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Diagnostic and Treatment Aspects of Actinic Keratoses

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Abbreviations used in the Thesis

AK	Actinic keratosis
Bcl-2	Protein of B-cell lymphoma 2 gene
CD31	Cluster of differentiation 31
IEC	Intraepidermal carcinoma
Ki67	The nuclear protein Ki67
LSRs	Local site reactions, synonymous to application site reactions
p16	Protein p16
p53	Tumour protein P53
p63	Protein p63
SCC	Squamous cell carcinoma
SWS	Shiny white streaks, also known as chrysalis or crystalline structures in dermatoscopy
UV	Ultraviolet light

Introduction

Actinic keratoses (AK) are one of the most common dermatological conditions seen in everyday general and dermatological practice. The usual clinical presentation is multiple, erythematous, ill-defined, scaly macules, papules, or plaques on a chronically sun-damaged skin of older fair-skinned individuals (Dziunycz et al., 2018; Reinehr and Bakos, 2019; Schmitz et al., 2018; Steuer et al., 2020; Tizek et al., 2019). The greatest clinical importance of AK is the possible progression to invasive squamous cell carcinoma (SCC), the second most common type of skin cancer (Stratigos et al., 2020b), although for some patients the cosmetic burden is the reason for consultation. Morphologically, AKs are classified as intraepidermal neoplasms. They represent the in-situ stage of SCC and are considered to be the initial phase in the development of invasive SCCs (Reinehr and Bakos, 2019; Rowert-Huber et al., 2007). The development of SCC may occur directly from AK or as a new lesion on surrounding sun-exposed skin, as the presence of AK increases the overall risk of subsequent cutaneous malignancies, especially cutaneous SCC (Guorgis et al., 2020). Although it is difficult to calculate the exact progression rate, a recent study by Madani et al. (Madani et al., 2021) showed that the cumulative incidence of cutaneous SCC reached 17.1 % at 10 years in patients with AK. Clinical discrimination of AK and SCC in early stages of dermal invasion may be challenging and it has been shown that 1.5 % of clinically diagnosed AK lesions identified by board-certified dermatologist were SCCs with superficial invasion on histopathological assessment (Ehrig et al., 2006). The diagnosis of all skin lesions including AK and SCC is facilitated by knowledge of dermatoscopy. The latter is a common and easy-to-use tool in dermatological practice, and a recent systematic review and study showed a sensitivity rate of up to 98.7 % and a specificity of up to 95 % when AK is diagnosed by dermatoscopy (Huerta-Brogeras et al., 2012; Valdés-Morales et al., 2020).

Moreover, a dermatoscopic progression model from AK to invasive SCC has been published previously (Zalaudek et al., 2012). Although several studies have assessed correlations between clinical, dermoscopic and histological features of AK and SCC (Lee et al., 2019; Papageorgiou et al., 2022; Rstom et al., 2022) studies assessing immunohistochemical features are lacking.

Treatment of AK is indicated in all patients to prevent the progression and can be targeted at individual lesions or the entire actinic field. All treatment options, except surgery, aim to destroy atypical keratinocytes, resulting in temporary inflammatory reactions of varying severity. Discussing the treatment-associated local site reactions (LSRs) before therapy may lead to delay in treatment initiation. During treatment, such reactions cause discomfort and distress (Eisen et al., 2021; Neri et al., 2019) and affect health-related quality of life (Hanke et al., 2016). A study by Strohal et al. (Strohal et al., 2012) showed that 19.4 % of patients treated with the commonly used imiquimod 5 % cream, scheduled unplanned visits due to their concerns about LSRs. Moreover, LSRs lead to non-adherence, which may reduce treatment efficacy (Stockfleth et al., 2015). Topical therapy is indicated for most patients with AK and field cancerisation because it is easy to use, highly effective, physician time saving and the only treatment modality targeting the entire area. Nevertheless, treatment-related factors such as the prevalence of severe application site reactions and changes in dermatoscopic signs during and after therapy needed to be additionally studied, and this was done in this work.

Aims of the Thesis

1 To determine the clinical, dermatoscopic, histopathological and immunohistochemical features characterizing actinic keratoses and intraepidermal carcinomas;

2 To evaluate the changes of the dermatoscopic features of actinic keratoses during treatment-induced inflammation and post-treatment;

3 To determine the incidence of treatment induced severe local skin reactions of actinic keratoses and the associated frequency of treatment discontinuation.

