funded clinical studies conducted around the world. The protocol of the systematic review (chapter 2) was published on the international Prospective Register of Systematic Reviews in Health and Social Care (PROSPERO).

The data is stored following the Findable, Accessible, Interoperable and Reusable (FAIR) principles (Wilkinson *et al*. 2016, Scientific Data).

Findable: The primary and secondary data were stored at the Radboudumc department servers as well as CASTOR, a cloud-based clinical data management platform. Raw data from equipment (e.g. PET/CT/SPECT) are backed-up on university servers belonging to the department of Medical Imaging.
Accessible: Data is accessible by senior staff members who are involved in the research. The data presented in chapter 2,3,4,5,8,9 have been published in scientific articles. Additional data associated with the chapters in this thesis are available via the corresponding authors upon request. After the studies regarding chapter 6 and 7 are completed, the final data will be analysed, with the intention to also publish this data in scientific articles in the future.
Interoperable: All protocols and data are documented in English.
Reusable: All data will be saved for at least 15 years after publication of each study. All used data analysed during these studies are available upon request from the corresponding author.

LIST OF PUBLICATIONS

M.J.M. Uijen*, B.M. Privé*, C.M.L. van Herpen, H. Westdorp, W. A. van Gemert, M. de Bakker, M. Gotthardt, M.W. Konijnenberg, S.M.B. Peters, J. Nagarajah. Kidney absorbed radiation doses for [^{177}Lu]Lu-PSMA-617 and [^{177}Lu]Lu-PSMA-I&T determined by 3D clinical dosimetry. *Submitted.*

B.M. Privé, S. Peters, **M.J.M. Uijen**, M.J.R. Janssen, W.A.M. van Gemert, M.C. Kreissl, S. Ezzidin, M.W. Konijnenberg, J. Nagarajah. Letter to the Editor RE: Tumor sink effect in ⁶⁸Ga-PSMA-11 PET: Myth or Reality? *Journal of Nuclear Medicine*. 2022.

S.T.H. Reerds*, **M.J.M. Uijen***, A.C.H. Van Engen – Van Grunsven, H.A.M. Marres, C.M.L. van Herpen, J. Honings. Results of histopathological revisions of major salivary gland neoplasms in routine clinical practice. *Journal of Clinical Pathology* 2022.

M.J.M. Uijen, J.A.M Weijers, C.M.L. Driessen, C.M.L. van Herpen, J. Nagarajah. ^{68}Ga-Prostate-Specific Membrane Antigen-avid Malignant Pleural Effusion in a patient with Metastatic Adenoid Cystic Carcinoma and concordance with ^{18}F-FDG PET/CT. *Clinical Nuclear Medicine* 2022.

M.J.M. Uijen*, G. Lassche*, A.C.H. van Engen-van Grunsven, C.M.L. Driessen, C.M.L. van Herpen. Case series of docetaxel, trastuzumab, and pertuzumab (DTP) as first-line anti-HER2 therapy and ado-trastuzumab emtansine (T-DM1) as second-line for recurrent or metastatic HER2-positive salivary duct carcinoma. *Oral Oncology* 2022.

W. van Boxtel*, **M.J.M. Uijen***, S.D. Krens, T. Dijkema, S.M. Willems, M.A. Jonker, S.A.H. Pegge, A.C.H. van Engen-van Grunsven, C.M.L. van Herpen. Excessive toxicity of cabozantinib in a phase II study in recurrent and/or metastatic salivary gland cancer patients. *European Journal of Cancer* 2022.

I. Seçin, **M.J.M. Uijen**, C.M.L. Driessen, C.M.L. van Herpen, P.T.J. Scheepers. Case Report: Two Cases of Salivary Duct Carcinoma in Workers With a History of Chromate Exposure. *Frontiers in Medicine* 2021.

S.D. Krens, W. van Boxtel, **M.J.M. Uijen**, F.G.A. Jansman, I.M.E Desar, S.F. Mulder, C.M.L. van Herpen, N.P. van Erp. Exposure-toxicity relationship of cabozantinib in patients with renal cell cancer and salivary gland cancer. *International Journal of Cancer* 2021.

M.J.M. Uijen*, Y.H.W. Derks*, R.I.J. Merkx, M.G.M. Schilham, J. Roosen, B.M. Privé, S.A.M. van Lith, C.M.L. van Herpen, M. Gotthardt, S. Heskamp, W.A.M. van Gemert, J. Nagarajah. PSMA radioligand therapy for solid tumours: background, opportunities, challenges and first clinical reports. *European Journal of Nuclear Medicine and Molecular Imaging* 2021.

