CRAIOVA UNIVERSITY OF MEDICINE AND PHARMACY

DOCTORAL SCHOOL

THE STUDY OF THE CLINICAL FEATURES AND EVOLUTION OF THE BASAL CELL CARCINOMA IN RELATION TO THE ETIOLOGICAL DETERMINANTS AND/OR FAVORABLE FACTORS

THE SUMMARY OF THE DOCTORAL THESIS

SCIENTIFIC LEADER:

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Motto:

“NE REVEILLE PAS UN LION ENDORMI”

L'épithéliome basocellulair est un lion qui dorm.

CONTENT

I. Introduction; Keywords

The skin, located at the border between the body and the environment, is the first organ to suffer consequences.

Skin cancers have a rapid growth, primarily due to a change in society's attitude towards tanned skin. If 100 years ago people considered the white skin, untouched by sun, a social and aesthetic asset, nowadays tan is highly appreciated, being regarded as a sign of health or rather of beauty. The sun, our best ally, can cause the premature aging of the skin, burns or cancers, if the exposures are prolonged, during the wrong time slot (between 11- 16 during the summer months).

Romania, according to the data provided by the National Meteorological Administration, has an annual number of 160-180 clear days, with a sun brightness duration of 2100-2200 hours (the sun shines 10-11 hours during the summer months). Dobrogea and Romanian Plain are the sunniest areas. In these regions, activities that involve prolonged sun exposure (farmers, fishermen, navigator, etc.) persist.

From the data published we find that over 80% of skin cancers are represented by basal cell carcinoma, which is why we decided to choose it as a research topic for the present paper.

Described for the first time by Jacob in 1827, this type of cancer remains permanently current, due to the clinical polymorphism, the histological structure with a very varied architecture and its different evolution, from one case to another.

So, as it is, it is easy to understand why another study of the clinical and evolutionary features of these tumors was needed.

To summarize in a few key words, I will list: morphoclinical variability, immunohistochemical polymorphism, customized therapy.

I express my gratitude to Mr. Doctor Professor, PhD Ion Ţolea, the scientific coordinator of this paper, for the valuable guidance give during the doctoral studies, for the trust he has give me from the beginning. He is, for me, a model of conduct, perseverance, much needed in scientific research work.

I thank to all the people with whom I collaborated in the laborious activity which formed the scope of the present research.

II. Objectives

Basal cell carcinoma aroused my interest through complex issues, due to the high degree of morphoclinic validity, associated with so many clinical and therapeutic features.

After studying the existing data at the present moment, in international literature and from us in the country, I support the idea of a mixed pathogenesis of the basal cell carcinoma , genetic and environmental. This affects the morphoclinical aspects, and implicitly on therapeutic means and methods.

Carcinogenesis is a multistage process by which a cell develops the malignant phenotype, as a result of the interaction between endogenous factors (age, genetic predisposition, hormonal disorders) and exogenous factors (chemical, physical, biological).

The mechanism by which ultraviolet (UV) radiations cause skin cancer is direct (photoreaction process) and indirect, of destroying Langerhans cells. UVB action over the keratinocytes causes the formation of free arachidonic acid and prostaglandins. Prostaglandins are the tumor promoters or amplify the starting of the tumor process. The effect of UV radiation over the skin is irreversible, induces, promotes and sustains carcinogenesis.

Sun exposures play an essential role in the etiology of basal cell carcinoma, although the latency period of the carcinogenesis process lasts years from the time of exposure until the tumor emerges. UV rays act directly over the epidermal DNA, causing definitive chain damage, with mutations occurring in specific protein genes involved in BCC carcinogenesis (TP53, PTCH1, SHH, SMO). Through the generated photo-oxidases, UV acts indirectly over the DNA, but the result is the same. UVBs induce on the keratocyte level the Fas ligand (FasL or CD95L) and trigger an antiapoptotic response through the phosphatidylinositol 3-kinase-Akt pathway. UV doses influence the characteristics of basal cell carcinomas. Low doses of UV, but with high frequency, increase the ratio of basal cell carcinoma / spinocellular carcinoma in favor of BCC. Chronic exposure to UV determines the nodular shape, and intermittent exposure determines the superficial form of BCC.

