SOLAR RADIATION AND SKIN CANCER RISK – BIOPHYSICAL INSIGHTS

EMANUELA MICU

Dermatology Department, Humanitas Clinic, Bucharest, Romania, e-mail: ema.cicarma@gmail.com

Abstract. Solar radiation is the main environmental risk factor for skin cancer, whose incidence is rapidly increasing in the latest decades. Individuals carry different skin cancer risk, depending mainly on the skin color (melanin type and distribution) and therefore Fitzpatrick skin typing is a widely used method to predict skin cancer risk. However, this method has its limitations as other factors are involved in skin cancer susceptibility, like genetic factors (MC1R variants) or DNA damage repair mechanisms. Thus, there is an urgent need for implementing objective, non-invasive methods to assess skin sensitivity and reactivity to sun exposure. In this review, I will analyze the main factors involved in photoprotection and the new devices currently employed to evaluate the response to ultraviolet radiation (in terms of erythema and pigmentation) and thus to predict skin cancer risk.

Key words: skin cancer, ultraviolet radiation, melanin, reflectance spectrophotometer.

INTRODUCTION

Humans spend about half of their lives exposed to the sun. However, only in the beginning of the 20th century, carcinogenetic effects of sunlight on skin (photocarcinogenesis) started to be recognized [3]. Currently, sun exposure is the only known readily modifiable risk factor for skin cancer [27, 34, 36]. Sun-seeking behavior observed in the latest decades, along with an increased surveillance of pigmented lesions, are considered to be responsible for the rapid increase in skin cancer incidence [26].

On the other side, light has been used in photomedicine since ancient times and sun exposure is the main source for vitamin D in humans, with a long list of beneficial effects for human health [19]. As a consequence, the nowadays dilemma is: can we expose ourselves to the sun in order to photosynthesize vitamin D in the skin and without increased risk of skin cancer? Moreover, not all individuals respond in the same way when exposed to the sun. Thus, there is a need for a reliable tool that can assess skin sensitivity and reactivity to the sun, in other

Received: November 2014;
in final form December 2014.

ROMANIAN J. BIOPHYS., Vol. 24, No. 4, P. 269–280, BUCHAREST, 2014

words, skin cancer risk. This will help elaborate optimal screening programs and prevention campaigns in order to reduce the burden of skin cancer.

The aim of this review is to provide a concise overview of some of the main biophysical aspects concerning sun exposure and skin cancer risk, from the solar radiation wavelengths considered responsible for skin cancer and melanin role in photoprotection to biophysical methods used to assess skin sensitivity/skin cancer risk.

ACTION SPECTRA FOR PHOTOCARCINOGENESIS

The solar radiation that reaches the earth surface is composed of different wavelengths of electromagnetic radiation and it is divided into three main regions: ultraviolet (UV) region (100–400 nm), visible region (400–760 nm) and infrared region (>760 nm). UV region is further divided into UVC (100–280 nm), UVB (280–315 nm) and UVA (315–400 nm). The limit between UVB and UVA is not firmly established; in photodermatology, 320 nm is used. Most of the effects of sun exposure on the skin are due to wavelength in the range of 300–400 nm, although for some rare disorders like solar urticaria, visible radiation is also responsible. In cutaneous carcinogenesis, the effects of UV radiation (UVR) in causing DNA damage and immunosuppression are crucial [15]. However, the exact action spectrum (AS) for melanoma, the most dreadful form of skin cancer, is not known. In this case, substitutes are used, like erythema AS and immunosuppression AS, as skin erythema and local and systemic immunosuppression are among the main effects of acute UVR exposure [28].

Erythema (redness), an acute inflammatory reaction to the sun, is mainly what people consider when speak about sun sensitivity. The erythema AS taken from the work of Anders *et al.* [1] has a maximum peak in the UVB region (around 300 nm), like the standard erythema AS from the Commission Internationale de l'Eclairage (CIE) [6], but also a distinct maximum in the UVA region (around 360 nm). The chromophore (the molecule that absorbs the light and whose photochemical alteration causes the effect) for erythema is considered to be DNA, with the induction of cyclobutane pyrimidine dimers (CPDs) [49]. DNA is a strong absorber of UVB and a weak one for UVA [42].