B.M. Privé, S.M.B. Peters, C.H.J. Muselaers, I.M. van Oort, M.J.R. Janssen, M. Sedelaar, M.W. Konijnenberg, P. Zamecnik, **M.J.M. Uijen**, M.G.M. Schilham, A. Eek, T.W.J. Scheenen6, J.F. Verzijlbergen, W.R. Gerritsen, N. Mehra, L.G.W. Kerkmeijer, R. Smeenk, D.M. Somford, J.P.A. van Basten, S. Heskamp, J. Barentsz, M. Gotthardt, J.A. Witjes, James Nagarajah. Lutetium-177-PSMA-617 in low-volume hormone sensitive metastatic prostate cancer, a prospective pilot study. *Clinical Cancer Research* 2021.

W. van Boxtel, **M.J.M. Uijen**, G.W. Verhaegh, S.M. Willems, M.A. Jonker, PALGA Group, J.A. Schalken, I.C.H. van Engen-van Grunsven, C.M.L. van Herpen. Prognostic value of PSMA, c-MET and E-cadherin in salivary duct carcinoma. *Oral Oncology* 2020.

M.J.M. Uijen, W. van Boxtel, C.M.L. van Herpen, M. Gotthardt, J. Nagarajah. 68Ga-Prostate-Specific Membrane Antigen-11-Avid Cardiac Metastases in a Patient With Adenoid Cystic Carcinoma, A Rare Presentation of a Rare Cancer. *Clinical Nuclear Medicine* 2020.

M.J.M. Uijen, G. Lassche, A.C.H. van Engen-van Grunsven, Y. Tada, G.W. Verhaegh, J.A. Schalken, C.M.L. Driessen, C.M.L. van Herpen. Systemic therapy in the management of recurrent or metastatic salivary duct carcinoma: A systematic review. *Cancer Treatment Reviews* 2020.

L.A. Steeman, **M.J.M. Uijen**, F.M. Plat, L. Huibers, M. Smits, P.H.J. Giesen. Out-of-hours primary care in 26 European countries: an overview of organizational models. *Family practice* 2020.

C.M.L. Driessen, **M.J.M. Uijen**, W.T.A. van der Graaf, C.C.M. van Opstal, J.H.A.M. Kaanders, T. Nijenhuis, C.M.L. van Herpen. Degree of nephrotoxicity after intermediate- or high-dose cisplatin-based chemoradiotherapy in patients with locally advanced head and neck cancer. *Head & Neck* 2016.

E. Wallace*, **M.J.M. Uijen***, B. Clyne, A. Zarabzadeh, C. Keogh, R. Galvin,1,3 S.M. Smith, T. Fahey. Impact analysis studies of clinical prediction rules relevant to primary care: a systematic review. *BMJ Open* 2016.

* Both authors contributed equally to the manuscript.

PHD PORTFOLIO

Institute for Health Sciences
Radboudumc

PhD candidate: M.J.M. Uijen
Department: Medical Oncology
Graduate School: Radboud Institute for Health Sciences

PhD period: 01-04-2019 – 30-04-2022
Promotors: Prof. Dr. C.M.L. van Herpen, Prof. Dr. J. Nagarajah, Prof. Dr. M. Gotthardt
Co-promotors: -