Tasks of the Thesis

The following tasks were set to achieve the aim of the doctoral thesis:

1 To identify the clinical characteristics of actinic keratoses that have progressed to squamous cell carcinoma through a systematic review of published prospective studies.

2 To summarise the most common dermatoscopic signs characterising facial actinic keratoses and intraepidermal skin carcinomas and provide explanations of these signs.

3 To investigate which of the following immunohistochemical markers: p53, p63, p16, Ki67, cyclin D, Bcl-2 and CD31, are differentially expressed in clinically healthy sun-damaged skin compared to skin samples in which actinic keratoses and intraepidermal carcinomas have developed.

4 To investigate which of the clinical and dermatoscopic signs of actinic keratoses and intraepidermal carcinomas are associated with the altered expression of the following immunohistochemical markers: p53, p63, p16, Ki67, cyclin D, Bcl-2 and CD31.

5 To evaluate how the inflammation caused by topical therapy changes the dermatoscopic signs of actinic keratoses.

6 To evaluate the post-treatment dermatoscopic signs of actinic keratoses.

7 To collect literature data on the incidence of severe application site reactions caused by topical therapies for actinic keratoses and the related frequency of treatment discontinuation due to these local site reactions.

Hypotheses of the Thesis

1. The clinical and dermatoscopic signs of actinic keratosis correlate with the underlying histopathological characteristics and the expression of the following immunohistochemical markers: p53, p63, p16, Ki67, cyclin D, Bcl-2 and CD31.

Dermatoscopical signs of actinic keratoses are diminished during the treatment-induced inflammatory phase and no signs of actinic keratosis remain after treatment.

Treatment of actinic keratoses often induces severe application site reactions leading to discontinuation of treatment.

Novelty of the Thesis

This Thesis consists of several studies that are unique in several aspects. First, by updating the previous review by Quaedvlieg et al. (Quaedvlieg et al., 2006) in a systematic manner, we were able to identify clinical signs that characterised AK that later progressed to invasive SCC, namely, long-standing and large (> 1 cm) or merging AK. Second, we examined seven immunohistochemical marker (p53, p63, p16, Ki67, cyclin D, Bcl-2 and CD31) expressions in elderly sun-exposed skin and the AK/IEC group, thus identifying disease-specific changes. In this study we found that the prevalence of amorphous masses, the intensity of p53 staining, the expression of Bcl-2 and CD31 were different in clinically healthy sun-damaged skin compared to skin samples in which AK and IEC had developed. In continuation, due to prospective patient sample, we were able to explore associations between clinical and dermatoscopic signs with histopathological and immunohistochemical marker expressions. Such extensive correlations are still relatively rare in the literature, mostly in case reports. As a result, we were able to show that dermatoscopically typical AK, including lesions with characteristic small arborising vessels more

commonly had amorphous masses in histopathology. Moreover, dermatoscopically typical AK had lower expression of CD31 and the absence of follicles was associated with increased subepidermal Bcl-2 expression. These findings are consistent with the current knowledge of the markers involved in the progression of keratinocyte carcinomas.

In the subsequent section of the study, we explored different treatment associated factors for AK. First, the alterations in dermatoscopic signs observed during treatment, including the variability of shiny white streaks (SWS). Additionally, the coexistence of AK and basal cell carcinoma at the same anatomical site is rarely documented in the existing literature. Our photo documentation during AK therapy revealed that effective treatment of epidermal AK lesions can facilitate the diagnosis of more profound lesions, such as basal cell carcinoma. Additionally, we showed the variable nature of SWS. Furthermore, to the best of the Author's knowledge, as part of this Thesis in 2018, for the first time in Latvia, daylight photodynamic therapy was administered to a group of patients, and the outcomes were recorded. In the concluding part of the research, the treatment-induced LSRs were studied. It was discovered that patients are more inclined to discontinue therapies with long-term treatment regimens than those that elicit stronger LSRs. This finding emphasises the critical role of patient counselling in clarifying potential reactions to treatments, thereby improving compliance and the overall success of the therapeutic interventions.