The AS for immunosuppression taken from Damian *et al.* [9] shows a peak around 310 nm and a smaller peak at 370 nm (long-wave UVA) and it is likely that in this case, several chromophores may be involved: DNA, *cis*-urocanic acid, tryptophan and reactive oxygen species (ROS) for UVB, and different unknown chromophores (porphyrins have been proposed) for UVA that absorb in the 360–380 nm region and that lead to oxidative stress and consequently to activation of the alternative complement pathway [16]. Another mediator of UVA immunosuppression may be nitric oxide that interacts with ROS [23].

Fig. 1 shows AS for erythema and immunosuppression convoluted with standard solar spectrum (effective irradiances), in order to have a better understanding of the AS that are important in skin cancer.

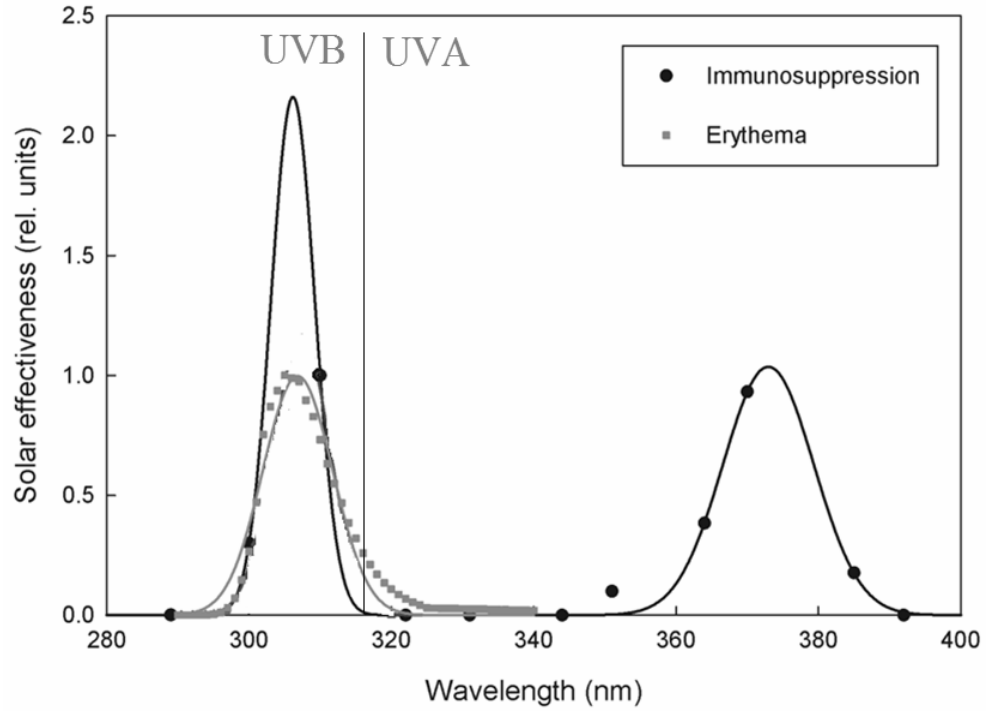

Fig. 1. Efficiency spectra for erythema and immunosuppression. The action spectra data are taken from [6] and [9] and convoluted with standard solar spectra, kindly provided by Mantas Grigalavicius, The Norwegian Radium Hospital, Oslo, Norway.

More robust evidence for skin cancer AS comes from mice experiments where UVB is shown to induce skin cancer in the absence of any other carcinogenetic agent by direct DNA damage, while the role of UVA is indirect, mainly by oxidative damage, involving melanin [10, 31]. Recently, Noonan *et al.* [31] put into discussion an UV-independent mechanism related to a dysregulation in melanin synthesis (endogenous oxidation). This has relevance for our discussion, to underline the role of melanin (meaning in general terms pigmentation) for skin cancer risk and to underline the complex network involved in skin cancer.