		Year(s)	ECTS
	TRAINING ACTIVITIES		
A.	***Courses & Workshops***		
	- RIHS Introductory course	2019	0.75
	- Basiscursus Regelgeving en Organisatie Klinisch onderzoekers (BROK) course	2019	1.5
	- Working with radionuclides course (TMS-VRS-D)	2019	2.0
	- Project Management for PhD Candidates	2019	2.0
	- Advanced conversation	2019-2020	2.5
	- Statistics for PhD candidates using SPSS	2020	2.0
	- Radboudtalks	2020	1.5
	- Scientific Integrity course	2020	1.0
	- Education in a nutshell	2020	1.0
	- Scientific Writing for PhD candidates	2020-2021	3.0
	- Mindfulness-Based Stress Reduction for PhD students	2021	1.0
	- NVVO Basiscursus oncologie	2021	2.5
B.	***Seminars & lectures***		
	- Summer school Nuclear Medicine (Berlin)	2019	2.0
	- Radboud Research Rounds	2019-2020	0.3
	- Research meeting Nuclear Medicine (weekly)	2019-2022	3.0
	- Research meeting Medical Oncology (weekly)	2019-2022	3.0
	- Multidisciplinary education Oncology	2019-2022	0.25
C.	***Symposia & congresses***		
	- RIHS PhD retreat (oral presentation)	2019	1.0
	- Jonge onderzoeksdag Nederlandse Werkgroep Hoofd-Hals Tumoren (NWHHT) (Amsterdam)	2019	0.25
	- Mini symposium Lustrum Centrum voor Hoofd-Hals Oncologie (Nijmegen)	2020	0.1
	- CaRe symposium (Virtual)	2021	0.25
	- DRCP symposium rare cancer (Virtual)	2021	0.25
	- PSMA forum (virtual; oral presentation)	2021	0.35
	- Annual European Molecular Imaging Meeting (EMIM) (Göttingen; poster presentation)	2021	1.25
	- Annual meeting European Society for Medical Oncology (ESMO) (Virtual; poster presentation)	2021	0.25
	- Jonge onderzoeksdag NWHHT (Groningen; oral presentation)	2021	0.25
	- Annual meeting European Association of Nuclear Medicine (EANM) (Virtual; oral presentation)	2021	0.25

D.	***Other***		
	- Member of RIHS PhD council (role of Education)	2020-2021	0.5
	- Medical Oncology journal club	2019-2022	3.0
	- Nuclear Medicine journal club	2019-2022	1.5
	- Global Prostate Cancer Foundation Journal Club (PSMA related topics)	2020-2021	0.3
	- Presentation Sponsorcafé Alpe d'Huzes (Nieuwegein)	2019	0.25
	TEACHING ACTIVITIES		
E.	***Lecturing***		
	- Lecture Biomedical students on Rare cancers	2019	0.1
	- Lectures Biomedical students on Medical Imaging	2020-2022	0.4
F.	***Supervision of internships / other***		
	- Research Project supervisor for (Bio)Medical students	2020-2021	2.5
	- Supervising Medisch Beeldvormende en Radiotherapeutische Technieken (MBRT) students (2)	2021	2.0
	- Supervising Bachelor student Biology	2022	1.0
TOTAL ECTS			45.05

Radboud University Radboudumc university medical center

DANKWOORD

Dit proefschrift was nooit tot stand gekomen zonder samenwerking, hulp en inzet van vele anderen die ik graag zou willen bedanken.
PS: zoals iedereen weet ben ik geen persoon van weinig woorden, dus een beknopt dankwoord zat er bij mij niet in ;).

Allereerst de **patiënten** die deel hebben genomen aan de klinische studies en/of op een andere wijze hebben bijgedragen aan onderzoek. Door de zeldzaamheid van speekselklierkanker moesten sommigen van jullie zelfs het hele land doorkruisen om naar het Radboudumc te komen. Het was motiverend en inspirerend om te ervaren dat velen van jullie, zelfs in deze moeilijke fase van het leven, nog zo graag wilden bijdragen aan nieuwe onderzoeken en behandelingen voor speekselklierkanker. Veelal namen jullie niet alleen deel aan onderzoek in de hoop daar zelf baat bij te hebben, maar deden jullie dit ook voor toekomstige patiënten. Zonder jullie was dit proefschrift nooit tot stand gekomen.
Dit geldt ook voor de **Patiëntenvereniging Speekselklierkanker**, een vereniging die zich met hart en ziel inzet voor (toekomstige) patiënten. Ik vond het bijzonder om deel te mogen zijn van de bijeenkomsten en de benefietevenementen, zoals het Stamppotbuffet en de Mountainbike4life!

Prof. dr. van Herpen, beste Carla, een promotieonderzoek stond eigenlijk niet echt op mijn planning. Toch benaderde jij me tijdens de borrel van de promotie van Chantal en wist je mij met al je enthousiasme over te halen. Jouw passie voor onderzoek voor zeldzame kankers wist je goed over te brengen. Makkelijk heb ik het niet altijd gehad tijdens mijn PhD, veelal omdat mijn onzekerheden en de zoektocht naar mijn eigen passie mij in de weg zaten, toch heb jij mij daar altijd heel fijn in ondersteund. Daarnaast vond ik het erg prettig dat je mij veel ruimte hebt gegeven tijdens mijn PhD. Naast dat jij me op wetenschappelijk gebied veel hebt geleerd, heb ik ook in de spreekkamer veel van je geleerd. Je bent een ontzettend warme dokter, die echt hart heeft voor patiënten en hun naasten. Veel dank voor alles.