Discussion

Exposure to ultraviolet (UV) light is the pivotal environmental cause of skin cancer that induces and promotes tumour expansion, and consequently sun-exposed sites, such as the face, are the most common location for skin cancers to arise (Reinehr and Bakos, 2019; Subramaniam et al., 2017; Van Der Pols et al., 2006a). Actinic keratoses (AK) are common UV light-induced lesions on chronically sun-damaged skin of light-skinned individuals that serve as a marker of UV damage and risk of cancer development, including direct progression. Although some authors state that it is not possible to predict which AKs will progress to invasive SCC (Stockfleth et al., 2012) and that these lesions are a general marker of SCC risk rather than true precursors (Stratigos et al., 2020a), a study published in 2006 by Quaedvlieg et al. (Quaedvlieg et al., 2006) identified several clinical findings associated with the malignant progression of AK and invented the acronym IDRBEU. In the acronym, "I" stands for inflammation/induration; "D", for a diameter > 1 cm; "R", for rapid enlargement; "B", for bleeding; "E", for erythema; and "U", for ulceration. Other minor clinical criteria identified in their study were pain, palpability, hyperkeratosis, pruritus, and pigmentation. In our study we updated the review of Quaedvlieg et al. and performed a systematic review of published prospective studies. We included studies from year 2005 onwards. The study was conducted according to the PRISMA guidelines and was registered in the PROSPERO database before the start of the study. As a result, we identified long-standing and large (> 1 cm) or merging AK as the most important risk factors for AK to progression into SCC in high-risk populations. This finding promotes the treatment of long-standing and large AK primarily, if all lesions cannot be treated at once.

In addition to identification of most important clinical signs, the most common dermatoscopic features of non-pigmented AK and facial intraepithelial carcinomas (IEC) were collected. These signs, along with their explanations and

pictures, were included in a chapter of the book “Dermatoscopy”, published in 2022. You can find this chapter in the list of publications.

We also compared histopathological and immunohistochemical markers between prospectively collected samples of normal skin and the study group of AK and SCC in situ (IEC). The included sample and control patients were of similar age to reduce possible confounding effects of age and UV exposure on immunohistochemical marker expression that have been previously shown to have an impact (Bakshi et al., 2020; Khodaeiani et al., 2013; Nasiri et al., 2021). In this study, we found differences in the prevalence of amorphous masses, the intensity of p53 staining, the expression of Bcl-2 and CD31 between study and control groups. Specifically, we found amorphous masses in most cases of AK/IEC and in none of the control biopsies. The replacement of dermal collagen with amorphous masses of elastotic material is characteric of severe solar elastosis and is the consequence of excessive cumulative UV damage (Karagas et al., 2007). This finding is consistent with UV radiation as the main cause of AK and with previous studies that have found severe solar elastosis in most cases of cutaneous SCC (Corbalán-Vélez et al., 2010; Karagas et al., 2007).

In our study we saw a higher staining intensity of p53 in the AK/IEC group, which is consistent with previous studies and the role of the p53 protein in the early stages of SCC development, allowing cells to bypass apoptosis (Bakshi et al., 2020; Berhane et al., 2002; Javor et al., 2021; Piipponen et al., 2021). At the same time, we saw no difference in the distribution/level of p53 expression in the epidermal layers between the two groups. Similar to the studies by Neto et al. (Neto Pimentel et al., 2013) and Piipponen et al. (Piipponen et al., 2020) – not all our AK expressed p53. More than 5 % immunoreactivity to p53 was observed in 75.8 % of the AK/IEC samples and the staining was considered strong in 44.8 % of the cases. Significantly higher p53 staining was observed with age and at sun-exposed sites, while the expression of p53 in the skin has

been shown to decrease with daily application of sunscreen (Van Der Pols et al., 2006b). The impact of these factors was minimised by sampling both the study and control groups from facial skin and by including patients whose age was not statistically different.