As a conclusion, erythema (sun sensitivity) depends on the wavelength (mainly UVB), dose of radiation and melanin quality and quantity and it may predict DNA damage.

SKIN COLOR AND PHOTOPROTECTION

Melanin is a mixture of biopolymers, produced in melanocytes (dendritic cells that in the skin are located in hair follicles and at the dermal-epidermal interface) from tyrosine. Its main function is to absorb UVR and to protect the skin against DNA damage. There are two broad classes of melanin: eumelanin which is brown/black and pheomelanin which is red/yellow [20]. It is mainly the eumelanin content that differs between different skin colors and offers more photoprotection than pheomelanin: it has higher resistance to degradation and quenches reactive oxygen species [5, 39, 44]. Melanin is packaged into melanosomes that have light-scattering and absorbing properties depending on its size and melanin content; they will protect the nucleus from the UVR.

Following UVR exposure, melanin content and melanosomes distribution increase in the upper layers of the skin and the skin becomes darker (a process commonly known as tanning) [43], thus protecting the lower layers like basal layer, where the highly dividing cells reside. It is important to underline that the effects on skin pigmentation differ between UVB and UVA: it is UVB that increases melanin synthesis, while UVA-induced tan is based on effects upon existing melanin (oxidation and distribution) [29]. A schematic representation of absorbing and scattering properties of the skin is shown in Fig.2. In people with lighter complexion which have lower amounts of eumelanin, more UV radiation penetrates through the epidermis and induces DNA damage. Several studies have shown a decreased DNA damage in darker skin types following UV exposure: the darker skin gets even darker upon UV exposure and becomes more photoprotective than lighter skin [11, 21]. Moreover, pheomelanin (of particular importance in red hair individuals), being more soluble than eumelanin [20], may leak out from the melanosomes, diffuse into the nucleus, and interact with DNA to promote mutagenesis especially in the context of UV radiation.

Melanogenesis is regulated via melanocortin 1 receptor (MC1R) of the α-melanocyte stimulating hormone (αMSH). The MC1R gene is highly polymorphic; loss-of-function variants are responsible for red hair and light skin phenotype (with poor tanning ability); these variants sensitize melanocytes to DNA damaging effects of UV radiation and confer a high risk for skin cancer [37]. This is supported by epidemiological studies and meta-analysis that show increased melanoma risk for certain MC1R variants [35]. Moreover, it is suggested that these variants may be directly linked to skin cancer, independently of skin color (direct MC1R non-pigment-related cell-signaling pathway) [32]. Even more, MC1R may influence the melanocyte ability to repair DNA damage following UV exposure [33, 38]. For a comprehensive review of the MC1R pathways the reader is referred to Nasti and Timares [30]. Thus, we cannot assume with certainty that an individual with darker skin, who does not burn easily is protected against deleterious effects of UVR, as he might be a carrier of one MC1R variant allele.

As a conclusion, photoprotection goes far beyond melanin/skin color. The key factor seems to be MC1R; however, other factors may be implicated, like immune system and DNA repair capacity. In clinical practice, this means that assessment of skin color is not enough to predict skin cancer risk. A more accurate approach should include evaluation of skin response after UV exposure, by objective methods that are able to quantify the reaction, ideally in relation with DNA damage, besides determination of pigmentary phenotype.