Prof. dr. Gotthardt, beste Martin, ik denk dat weinig promovendi kunnen zeggen dat de professor vaker koffie voor hen heeft gehaald dan andersom, maar wellicht dat dit heeft bijgedragen aan de relatief vlotte afronding van mijn proefschrift. Jouw begeleiding de afgelopen jaren heb ik als heel fijn ervaren. Jij was de eerste waar ik naar toe ging als iets even minder lekker liep. Verder heb je me veel vertrouwen gegeven in mijn eigen kunnen, veel dank daarvoor. Alhoewel onze overleggen soms meer filosofisch van aard waren en over van alles behalve de inhoud van mijn onderzoek gingen, waren ze niet minder waardevol. Ook fijn dat je na mijn tientallen mailtjes en appjes (sommige vol hoofdletters) uiteindelijk mijn proefschrift door had genomen voordat ik op reis ging (@Toekomstige promovendi, deze tactiek werkt!). Martin, dank voor de gezellige samenwerking.

Prof. dr. Nagajarah, beste James, de PSMA-expert van de afdeling en een pure wetenschapper! Bij jou kon ik met al mijn PSMA-gerelateerde vragen terecht. Jij was altijd op de hoogte van de meest recente ontwikkelingen en kon deze goed vertalen naar de toekomst. Ook voor het beoordelen van scans van studiepatiënten maakte jij altijd tussendoor tijd om daar samen naar te kijken. Bedankt voor al je inhoudelijke en praktische hulp bij de klinische studies. Verder heb ik ook veel gehad aan je relativeringsvermogen, als ik hier en daar gestrest was, hielp jij om zaken in perspectief te plaatsen. Bedankt voor al je begeleiding.

Dr. Driessen, beste Chantal, tijdens de bachelor begon ik al met onderzoek, toen jij nog promovenda was. Onze samenwerking was erg gezellig en nadien bleven we contact houden. Toen ik bij jou aan de keukentafel jouw promotieboekje zag, had je de twinkeling in mijn ogen goed gespot… toch wel echt gaaf zo'n promotietraject en promotieboekje. Alhoewel ik daar de afgelopen jaren ook vaak genoeg anders over heb gedacht, ben ik toch wel echt heel blij en trots dat ik nu zelf ook zo'n tof boekje heb. Onwijs leuk en toevallig dat jij je in de tussentijd op de hoofd-halsoncologie had gespecialiseerd en we op die manier opnieuw samen hebben gewerkt. Ook deze keer was weer reuze gezellig. Thanks voor alles, Chantal.

Dr. van Engen-van Grunsven, beste Ilse, dé speekselklierpatholoog, met jou heb ik ontzettend fijn samengewerkt. Bedankt voor al je waardevolle input bij de studies en manuscripten. En ook heel erg bedankt voor de gezelligheid eromheen, zoals toen we samen in de auto zaten naar het Stamppotbuffet.

Natuurlijk wil ik ook graag de overige co-auteurs bedanken, die hebben meegewerkt aan een of meerdere hoofdstukken. In het bijzonder nog **Dr. Jonker**, Marianne, voor al je statistische hulp. Zowel bij het opzetten van nieuwe studies, als het uitwerken van de data van reeds uitgevoerde studies.

Prof. dr. Swinkels, beste Dorine, mijn mentor tijdens mijn promotietraject. Bedankt voor de fijne gesprekken!

Veel dank aan de leden van de manuscriptcommissie, **Prof. dr. Grünberg**, **Prof. dr. Takes** en **Dr. Vogel**, voor het beoordelen van mijn proefschrift.

Dan het **speekselklierkanker-onderzoeksteam**, begonnen met **Eline**, en verder uitgebreid door mijn voorganger **Wim**. Jullie hebben samen een mooie onderzoekslijn uitgezet! Wim, met jou heb ik nog een tijd samengewerkt en in die tijd heb ik veel van je mogen leren, waarvoor dank. Gerben en ik hadden samen het stokje van je overgenomen. **Gerben**, tegelijk met jou een PhD doen was zeker niet gemakkelijk voor mijn ego, dat geef ik eerlijk toe. Als iemand het '*Professor*-gen' heeft dan ben jij het wel, en mezelf aan jou meten kon ik dus maar beter niet doen. Toch vond ik het heel fijn om met je samen te werken, ik heb veel van

je geleerd en je had altijd waardevolle input. Ik weet zeker dat jij een glansrijke toekomst binnen de medische wereld tegemoet gaat. Lieve **Jetty**, Gerben en ik geven het stokje weer aan jou door. Ik heb onwijs veel vertrouwen in jouw capaciteiten om het onderzoek naar speekselklierkanker verder voort te zetten. Je gaat je PhD rocken. En wat hebben we gelachen in o.a. Groningen hè ;).