Our results showed that the AK and IEC samples had higher Bcl-2 levels and quantitative expression in the epidermal layers and dermis. Bcl-2 is an antiapoptotic molecule located in the mitochondrial membrane. Alteration in the function of the Bcl-2 protein can promote cancer development. Whereas in normal skin, Bcl-2-positive basal cells are a reservoir for squamous epithelial regeneration, and in sun-exposed skin, Bcl-2 prevents UV-induced cell death (Hussein and Ahmed, 2022; Onder et al., 2019). Increased expression of Bcl-2 has previously been observed in AK and Bcl-2-expressing tumour cells are significantly more numerous in SCC compared to asymptomatic AK (Berhane et al., 2002; Woo et al., 2017). Furthermore, Bcl-2 can stain dermal inflammatory cell infiltrates and inflammation is associated with a progression of AK to malignant SCC through the production of reactive oxygen species, the promotion of further immune responses, cell transformation, survival, proliferation, invasion, angiogenesis, and metastasis (Farshchian et al., 2017; Singh et al., 2019; Woo et al., 2017). Furthermore, a stepwise gradation of Bcl-2 expression has been shown from asymptomatic AK, through inflamed AK to SCC, consistent with progression through inflamed AK (Berhane et al., 2002). Our results showed that both the Bcl-2 subepidermal infiltrate and CD31 (the most sensitive and specific endothelial marker in paraffin sections (Stuart, 2013)) are increased in the AK/IEC sample compared to controls. This finding is consistent with the previously discussed role of inflammation in the pathogenesis of AK progression to SCC.

As a strength of our clinical study, we would like to mention that this is one of the few studies where all biopsies were taken from face.

Our patient sample was predominantly female. Although AKs are usually diagnosed more often in male patients, and considering the prevalence of baldness, males have larger sun-exposed areas, other studies have also had a higher female prevalence (Kohl et al., 2017; Lee et al., 2019; Madani et al., 2021). The female predominance in our sample could be explained by the average life expectancy in Latvia. It reached 68.2 years for men and 77.9 years for women in year 2021, reminding that the mean age of the included patients was 78.1 years (Central Statistics Bureau of Latvia, 2021).

Further analysis of the obtained histopathological and immunohistochemical marker changes in relation to the dermatoscopic features revealed that dermatoscopically typical AKs more often had amorphous masses and a lower expression of the CD31 marker. The latter indicates a lower density of blood capillaries. In addition, subepidermal expression of Bcl-2 was associated with follicular loss. These preliminary findings highlight blood capillary changes and follicular loss as essential dermatoscopical steps in the progression of AK to SCC.

Dermatoscope is an easy-to-use handheld device, that provides magnification and removes reflections from the skin surface to visualise morphological structures invisible to the naked eye (Pan et al., 2008). It has been shown to improve the diagnostic accuracy of both pigmented and non-pigmented skin lesions, sometimes providing relevant prognostic information through known dermoscopic-histopathologic correlations (Sinz et al., 2017). Common dermoscopic signs of AK are white-to-yellow surface scale, red pseudonetwork, which is formed by perifollicular erythema often combined with linear-wavy telangiectasia, targetoid-like hair follicles which are formed by yellowish keratotic plugs filling and a white halo surrounding the hair follicles, and rosette sign (Lee et al., 2014; Zalaudek et al., 2006).

Within this Thesis the correlation between the dermatoscopic characteristics of individual lesions and their histological and immunohistochemical changes, as well as the evolution of these dermatoscopic features throughout the treatment process were studied. In the latter part, clinical and dermatoscopic images were captured using digital dermatoscopy at three distinct stages: prior to treatment, during the inflammatory phase induced by the treatment, and post-treatment. Patients were categorised into two groups based on the prescribed treatment modality: either topical application of 5 % 5-fluorouracil cream or daylight photodynamic therapy using methylaminolevulinate (brand name Metvix®, produced by Galderma). The study revealed that shiny white streaks (SWS) were more prevalent than previously reported in the literature (Balagula et al., 2012; Liebman et al., 2012) and that they are variable dermatoscopic structures that may disappear during therapy-induced inflammation and reappear after therapy. SWS, also known as chrysalis or crystalline structures, are white, perpendicular lines a few millimetres long that are only visible in a polarised light dermoscopy (Kittler et al., 2016). These structures, considered as a dermoscopic sign of dermal fibrosis, are caused by polarisation of thickened hyaline fibrous bundles (Haspeslagh et al., 2016; Pizzichetta et al., 2014) and have been reported in a variety of skin lesions, including AK (Balagula et al., 2012; Liebman et al., 2012). In our case series, SWS were observed in 47 % of AKs before starting therapy. In previously published studies, the incidence of these structures was observed in 1.7 %–29.4 % of AK (Balagula et al., 2012; Liebman et al., 2012). This observation could be explained by the selection of AK lesions, as lesions with scales that could possibly hide SWS, were excluded from our study. Noteworthy, scale is a common feature of AK, with a prevalence of 79.4-85 % (Lee et al., 2014; Zalaudek et al., 2006). In addition, scales, crusts and erosions commonly develop with topical treatment due to destruction of atypical keratinocytes, and such