Ionizing radiations generate electrons, causing direct / indirect DNA damages.

Smoking, HIV / AIDS and immunosuppressive therapies used in people with organ transplants or with other medical conditions, by diminishing the body's defense capacity, predispose to the development of basal cell carcinomas.

People (agricultural workers, seamen, fishermen, mechanizers, gardeners, builders) who, by the nature of their job, are required to work outdoors have an increased risk of developing BCC.

People with phototype I and II have a increased incidence of basal cell carcinoma, due to the absence of the protective effect of melanin.

In the context of the above, the objective set at the beginning of this work was to identify the existence or not of some correlations/ particularities between the etiological factors and the clinical aspect, the consequences on the architecture of the histological structure, as well as the evolutionary behavior of the lesion in relation to the therapeutic method.

III. Material and Method

To highlight the clinical features and evolution in relation to the etiological factors, , I conducted a retrospective study, trying to highlight the existence or not of several clinical and evolutionary features of the basal cell carcinoma related to the etiological factors. I used the data achieved over a period of 5 years (1.01.2014-31.12.2018), comprising a number of 299 patients who came to the Dermatology Clinic and to Constanta Specialty Outpatient Unit, diagnosed with basal cell carcinoma, who presented one or more (epitheliomatosis) tumor formations, confirmed histopathologically.

For each case I prepared an individual sheet, which included personal anamnestic data (age, sex, profession), pathological and heredocolateral history, living conditions, the history of carcinoma development (the onset form, the evolution until the specialty examination), the histopathological result and the immunohistochemical result (if indicated). All patients consented to the participation and processing of personal data, observing the principles of the declaration from Helsinki, signing an informed consent form.

Data statistical processing aimed at: the clinical-statistical study of the territorial distribution, of the distribution by age, sex, profession and the evolution of the tumor formation.

The histopathological examination included the following standard steps: macroscopic examination, sampling, histoprocesarea (formal fixation 10%, paraffin inclusion, sectioning of paraffin blocks, fixing on blades, standard hematoxylin- eosin staining) and microscopic examination of de blades. In most cases 1-2 sections were processed for establishing the diagnosis of basal cell carcinoma. In the case of intricate, pigmented forms, serial section were examined.

For each pacient an individual record was prepared, which included in addition to personal data, indicators followed in study:

a. The classification of tumors into histological subtypes according to the tumor growth pattern, depending on the predominant aspect of the tumor;
b. The histological aspect compared to the clinical aspect;
c. The invasion degree of the neighboring structures.

From the desire to identify in more detail the basal cell carcinoma behavior in antibody action I carried out a special research in 10 cases, chosen randomly from the total number studied, comprising different histological types.

Tissue blocks, with confirmed cases of basal cell carcinoma, were randomly selected from the Archive of the Pathological Anatomy Section of Constanța County Emergency Hospital, between 2017-2018. The case study included tumors of the types with tricoblastic differentiation (2 cases), solid (3 cases), collisional solid + infiltrative (1 case) and superficial multicentric (4 cases). Skin fragments, sampled by skin biopsy, were rapidly fixed in 10% formalin (for 5-10 minutes). The collected tissue cups were processed in paraffin and processed automatically (DIAPATH Platform). Sections of 5 microns were practiced, which were stained by conventional techniques with usable kits (hematoxylin-eosin, progressive method, DIAPATH). After reconfirming the diagnosis, the immunohistochemical tests were performed by the polymer method, with HRP / DAB detection and viewing system, on the semi-automatic BIOCARE platform, with the Bcl2, Ki67 and CD3 antibodies, in strict compliance with the protocols. The intensity of the immunohistochemical reaction was noted as it follows: weak +, moderate ++, intense +++. For Bcl2 we used as positive internal control the active B lymphocytes, for Ki67 the basal keratinocytes, and for CD3 the external control was the palatine amygdala.