Verder wil ik graag van de **Medische Oncologie** alle **Oncologen**, **Arts-assistenten**, **Verpleegkundigen**, **Doktersassistenten**, **Postdocs**, **het Secretariaat**, **het Trialbureau** en de **Datamanagers** bedanken.
In het bijzonder wil ik de mede-(ex)PhD'ers van de Onco bedanken, beste **Sarah**, **Jorien**, **Iris**, **Stefan**, **Dide**, **Kim**, **Wouter, Sandra**, **Sophie**, **Lotte**, **Vicky**, **Minke,** bedankt voor alle gezelligheid! Eerst in 'the Dungeon' en later op 'vier hoog'. Sorry dat ik het laatste jaar vaker bij de NucMed neer was gestreken; ik was lui geworden en die afdeling was nou eenmaal een stuk dichterbij. Maar als ik weer bij jullie kwam zitten, dan was het altijd reuze gezellig bijkletsen.
Hettie, trialmanager en tevens mijn soort-van-buurvrouw, erg bedankt voor al je hulp rondom het opzetten en uitvoeren van klinische studies. Soms zag ik met alle documenten door de bomen het bos niet meer, maar jij gelukkig wel ;). Leuk dat we elkaar nog blijven spreken.
Kim, bedankt voor de fijne samenwerking op de speekselklierkanker-poli.
Sietske, bedankt voor het plannen van de afspraken in de overvolle agenda van Carla!

Over naar de afdeling **Nucleaire Geneeskunde** (de NucMed). Ik heb het vaak vermeld: "mijn contract is officieel bij de Medische Oncologie", maar ik was toch aanzienlijk vaker bij de NucMed te vinden!

Allereerst veel dank aan het (door-mij-nu-zo-even-genoemde) **PSMA-therapieteam**. **Bastiaan** en **Steffie**, jullie hadden tijdens jullie PhD's al veel van de complexe logistiek van radionuclide-therapiestudies uitgewerkt. Hierdoor was het opzetten van mijn studies een stuk gemakkelijker, dat was erg fijn. Dank voor het uitgebreid delen van jullie ervaringen.
Krisja, veel dank voor je hulp en betrokkenheid bij de LUPSA-studie, erg fijn om de eerste studiepatiënten samen met jou te hebben gezien! Veel dank aan de **Nucleair Geneeskundigen**, in het bijzonder **James**, **Martin**, **Marcel** en **Willemijn**, voor de hulp tijdens de therapie-toedieningen. Ook veel dank aan alle **laboranten**, voor de logistiek en uitvoering van scans en therapietoedieningen. In het bijzonder **Maarten**, **Peter K.** en **Marjo**, zoals jullie weten zijn PSMA-therapiestudies logistiek soms best een uitdaging, maar jullie waren altijd bereid om mee te denken en de planning passend te maken. Ik heb jullie betrokkenheid als zeer fijn ervaren! Dat geldt ook voor het **Hotlab**, en ik het bijzonder **Maichel** en **Peter L.**. Ondanks dat we wat pech hadden bij de eerste labellingen (waar ik wat onrustige nachten van heb gehad), weet ik dat jullie altijd jullie uiterste best deden om alles zo goed mogelijk te laten verlopen, dank! Dank aan de **Klinisch Fysica**; **Steffie**, **Daphne L.**

en **Antoi** voor het helpen met de stralingsveiligheid (waar deze dokter toch maar weinig kaas van had gegeten). Last-but-not-least, het **Trialbureau**, specifiek **Michel**, veel dank voor alle hulp bij het opzetten, uitvoeren en coördineren van de klinische studies.