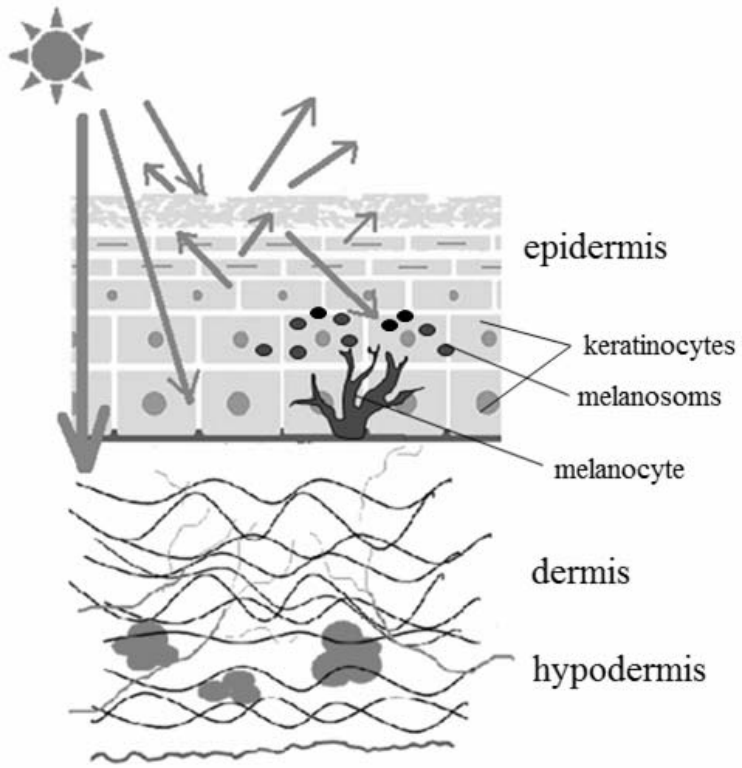

Fig. 2. Light-skin interaction. There are two fundamental light-tissue interactions: scattering and absorption (by chromophores, like melanin). The ticker the skin is, the more it scatters and absorbs light (electromagnetic radiation). However, the main photoprotective mechanism is melanin synthesis: following sun exposure, melanin synthesis and melanosomes distribution increase in the upper layers of the skin, protecting the keratinocytes nucleus.

METHODS TO ASSESS UVR SKIN REACTIVITY

How do we assess a person reactivity to sun exposure and, consequently, to skin cancer risk? Not all individuals carry the same risk; some of them will never develop skin cancer, while others are at high risk. As I exemplified above, MC1R

genotype seems to be a more reliable indicator than skin color or the answer to the question: Do you tan or burn when exposed to the sun? However, this is far from becoming a practical tool in clinical practice and we have to rely on other methods.

There are several methods used to quantify the skin response to sun exposure (both redness and tanning) in order to assess sun sensitivity and reactivity (Table 1). In the following, non-invasive methods currently employed or under investigation are detailed:

1. Fitzpatrick classification of the tendency of burn and tan [13]; it is a self-estimated UV sensitivity based on a questionnaire, with six possible answers (Table 2). It is widely used both in clinical practice and in clinical trials. Several epidemiological studies have found a correlation with skin cancer risk [14, 46].

2. Phototesting; this is performed in a clinic or a laboratory that has the means of UVR stimulating; it evaluates visually the tendency to burn upon exposure to a certain amount of UVR and minimal erythema dose (MED) is approximately calculated. This is mainly used for phototherapy and photo patch testing. Although several studies have found that MED correlates with Fitzpatrick skin type [2, 4], other studies could not find an accurate relation between erythema as quantified by MED and skin type [40, 47]. Thus, there is a need for more objective methods to quantify erythemal response after UV exposure.

3. Objective methods based on reflectance spectroscopy: scanning reflectance spectrophotometers (very expensive, used in fundamental research laboratories) and portable devices: narrow-band reflectance spectrophotometers (based on the difference in absorption between melanin (skin pigmentation) and hemoglobin (redness) at well-chosen wavelengths) and diffuse reflectance spectrophotometers.

4. Biomarkers of UV exposure (like DNA damage repair products) – projects in progress to be validated.

Table 1

Skin reactivity to ultraviolet radiation

Methods	**Parameters measured**	**Observations**
Questionnaires	**Fitzpatrick skin type** It evaluates erythema and induced pigmentation and classifies the responses into six skin types.	It is a subjective, anamnestic method.
Phototests UV* sources: Solar simulator, nUVB*, UVA	**Minimal Erythema Dose (MED)** It predicts the UV dose to elicit just perceptible erythema 24 hours after UV-exposure.	Erythema/pigmentation is graded visually according to a scale. It is widely used in phototherapy.
	Minimal Melanogenic Dose (MMD) It predicts the UV dose to elicit pigmentation 7 days after UV-exposure.	