lesions were also excluded. Another reason for high prevalence of SWS in our study was that even a small number of SWS was considered a positive feature and the FotoFinder Systems medicam 1000 device offers higher magnification and resolution in comparison with handheld devices.

In our study, despite the small sample size, it was possible to determine several possible scenarios of treatment impact on SWS. First, although successful therapy is usually associated with the disappearance of dermoscopic signs, SWS can remain present throughout all treatment stages or even appear at a 1-month post-treatment visit without other dermoscopic signs of AK. As SWS is not a required feature for AK, permanence of SWS is not a counter-condition to treatment success. Second, as other dermoscopic structures, SWS can disappear during or after therapy and finally, SWS can temporarily disappear during treatment induced inflammation and reappear afterwards. This last observation was present in 13 % of the lesions analysed, and although the exact reason for this phenomenon is not yet clear, it leads us to speculate that SWS might also be hidden in other clinically clearly erythematous lesions, not limited to AK.

The final part of the work investigated the prevalence of severe local site reactions (LSRs) using topical field treatment modalities – daylight photodynamic therapy, 5 % imiquimod cream, 3.75 % imiquimod cream, 0.015 % ingenol mebutate gel, 3 % diclofenac gel with 2.5 % hyaluronic acid, 0.5 % fluorouracil gel, and 0.5 % fluorouracil and 10 % salicylic acid combined gel. To achieve this objective, a systematic search of articles was conducted using the PubMed database, focusing on the incidence of severe reactions at the application site in relation to the medication used and the discontinuation rates of therapy due to these reactions. Studies that required the use of specialised equipment, such as conventional photodynamic therapy, were excluded from consideration. Consequently, data from 19 studies were aggregated,

demonstrating that the prevalence of severe reactions at the application site varied widely, ranging from 0 % to 58.5 %. Severe reactions were rarely observed with daylight photodynamic therapy, whereas they were most reported with imiquimod treatment. Additionally, the exclusive use of imiquimod was associated with the development of systemic symptoms. An additional set of 14 articles reported on the discontinuation of treatment due to application site reactions. The highest discontinuation rates, ranging from 4.9 % to 13.6 %, were during treatment with 3 % diclofenac, which is typically prescribed for twice-daily application over a period of 3-6 months. In studies involving 5 % imiquimod, treatment cessation occurred in up to 3.2 % of cases, a lower rate that was attributed to thorough patient education about potential reactions at the application site. Furthermore, non-treatment related risk factors for more severe reactions at the application site were identified, including lighter skin colour (phototype I–II), being female, being under 70 years of age, and higher ambient temperatures during daylight photodynamic therapy (Fargnoli et al., 2015; Galvão et al., 2017; Ortega del Olmo and Salido-Vallejo, 2018; Ricci et al., 2016).

Conclusions

1 Based on findings from published prospective studies, invasive squamous cell carcinomas most frequently originate from actinic keratoses that are either longstanding, large, or merging, particularly in high risk populations. Consequently, these clinical manifestations can be regarded as risk factors for an increased likelihood of developing squamous cell carcinoma.

2 The most frequent dermatoscopic signs characteristic of facial non-pigmented actinic keratoses are background erythema, white follicular openings, surface scales, rosettes, fine, linear, wavy vessels, microerosions, and sun-damaged surrounding skin. In comparison, facial intraepidermal carcinomas have additional features such as red starburst pattern, centrally located scales or keratin, dotted or glomerular vessels, hairpin vessels and ulcerations.

