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ASPECTS OF DOSE, DOSE RATE AND RADIOISOTOPES IN BRACHYTHERAPY OF UVEAL MELANOMA

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By

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The thesis will be defended in public at Aula, St. Erik Eye Hospital, Eye Center of Excellence, Eugeniavägen 12, Solna, Friday 15th of January 2021, 9:00 AM.

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To all patients with uveal melanoma.

POPULAR SCIENCE SUMMARY OF THE THESIS

Uveal melanoma is a rare malignant tumor in the eye which affects 5-10 persons per million inhabitants per year. People with northern appearance have a higher risk of the disease, but the correlation to ultraviolet exposure is not as certain as it is to malignant melanoma in the skin. The age of diagnosis is generally around 60 years old and it is equally common in males and females. Uveal melanoma rarely affects children. Unfortunately, the cancer has spread to other organs in up to one half of patients within 15 years from diagnosis. After the tumor has spread, most patients die within a year.

In the late 60's local radiation treatment (brachytherapy) became more and more available as an alternative treatment to removal of the entire eye. It meant a possibility for the patient to keep the eye and preserve vision. Brachytherapy gained even more popularity as a beneficial treatment option for medium sized tumors when it was shown that these patients have the same survival rate as patients who have the eye removed. In Sweden we use the radionuclide ruthenium-106 for tumors thinner than 6-7 mm, and iodine-125 for tumors 7-10 mm thick. Tumors thicker than 10 mm are treated with eye removal. Between 1979 and 1999 only ruthenium-106 was available, and thus there are some tumors thicker than 7 mm that have been treated with ruthenium-106 in the past.

In paper I we evaluated the radiation dose rate, i.e. radiation dose per time unit in ruthenium-106 brachytherapy. We investigated if there was an association between dose rate and the risk of having to remove the eye after brachytherapy, due to insufficient effect of the treatment/tumor relapse or due to a blind and/or painful eye because of radiation damage to normal ocular tissue surrounding the tumor. We found no such correlation.

In paper II we compared ruthenium-106 and iodine-125 for the treatment of tumors 5.5 mm or thicker. We compared matched subgroups and found no difference in risk of dying from the melanoma, but the patients treated with ruthenium-106 had a higher risk of needing another round of brachytherapy due to insufficient effect of the treatment or tumor recurrence.

We thereby confirmed that iodine-125 is preferable for thicker tumors, in accordance with today's treatment regime.

In paper III we investigated whether there are any gender differences regarding tumor characteristics, effect of treatment or patient survival. We found no differences between men and women that would indicate that the patient's gender should be considered when handling patients with uveal melanoma.

In paper IV we analyzed whether radiation dose or dose rate in brachytherapy with ruthenium-106 and iodine-125 correlated to the risk of developing subsequent tumors and death. We found no such correlation.

ABSTRACT

Uveal melanomas constitute melanomas arising from the choroid, the ciliary body or the iris, and in adults stands for the most common primary malignant intraocular tumor. It has a high risk of metastatic spread followed by inevitable death, and unfortunately the prognosis has not improved over the last decades.

In the beginning the only considerable treatment was enucleation, i.e. removal of the entire eye. However, brachytherapy has since the 70's become the primary treatment for medium sized tumors, and the survival rate has been shown to be equal compared to primary enucleation.

The aim of our studies included in this thesis was to investigate the efficacy of today's brachytherapy treatment regime and if any differences in ocular or patient survival could be found considering different brachytherapy aspects. When investigating only ruthenium-106 brachytherapy, no association was found between high or low dose rate and the risk of secondary enucleation, i.e. enucleation due to extensive unwanted side effects, insufficient treatment effect on the tumor or tumor relapse. In further analysis with both ruthenium-106 and iodine-125 brachytherapy included in the study and the risk of tumor related mortality considered, there was no statistical difference in outcome related to either dose or dose rate applied and we found no negative consequences for patients that received lower dose and dose rates than intended.

As there is a gender difference in survival outcome for some cancers, we investigated this issue regarding uveal melanoma, but no differences could be found indicating this should be a factor to consider when planning treatment.

Ruthenium-106 brachytherapy is generally used throughout Europe. In Sweden, iodine-125 for larger medium sized tumors became available 20 years after ruthenium-106 brachytherapy had begun. We therefore had a great opportunity to analyze the larger tumors treated with ruthenium-106 in the earlier years and compare them to same size tumors treated with iodine-125. There was no difference in survival between

matched groups, but ruthenium-106-treated patients had a significantly higher risk of needing retreatment, thus making iodine-125 the preferred nuclide for thicker tumors.

In conclusion gender is not a factor that needs considering when planning treatment for uveal melanoma. Treatment with brachytherapy is both safe and effective within the current recommended doses and dose rates. Our findings suggest that lower doses and dose rates still would be adequate, and this should be investigated further. The current protocol for radioisotope selection based on tumor thickness also seems adequate for the best possible ocular and survival outcome.

Key words: uveal melanoma, brachytherapy, iodine-125, ruthenium-106, dose, dose rate

LIST OF SCIENTIFIC PAPERS

I. Fili, M., Lundell, G., Lundell, M., & Seregard, S. (2014). High dose rate and low dose rate ruthenium brachytherapy for uveal melanoma. No association with ocular outcome. *Br J Ophthalmol, 98*(10), 1349-1354. doi:10.1136/bjophthalmol-2014-305055

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√III.

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LIST OF ABBREVIATIONS

AJCC   American Joint Committee on Cancer

BAP1   BRCA associated protein 1

BRUM   Brachytherapy Registry of Uveal Melanoma

COMS   Collaborative Ocular Melanoma Study

EBRT   external beam radiation therapy

EIF1AX   encoding eukaryotic translation initiation factor 1 A X-linked

eV   electron Volt

GEP   gene expression profile

GNA11   guanine nucleotide-binding protein subunit alpha 11

GNAQ   guanine nucleotide-binding protein subunit alpha q

Gy   Gray

IDO   indoleamine 2,3-dioxygenase

MAPK   mitogen-activated protein kinase

OCT   optical coherence tomography

OOTF   Ophthalmic Oncology Task Force

PAS   periodic acid-schiff

SF3B1   encoding splicing factor 3B subunit 1A

TIGIT   T cell Ig and ITIM domain

TTT   transpupillary thermotherapy

UBM   ultrasound biomicroscopy

UV   ultraviolet

VEGF   vascular endothelial growth factor

1 INTRODUCTION

Uveal melanoma is the most common primary malignant intraocular tumor, with a high risk for metastasis and tumor-related death.(A. D. Singh, Turell, & Topham, 2011) The tumor's tissue of origin - the uvea - consists of three anatomical structures: the choroid, the ciliary body and the iris. The melanoma originates from melanocytes that normally occur in these structures, and spreads hematogenously predominantly to the liver. Historically, the primary treatment was surgical removal of the entire eye - *enucleation* . Enucleation is still the primary treatment for large tumors, but for small to medium-sized tumors, eye-preserving local radiation treatment - *plaque brachytherapy* – has been the standard treatment for four decades (American Brachytherapy Society - Ophthalmic Oncology Task Force. Electronic address & Committee, 2014) without negative survival consequences.(Diener-West et al., 2001b) Brachytherapy treatment recommendations generally have been based on consensus, as studies on dosimetric issues have been low in number. Patient and ocular factors that might influence the effect of brachytherapy is also a field highly unexplored.

2 LITERATURE REVIEW

2.1 UVEAL MELANOMA

2.1.1 Anatomy

Uveal melanoma is located in the iris and ciliary body in 4 % and 6 % of cases respectively. The absolute majority, 90 %, originate from the choroid.(Kaliki & Shields, 2017) The choroidal melanomas are mostly dome shaped or mushroom-shaped and greyish in tone. Iris melanomas are mostly located inferiorly.(Shields, Kaliki, Shah, et al., 2012)
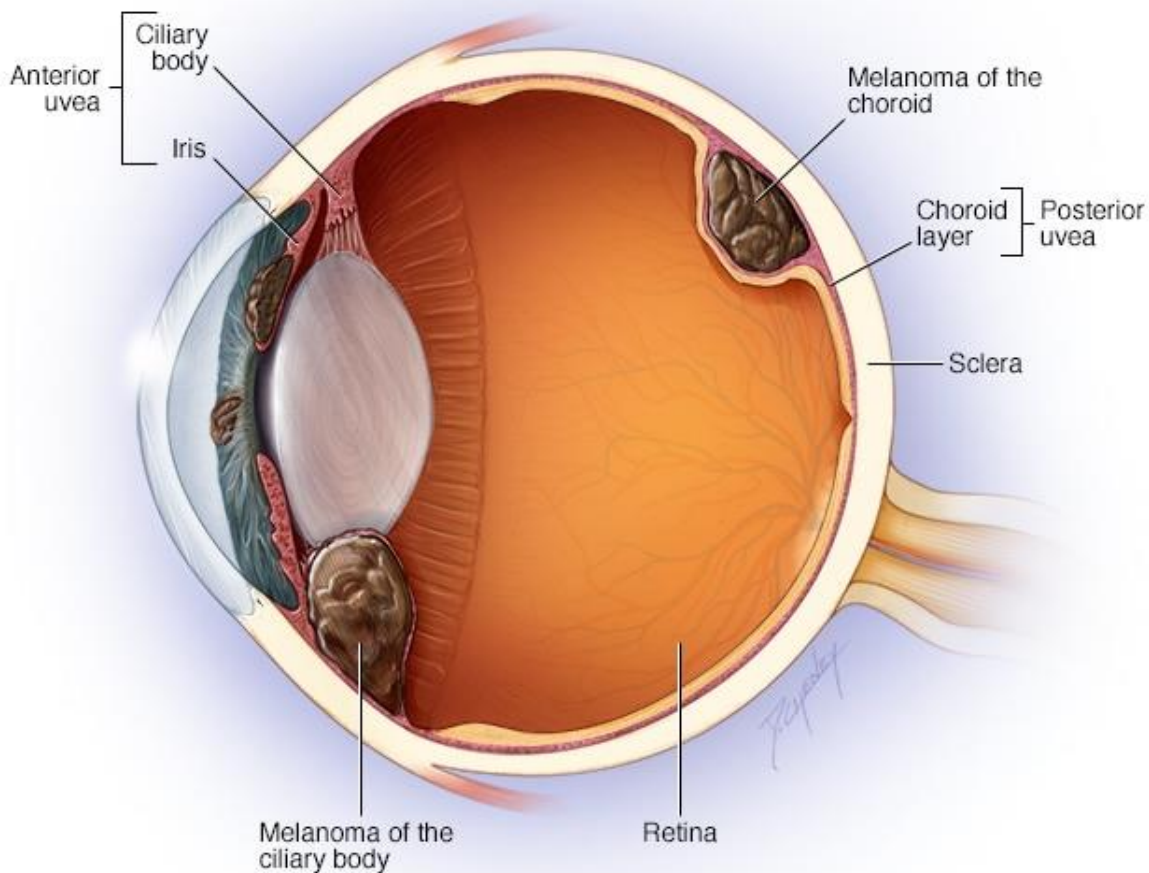

Figure 1. Locations of uveal melanoma. Used with permission of Mayo Foundation for medical education and research. All rights reserved.