	Pigment Protection Factor (PPF) It predicts the UV dose to produce 1 MED.	The equations for calculation of redness percent and pigmentation percent are built into the instrument.
Tristimulus colourimeters	**Skin colour** CIE L*a*b* values; colour is expressed in a 3-dimensional coordinate system, in terms of 3 units L* (white-black) a* (red-green) and b* (blue-yellow).	
Portable reflectance spectrophotometers Different artificial light sources (visible range)	**Erythema index and melanin index** It assesses skin colour by providing a read-out of the hemoglobin / melanin index.	
	Skin reflectance spectra The amounts of melanin, oxy-Hb and deoxy-Hb are calculated from the spectra.	
Biomarkers (measured in body fluids)	**Urinary excreted T=T dimers** (high-performance liquid chromatography) It aims to evaluate DNA damage following UV exposure (projects in progress, like Icepure project [50].	
	Urinary 8-hydroxydeoxyguanosine (8OHdG) It evaluates oxidative DNA damage.	
Genotyping (sequencing of MC1R gene from blood sampling)	**MC1R variants**	MC1R gene is considered a susceptibility gene for sunburn, photoageing and skin cancer.
Chemical analysis (hair melanin)	**Derivatives of pheomelanin**	These pheomelanin markers may predict high risk individuals.
Invasive methods Skin biopsy	**"Solar-signature" mutations** In specific genes, like TP53 or CDKN2A.	
	Melanocyte and CPDs detection (indirect immunofluorescence) It quantifies DNA damage after UV exposure.	
	Melanin content (high-performance liquid chromatography) It quantifies different types of melanin in different skin types.	

*UV – ultraviolet, nUVB – narrowband UVB

Table 2

Fitzpatrick skin type and correlation with skin cancer risk

Skin type	Skin reaction to sun exposure	Eumelanin content	MED*	MC1R* variants	Skin cancer risk
I	Always burn, never tan	Low	Low	Loss-of-function variants	High
II	Usually burn, tan less than average (with difficulty)				
III	Sometimes mild burn, tan about average				
IV	Rarely burn, tan more than average (with ease)				
V	Very rarely burns, tans very easily (brown-skinned persons)				
VI	Never burns, tans very easily (black-skinned persons)	High	High	Functional variants	Low

*MED – minimal erythema dose, MC1R – melanocortin 1 receptor.

I will focus on recently designed hand-held spectrophotometers, as they are easy to use, real-time and sensitive, specifically designed for dermatological applications [7, 41]. Reflectance spectrophotometers at selected spectral bands or at visible narrow-band rely on hemoglobin and melanin, the main chromophores in the visible region. Currently, there are several such instruments that measure "erythema index" and "melanin index": Erythema/Melanin Meter (DiaStron Ltd, Andover, Hampshire, UK) Dermaspectrometer, (Cortex Technology, Hadsund, Denmark), Mexameter (Courage & Khazaka GmbH, Koln, Germany), and UV-Optimize (Matic, Naerum, Denmark). The UV-Optimize 555 measures skin erythema and skin pigmentation independently, correlates these measurements to the UV sensitivity determined by a MED test performed with a broadband UVB-source and then Pigment Protection Factor is calculated [48].

However, these narrow-band spectrophotometers have limitations, as they do not distinguish between different types of hemoglobin and types of melanin and do not take into account other chromophores [41]. Thus, diffuse reflectance spectrophotometers have been developed, like: OceanOptics (Boca Raton, FL, USA), Newport (Irvine, CA, USA), B&W Tek (Newark, DE, USA), etc.