Images with tumor areas and the adjacent epidermis were photographed with a LEICA ICCHD 50HD camera with a LEICA DM 750 microscope, using the LAS V4.6 software, on a LEICA DM 750 microscope, under conditions of constant brightness and contrast.

IV. Results and Discussions

According to the data achieved following the statistical processing, the ratio between genders is relatively equal, existing a slight predominance of the male gender (153 patients). Gender ratio M / W = 1.04.

Cases distribution per year does not show an increase in the number of cases, there being insignificant variations (10-20 cases plus or minus) from year to year, mentioning that these data were collected only from the information available in the national health network.

Basal cell carcinoma was most commonly diagnosed on people of the group 70-79 years old (87 patients in 299 cases) representing about 1/3 (29.10%). The tumor prefers the third age, when histopathological changes can be discussed within the context of the degeneration process of the cutaneous organ (prolonged and repeated exposure to the sun, phototype, life habits).

Patients from rural areas predominate in this study. These data can be explained from the point of view of occupations that require a prolonged exposure to the sun (farmers, gardeners, mechanizers, etc.).

The overwhelming percentage of 76.89% of cases with face and neck location, this underlines the decisive significance of UV exposure in the development of skin cancer.

In most cases the time run from onset to coming to the physician is 1-3 years.

The pearly form was the most frequent (98 cases out of 293), followed closely by the nodular form (29%). The pearly form was diagnosed, most commonly, in the age group 50-59 years (25 cases out of 98 pearly shaped carcinomas).

The forms of non-infiltrative evolution predominated in our study.

In our study the nodular form was more frequent in the age group 70-79 years (31 cases out of 86). The superficial form predominated in the age group 40-49 years (9 cases out of 28). The distribution of the superficial shape according to the age groups is almost uniform between 40-79 years. We could not compare our results with those of the literature, because we did not find studies conducted.

In case of the infiltrative form we obtained a maximum at the age range 60-69 years (7 cases out of 23), and the micronodular form presented a maximum at 70-79 years (4 cases out of 9) followed closely by the age of 50-59 years (2 cases out of 9).

We correlated the data of the clinical forms with the age groups, in order to observe, if there is a connection between the clinical aspect and the body defense (considering that aging is synonymous with the defense degradation). Aggressive forms predominated in the 6th and 7th decades, and pearly and superficial forms were more frequent in the 4th and 5th decades. We cannot a sustain a connection of the clinical aspect with the immunological status of the body, because the nodular form (considered a less aggresive form) has a peak on the chart at the age range 70-79 years.

Out of a total of 297 cases, the solid form was predominant (111 cases), without a significant gender difference.

The solid subtype, in turn, presented purely solid forms and forms with adenoid areas (8 cases), cystic areas (7 cases), keratinization areas (2 cases). Our group did not include solid

forms with pigmentation or sclerodermiform areas. The invasive character was present in 15 cases.

Differentiating carcinomas were more frequently diagnosed in males (over 2/3 of cases), without being able to give an explanation.

Of the group studied, 68% (59 patients) had infiltrative character, of which 37 cases were on males (63%).

The cytoplasmic and / or nuclear reaction to Bcl2 was variable, BCC infiltrating and superficial having a cytoplasmic reaction with maximum intensity.

- The components of the collision type reacted differently, the solid component having a low intensity (60%), compared to the infiltrative component (intensely positive), as in the pure solid forms. In one case of BCC with tricoblastic differentiation we obtained a moderately positive to intensely positive reaction (80%), and in the other case a negative reaction, however the internal control was unconvincing. The superficial multicentric forms reacted to an intense positive proportion of 80%.

- The reaction to Ki67 was generally intensely positive in variable proportion (10-40%), the maximum value being observed in the solid-infiltrative collisional form. Inside the non-infiltrating solid tumors, a depressive reaction to Ki67 was observed.

-Reaction to CD3 was intensely positive, of brisk type in the case of the 35-year-old patients and non-brisk type in the case of patients over 50 years old.