3 The prevalence of amorphous masses, the intensity of p53 staining, the expression of Bcl-2 and CD31 are different in clinically healthy sun-damaged skin compared to skin samples in which actinic keratoses and intraepidermal carcinomas have developed.

4 Dermatoscopically typical actinic keratoses more commonly show amorphous masses in histopathology and have lower expression of CD31. Absence of follicles was associated with increased Bcl-2 subepidermal expression.

5 Shiny white streaks seem to be variable dermatoscopic structures that can be unseen in lesions with therapy-induced inflammation, disappear following topical treatment and sometimes appear for the first time after treatment. It could be important to take into consideration the dynamics of shiny white streaks when assessing their presence.

6 Treatment of actinic keratoses identifies treatment-resistant lesions which retain their dermoscopic features and allows better clinical and dermatoscopic visualisation of the underlying skin, thus further promoting early diagnosis of skin cancer.

7 Local site reactions of severe intensity seem to be extremely common with topical therapies for AK, especially with imiquimod. The only therapeutic modality with a low prevalence of severe local site reactions is daylight photodynamic therapy. Treatment discontinuation due to local site reactions is also common, although the highest prevalence of treatment discontinuation due to local site reactions is reported in studies with the longest treatment regimens, such as diclofenac, and not in studies reporting the highest prevalence rates of severe local site reactions.

Proposals

1 If it is not possible to treat all actinic keratoses at the same time, priority should be given to long-standing and large or confluent actinic keratoses as they may have a higher risk of progression to invasive squamous cell carcinomas.

2 Dermatoscopy should be used in evaluation of all actinic keratoses before and after therapy.

3 All actinic keratoses with any atypical dermatoscopic signs warrant biopsy to exclude intraepidermal carcinoma or early invasive squamous cell carcinoma.

4 Careful explanation of expected application site reactions should be given to all patients as it reduces the treatment discontinuation rate.

5 Further research on daylight photodynamic therapy for actinic keratosis in Latvia is recommended, as in the context of this particular therapy, the medication is administered in a single application and it is associated with a relatively rare occurrence of severe local site reactions.

List of publications and reports on the topic of the Thesis

Publications:

1. Balcere, A., Sperga, M., Čēma, I., Lauskis, G., Zolovs, M., Rone-Kupfere, M., Krūmiņa, A. 2023. Expression of p53, p63, p16, Ki67, Cyclin D, Bcl-2, and CD31 Markers in Actinic Keratosis, In Situ Squamous Cell Carcinoma and Normal Sun-Exposed Skin of Elderly Patients. J. Clin. Med. 2023, 12, 7291. doi: 10.3390/jcm12237291.

2. Balcere, A., Konrāde-Jilmaza, L., Pauliņa, L. A., Čēma, I., Krūmiņa, A. 2022. Clinical Characteristics of Actinic Keratosis Associated with the Risk of Progression to Invasive Squamous Cell Carcinoma: A Systematic Review. J Clin Med. 2022 Oct 6;11(19):5899. doi: 10.3390/jcm11195899.

3. Balcere, A. 2021. Dermatoscopy of Facial Non-Pigmented Actinic Keratosis and Intraepidermal Carcinoma. In: Pietkiewicz, P., editor. Dermatoscopy [Internet]. London: IntechOpen; Available: https://www.intechopen.com/chapters/77640. doi: 10.5772/intechopen.98875.

4. Balcere, A., Ozola, E., Karls, R., Čēma, I., Rone-Kupfere, M., Krūmiņa, A. 2020., Dermoscopic monitoring of shiny white streaks during topical treatment of actinic keratosis. Proceedings of the 62nd International Scientific Conference of Daugavpils University, 139-143. Available: https://dukonference.lv/files/978-9984-14-925-7_62_konf_kraj_A_Dabaszin.pdf.

5. Balcere, A., Rone-Kupfere, M., Čēma, I., Krūmiņa, A. 2019. Prevalence, Discontinuation Rate, and Risk Factors for Severe Local Site Reactions with Topical Field Treatment Options for Actinic Keratosis of the Face and Scalp. Medicina (Kaunas). 2019 Apr 4;55(4):92. doi: 10.3390/medicina55040092.