The tumors are categorized as small, medium or large in size by a widely accepted set of criteria established by the Collaborative Ocular Melanoma Study group (COMS) in the late 1980's. Small tumors are defined as < 2.5 mm in height and >5 mm in diameter. Medium sized tumors are 2.5-10 mm thick and a maximum of 16 mm in diameter. Tumors exceeding 10 mm in thickness, or 16 mm in diameter at the base are defined as large.("Design and methods of a clinical trial for a rare condition: the Collaborative Ocular Melanoma Study. COMS Report No. 3," 1993) If the melanoma covers more than one

quarter of the choroid but still is not thicker than 5 mm it is referred to as a diffuse melanoma.(Reese & Howard, 1967) Men have been reported to have a larger and more posterior located tumors than women.(B. E. Damato & Coupland, 2012)

Figure 2. Typical appearance of a choroidal melanoma. Picture from patient at St. Erik Eye Hospital.

Histologically, uveal melanoma consists of tumor cells, fibroblasts, macrophages, lymphocytes and blood vessels. There are two types of tumor cells; spindle cells and epithelioid cells. The most common tumor cell type is type B spindle cells, which are fusiform with a spindle shaped nucleus and a clearly defined nucleolus (Figure 3). Type A spindle cells have a less clearly defined or absent nucleolus, but these cells are now considered nevus cells. The epithelioid cells are round or polygonal and the large nucleus contains one or two prominent nucleoli (Figure 4).(McLean, Foster, Zimmerman, & Gamel, 2018) The most common tumor type is the mixed cell type, as the tumor need to be composed of at least 90 % of spindle or epithelioid cell type to be classified as one or the other.("Histopathologic characteristics of uveal melanomas in eyes enucleated from the Collaborative Ocular Melanoma Study. COMS report no. 6," 1998; McLean et al., 2018) Spindle cell tumors have been associated with higher age at diagnosis.(Andreoli, Mieler, & Leiderman, 2015)
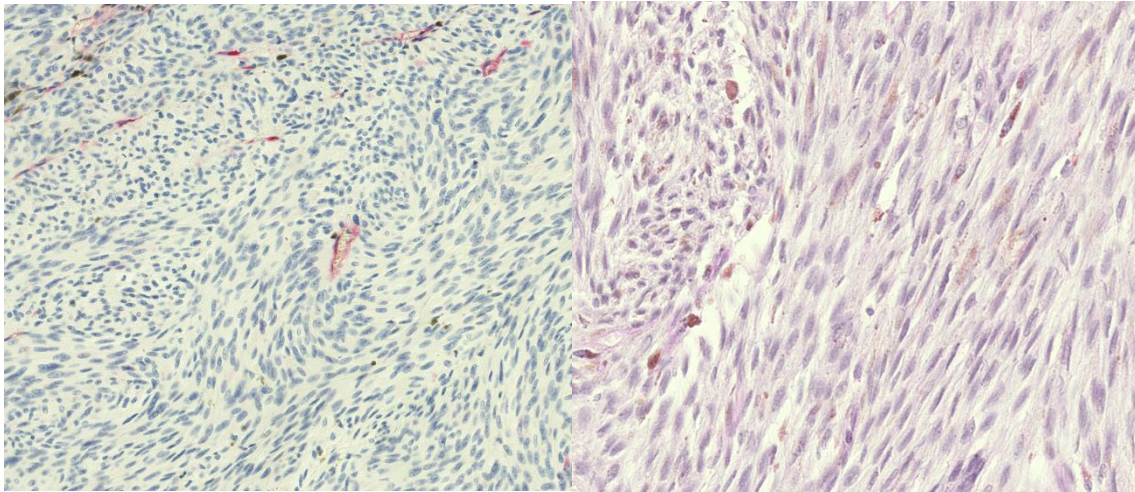
Figure 3. Spindle cell-type tumor. Laminin staining (left, ×20) and PAS staining (right, ×40). Picture from patient at St. Erik Eye Hospital.
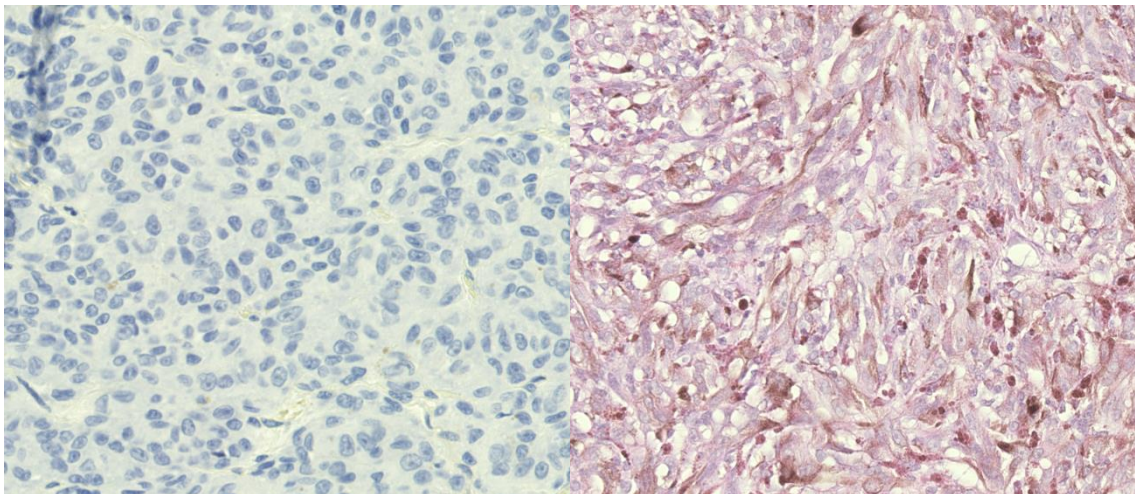

Figure 4. Epithelioid cell-type tumor. Laminin staining (left, ×40) and PAS staining (right, ×40). Picture from patient at St. Erik Eye Hospital.

2.1.2 Epidemiology

Uveal melanoma comprise 85 % of all ocular melanomas, which in turn represents 5 % of all melanomas. (A. D. Singh, Bergman, & Seregard, 2005) The natural course including recurrences and metastases was first described in parallel by Scottish surgeon James Wardrop (1782-1869) and Scottish ophthalmologist-anatomist Allan Burns (1781-1813). (Kivela, 2018)

The age of diagnosis is in general just over 60 years old (Andreoli et al., 2015; Aronow, Topham, & Singh, 2018; Bergman et al., 2002) but around a decade younger for Hispanics and Asians.(Shields, Kaliki, Cohen, et al., 2015) Some studies show a slight overrepresentation in men,(Bergman et al., 2002; Mahendraraj, Lau, Lee, & Chamberlain,

2016) while others don't.(Aronow et al., 2018; Shields, Kaliki, Cohen, et al., 2015) Uveal melanoma in children and adolescents is very uncommon.(Al-Jamal et al., 2016) .

The incidence in Europe is increasing from south to north with an incidence of less than 2 per million in Italy and Spain, increasing to more than 8 per million in the Nordic countries.(Bergman et al., 2002; Virgili et al., 2007) (Krohn, Monge, Skorpen, Mork, & Dahl, 2008) In the US, the general incidence has been estimated to 5.1-5.2(Aronow et al., 2018; Mahendraraj et al., 2016) with a small but significant increase of 0.5 % in whites.(Aronow et al., 2018) The incidence in Canada is 3.8 per million with a steady slow increase.(Ghazawi et al., 2019) Uveal melanoma is very uncommon in Hispanic, Asian and Black populations, representing only 2-5 percent of all affected individuals while Caucasians stand for the remaining 95-98 %.(Aronow et al., 2018; Mahendraraj et al., 2016; Phillpotts et al., 1995; Shields, Kaliki, Cohen, et al., 2015) High incidences of 9.5 and 8.0 per million have been reported in Ireland and Australia, respectively. (Baily et al., 2019) (Vajdic et al., 2003) A very low incidence of 1.9 per million has been reported from Japan.(Tomizuka, Namikawa, & Higashi, 2017) Generally, incidences increase with the latitude.(Yu, Hu, & McCormick, 2006)

2.1.3 Pathogenesis

Risk factors for developing choroidal or ciliary body melanoma is having fair skin and light-colored eyes. Having congenital ocular melanocytosis or melanocytoma has also been identified. Further, the BAP1-tumor disposition syndrome is a risk factor, (Walpole et al., 2018) which is a cancer syndrome including mesothelioma, cutaneous melanoma, uveal melanoma and renal cell carcinoma. (Masoomian, Shields, & Shields, 2018) Whether there is a correlation between exposure to ultraviolet (UV) radiation and uveal melanoma is still not clearly understood, some studies show weak association and some studies show no association.(Mallet, Gendron, Drigeard Desgarnier, & Rochette, 2014; A. D. Singh, Rennie, Seregard, Giblin, & McKenzie, 2004) Very little UV radiation reaches the retina and underlying choroid after passing the cornea, lens and vitreous body in adults(Sliney, 2002) and uveal melanomas show low presence of mutations associated with UV damage.(Mallet et al., 2014) As the incidence of uveal melanoma and cancers in the periocular region remain stable, while cutaneous melanoma is steadily increasing in incidence, it has been suggested that UV exposure differs in the periocular region from the rest of the body.(Weis et al., 2019) However, as fair skin color with easy sunburn, as well as freckles and light eye color have been identified as risk factors for uveal melanoma in a large meta-analyses, the correlation to UV light is still under investigation. Cutaneous and iris nevi were also risk factors as well as welding and occupational cooking. In contrast, free time activities outside, work-related sunlight exposure, latitude of birth, and hair color had no significant correlation.(Nayman, Bostan, Logan, & Burnier, 2017)

Choroidal melanoma can arise de novo or from malignant transformation of choroidal nevi. The rate of choroidal nevi transforming in to melanomas has been estimated to less than 1 % in white Americans.(A. D. Singh, Kalyani, & Topham, 2005) Choroidal nevi are round to oval, flat and usually pigmented lesions and present in approximately 5 % of the adult population (>40 years, Americans).(Chien, Sioufi, Surakiatchanukul, Shields, & Shields, 2017) A commonly used mnemonic to remember risk factors for transformation of nevi into melanomas was created by professor Carol Shields (Wills Eye Hospital, Philadelphia, USA); "To find small ocular melanoma using helpful hints daily", where T represents thickness exceeding 2 mm, F represents subretinal fluid, S represents symptoms of the vision being disturbed, O represents orange lipofuscin pigment, M represents margin with a shorter distance than 3 mm from the optic disc, U represents ultrasonographic hollowness, H represents absence of halo on ultrasound, and D represents absence of drusen.(Shields, Dalvin, et al., 2018; Shields, Furuta, Berman, et al., 2009) Melanomas less than 3 mm thick rarely spread, but share the same risk factors as larger melanomas.(Jouhi et al., 2019)

Risk factors for developing iris melanoma from a nevus include being younger than 40 years old, episodes of hyphema (blood in the anterior chamber of the eye), location clockwise between 4 and 9, diffuse tumor involving the entire iris surface and feathery margins.(Shields, Kaliki, Hutchinson, et al., 2013)

Metastases spread hematogenously, with 90% spreading to the liver but also to the lungs (28-31%) and bone (18-23%). Brain metastases and spread to lymph nodes, subcutaneous tissue or the gastrointestinal tract is less common. More than half of the patients have only 1 organ involved, around 25 % 2 organs and very few 3 organs or more. (Diener-West et al., 2004; Lorigan, Wallace, & Mavligit, 1991)

2.1.4 Genetics

In 2004 uveal melanoma was categorized in two different gene expression profiles (GEP) and a classifier for clinical use is available.(Onken, Worley, Ehlers, & Harbour, 2004; Onken, Worley, Tuscan, & Harbour, 2010) GEP1 is associated with a more favorable prognosis than GEP2.(Onken et al., 2004) Lately however four categories of uveal melanoma has been defined (A-D) where GEP1 are A and B tumors and GEP2 are C and D tumors, divided further after their chromosome 6 and 8 status and inflammatory phenotype.(Robertson et al., 2017) (Jager, Brouwer, & Esmaeli, 2018)