Lieve **mede-(ex)PhD'ers van de NucMed**, met wie ik het grootste deel van mijn PhD samen heb gewerkt; **Joey**, **Melline**, **Yvonne**, **Tom**, **Peter W.**, **Fortuné**, **Robin**, **Bastiaan**, **Gerwin**, **Jan-Marie**, **Nienke**, **Massis**, **Estel, Daphne D.**, **Daphne L.**, **Simone**, **Esther**, **Evelien**, **Steffie**, onwijs bedankt voor al jullie steun, humor en gezelligheid. Gedeelde smart is halve smart ;). Het was fijn om alle highs en lows (die laatste waren er bij mij statistisch significant meer) met jullie te kunnen delen. Op de momenten dat ik niet "ziek in de data" was, was ik aan het genieten van de fijne sfeer die wij hadden. Ik vind het eerlijk gezegd het moeilijkste om onder woorden te brengen hoe jullie mij door deze PhD hebben geholpen. Zonder jullie was dit boekje er zéker niet geweest. Onwijs veel dank!
In een poging om mijn dankwoord nog enigszins behapbaar te houden wil ik slechts twee van jullie nog even in het bijzonder noemen.
Lieve **Melline**, wij zijn ongeveer tegelijk onze PhD's gestart en vanaf het moment dat we in het 1e jaar samen op Summer school zijn geweest in Berlijn, was een mooie vriendschap ontstaan. Leuk om samen met jou de PhD-periode te hebben beleefd, zowel op het werk als erbuiten ;).
Lieve **Yvonne**, jij was voor mij wellicht wel het beste voorbeeld van het combineren van hard werken met gezelligheid en humor. Samenwerken met jou verliep uiterst soepel en je hilarische uitspraken zal ik niet snel vergeten.
Ook met de PhD'ers die rond mijn laatste jaar gestart zijn, was het reuze gezellig op het werk en op leuke uitjes zoals de NucMed-skireis. Beste **Meike**, **Anne**, **Tess**, **Daan**, **Lovisa**, **Ysbrand**, **Sevilay**, **Kim**, **Sylvia**, **Eva**, veel succes met jullie PhD's!

Verder wil ik van de Nucmed ook nog graag de **Post-docs** (**René**, **Lieke, Sanne** en **Marti**), de **Analisten** (**Milou**, **Gerben**, **Cathelijne** en **Janneke**) en alle **Senior-Onderzoekers** bedanken. De deur stond bij iedereen altijd open, en het was fijn om te kunnen sparren wanneer nodig.
Dank voor de fijne sfeer die er op de afdeling hangt doordat jullie die met z'n allen creëren.
In het bijzonder wil ik graag **Peter L.** en **Sandra** bedanken, voor jullie betrokkenheid bij mijn projecten, maar vooral ook bij mijn persoonlijke en sportieve ontwikkelingen ;).
Cathelijne, ook nog erg bedankt voor je betrokkenheid bij het stageproject van Carlo. **Jur**, bedankt voor de gezelligheid op het werk en op de wielrenfiets! Die keer dat jij tijdens het wielrennen spontaan bij mijn ouders in de (thuis)kappersstoel zat, blijft een grappig moment om aan terug te denken.

Sam, mijn KNO-buddy, ik vond het heel leuk dat we samen projecten hebben gedaan. Samenwerken met jou verliep heel soepel. En vooral ook leuk dat we een vriendschap daaromheen hebben opgebouwd. Helemaal tof dat je ook in 'Nimma' bent komen wonen.

Imran, wat was jij gemotiveerd om naast je drukke 'spoedarts' baan ook nog betrokken te raken bij wetenschappelijk onderzoek. Ook jij hebt ervaren dat onderzoek niet altijd gemakkelijk/soepel verloopt. Maar wat tof dat je toch je 1e publicatie hebt gehaald! Verder was samenwerken erg gezellig en leuk.

Dank aan de **studenten** die ik heb begeleid tijdens mijn onderzoekstijd. Ik vond het erg leuk, maar ook zeer leerzaam om jullie te begeleiden. In het bijzonder **Julia** en **Tino**, jullie tegelijkertijd begeleiden was met momenten wel wat pittig, maar ik heb ook erg kunnen lachen om jullie verschillende manier van aanpakken. **Carlo**, in de laatste maanden van mijn PhD begon jouw stage nog, dat was achteraf gezien misschien niet het meest praktisch, toch zijn 'de boys' & ik er erg trots op je dat je je stage hebt afgerond!

Veel dank aan mijn **geweldige vrienden**, het chillen, lachen, relaxen en gek doen met jullie heeft ervoor gezorgd dat mijn batterij telkens weer werd opgeladen wanneer deze door het werk wat leeg was gelopen.