There are also colorimetric instruments that integrate a spectrometer (chromameters), like the Minolta Spectrophotometer (Minolta, Osaka, Japan). Artificial light is delivered to the skin and reflected light is measured by a photodiode, at different wavelength intervals in the 400–700 range. Results are then converted and displayed according to the L*a*b* colorimetric system adopted by CIE in 1976 [45)]. The L* value gives the relative lightness ranging from total black (L* = 0) to total white (L* = 100); the a* value represents the balance between red (positive value) and green (negative value); and the b* value represents the balance between yellow (positive value) and blue (negative value).

Using these non-invasive, real-time modern devices, researchers have shown that indeed total melanin content is insufficient to adequately explain all the variations in UVR sensitivity of human skin [8].

NON-INVASIVE BIOMARKERS OF DNA DAMAGE

The latest research area in the field is represented by biomarkers of UV exposure measured in urine, such as DNA damage repair products.

When we go to the sun, we get DNA damage. The main damage induced by UVR is the dimers formation between DNA bases pyrimidines, that lead to a covalent ring structure, the CPD (in particular, thymine (T) dimer) and also to covalent 6-4 linkage (6-4 pyrimidine (6-4) pyrimidone photoproducts (6-4 PPs)). If these lesions are not repaired, UV-specific mutations occur. UV DNA damage is repaired through the nucleotide excision repair (NER) mechanism that involves at least 23 genes/proteins [12]. The basic steps involved in NER pathway are: 1) recognition of damage and recruitment of a 10 subunits multiprotein repair complex to the damaged site, 2) nicking (demarcating) the damaged strand and excision of the damaged region, along with a fragment of about 30 nucleotides, between the two nicks, 3) filling the resultant gap by a DNA polymerase and 4) ligating the final nick to seal the strand, using ligases. The relevance of NER to skin cancer is demonstrated by the dramatically increased risk of all three types of skin cancer in individuals lacking this form of repair (xeroderma pigmentosum) [22]. Moreover, single nucleotide polymorphisms (SNPs) in NER genes may be associated with increased risk of melanoma [24] and decreased melanoma survival in the general population [12].

As an example of biomarker, fragments containing cyclobutane thymine dimer (T=T) are excised by dual incisions from DNA and excreted in urine [18, 25]. At the present, the relationship between DNA photodamage in the skin and in the urine is not known and this relationship will be determined in future projects. Of note, the rate of DNA damage repair is highly variable among individuals and some studies revealed no correlation between skin type and DNA damage repair rates [17, 43].

CONCLUSION

This article describes skin sensitivity (in terms of erythema) and reactivity (erythema, tanning, DNA repair) to sun exposure, and discusses the need for a reliable biophysical method to assess the mentioned parameters as indicators of skin cancer risk. The article underlined the idea that skin cancer susceptibility is not only about skin color and thus screening programs and prevention campaigns

need to take into account multiple factors. As techniques are becoming more refined, faster, and more affordable, we expect that objectively evaluation of skin UVR sensitivity to be more accurate and easy to use in larger clinical trials.