There are five different genes identified to contain mutations important for the prognosis of uveal melanoma; BAP1, EIF1AX, SF3B1, GNAQ and GNA11. The notion that there are consistent alterations in a few chromosomes is known from cytogenetic studies from over 30 years back.(Sisley et al., 1990; Sisley et al., 1997) Lack of the whole chromosome 3; monosomy 3, occurs in around half of patients with uveal melanoma and is the most significant chromosome abnormality with a connection to worse prognosis,(B. Damato et al., 2007) and has also been associated with epithelioid cell type tumors.(Scholes et al., 2003)

Amplification of the long arm of chromosome 8q (around 40 % of patients), or loss of 8p (around 25 %) is also associated with shorter survival,(van den Bosch et al., 2012) as well as loss of chromosome 6q which is seen in one quarter of patients. Gain of chromosome 6p is present in around 25 % and is on the contrary a positive alteration. Whether it depends on the aberration itself or the fact that gain of chromosome 6p is associated with a lack of monosomy 3, is yet not clear.(Nichols, Richmond, & Daniels, 2016) Mutation in the BAP1 tumor suppressor gene is strongly associated with high risk of metastatic disease and tumor-related death.(Decatur et al., 2016; Harbour et al., 2010; Jager et al., 2018) BAP1 is a tumor suppressor located at chromosome 3p21 and has been suggested as explanation to why monosomy 3 is associated with poorer prognosis.(Harbour & Chao, 2014; Robertson et al., 2017) Mitogen-activated protein kinase (MAPK) is a pathway which in the end affects cell proliferation, differentiation and apoptosis. The vast majority of uveal melanoma have an activated MAPK pathway, which is seldom caused by a BRAF mutation but rather from mutations in the mutually exclusive driver mutations in the guanine nucleotide-binding protein GNAQ and GNA11.(Van Raamsdonk et al., 2009; Van Raamsdonk et al., 2010) GNAQ or GNA11 is present in 85 % of uveal melanomas, whereas BRAF mutations, which are quite common in cutaneous melanomas, are rare (Davies et al., 2002; Patel et al., 2011). It has also been shown that a majority of choroidal nevi carry a mutation in GNAQ or GNA11.(Vader et al., 2017) Mutations in EIF1AX and SF3B1 has been identified to be frequently present in uveal melanomas with disomy 3, which carry a positive prognosis.(Martin et al., 2013)

In contrary to most tumors in which the metastases carry similar mutations to the primary tumor,(Reiter et al., 2018; Vogelstein et al., 2013) uveal melanoma metastases have quite many mutational changes compared to the primary tumor.(Shain et al., 2019)

2.1.5 Diagnosis

The diagnosis of choroidal melanoma is established with fundus examination through slit lamp biomicroscopy and/or indirect ophthalmoscopy in conjunction with B-scan ultrasonography, which provides a better possibility than optical coherence tomography (OCT) to determine the shape of the tumor and also see acoustic hollowness and choroidal excavation.(Coleman et al., 2004) A-scan ultrasonography is also performed to evaluate internal reflectivity, being low in choroidal melanoma as opposed to choroidal hemangioma, which can have a similar shape but have high internal reflectivity. In case of intermediate internal reflectivity, choroidal metastasis should be suspected. These examinations are sufficient for diagnosis of choroidal melanoma and initiation of treatment, as the accuracy has been shown to be almost 100 %.("Accuracy of diagnosis of choroidal melanomas in the Collaborative Ocular Melanoma Study. COMS report no. 1," 1990; "Histopathologic characteristics of uveal melanomas in eyes enucleated from the Collaborative Ocular Melanoma Study. COMS report no. 6," 1998)

Transscleral or transvitreal biopsy is indicated when diagnostic uncertainty occurs, usually between amelanotic melanoma and metastatic adenocarcinoma, or other even less frequent tumors. To visualize iris and ciliary body tumors ultrasound biomicroscopy (UBM) provides a superior image of the tumor than anterior segment OCT.(Bianciotto et al., 2011)

Figure 5. Iridociliary melanoma (middle) visualized by AS-OCT (left) versus UBM (right). Note how the margins are much more clearly visible in UBM. Reprinted with permission by Elsevier.(Bianciotto et al., 2011)

Fluorescein and/or indocyanine green angiography are not needed for diagnosis but can be of value when there are differential diagnostic considerations from other choroidal tumors.(Shields, Shields, & De Potter, 1995) MRI can be valuable when extraocular tumor growth needs to be excluded.(Beenakker et al., 2016) Autofluorescence images show hyperfluorescence if there is presence of orange pigment in the melanoma, while benign nevi generally show hypo- or isofluorescence.(Shields, Bianciotto, Pirondini, et al., 2008; Shields, Pirondini, et al., 2008)

Specific biomarkers for early detection of uveal melanoma and also detection of metastatic disease is a hot research field, but no markers are yet clinically in use.(Bande Rodriguez et al., 2020; Triozzi & Singh, 2012)

2.1.6 Prognosis

While only 2 % of patients have metastatic disease at presentation, unfortunately 15 % and 25 % of the patients suffer from metastatic spread after 5 and 10 years respectively, when considering all locations.(Shields, Furuta, Thangappan, et al., 2009)

Ciliary body melanomas and diffuse melanomas are associated with a higher risk of metastatic disease compared to choroidal melanomas, while iris melanomas have a lower risk, which can be attributed to the hidden or clearly visible location of these respective lesions which affects the time to discovery.(Kaliki, Shields, & Shields, 2015; Nichols et al., 2016; A. D. Singh, Shields, & Shields, 2001) More specifically at 5 and 10 years, iris

melanomas metastasize in 4 % and 7 %, choroidal melanomas in 15 % and 25 % and ciliary body melanomas in 19 % and 33 % respectively.(Shields, Furuta, Thangappan, et al., 2009)

Every mm increase in thickness and diameter is associated with higher risk of tumor spread (Diener-West et al., 2004; Shields, Furuta, Thangappan, et al., 2009; Shields, Sioufi, et al., 2018). Small melanomas up to 3 mm thick have a 5-, 10- and 20-year risk of 6 %, 10-12 % and 20 % for metastatic disease, medium sized tumors 3 to 8 mm have a 14 %, 26 % and 37 % risk respectively, and large tumors more than 8 mm thick a considerably higher risk of 35-38 %, 49 % and 67 %.(Shields, Furuta, Thangappan, et al., 2009)

Higher age at diagnosis is also a risk factor for metastatic disease.(Shields, Kaliki, Furuta, Mashayekhi, & Shields, 2012) Tumors in non-Caucasian patients are generally thicker, more hemorrhagic and more peripheral at diagnosis, but the risk of metastatic spread and overall survival does not differ from Caucasians.(Andreoli et al., 2015; Shields, Kaliki, Cohen, et al., 2015) Patients with ocular or oculodermal melanocytosis, which have an increased risk of uveal melanoma, have a doubled risk for metastatic spread compared to uveal melanoma patients without melanocytosis.(Shields, Kaliki, Livesey, et al., 2013)

In a study of pediatric uveal melanoma, that our group contributed to as part of the European Ophthalmic Oncology Group, uveal melanoma has been described in patients as young as less than 3 years old. In young patients with uveal melanoma, the prognosis seems to be more favorable for children younger than 10 years old, with no metastases found after 10 years follow up. In children 10 to 17 years, the proportion of patients suffering from metastatic disease was 9 %, and in the group of young adults 18 to 25 years old it was 17 %. In patients older than 10 years old there was also a worse prognosis for female patients, which was not observed in the group younger than 10 years old.(Al-Jamal et al., 2016)

If the melanoma consists almost exclusively of spindle cells the risk of metastatic disease is lower than if the melanoma is mixed with epithelioid cells. The higher the percentage of epithelioid cells and the bigger they are, the higher the risk of tumor spread.(B. Damato et al., 2007; McLean et al., 2018)

Local tumor recurrence after treatment is associated with a higher risk of metastatic spread, demonstrated in a few small studies(Caujolle et al., 2013; Gragoudas, Lane, Munzenrider, Egan, & Li, 2002; Jampol et al., 2002; Vrabec et al., 1991) but later confirmed by a large multicenter study.(Ophthalmic Oncology Task, 2016)

After discovery of a metastasis, the median survival time is 4-15 months in different reports, with a widely ranging survival rate of 15-50 % and 8-30 % after 1 and 2 years, respectively.(Augsburger, Correa, & Shaikh, 2009; Collaborative Ocular Melanoma Study, 2006; Diener-West et al., 2004; Khoja et al., 2019; Kuk et al., 2016; Rietschel et al., 2005) Rietschel et al noted that surviving patients after 2 years also seem to have a less steep mortality rate and 22 % of patients included in the cohort were alive after 4 years. They also

reported a longer survival time in females (Rietschel et al., 2005) with metastatic uveal melanoma, which was supported by a meta-analysis by Khoja et al.(Khoja et al., 2019)

In Sweden the 5 year relative survival after ruthenium-106 brachytherapy is reported to 95 %.(Bergman, Nilsson, Lundell, Lundell, & Seregard, 2005) Overall, the 5-year relative survival of uveal melanoma of around 80 % remains unchanged in most countries.(Aronow et al., 2018; Mahendraraj et al., 2016) (Chew, Spilsbury, & Isaacs, 2015; Krohn et al., 2008)

2.2 TREATMENT

2.2.1 Brachytherapy

The first eye-sparing treatments were carried out by ophthalmologist Richard Deutschmann in Hamburg, Germany, 1915 by implanting capsules with mesothorium (radium-228) in fractioned doses. Later on in London, 1929, R Foster Moore implanted radon-222 seeds directly in a choroidal sarcoma and thereby managed to treat the tumor while still preserving vision.(Moore, 1930) Also in London, Hyla Bristow Stallard started treating rhabdomyosarcoma with seeds of cobalt-60 in the 1940s and improved his technique by using first wax and then later silver or gold shielded cobalt-60 placed on the episcleral surface. (Stallard, 1948, 1966) In the 1960s Peter Lommatzsch started using ruthenium-106 successfully in Leipzig, Germany. (Lommatzsch, 1986) By the work of Samuel Packer and Marvin Rotman in New York, iodine-125 replaced cobalt-60 resulting in less damage to healthy tumor-surrounding tissue, a consequence from iodine-125s faster dose fall off. (Packer & Rotman, 1980) Brachytherapy with radioactive nuclides has over the years become the predominantly used treatment for small to medium-sized tumors.(Aronow et al., 2018)

In 1984, 43 clinical centers in North America formed the Collaborative Ocular Melanoma Study Group (COMS) which performed the first randomized controlled studies on ocular melanoma treatment.(Arun D. Singh & Kivelä) In one of the studies, patients with medium sized tumors were enrolled between 1987 and 1998 and randomized to either iodine-125 brachytherapy or primary enucleation.(Jampol et al., 2002) Iodine-125 was chosen over cobalt-60 as it had comparable and sufficient tissue penetration up to 15 mm thickness, however requiring only 0.5 mm of a gold shield to shield off the radiation to less than 0.1 %, while 60-cobalt had 50 % radiation left with a shield of 12 mm lead. Iodine-125 is a photon emitter, its half-life extending to 60 days. It decays through electron capture to an excited state of tellurium-125 which, while emitting a 35.5 keV gamma photon, decays to its ground state.(Khan & Gibbons, 2014; Marwaha, Macklis, Singh, & Wilkinson, 2013) The prescription dose to the tumor apex was set to 100 Gy, (Earle, Kline, & Robertson, 1987) revised to 85 Gy by the Radiation Therapy Committee of the American Association of Physicists in Medicine in 1995(Nath et al., 1995) The American Brachytherapy Society has later on updated the recommendations in 2003 stating a minimum dose rate of 0.60 Gy/h and a dose still of 85 Gy for iodine-125, based on the COMS standard. A further update in 2014 supplied a dose recommendation range of 70-100 Gy. No specific dosimetric recommendations are available for other radionuclides. (American Brachytherapy Society - Ophthalmic Oncology Task Force. Electronic address & Committee, 2014)