Lieve **Stephan**, naast alle gezellige avondjes waren het ook onze gezamenlijke sportieve activiteiten die me 'sane' hebben gehouden de afgelopen jaren. In het 1e jaar hebben we samen de Alpe d'Huez gefietst en het laatste jaar sloten we af met onze eerste Marathon, wat waren dit toffe ervaringen! Lieve **Max**, wat een top-chick ben je toch! Met jou kan ik áltijd álles delen, bedankt voor al je liefde en steun. Lieve **Bob**, bedankt voor de gezelligheid o.a. tijdens zeilen op de donderdagavonden met mijn paps. Lieve 'G&T-matties', dank voor de avonden met alcoholische versnaperingen, jullie hebben mij veel ontspanning gegeven ;).

Lieve 'Lovely Ones', **Marleen**, **Emily**, **Yara** en **Inge**, in onze studententijd hebben we mooie vriendschappen opgebouwd, fijn dat we dit nog steeds tijdens het drukke werkende leven voortzetten.

Lieve **Tanja**, **Barbara**, **Anika**, **Koen** en **Huub** (ik durf onze whatsapp-groeps-naam maar niet te noemen hier ;)), bedankt voor alle gezellige avondjes en weekendjes weg! Gelukkig zitten jullie allemaal niet in de medische hoek (behalve Bar dan een beetje) en heb ik het met jullie over hele andere dingen kunnen hebben de afgelopen jaren.

Mijn vriend van kleins af aan, **Stefan**, we hebben het afgelopen jaar een beetje in hetzelfde schuitje gezeten. Terwijl ik mijn PhD afrondde, was jij je masterscriptie aan het afronden. Ik ben onwijs trots op je dat je opnieuw bent gaan studeren en je droomstudie glansrijk hebt

afgerond! Alleen jammer dat we niet meer in hetzelfde dorp/stad wonen. We moeten maar snel een weekend weg om onze prestaties te vieren.

Lieve **Jordy**, mijn brother-from-another-mother-&-father, ook jij veel dank voor alle gezelligheid. Onze surfvakantie in Spanje was een mooie afsluiting!

Lieve **Lisanne**, van mijn vrienden ken jij de 'struggles' van klinisch wetenschappelijk onderzoek het beste. Het was fijn om met jou te kunnen sparren, maar ook vooral om gezellig te wandelen en bij te kletsen. Ik heb onwijs veel respect dat jij je hart bent gaan volgen en nu een baan hebt waar je helemaal happy van wordt. Je hebt me geïnspireerd!

Lieve **Ceca**, wat woon je toch ver weg helaas. Toch hebben we via skypegesprekken gelukkig veel gelachen. Paik&Pees-4-ever.

Beste **Wouter**, nu mijn PhD is afgerond heb ik wellicht meer tijd om toch echt samen voor die triathlon te trainen. Maar waarschijnlijk blijft het bij gezellig chillen, waarbij jij me updates kan geven over het wel daadwerkelijk trainen ervoor ;).

Verder ook nog dank aan mijn leuke ex-collega's van de Bedrijfspoli, die ik inmiddels ook fijne vrienden mag noemen. Dank **Jeanne Margot** en **Frank**, leuk dat we elkaar nog steeds spreken!

Lieve Paranimfen, Marleen & Joey, wat fijn dat jullie mijn paranimfen zijn.
Marleen, mijn lieve, gekke, fijne vriendin ;). Met jou kan ik altijd 100% mezelf zijn, mede omdat jij net zo bijzonder (gek) bent als ik. Je betekent ontzettend veel voor mij.
Joey, mijn cogroepgenoot & kamergenoot, in tegenstelling tot Marleen, verschillen wij juist heel erg. Ik heb erg veel gehad aan jouw relaxedheid en nuchterheid tijdens mijn PhD. Je hebt me vaak zat geholpen en wijze adviezen gegeven. Wat ben je toch een fijn persoon!

Lieve **Familie Driessen**, **Familie Uijen**, **de Ravenberg-Familie** (in het bijzonder **Jos** & **Dominique** en **Marc** & **Marleen**) en **Familie Wijnhoven**, bedankt voor al jullie interesse, vertrouwen en steun tijdens mijn promotietraject. Maar vooral ook dank voor alle liefde die ik van jullie krijg!