REFERENCES

1. ANDERS, A., H.J. ALTHEIDE, M. KNALMANN, H. TRONNIER, Action spectrum for erythema in humans investigated with dye lasers, *Photochem. Photobiol*., 1995, **61**, 200–205.
2. ANDREASSI, L., S. SIMONI, P. FIORINI, M. FIMIANI, Phenotypic characters related to skin type and minimal erythemal dose, *Photodermatol.,* 1987, **4**, 43–46.
3. BOLANCA, Z., I. BOLANCA, M. BULJAN, I. BLAJIC, Z.J. PENAVIC, M. SITUM, Trends, habits and attitudes towards suntanning, *Coll. Antropol*., 2008, **32**, 143–146.
4. CARRETERO-MANGOLIS, C., H.W. LIM, Correlation between skin types and minimal erythema dose in narrowband UVB (TL-01) phototherapy, *Photodermatol. Photoimmunol. Photomed*., 2001, **17**, 244–246.
5. CHEDEKEL, M.R., Photochemistry and photobiology of epidermal melanins, *Photochem. Photobiol*., 1982, **35**, 881–885.
6. CIE, Action spectrum for the production of previtamin D3 in human skin, *Technical Report 174*, *International Commission on Illumination*, 2006.
7. CLARYS, P., K. ALEWAETERS, R. LAMBRECHT, A.O. BAREL, Skin color measurements: comparison between three instruments: the Chromameter(R), the DermaSpectrometer(R) and the Mexameter(R), *Skin. Res. Technol*., 2000, **6**, 230–238.
8. COELHO, S.G., B.Z. ZMUDZKA, L. YIN, S.A. MILLER, Y. YAMAGUCHI, T. TADOKORO, V.J. HEARING, J.Z. BEER, Non-invasive diffuse reflectance measurements of cutaneous melanin content can predict human sensitivity to ultraviolet radiation, *Exp. Dermatol.*, 2013, **22**, 266–271.
9. DAMIAN, D.L., Y.J. MATTHEWS, T.A. PHAN, G.M. HALLIDAY, An action spectrum for ultraviolet radiation-induced immunosuppression in humans, *Br. J. Dermatol*., 2011, **164**, 657–659.
10. DE GRUIJL, F.R., H.J. STERENBORG, P.D. FORBES, R.E. DAVIES, C. COLE, G. KELFKENS, H. VAN WEELDEN, H. SLAPER, J.C. VAN DER LEUN, Wavelength dependence of skin cancer induction by ultraviolet irradiation of albino hairless mice, *Cancer Res*., 1993, **53**, 53–60.
11. DEL, B.S., J. SOK, F. BERNERD, Assessment of ultraviolet-radiation-induced DNA damage within melanocytes in skin of different constitutive pigmentation, *Br. J. Dermatol*., 2013, **168**, 1120–1123.
12. EMMERT, S., K.H. KRAEMER, Do not underestimate nucleotide excision repair: it predicts not only melanoma risk but also survival outcome, *J. Invest. Dermatol*., 2013, **133**, 1713-1717.
13. FITZPATRICK, T.B., The validity and practicality of sun-reactive skin types I through VI, *Arch. Dermatol.,* 1988, **124**, 869–871.
14. GOGIA, R., M. BINSTOCK, R. HIROSE, W.J. BOSCARDIN, M.M. CHREN, S.T. ARRON, Fitzpatrick skin phototype is an independent predictor of squamous cell carcinoma risk after solid organ transplantation, *J. Am. Acad. Dermatol*., 2013, **68**, 585–591.
15. HALLIDAY, G.M., S.N. BYRNE, J.G. LYONS, D.L. DAMIAN, Photocarcinogenesis nonmelanoma skin cancer, in *Handbook of Photomedicine*, Hamblin MR, Huang Y-Y, eds. New-York, Taylor & Francis Group, 2014, pp. 69–79.
16. HALLIDAY, G.M., S. RANA, Waveband and dose dependency of sunlight-induced immunomodulation and cellular changes, *Photochem. Photobiol*., 2008, **84**, 35–46.
17. HEMMINKI, K., G. XU, L. KAUSE, L.M. KOULU, C. ZHAO, C.T. JANSEN, Demonstration of UV-dimers in human skin DNA in situ 3 weeks after exposure, *Carcinogenesis*, 2002, **23**, 605–609.