In 2001 results were published, which showed no difference in survival when comparing treatment of medium-sized melanomas with iodine-125 to primary enucleation.(Diener-West et al., 2001b) This put an end to concerns both that enucleation caused seeding of melanoma cells and an increasing risk of metastatic disease, as well as concerns that brachytherapy would not be as effective as primary enucleation.(A. D. Singh, Rennie, Kivela, Seregard, & Grossniklaus, 2004) These data were confirmed with a longer

follow up of 12 years in 2006.("The coms randomized trial of iodine 125 brachytherapy for choroidal melanoma: V. twelve-year mortality rates and prognostic factors: coms report no. 28," 2006) Local treatment failure was 10 % after 5 years in the brachytherapy group and 15 % had secondary enucleation.(Jampol et al., 2002) Patients included in the report did not have melanomas located contiguously with the optic disc or melanomas extending more than a 90 degree angle of the optic disc if they were located within 2 mm.(Diener-West et al., 2001a) Reports on brachytherapy treatment of juxtapapillary choroidal melanomas both with iodine-125 and ruthenium-106 have however showed comforting results of similar recurrence rates as with treatment of melanomas further from the optic disc.(Lommatzsch & Lommatzsch, 1991; Sagoo et al., 2011)

While iodine-125 is the most commonly used nuclide overall and foremost in the US, ruthenium-106 is widely used in Europe, (Pe'er, 2012; Stefan Seregard & Damato, 2014) Both radionuclides offers a highly safe option for plaque surgeons, albeit surgery time should be kept to a minimum and the number of procedures should not exceed 200 per year.(Laube, Fluhs, Kessler, & Bornfeld, 2000)

Ruthenium-106 is a β ray electron emitter with a half-life of 373 days (Pe'er, 2012). It decays to rhodium-106 which has a half-time as short as 30 seconds, thus almost immediately decaying further to its ground state of palladium-106, while emitting beta radiation with a maximum energy of 3.5 MeV.(Krohn, Chen, Stabo-Eeg, & Hamre, 2020)
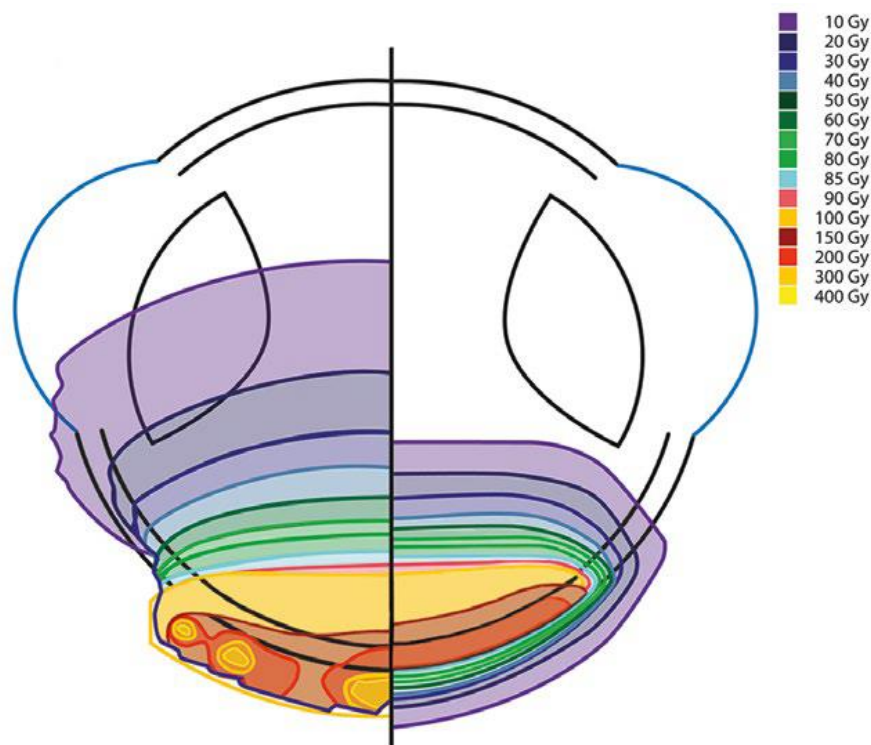
A
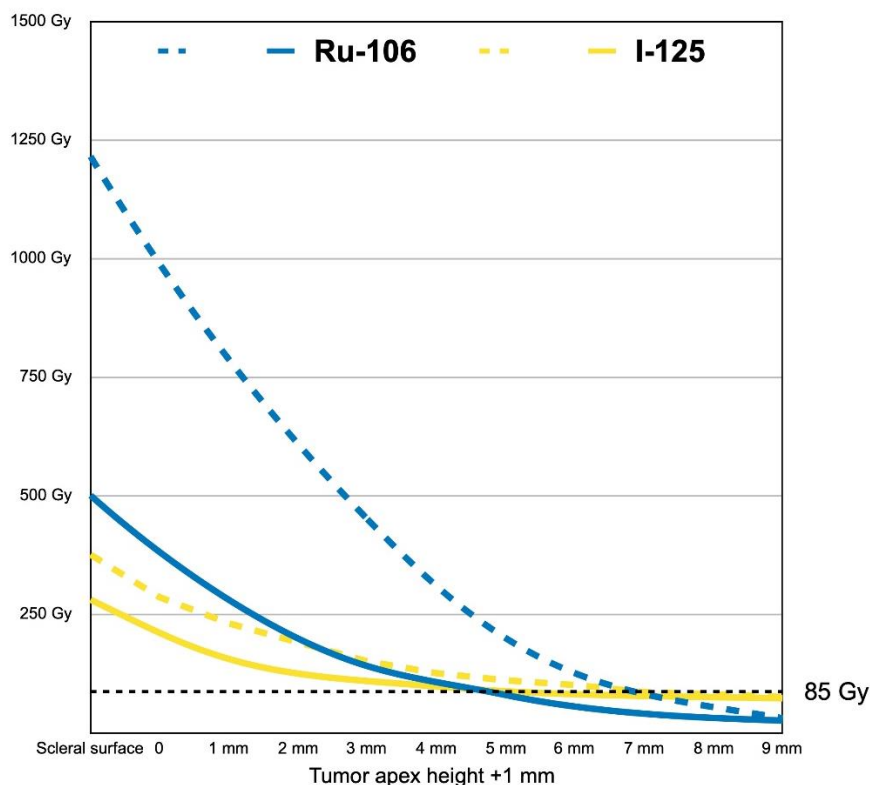
B

Figure 6. A) Approximate dose fall-off curves for ruthenium-106 (106Ru, blue) and iodine-125 (125I, yellow) plaques when 85 Gy is prescribed at the apex +1 mm of 5 mm thick (solid lines) and 7 mm thick (dotted lines) tumors. For every millimeter of penetration into the tissue, the dose fall-off for the β-emitting 106Ru is approximately threefold faster than for the γ-emitting 125I. This implicates that scleral doses have to be significantly higher with 106Ru than 125I for equivalent doses at the tumor apex. **B)** Comparison of theoretical two-dimensional dose distributions of a 125I plaque (left) and a 106Ru plaque (right), both planned with a dose of 85 Gy to the apex of a thin tumor. 125I delivers higher doses to tissues beyond the tumor, and to tissues lateral to the plaque margin. B reprinted with permission from Eckert & Ziegler BEBIG, Berlin, Germany.(Fili et al., 2020)

Other nuclides that have been used for plaque brachytherapy include irridium-192 and cobalt-60, not preferred as both are high-energy gamma emitters that deliver large radiation doses to healthy tumor-surrounding tissue and also present a risk of radiation exposure to caregivers. Palladium-103 has a faster dose fall of than iodine-125, but not as fast as ruthenium-106, and is used in some centers.(Finger, Chin, Duvall, & Palladium-103 for Choroidal Melanoma Study, 2009; Nag et al., 2003) In the most recent recommendations from the American Brachytherapy Society (ABS) – OOTF, the radionuclides used were summarized as iodine-125 and palladium-103 in North America, iodine-125 or ruthenium-106 in Europe, only ruthenium-106 in Japan, and in Russia ruthenium-106 or strontium-90.(American Brachytherapy Society - Ophthalmic Oncology Task Force. Electronic address & Committee, 2014)

2.2.2 Ophthalmic applicators

Ophthalmic applicators used in Sweden are produced in Germany by Eckert & Ziegler BEBIG, Berlin, Germany. They manufacture the COMS gold coated iodine-125 plaques as well as silver coated ruthenium-106 plaques, being the sole producer globally for the latter. They are spherically shaped and plaques up to 25 mm in diameter are available, but most centers use plaques with diameters of up to 20 mm. Since a safety margin of 2 mm is recommended around the tumor, the maximum tumor diameter treatable with plaque brachytherapy is 16 mm in most centers. (American Brachytherapy Society - Ophthalmic Oncology Task Force. Electronic address & Committee, 2014; Nag et al., 2003) In posterior tumors the safety margin might be omitted in some cases, in order to save the macula and optic disc from radiation, as long as the dose to the scleral surface is at least 300 Gy.(B. Damato, Patel, Campbell, Mayles, & Errington, 2005)

Iodine-125 plaques are loaded with iodine seeds for each treatment, the number of seeds needed calculated based on present radioactivity in the seeds and tumor size. The seeds are encapsulated in 0.05 mm titanium tubes, which are welded at each end creating an uneven dose distribution with the lowest dose at the ends. Up to 24 seeds can fit into each plaque.

Figure 7. Iodine-125 plaques. Reprinted with permission from Eckert & Ziegler BEBIG GmbH.

In ruthenium-106 plaques, on the concave side of the plaque there is a thin film of ruthenium-106 covered by a 0.1 mm silver foil that works as a radiation window. The back of the plaque has a 0.7 mm thick silver foil and functions as a radiation shield, which absorbs about 95 % of the beta radiation. According to Eckert & Ziegler, each plaque is calibrated according to a certified reference dose rate from the National Institute of Standards and Technology, USA (NIST). The reference point is at the plaque axis 2 mm from the surface and the absorbed dose rate to water is 4.8 Gy/h, which equals approximately 7.2 Gy/h at the scleral surface. The dose absorbed in tissue decreases to one tenth of its initial value after 7 mm. The plaques are used multiple times within one year after manufacturing.

silver window (0.1 mm)

Ru-106 coated target foil (0.2 mm)

silver backing (0.7 mm)

Figure 8. Ruthenium-106 plaques. Reprinted with permission from Eckert & Ziegler BEBIG GmbH.

2.2.3 Brachytherapy treatment procedure

Treatment is performed under general anesthesia. The conjunctiva is opened and a light source is put to the pupil, locating the tumor which appears as a dark shadow in the sclera. The plaque is placed over the tumor base with a safety margin of 2-3 mm. If necessary, extraocular muscles are temporarily disinserted. The conjunctiva is then sewn back over the

plaque. In the event of a tumor located close to the optic disc, adjunctive treatment with transpupillary thermotherapy (TTT) is sometimes administered. After planned treatment duration is reached, the plaque is removed, muscles are reinserted and the conjunctiva sewn back in place.