V. Final conclusions

1. Increase in the number of cancer patients worldwide, despite the prevention rules implemented lately, is a real challenge, both for medical researchers, and especially for clinicians.

2. The malignant skin lesions being exposed on sight, with the possibility of detection even from the onset, with slow evolution, sometimes misleading, as well as their occurrence, in more than 50% of cases, on other skin lesions, called precancerous, led to doubling, or even tripling the number of those who go to the physician for the suspicion of a malignant process.

3. Of the malignant skin tumors, the most frequent ones are epithelioomas, accounting for 90-95% of the total, but also the least serious ones.

4. In this paper I tried to identify the existence, or not, of certain correlations between the clinical and architectural aspect of the histological structure, as well as the evolving behavior of the skin lesion in relation to the optional therapeutic method.

5. As regards the etiopathogenesis of skin cancer, exposure to ultraviolet radiations is the most important triggering factor, due to their cumulative risk. Thus, excessive exposure, since childhood, to ultraviolet radiations, explains the emergence of skin tumors in young people.

6. Our study showed us that basal cell carcinomas occur in both genders, at any age, with a maximum incidence between 70-79 years, confirming the theory that the changes undergone by the skin organ, once with passing of the years, also represents a significant etiopathogenic factor.

7. The case study studied shows a very small difference between the number of patients from the urban and rural areas, so we cannot afford to sustain the idea of the low incidence in the urban area. The difference of 7.68% is not true, due to the small group of patients included in the study.

8. The preferred location of BCC in the cases studied was at the level of the cephalic extremity, the nasal pyramid having the weight, followed closely by the cheek area and the internal angle of the eye.

9. Basal cell carcinoma has an atypical, deceptive onset, which is why the patient goes late to the physician. The characteristic elements of this tumor appear, according to our study, after 1-3 years.

10. The basocellular epithelioma presents a clinical polymorphism, from superficial forms to invading, destructive forms, the clinician having difficulties in issuing the prognosis and especially in establishing the therapeutic conduct. Superficial forms, often neglected, can occur at any age. The pearly form was more frequent in the age group 50-59 years, the nodular form at 70-79 years, and the invading forms had a significant increase after the fifth decade, the infiltrative form with a maximum between 60-69 years.

11. Often, the clinical aspect does not correspond to the histological aspect, in the same tumor there may be several histological types, the behavior being different, which is why the therapeutic approach must be differentiated, depending on the case particularity.

12. Ulceration and pigmentation are a sign of a long evolution of the tumor. However, pigmentation is not a common feature, only 11% of the cases studied presented it, but ulceration was more common, 20% of cases, especially in males.

13. As regards the histological aspect of BCC, polymorphism is also present, defining remaining the basaloid cell.

14. The histopathological examination is mandatory for establishing the certain diagnosis. There are situations where further investigations, such as immunohistochemical studies, are required, in our study being used only in two cases.

15. In order to establish certain specificities regarding the sensitivity of cellular reactions, we undertook a randomized 10-cases study. The cytoplasmic and / or nuclear reaction is variable on Bcl-2, with maximum in case of infiltrative and superficial forms and a weak reaction in the solid forms. The reaction to the KI67 marker is nonspecific, and the use of CD3 in our study helped us to highlight the presence of the peritumoral inflammatory infiltrate in all cases, but the distribution was different. This would explain the different evolution of the same histological form.

16. In the basal cell carcinoma, there have not been identified enough data to convince us of the presence of a certain reaction of the immune system.

17. An almost unpredictable evolution of BCC has led to the development of a wide range of therapeutic methods, all aimed at destroying the tumor, but none is perfect. Electrocautery remains the most used method, although surgical excision is considered the standard method. Local chemotherapy is the therapeutic approach in patients who have comorbidities that contravene to the other methods.

18. Regardless of the method used, the recurrence rate is present. In our study, electrocautery and chemotherapy with imiquimod 5% had the lowest recurrence rates on the number of patients studied

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