6. Balcere, A., Karls, R., Čēma, I., Rone-Kupfere, M., Vīksna, L., Krūmiņa, A. 2019. Treatment of Actinic Keratoses Facilitates Dermatoscopic Diagnosis of Early Basal Cell Carcinoma: A Case Report and Review. Case Reports in Dermatology. 2019. 11 (1), 16-22; doi: 10.1159/000496329.

7. Balcere, A., Karls, R. 2016. The prevalence of actinic keratosis and basal cell carcinoma and their association with UV radiation in the elderly Latvian population. Abstracts from the 16th World Congress on Cancers of the Skin 2016. Melanoma Research. 2016, 26 e-Supplement 1:e61–e62.

8. Balcere, A., Karls, R., Zabludovska, K. 2015. The Prevalence of Actinic Keratoses and Its Associated Risk Factors in Elderly Latvian People. Collection of Scientific Papers: Research articles in medicine & pharmacy, 2015: Internal Medicine. Surgery. Medical Basic Sciences. Stomatology. Pharmacy. Riga Stradiņš University, 2015; pp:29-34.

Reports and theses at international congresses and conferences:

1. Balcere, A., Sperga, M., Čēma, I., Rone-Kupfere, M., Krūmiņa, A. Presence of amorphous masses is more common in dermatoscopically classical actinic keratosis than more advanced lesions. 2023. 32nd European Academy of Dermatology and Venereology Congress. Berlin, Germany, 11.-14. October, 2023. (Poster presentation).

2. Balcere, A., Sperga, M., Čēma, I., Lauskis, G., Zolovs, M., Rone-Kupfere, M., Krūmiņa, A. 2023. Dermatoscopic and histopathologic correlations of facial keratinocyte neoplasia. 19th Congress of Baltic Association of Dermatovenerologists. Riga, Latvia, 14.-16. September 2023. (Oral presentation).

3. Balcere, A., Sperga, M., Čēma, I., Rone-Kupfere, M., Krūmiņa, A. 2022. Šūnas cikla regulējošo marķieru un CD31 marķiera ekspresija klīniski veselā pusmūža un gados vecāku pacientu sejas ādā. 9th Latvian Congress of Physicians, Latvian Medical Association, Riga, Latvia, 22.-24. September 2022. (Poster presentation).

4. Balcere, A., Čēma, I., Ščerbuks, M., Rone-Kupfere, M., Krūmiņa, A. 2022. Multiple forms of squamous cell carcinoma in a patient with a field cancerisation. A case report. 18th Congress of Baltic Association of Dermatovenerologists. Riga, Latvia, 22.-24. September 2022. (Oral presentation).

5. Balcere, A., Konrāde-Jilmaza, L., Pauliņa, L. A., Čēma, I., Rone-Kupfere, M., Krūmiņa, A. 2022. Kuras aktīniskās keratozes progresē par plakanšūnu karcinomām? International Summer School "Dermatology Without Borders". Liepaja, Latvia, 29.-30. July 2022. (Oral presentation).

6. Erta, A., Balcere, A. 2022. Knowledge about ultraviolet radiation and tanning behaviour among university students: comparison of healthcare and non-healthcare related study programs. International Student Conference 2022, Rīgas Stradiņa universitāte, Riga, Latvia, 24.-25. March 2022. (Oral presentation).

7. Balcere, A., Sperga, M., Ščerbuks M., Čēma, I., Rone-Kupfere M., Krūmiņa, A. 2021. Clinical, dermoscopic, and morphological correlations of actinic keratosis. European Academy of Dermatology and Venereology Spring Symposium 2021. Online, 06.–07. May 2021. (Poster presentation).

8. Konrade-Jilmaza, L., Balcere, A. 2021. Which are the most effective educational skin cancer prevention programs to induce behavioural change in children? European Academy of Dermatology and Venereology Spring Symposium 2021. Online, 06.–07. May 2021. (Poster presentation).

9. Balcere, A., Sperga, M., Čēma, I., Rone-Kupfere, M., Krūmiņa, A. 2021. Immunohistochemical marker correlation with dermoscopic findings in actinic keratosis and squamous cell carcinoma in situ. Virtual 10th World Congress of Melanoma in conjunction with the 17th European Association of Dermato Oncology (EADO) Congress. Online, 15-17. April 2021. (Poster presentation).

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