2.2.4 Ophthalmic brachytherapy in Sweden

In Sweden, brachytherapy treatment with ruthenium-106 started up in 1979 and 20 years later iodine-125 became available as treatment of choice for tumors more than 6-7 mm thick. Large tumors more than 10 mm thick are still treated with primary enucleation due to concerns for insufficient radiation doses. Similarly, lesions located adjacent to the optic nerve may be treated with primary enucleation regardless of tumor size, due to surgical inaccessibility.

A standard dose of 100 Gy to the tumor apex was prescribed when ruthenium-106 was introduced in 1979, and a maximum dose of 1000 Gy was set to the scleral surface. In 1999 the maximum scleral dose was increased to 1500 Gy and iodine-125 was introduced with an estimated treatment dose of 85 Gy to the tumor apex. During the 20 years when only ruthenium-106 was available, some thicker tumors than 7 mm were treated, when eye preserving treatment was highly desirable.

2.2.5 Radiation biology

Brachytherapy constitutes implantation of radioactive sources inside or next to the tumor, brachy meaning short distance in Greek. (Hall & Giaccia, 2011) A nuclide is defined by an atoms combination of protons and neutrons, in conjunction with its energy state. An isotope is defined by one elements differing number of neutrons. The ionizing radiation which is emitted when subatomic bonds break and radionuclides and radioisotopes decay into more stable forms, can create ions by displacing electrons from atoms or molecules. The tissue that absorbs ionizing radiation forms free radicals and ions when chemical bonds break between molecules. These oxidize DNA and other contents in the cells. The cell loses its capacity to reproduce and, if repair mechanisms are insufficient, undergo apoptosis. The SI unit for absorbed radiation is Gray. One Gray equals 1 Joule of energy absorbed by 1 kg tissue.(Khan & Gibbons, 2014; A. D. Singh, Pelayes, Seregard, Macklis, & Bandello, 2013)

2.2.6 Radiation effects on ocular tissue

After brachytherapy of a choroidal melanoma, radiation retinopathy occurs in over 60 % of patients. (S. Seregard, Pelayes, & Singh, 2013) Dose rates more than 2.6 Gy/h to the tumor base as well as if the margin of the melanoma base is under 4 mm from the fovea, are both risk factors associated with a higher risk of this complication. (Gunduz et al., 1999b) Exudative retinal detachment due to increased vascular permeability can be seen directly after radiation, sometimes resolving, but chronic retinal detachment is not uncommon. If the patient suffers from diabetes, there is a higher risk of radiation induced proliferative retinopathy which later on can cause rubeosis and neovascular glaucoma, which together with

exudative retinal detachment is referred to as a type of toxic tumor syndrome.(Groenewald, Konstantinidis, & Damato, 2013; Gunduz et al., 1999b) Radiation induced macular edema is treated with intravitreal corticosteroids and/or anti-VEGF, both treatments leading to improvement. However, spontaneous improvement may occur why more studies on the effect is needed.(Giuliari, Sadaka, Hinkle, & Simpson, 2011; Groenewald et al., 2013) Edema is the earliest sign of radiation maculopathy, a complication which not surprisingly corresponds to the distance between the fovea and the tumor margin as well as radiation doses exceeding 50 Gy.(Gunduz et al., 1999b; Pagliara et al., 2018)

A dose of more than 50 Gy received by the optic nerve constitutes a risk for optic neuropathy. Treatment with intravitreal steroids has little effect on vision improvement, and systemic corticosteroids have no effect. Hyperbaric oxygen therapy may have effect if administered early.(Groenewald et al., 2013) Developing cataract is a risk if the irradiation to the lens is more than 10 Gy. Treatment is with standard cataract surgery with good results.(Wachtlin et al., 2000)The reported incidence of radiation induced cataract, maculopathy and optic neuropathy varies widely from just a few percent to more than 80 %, 60 % and 30 % respectively. (Naseripour et al., 2016; Takiar et al., 2015; Wen, Oliver, & McCannel, 2009) In comparing 106-ruthenium with 125-iodine Wilkinson et al found an 18 % lower radiation dose to the macula, 53 % lower radiation dose to the optic disc, and as low as 89 % lower radiation dose to the lens for tumors < 5 mm thick.(Wilkinson, Kolar, Fleming, & Singh, 2008) Scleral necrosis is a rare complication seen in less than 10 % of treated patients at 10 years after brachytherapy. There is an increased risk for thicker tumors, if there is ciliary body involvement and if there is higher intraocular pressure. (Gunduz et al., 1999a; Radin et al., 2008)

2.2.7 Radiation effects on uveal melanoma

Irradiated tumor cells undergo necrosis or apoptosis, and tumor blood vessels loose endothelial cells which leads to vascular occlusion followed by fibrosis. Less subretinal fluid and increased pigmentation as well as increased internal reflectivity on ultrasound examination are clinical findings after treatment. (Maheshwari & Finger, 2018; A. D. Singh, Rennie, Kivela, et al., 2004) The rate of tumor regression depends on which type of radiotherapy that has been used (ruthenium faster than iodine) and tumor thickness (thicker tumors regress faster).(Rashid, Heikkonen, Singh, & Kivela, 2018) These findings support the notion that in ocular brachytherapy, fast regression is not only due to shorter doubling times in aggressive tumors, but is also related to the very high radiation doses administered to the tumor base, causing immediate cell disruption, vessel obliteration and tumor infarction.(B. Damato, 2018) A rapid regression of tumor size after radiation correlates to worse prognosis according to some(Augsburger, Gamel, Shields, Markoe, & Brady, 1987; Kaiserman et al., 2004), but not all authors,(Pepin et al., 2019) and some have found that rapid regression is associated with earlier metastases and slow regression with late metastases (after 2 years).(Glynn, Seddon, Gragoudas, Egan, & Hart, 1989) A great number of accumulated melanomacrophages or extensive fibrosis after radiation can slow regression (B.

Damato, 2018), as well as a dark pigmented tumor.(Rashid et al., 2018) Consideration must also be taken in regression pattern, as oval to dome shaped or mushroom shaped tumors seem to regress more in area than thickness.(Rashid, Heikkonen, & Kivela, 2015)

Since monosomy 3 is associated with worse prognosis, fast regression rate and the presence of monosomy 3 have been investigated, not showing such correlation in some studies (Chiam et al., 2014; Gupta et al., 2016; Salvi et al., 2017) but indeed in others.(Shields, Bianciotto, Rudich, et al., 2008) GEP1 has been linked to faster regression than GEP2 the first 6 months after iodine-125 brachytherapy.(Mruthyunjaya, Seider, Stinnett, Schefler, & Ocular Oncology Study, 2017) As irradiated tumors show a higher frequency as well as a higher complexity of chromosomal aberrations than untreated tumors,(Dogrusoz et al., 2015) questions have been raised whether the treatment itself allows or induces metastatic progress when local failure occurs.(B. Damato, 2018)
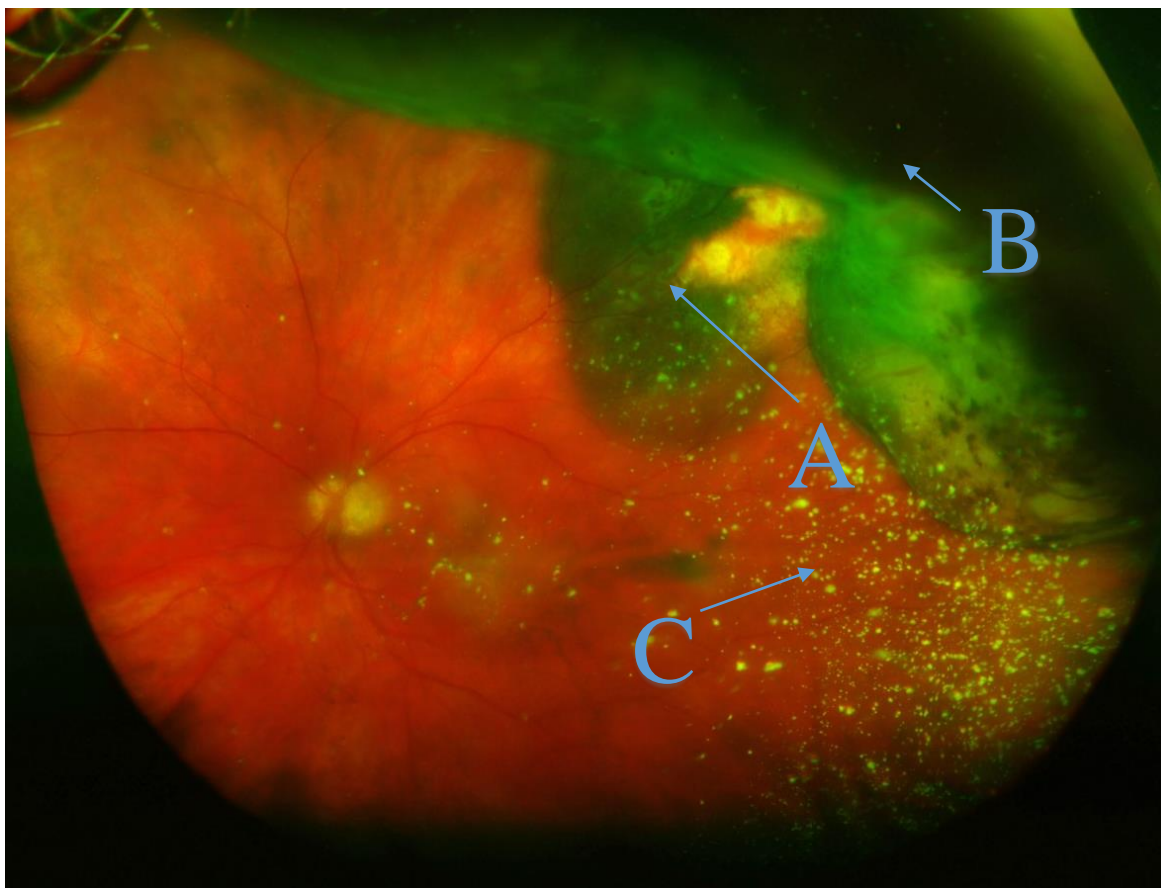

Figure 9. Choroidal melanoma treated with brachytherapy. A) Successfully treated area with a yellowish atrophic appearance. B) Large area of tumor relapse. C) Asteroid hyalosis in the vitreous body, incidental finding not related to uveal melanoma.(Burris et al., 2017) Picture from patient at St. Erik Eye Hospital.

2.2.8 Other treatments

As mentioned earlier, large tumors 10 mm thick or more are still primarily treated with enucleation. If there is major extraocular growth, exenteration is the only option, where all tissue in the orbital cavity is removed.