Beste **Rob** (Knoet), tijdens mijn PhD heb ik van professoren wetenschappelijke vaardigheden geleerd. Maar gedurende dezelfde periode heb jij mijn vriend vele praktische vaardigheden geleerd, veel dank voor al je hulp om van ons huisje nog meer ons droomhuisje te maken.

Beste **Robert** en **Madeleine**, veel dank voor al jullie interesse in mijn promotietraject.

Lieve **Schoonfamilie**; **Frank**, **José** en **Julie**, wat bof ik toch met zo'n fijne schoonfamilie! Er zijn er maar weinig die de Vierdaagse zullen lopen met hun schoonmoeder en schoonzusje. En er zijn maar weinig schoonvaders die het halve promotieboekje van hun schoondochter kunnen en zullen doornemen om de spelling en grammatica te controleren. Veel dank voor alles en in het bijzonder de heerlijke vakanties in Bretagne!

Lieve **papa** en **mama**, **Ivo** en **Jolanda**, jullie stimuleren mij altijd om te doen wat ik leuk vind. Aangezien ik zelf nog steeds niet zo goed weet wat ik leuk vind, zien jullie mij soms worstelen en onverwachte keuzes maken (hoppa, ineens een promotietraject). Ik besef dat dit voor jullie zeker niet altijd gemakkelijk is. Maar jullie zijn altijd mijn steun en toeverlaat en ben ik jullie onwijs dankbaar voor alles! Ik hou onwijs veel van jullie.
Lief zusje, **Femke**, bedankt voor al je steun en liefde. Zo fijn dat jij en **Bart** zo dichtbij wonen, en ik altijd bij jou langs kan gaan om te chillen, ijsjes te halen of een wandelingetje te maken zodat jij je aantal stappen haalt. Je bent gek dat je ondanks de ervaringen tijdens mijn PhD zelf ook nog een promotietraject wil starten ;), maar het zal jou vast en zeker soepeler afgaan. I love you.

Allerliefste **Yannick**,
De afgelopen jaren zijn een beetje een rollercoaster geweest. Ik besef maar al te goed dat het promotietraject een grote rol in ons leven heeft gespeeld de afgelopen jaren. Ondanks dat ik in mijn loopbaan nog een beetje zoekende ben, ben ik dat verder gelukkig niet. Je bent de liefde van mijn leven, mijn beste vriend en mijn thuis, kortom: je bent mijn alles!
Ik hou van je! Bedankt voor al je steun en liefde.

CURRICULUM VITAE

Maike Uijen werd op 5 september 1993 geboren te Ubbergen. In 2011 voltooide zij de middelbare school aan het Stedelijk Gymnasium Nijmegen en in datzelfde jaar begon zij aan haar studie geneeskunde aan de Radboud Universiteit in Nijmegen. Tijdens haar studie verrichtte zij twee stages in het buitenland, te weten een extracurriculaire onderzoeksstage bij het HRB Centre for Primary Care Research in Dublin (Ierland) en een Tropencoschap in Biharamulo (Tanzania).

Haar onderzoeksstage in het kader van haar Master verrichtte zij bij IQ healthcare (Radboudumc) onder supervisie van Dr. Paul Giessen, Dr. Marleen Smits en Dr. Erik Plat.

Zij rondde haar studie geneeskunde in 2018 cum laude af en begon nadien als arts in de Bedrijfsgeneeskunde bij de Bedrijfspoli in Nijmegen.

In 2019 startte ze met haar promotieonderzoek zoals beschreven in dit proefschrift onder leiding van promotoren Prof. dr. Carla van Herpen, Prof. dr. James Nagarajah en Prof. dr. Martin Gotthardt. Tijdens haar promotietraject presenteerde zij haar werk op verschillende nationale (NWHHT Jong Onderzoeksdag) en internationale (ESMO, EANM, EMIM) congressen en bijeenkomsten van de Patiëntenvereniging Speekselklierkanker. Daarnaast begeleidde zij studenten van diverse studies met hun wetenschappelijke stages.

Maike woont samen met Yannick Gilleit in Nijmegen.
Haar vrije tijd besteed ze graag wandelend (vooral tijdens de Nijmeegse 4-daagse), hikend (liefst in Ierland en Schotland met Yannick), hardlopend (alhoewel ze na het voltooien van haar eerste marathon in 2021 sindsdien haar motivatie is verloren), fietsend, of op het water (zeilend met haar vader).