Local resection of medium sized and large tumors is performed by internal evacuation or external lamellar sclerouvectomy in some ophthalmic oncology centers. Various results have been described, some report a high local recurrence rate of 30 % (Hong, Wei, Hua, Xu, & Shao, 2014) after 5 years, while others report 10 % in the presence of neoadjuvant proton beam therapy in addition to local resection. The rate of complications requiring additional vitreoretinal surgery in the latter study was however very high (70 %).(Willerding et al., 2016) In a comparison between neoadjuvant proton beam therapy and adjuvant ruthenium-106 brachytherapy together with local resection for large tumors there was an eight fold higher risk of local recurrence with brachytherapy, but no difference in risk of enucleation or metastatic spread.(Boker et al., 2018) Concerns about these techniques are that external approach requires hypotensive anesthesia and there is also worries about tumor seeding. Further analysis on these risks and also the advantages to brachytherapy is needed.(B. E. Damato, 2012)

Charged particle therapy is an external radiation technique using protons, helium or carbon. Protons are most commonly used and they produce low irradiation when traveling through a mass until they stop, where they deliver the maximum of their energy, called the Bragg peak. Treatment for choroidal melanoma is generally 56-60 Gy delivered to the prescription point in 4 daily fractions, but doses up to 70 Gy in 5 daily fractions are also within treatment standards.(Hrbacek et al., 2016) The anterior segment complications and secondary enucleation rates have been reported to be of higher frequency than for iodine-125 treatment,(American Brachytherapy Society - Ophthalmic Oncology Task Force. Electronic address & Committee, 2014) but other studies including a large meta-analysis covering over 8000 patients showed no survival benefit or difference in eye retention rate compared to brachytherapy.(Abrams, Gagne, Melhus, & Mignano, 2016; Wang et al., 2013) The rate of local recurrence after charged particle therapy is however reported to be lower than after plaque brachytherapy.(B. Damato, Kacperek, Chopra, Campbell, & Errington, 2005; Desjardins et al., 2012; Wang et al., 2013)

Other treatments like stereotactic photon beam therapy and endoresection are less common but also provide a high degree of local control. (Dunavoelgyi et al., 2011) (Konstantinidis, Groenewald, Coupland, & Damato, 2014)

2.2.9 Follow up and screening for metastases

In Sweden ocular examination is performed 1,3 and 6 months after brachytherapy or enucleation and thereafter annually for the rest of the patients life, as local recurrence can

occur many years after treatment.(Ophthalmic Oncology Task, 2016) Screening for metastasis is done by liver ultrasound or CT semiannually for 5 years. Ultrasound offers a high sensitivity and specificity for suspected metastatic lesions and excellent specificity is achieved with confirmatory CT scan.(Choudhary, Gupta, Bena, Emch, & Singh, 2016) Liver function blood tests are not used in Sweden, they are not effective for early detection of metastases but have a high negative predictive value (Mouriaux, Diorio, Bergeron, Berchi, & Rousseau, 2012) and high LDH was associated with shorter survival in a Dutch nationwide cohort of advanced disease.(Jochems et al., 2019) Other blood biomarkers for detection of metastases have not been proven reliable enough for clinical use.(Bande Rodriguez et al., 2020; Triozzi & Singh, 2011, 2012)

2.2.10 Adjuvant treatment and treatment of metastatic disease

TTT is an accepted adjuvant treatment for juxtapapillary tumors but is not advised as primary treatment even for small melanomas. (Harbour, Meredith, Thompson, & Gordon, 2003; Mashayekhi et al., 2015; Win, Robertson, Buettner, McCannel, & Bennett, 2006)

Pre-enucleation radiation is seldom used as it has not shown any survival benefits.(Hawkins & Collaborative Ocular Melanoma Study, 2004) However, post exenteration or post enucleation of a tumor with some extrascleral growth is occasionally treated with external beam radiation therapy (EBRT, 50 Gy in 180-200 cGy fractions). To avoid radiation damage to eyelids, intraorbital brachytherapy is also an option.(Finger, 2009) Local resection is sometimes conducted with preoperative proton beam radiation or plaque radiation after the resection as described earlier.(Boker et al., 2018)

Adjuvant treatment has not been proven effective and efforts using treatments for cutaneous melanoma for metastatic uveal melanoma has shown poor results, including antibodies inhibiting the programmed death receptor 1 (PD-1).(Algazi et al., 2016; Carvajal et al., 2017; Triozzi & Singh, 2014) Liver screening has been questioned since no effective treatment is available. However, since high diagnostic accuracy can be achieved by non-invasive methods, the benefits are clear. By early detection of metastases, the patient may be offered the opportunity to make life altering decisions about palliative treatment. Early detection also increases the likelihood that a patient can be included in a clinical trial. (Choudhary et al., 2016)

Tebentafusp is a new form of immunotherapy based on the monoclonal T cell receptor against cancer (ImmTAC) platform. It has an immune mobilizing function and targets cells that express HLA A0201 which has complexed with the peptide gp100. In melanoma Gp100 is expressed strongly, while it has only a weak expression in melanocytes, and in other tissue there is a minimal expression. Results are promising and further trials are ongoing.(B. E. Damato, Dukes, Goodall, & Carvajal, 2019)

Local treatment of liver metastases includes metastasectomy, local chemotherapy by arterial liver infusion and liver embolization, none of which has been proven to prolong survival.(Augsburger et al., 2009)

3 RESEARCH AIMS

In paper I we studied whether dose rate applied to the scleral surface or tumor apex was associated with the risk of secondary enucleation in brachytherapy of uveal melanoma with ruthenium-106.

In paper II we compared ruthenium-106 and iodine-125 regarding treatment outcome and survival in treatment of uveal melanomas 5.5 mm or thicker.

In paper III we investigated gender differences in long-term survival after brachytherapy of uveal melanoma with ruthenium-106 and iodine-125.

In paper IV we evaluated the effect of dose and dose rate on disease-specific mortality after brachytherapy of uveal melanoma with ruthenium-106 and iodine-125.

4 MATERIALS AND METHODS

4.1 PATIENTS AND METHODS

In paper I the computerized Medlog V.2012-8 (Information Analysis, Crystal Bay, Nevada, USA) registry of all patients in Sweden treated with brachytherapy for uveal melanoma was searched. Later on, in paper II-IV, the Medlog registry had been replaced by the Brachytherapy of Uveal Melanoma registry (BRUM) (Registry Center South, EyeNet Sweden), from were all patient data was retrieved.

Date and cause of death was retrieved from the National Mortality Registry. This registry contains data on cause of death of all diseased persons in Sweden. As misclassifications of the actual cause of death is frequent between cutaneous and uveal melanoma,(Bergman et al., 2003) we cross-checked the National Mortality Registry's data with our own clinical records of patients' metastases. If a patient had metastasized uveal melanoma and later died from metastasized melanoma without any records of diagnosed or treated cutaneous melanoma, we considered the cause of death set as cutaneous melanoma as misclassified.

In paper I, 962 patients treated with ruthenium-106 from 1 November 1979 to 31 December 2012 was included. In total eight groups - two sets of approximate quartiles based on dose rates at the tumor apex and the scleral surface, respectively, were created. Data regarding age at diagnosis, gender, eye laterality, tumor thickness, tumor diameter, AJCC T-category(Edge et al., 2010), tumor distance to the optic disc, tumor distance to the macula, dose rate to the scleral surface, dose rate to the tumor apex, enucleation after brachytherapy and reason for enucleation was analyzed.

In paper II, 1493 patients treated with either ruthenium-106 or iodine-125 brachytherapy from 1 November 1979 to 31 December 2015 were included and data regarding tumor thickness, tumor diameter, brachytherapy nuclide (ruthenium-106 or iodine-125), retreatment, rates of TTT, enucleation after brachytherapy and survival was analyzed. Matched subgroups were created in consideration of recommendations for 1:1 nearest neighbor matching. Values with the highest impact on the means of age at diagnosis, gender, pretreatment visual acuity, tumor thickness, tumor diameter, AJCC T-category(Kivelä et al., 2017) and follow up years were excluded until all differences became non-significant in Mann-Whitney U tests(Stuart, 2010). Two pairs of matched subgroups were made, one pair of 55+55 patients with tumors 5.5-7.4 mm thick and one pair of 45+45 patients with tumors ≥7.5 mm thick, treated with ruthenium-106 and iodine-125 respectively.

In paper III, 1541 patients treated with either ruthenium-106 or iodine-125 brachytherapy for uveal melanoma from 1 November 1979 to 20 November 2017 were included and data regarding gender, age at diagnosis, pretreatment best corrected visual acuity, symptom duration before presentation, type of symptoms, tumor origin, tumor thickness and diameter, AJCC T-category(Kivelä et al., 2017), brachytherapy nuclide

(ruthenium-106 or iodine-125), prescribed radiation dose, treatment duration, TTT before or after brachytherapy, enucleation after brachytherapy, date of metastasis and follow up years was analyzed.

In paper IV, 1238 patients treated with brachytherapy for uveal melanoma from 25 April 1996 to 16 March 2016 were included and data regarding age at diagnosis, gender, pretreatment best corrected visual acuity, symptom duration before presentation, type of symptoms, tumor thickness and diameter, AJCC T category(Kivelä et al., 2017), brachytherapy nuclide (ruthenium-106 or iodine-125), prescribed radiation dose, planned treatment duration, actual treatment duration, TTT before brachytherapy, date of death (melanoma-related, from other causes or unknown cause) and follow up years was analyzed.

For all patients in paper I-IV, tumor thicknesses and diameters were measured pre-operatively by standardized A and B-scan ultrasonography. From 1979-1999 ruthenium-106 was the only nuclide available. Maximum dose at the tumor apex was 100 Gy and to the scleral surface 1000 Gy. In 1999, iodine-125 was introduced as the treatment standard for thicker tumors more than 6-7 mm, with a maximum apex dose of 100 Gy which was lowered to 80 Gy in 2003 due to concern for surrounding tissue. Also, in 1999, the maximum dose to the scleral surface was increased to 1500 Gy.

Ruthenium-106 plaques were of the CCA, CCB, CCX, CCZ, COB, CIA or CIB types (Eckert & Ziegler BEBIG, Berlin, Germany) with diameters up to 20 mm. Iodine-125 plaques were custom made by a goldsmith in 18 carat gold, with 20 iodine-seeds (Eckert & Ziegler) glued to the concave surface. Radioactivity data for each plaque were independently verified by medical physicists at the Karolinska University Hospital, Solna, Sweden. Dosimetry was performed by using a p-type silicon semiconductor detector (Scanditronix, Sweden), estimating dose rate at different depths from the applicators convex surface with an estimated accuracy of ±4.5 %.(Lax, 1991) Silicon diodes have been used for dosimetric calculations of high-energy photon and electron beam radiation since the 60's.(Rikner & Grusell, 1987) Based on tumor size and the current radiation activity in the specific plaque, the exact individual treatment time for each patient was calculated by the medical physicists.

Surgery was performed under general anesthesia. The tumor was located by transillumination, followed by plaque positioning with a minimum 2 mm margin around the melanoma. Tumors close to the fovea had eccentric plaque fixation and adjunctive laser therapy to the compromised tumor margin. Juxtapapillary melanomas were either treated with a notched plaque or standard plaque combined with adjunctive laser therapy. Some patients received pre-, peri-, or postoperative adjunctive argon laser photocoagulation or TTT. Follow up was scheduled at 1, 3, 6 and 12 months after treatment, and then annually for the rest of the patient's life. Screening for liver metastases by ultrasonography or CT was performed semiannually for 5 years after diagnosis. After a few years, patients not from the Stockholm region were referred back to their home clinics. In case of tumor relapse, the patient was however referred back to our clinic as we are the sole treating clinic in the country. In case of enucleation this was also known to us as all specimens are sent to our clinic for

histopathological analysis. Therefore, all data on relapse and enucleation are accurate. Date of last follow up might be lost for some patients, but most clinics in the country have a well-established routine of sending copies of clinical records to our clinic for registration in the BRUM registry.

All studies received approval from the Central Ethical Review Board, Stockholm, Sweden.

4.2   STATISTICAL ANALYSIS

Paper I: Statistical analysis was performed using Statistica V.12 software (Statsoft Scandinavia AB, Uppsala, Sweden). Level of statistical significance was set to p <0.05. Frequency distributions were studied by the median value and interquartile range as frequencies were not normally distributed. To evaluate differences between baseline characteristics in the groups, independent t test and one-way analysis of variance (ANOVA) were performed for continuous variables and for categorical variables the Pearson χ^2 test was used. Kaplan Meier survival curves were generated for each dose rate quartile and compared with the log rank (Mantel-Cox) test for secondary enucleation analysis. Univariate and multivariate analyses using Cox regression modelling was also performed. Data was consistently treated linearly in the Cox regression analyses.

Paper II: Statistical analysis was performed using SPSS statistics V.25 (IBM, Armonk, New York, USA). In power calculations we studied the largest similar cohort published to date, in which the relative risk for tumor recurrence and enucleation after ruthenium-106 versus iodine-125 brachytherapy was 13.75 and 3.12 respectively.(Shields et al., 2002) To detect the smallest of these differences (risk for enucleation) with 80 % power at a significance level of 0.05, the sample size for each arm was estimated to 9 patients. Reversely, in our sample size we were able to detect differences of down to approximately 8 % with 80 % power and 9 % with 90 % power. The Mann-Whitney U test was used for tests of the null hypothesis in non-parametrically distributed group level data. The Pearson χ^2 test was used for comparisons of proportions in two-by-two tables. For analyses of hazard for post brachytherapy enucleations, repeated brachytherapy and melanoma-related mortality, multivariate Cox regressions with tumor thickness and diameter as covariates were used. Kaplan-Meier survival curves were computed for matched subgroups and the log-rank (Mantel-Cox) test of equality of survival distributions was applied for analyses of disease-specific survival and overall survival.

Paper III: Statistical analysis was performed using SPSS statistics V.25 (IBM, Armonk, New York, USA). In power calculations for 90 % power, the sample size for each arm was estimated to at least 541 patients at a significance level of 0.05 and the smallest detectable difference between the groups of 10 %. Reversely, in our sample size of 775 men and 766 women, we would be able to detect differences of down to 8-9 % with 90 % power.(Tunes da Silva, Logan, & Klein, 2009) The Mann-Whitney U test was used for tests

of the null hypothesis in non-parametrically distributed group level data. The Pearson χ^2 test was used for comparisons of proportions in two-by-two tables. Bi- and multivariate Cox regression analysis with tumor thickness and diameter were used for analyses of hazard for post-brachytherapy enucleations, repeated brachytherapy and melanoma-related mortality. To test weather our survival data met the proportional hazard assumption, we built a Cox regression model with a time-dependent versus a time-independent treatment variable (man or woman). Kaplan-Meier curves for disease-specific and overall survival were computed and the log rank (Mantel-Cox) test of equality of survival distributions was applied. Equivalence between men and women was tested as the relative risk for melanoma-related death with modified two-sided 95 % CIs as for binary endpoints.(Tunes da Silva et al., 2009)

Paper II-IV: Visual acuity was measured on a decimal scale chart at a distance of 5 m (Monoyer Granström) ranging from 1.0 (corresponding to 20/20) to 0. Modified from standards in the Swedish National Quality Registry for Cataracts, counting fingers at a distance of 4 m was recorded as 0.08, hand movements as 0.04, perception and localization of light as 0.01 and amaurosis as 0. Average visual acuities were calculated on -1 LogMar converted values, and then converted back to the decimal scale.

AJCC T-categories were codified as T1=1, T2=2, T3=3 and T4=4, to be able to compare the categories statistically. Actual radiation dose given was defined as: (actual brachytherapy duration / planned brachytherapy duration) x prescribed dose = actual dose.

4.3   ETHICAL CONSIDERATIONS

As all the studies have been based on retrospective data from an already existing registry and no changes were made to diagnostic procedures, treatment or follow-up of patients, informed consent was waived in accordance with the permissions from the regional ethical review board in Stockholm and the Swedish Ethical Review Authority. All studies adhered to the tenets of the declaration of Helsinki.

To be reminded of an event that may have happened many years ago might be very stressful for the patient, and as the results are presented on a group level there is no way to identify single patients. Thus, the risk of harm to the patient has been considered higher than the benefit ifwe were to contact every single patient for approval.

5 RESULTS

5.1 PAPER I

"High dose rate and low dose rate ruthenium brachytherapy for uveal melanoma- no association with ocular outcome."

We investigated if dose rate to the scleral surface or tumor apex correlated to the risk of secondary enucleation after ruthenium-106 brachytherapy of uveal melanoma. Tumor and patient characteristics were similar across the quartiles. As expected, thicker tumors treated with a standard dose to the tumor apex consequently had a lower dose rate to the tumor apex. There was a wide frequency distribution of both dose rate to the tumor apex (0.01-4.76 Gy/h) and to the scleral surface (1.66-11.11 Gy/h). Dose rate to the tumor apex was clearly skewed, again as expected as thinner tumors receive a higher dose per time unit to the apex. Dose rate to the scleral surface had a near normal distribution.

Most patients got to keep their treated eye, the eye retention rate was 80 % 5 years after treatment and 72 % 10 years after treatment. When secondary enucleation did occur, 75 % of these were due to tumor progression and the rest due to extensive damage to ocular tissue which could lead to secondary glaucoma, optic neuropathy, radiation retinopathy and other negative conditions that caused a blind and/or painful eye. Enucleation only because of the patients wish was rare.

No statistical difference between the quartiles could be detected in Kaplan-Meier survival curves neither when analyzing dose rate at the tumor apex or dose rate at the scleral surface. In univariate analysis, independently predictive clinical parameters of secondary enucleation were tumor diameter, tumor thickness and distance to the optic disc, but in further multivariate analysis only tumor diameter was statistically significant (p=0.049). There were no significant correlations between high or low dose rate and the risk of secondary enucleation, neither at the scleral surface nor at the tumor apex.

5.2 PAPER II

"Ruthenium-106 versus iodine-125 plaque brachytherapy of 571 choroidal melanomas with a thickness of ≥5.5 mm."

Of 571 patients, 317 (56 %) were treated with ruthenium-106. The mean apex dose was 91.6 Gy for ruthenium-106, as expected a higher mean dose than for iodine-125, which was 81.4 Gy, and the difference was statistically significant (p<0.001). There was also a statistically significant difference in tumor thickness between the groups, the mean thickness for ruthenium-106 treated tumors being 6.6 mm versus 8.5 mm for iodine-125 treated tumors (p<0.001), and the former group having a significantly better pretreatment visual acuity (p=0.03). There was no significant difference in rate of pre- peri- or postoperative TTT (p=0.97) between the radioisotopes.

There was a statistically significant difference in need for a second round of brachytherapy (p<0.001), being 8 % (14 patients) in the ruthenium-106 group with a mean treatment interval of 2.2 years, and 1 % (3 patients) in the iodine-125 group with a mean treatment interval of 1.25 years (p=0.51). The main reason was tumor relapse for both groups, except for 2 ruthenium-106 treated patients who had insufficient regression. Of the 17 patients treated twice, 8 died from metastatic disease.

Twenty percent of the ruthenium-106 treated patients underwent secondary enucleation, of which 89 % was due to tumor relapse and 3% due to insufficient treatment effect. The remaining patients had blind eyes with or without pain. Of the iodine-125 treated patients, 17 % underwent secondary enucleation, of which 70 % was due to tumor relapse. The remaining patients had unwanted side effects like secondary glaucoma, chronic uveitis or total retinal detachment, except for 1 patient who preferred to have the eye enucleated.

Multivariate Cox regression analysis with tumor diameter, tumor thickness, ruthenium-106 and iodine-125 as covariates revealed a significantly increasing hazard for secondary enucleation with increasing tumor thickness (p<0.001), but not for the other covariates. Overall, there was no difference in disease-specific survival between the radioisotopes. In the matched subgroups with tumors 5.5-7.4 mm, the mortality rate was 30 % and 45 % after 5- and 10-years post treatment with ruthenium-106, and 23 % and 38% after iodine-125 treatment. The subgroups with tumors ≥7.5 mm had a 5- and 10-year mortality rate of 25 % and 51% after ruthenium-106 treatment, and 33% and 54% after iodine-125 treatment respectively. There were no statistically significant differences in 5- and 10-year survival between any of the matched subgroups.

5.3   PAPER III

"No Gender Differences in Long-Term Survival after Brachytherapy of 1,541 Patients with Uveal Melanoma."

Of the patients treated with ruthenium-106 there were 626 men and 619 women. One hundred and forty nine men and 147 women were treated with iodine-125. There were no statistical difference between men and women regarding age at diagnosis, visual acuity before treatment, symptom duration or type before diagnosis, which uveal structure the tumor originated from, AJCC T-category, radionuclide used, prescribed radiation dose, intended or actual treatment duration, proportion of TTT before or after brachytherapy, or follow up years. Tumor thickness and diameter were borderline significantly larger for men (p=0.06) than for women (p=0.08). The proportional hazard assumption was met in analysis of time from brachytherapy to death or last follow-up (p=0.83).

The deviation from planned treatment time to actual treatment time was similar between men and women. There was no statistical difference in hazard ratio for disease-specific death in a multivariate Cox regression analysis where tumor thickness and dose in

categories of decreasing dose by 10 Gy where included as covariates (dose >90 Gy, 80-90 Gy, 70-80 Gy and <70 Gy).

One hundred and sixteen (15 %) men and 121 (16 %) women suffered from tumor relapse (p=0.65). Twenty-one men had brachytherapy retreatment because of tumor relapse (19 patients) or insufficient treatment effect (2 patients). Twenty-seven women had brachytherapy because of retreatment tumor relapse (24 patients) or insufficient treatment effect (3 patients) There was no statistically significant difference in risk of retreatment (p=0.36) which, if it did occur had a mean interval of 2.2 years between the treatments for men and a mean interval of 2.8 years for women (p=0.45). In a multivariate Cox analysis with tumor thickness and tumor diameter added as covariates, there was no significant difference in hazard ratio for repeated brachytherapy for men and women (p=0.47). Of the in total 48 retreated patients, 17 (35%) eventually died from metastatic disease.

Secondary enucleation was performed on 107 (14 %) of the men and 111 (14 %) of the women. The main reason was tumor relapse in 91 % for men and 90 % for women including 2 % because of insufficient treatment effect. The remaining were because of extensive side effects. The enucleation-free Kaplan-Meier survival was similar between men and women (p=0.86) and in total 33 % of the secondary enucleated patients eventually died from their melanoma. The median survival after diagnosis of metastatic disease of 0.9 years for men was not statistically different from the 1.1 years for women (p=0.12).

The 5-year melanoma-related mortality rate for men and women were 14 % and 15 %, and the 10-year mortality rate were 24 % and 26 % respectively. Lastly, the 15-year mortality rate were 27% and 32 % respectively. When adding tumor diameter and sex as covariates in a bi-variate Cox regression analysis, there were no significant difference in risk of disease-specific death between men and women (p=0.32). For men, the disease-specific survival was 18.2 years, and for women 15.5 years, with no statistically significant difference in Kaplan-Meier analysis (p=0.22). Overall survival of 13.5 years for men and 15.3 years for women was also similar (p=0.60). The relative risk of disease-specific death for men versus women was 0.9 in test of equivalence.

5.4   PAPER IV

"Effect of plaque brachytherapy dose and dose rate on risk for disease-related mortality in 1238 patients with choroidal melanoma"

The mean actual radiation dose ranged from 73.0 to 108.6 Gy in the first to the tenth decile, and the dose rate ranged from 0.5 Gy/hour to 2.8 Gy/hour. Across the deciles, symptoms and patient characteristics had an equal distribution for both dose and dose rate. Tumor distance to the macula was not equally distributed (p<0.001), being longer for lower dose and dose rate deciles. Distance to the optic disc was also longer for lower dose rate deciles (p=0.006), but equally distributed for dose deciles. As expected, factors related to tumor size were not

equally distributed. Larger tumors were more frequent in deciles with lower doses and dose rates, in concordance with the limited dose to the scleral surface of 1500 Gy. Iodine-125 was more frequent in the lower dose deciles (p<0.001) since the dose aim to the tumor apex is 80 Gy for iodine-125 and 100 Gy for ruthenium-106. Regarding dose rate ruthenium-106 was the radionuclide more frequent in the high and low deciles and iodine-125 in the middle groups (p<0.001). Pretreatment best corrected visual acuity and treatment with TTT before brachytherapy were not equally distributed but did not show a certain direction of distribution for dose (p=0.004 and p<0.001) or dose rate (p<0.001 for both).

There was a median follow up time for metastatic-free patients of 7.5 years (IQR 5.8). The total mortality rate by the end of follow up was 37 % (462 patients). The cause of death was unknown for 18 % of these 462 patients, while 58 % had died from the melanoma and 24 % had died from other causes. Kaplan-Meier curves showed a significant difference in tumor-related mortality between high and low dose and dose rate (p<0.001), but in further analysis with a multivariate Cox regression analysis were tumor thickness, tumor diameter and distance to optic disc were added as covariates, there was no association between dose and melanoma-related mortality (p=0.4) or dose rate and melanoma-related mortality (p=0.6). For both dose and dose rate, tumor diameter was independently associated with melanoma-related mortality (p<0.001), but not tumor thickness (p=0.04 and p=0.07 respectively) or distance to the optic disc (p=0.09 and p=0.1 respectively).

6 DISCUSSION

In Sweden, the first treatment with plaque brachytherapy for uveal melanoma was performed in November 1979. Ever since that very first patient, extensive data on patient and tumor characteristics has been continuously collected along with treatment parameters and follow-up. As St. Erik Eye Hospital in Stockholm is the only center in the country performing the treatment, this registry has full coverage. Furthermore, personal identification numbers allow for near complete follow-up data, regardless of the patients' region of residence. For almost three decades, all data was registered by one single devoted person, Professor Göran Lundell, and after that only a handful others, ensuring high that every involved individual has a high degree of insight, knowledge and professional investment in the data. These circumstances form the backdrop for a unique research environment that is virtually unmatched in the world.

The treatment parameters collected include dosimetric aspects such as dose and dose rate at both the tumor base and tumor apex. Previous studies have shown that with dose rates that are too low, tumor cells may recover after being irradiated while dose rates that are too high will cause unnecessary damage to normal tissue.(Yang et al., 1993) In the first study included in this thesis (paper I), by analyzing the rate of secondary enucleation in relation to widely ranging dose rates in ruthenium-106 brachytherapy, we came to the conclusion that the dose rates commonly used in our practice are neither causing too much damage to normal surrounding ocular tissue, nor causing unacceptable rates of local recurrence. Naseripour et al studied 51 patients with melanomas 7-11 mm thick, treated with ruthenium-106 and found a lower secondary enucleation rate of 10 % than our results, however with a shorter mean follow up time of 3 years. Radiation dose rates to tumor apex and to the scleral surface were 0.37 (0.23 - 0.51) Gy/h and 6.44 (5.94 – 7.94) Gy/h respectively.(Naseripour et al., 2016) Mossböck et al reported in a small number of patients a significantly lower metastatic rate in patients treated with plaques with calculated dose rates > 4 Gy/h than patients treated with plaques less than 4 Gy/h.(Mossbock, Rauscher, Winkler, Kapp, & Langmann, 2007). The absence of data on dose rate at the tumor apex or scleral surface however makes comparison challenging. One must also take into account that differing results in eye retention rate is influenced by what is traditionally recommended in each clinic, in case of local recurrence – retreatment or secondary enucleation.

As ruthenium-106 was the only radionuclide used in our center between 1979 and1999, we had an opportunity to compare ruthenium-106 and iodine-125 treatment for thicker tumors than 5.5 mm in subgroups matched regarding patient demographics and clinical features, paper II. We found no significant differences in risk of secondary enucleation or of melanoma-related death in contrast to Tagliaferri et al. who found a significantly higher rate of metastatic disease after treatment with ruthenium-106 compared to iodine-125 in patients with tumors 5-7 mm thick, but no difference in local control, albeit only in 26 patients.(Tagliaferri et al., 2012) We did however find that ruthenium-106 brachytherapy of thicker tumors is significantly more often associated with the event of

retreatment than iodine-125 brachytherapy. This alone we consider sufficient reason to avoid ruthenium-106 for thicker tumors. The reason for retreatment is local recurrence and occasionally insufficient regression. Even if we found no such relation in our study, local recurrence has been associated with higher risk of metastatic disease in a large multicenter study by the Ophthalmic Oncology Task Force.(Ophthalmic Oncology Task, 2016) In this context, it should however be mentioned that the prognostic implications of tumor recurrence may be caused by the fact that aggressive tumors recur more often, rather than that insufficient brachytherapy cause increased aggressiveness. Consistent with our findings in both paper I and II, Papageorgiou et al. didn't find a correlation between dose rate and risk of local recurrence, but indeed a higher risk for local recurrence for tumors >5 mm thick treated with ruthenium-106.(Papageorgiou, Cohen, Bunce, Kinsella, & Hungerford, 2011) Tarmann et al. analyzed 30 patients with tumors >7 mm thick that received ruthenium-106 treatment, and found a significant correlation to secondary enucleation but not to tumor recurrence.(Tarmann et al., 2015) Again, differing results are influenced by local decisions on retreatment versus secondary enucleation in cases of tumor relapse. Not least, avoidance of retreatment is of course desirable for the patient and also for economic and health care reasons.

For thinner tumors, ruthenium-106 seems to be a better treatment choice. It has a high local control rate,(B. Damato, Patel, et al., 2005; Takiar et al., 2015) and its preferred usage for thinner tumors is supported by the findings by Takiar et al, who found a significantly higher risk of unwanted side effects when comparing iodine-125 against ruthenium-106 for tumors <5 mm thick.(Takiar et al., 2015)

As described earlier, men have been reported to have larger tumors than women at presentation. (B. E. Damato & Coupland, 2012) In addition, men have been reported to have a higher frequency and earlier debut of metastatic disease than women, (Zloto, Pe'er, & Frenkel, 2013) and also a shorter survival rate after the diagnosis of metastatic disease. (Rietschel et al., 2005) In paper III we analyzed tumor and treatment parameters in relation to gender, as far as we know not investigated before from a treatment perspective. We found no significant differences between men and women in survival or ocular outcomes that would indicate that the treatment strategy should be adjusted in any way depending on the patient's sex.

The dose of 85 Gy to the tumor apex, at least for iodine-125, has traditionally been accepted since the COMS trials, and in the same time period our clinic lowered the apex dose from 100 Gy to 80 Gy for iodine-125, i.e 5 Gy lower than the American Brachytherapy Society's recommendations. Throughout the years, the great majority of patients have received the intended dose, but for natural reasons, final doses have been lower and higher than prescribed in some cases. In paper IV we found no adverse effects on treatment effectivity or side effects from a dose range of 73-108 Gy. This finding is in concordance with Perez et al. who did not find lower rates of local control, survival, metastases with iodine-125 doses down to 63-69 Gy to the apex. They did however note significantly worse visual acuity and radiation complications with higher doses up to 104 Gy (Perez et al., 2014) and this report was included in a large meta-analysis of 15 retrospective and prospective

studies by Echegaray et al. who also concluded that there are no certain relation between local recurrence and lower iodine-125 doses to the apex, opening up for randomized trials investigating this further.(Echegaray, Bechrakis, Singh, Bellerive, & Singh, 2017) Quivey et al found a correlation between lower dose and dose rate to the tumor apex, lower dose rate to the base, long treatment duration and increased risk of local failure when analyzing 150 patients treated with iodine-125 retrospectively, however treatment dose to the tumor apex ranged all the way down to 29.5 Gy.(Quivey, Augsburger, Snelling, & Brady, 1996)

In our study we included both ruthenium-106 and iodine-125, a decision based on our finding in paper II, that the use of ruthenium-106 or iodine-125 is not an independent predictor of melanoma-related death.

The fact that uveal melanoma can metastasize decades after primary treatment raises concerns that it doesn't really matter how and if we try to treat the tumor. However, it is well known that larger tumors carry a worse prognosis.(Arnljots et al., 2018; Shields, Furuta, Thangappan, et al., 2009; Shields, Kaliki, Furuta, et al., 2015) It has also been shown that older age at treatment is associated with worse prognosis and most risk factors for tumor spread. As the mean tumor diameter is larger for older patients, and larger tumors carry a higher degree of for example monosomy 3, earlier treatment prevents not only local tumor growth but also negative genetic alterations, dedifferentiation and metastatic spread.(B. E. Damato, Heimann, Kalirai, & Coupland, 2014) Over the last years the importance of tumor surrounding microenvironment for the metastatic process has been paid more and more attention. Uveal melanoma primary tumors which express a high level of immune checkpoints T cell Ig and ITIM domain (TIGIT) and indoleamine 2,3-dioxygenase (IDO) carry a higher risk of metastatic spread,(Robertson et al., 2017; Stalhammar, Seregard, & Grossniklaus, 2019) as well as tumors with a high rate of infiltrating macrophages and a dense extracellular matrix, (Makitie, Summanen, Tarkkanen, & Kivela, 2001; Stalhammar, See, Phillips, & Grossniklaus, 2019) all of which have more time and opportunity to develop in the happening of insufficient and/or late treatment. That being said, brachytherapy for uveal melanoma is a necessary treatment and our findings support that lower treatment doses and dose rates are still effective, with the benefit of fewer ocular complications.

7 CONCLUSIONS

Uveal melanoma is a rare cancer, but still the most common primary malignant intraocular tumor in adults. The prognosis is similar after enucleation and eye-preserving plaque brachytherapy. A high proportion of patients develop metastases, after which the prognosis is dismal.

Plaque brachytherapy of uveal melanoma with ruthenium-106 and iodine-125 is well established and these radionuclides are widely used in most developed countries. The aim of this thesis was to analyze different aspects of brachytherapy of uveal melanoma and its correlation with ocular and patient outcomes. Both radionuclides have their place in the treatment regime, with ruthenium-106 being more advantageous for thinner tumors and iodine-125 for thicker tumors. The current dose and dose rate recommendations are effective but our findings support that lower doses and dose rates are also safe and effective, which opens up for further investigation in randomized trials. We found no indications that the patient's gender needs to be taken into account when planning the brachytherapy treatment, or when estimating prognosis for the patient.

8 POINTS OF PERSPECTIVE

The introduction of brachytherapy as a treatment alternative for uveal melanoma has meant a sight preserving alternative for countless patients. In the initial era of brachytherapy, treatment was based mainly on consensus. Increasing number of studies analyzing the optimal radionuclide, dose and dose rate as well as which patient features and genetic aspects are most relevant for outcome, takes us further and further towards optimizing the brachytherapy treatment for each individual patient. To be able to tell the patients that we are giving the best treatment available not only based on experience, but also based on extensive research, is of essence.

My studies are retrospective and not randomized. Even if a prospective randomized trial is always preferable, the large number of patients that we are able to analyze in our extensive BRUM directory reaching four decades back, makes our findings a very high powered valuable contribution to the knowledge about uveal melanoma and brachytherapy we have today.

Hopefully, one day brachytherapy will be a treatment that was used in the past. Hopefully, as we understand more and more of the molecular pathways and the genetics of uveal melanoma, a tumor specific treatment effective on both primary tumor and micrometastases, not harmful for normal tissue, will become a reality. Until then, plaque brachytherapy will be a mainstay in uveal melanoma treatment and consequently, further studies in the field of brachytherapy in parallel with studies of uveal melanoma on a molecular level are warranted.

9 ACKNOWLEDGEMENTS

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