18. HEMMINKI, K., G. XU, C.F. LE, Ultraviolet radiation-induced photoproducts in human skin DNA as biomarkers of damage and its repair, *IARC Sci. Publ*., 2001, **154**, 69–79.
19. HOLICK, M.F., Sunlight and vitamin D for bone health and prevention of autoimmune diseases, cancers, and cardiovascular disease, *Am. J. Clin. Nutr*., 2004, **80**, 1678S–1688S.
20. ITO, S., K. WAKAMATSU, H. OZEKI, Chemical analysis of melanins and its application to the study of the regulation of melanogenesis, *Pigment. Cell. Res*., 2000, **13**, 103–109.
21. KADEKARO, A.L., R.J. KAVANAGH, K. WAKAMATSU, S. ITO, M.A. PIPITONE, Z.A. BDEL-MALEK, Cutaneous photobiology. The melanocyte vs. the sun: who will win the final round? *Pigment. Cell. Res*., 2003, **16**, 434–447.
22. KRAEMER, K.H., M.M. LEE, J. SCOTTO, *Xeroderma pigmentosum*; cutaneous ocular, and neurologic abnormalities in 830 published cases, *Arch. Dermatol.*, 1897, **123**, 241–250.
23. KUCHEL, J.M., R.S. BARNETSON, G.M. HALLIDAY, Nitric oxide appears to be a mediator of solar-simulated ultraviolet radiation-induced immunosuppression in humans, *J. Invest. Dermatol*., 2003, **121**, 587–593
24. LI, C., Z. HU, Z. LIU, L.E. WANG, S.S. STROM, J.E. GERSHENWALD, J.E. LEE, M.I. ROSS, P.F. MANSFIELD, J.N. CORMIER, V.G. PRIETO, M. DUVIC, E.A. GRIMM, Q. WEI, Polymorphisms in the DNA repair genes XPC, XPD, and XPG and risk of cutaneous melanoma: a case-control analysis, *Cancer Epidemiol. Biomarkers Prev*., 2006, **15**, 2526-2532.
25. LILJENDAHL, T.S., A. BLOMQVIST, E.M. ANDERSSON, L. BARREGARD, D. SEGERBACK, Urinary levels of thymine dimer as a biomarker of exposure to ultraviolet radiation in humans during outdoor activities in the summer, *Mutagenesis*, 2013, **28**, 249–256.
26. LINOS, E., S.M. SWETTER, M.G. COCKBURN, G.A. COLDITZ, C.A. CLARKE, Increasing burden of melanoma in the United States, *J. Invest. Dermatol*., 2009, **129**, 1666–1674.
27. LUCAS, R.M., A.J. MCMICHAEL, B.K. ARMSTRONG, W.T. SMITH WT, Estimating the global disease burden due to ultraviolet radiation exposure, *Int. J. Epidemiol.*, 2008, **37**, 654–667.
28. MATSUMURA, Y., H.N. ANANTHASWAMY, Toxic effects of ultraviolet radiation on the skin, *Toxicol. Appl. Pharmacol*., 2004, **195**, 298–308.
29. MIYAMURA, Y., S.G. COELHO, K. SCHLENZ, J. BATZER, C. SMUDA2, W. CHOI, M. BRENNER, T. PASSERON, G. ZHANG, L. KOLBE, R. WOLBER, V. J. HEARING, The deceptive nature of UVA-tanning versus the modest protective effects of UVB-tanning on human skin, *Pigment Cell Melanoma Res.*, 2011, **24**, 136–147.
30. NASTI, T.H., L. TIMARES, MC1R, Eumelanin and pheomelanin: Their role in determining the susceptibility to skin cancer, *Photochem. Photobiol*., 2014, doi: 10.1111/php.12335 (epub ahead of print).
31. NOONAN, F..P, M.R. ZAIDI, A. WOLNICKA-GLUBISZ, M.R. ANVER, J. BAHN, A. WIELGUS, J. CADET, T. DOUKI, S. MOURET, M.A. TUCKER, A. POPRATILOFF, G. MERLINO, E.C. DE FABO, Melanoma induction by ultraviolet A but not ultraviolet B radiation requires melanin pigment, *Nat. Commun*., 2012, **3**, 884.
32. PALMER, J.S., D.L. DUFFY, N.F. BOX, J.F. AITKEN, L.E. O'GORMAN, A.C. GREEN, N.K. HAYWARD, N.G. MARTIN, R.A. STURM, Melanocortin-1 receptor polymorphisms and risk of melanoma: is the association explained solely by pigmentation phenotype?, *Am. J. Hum. Genet.,* 2000, **66**, 176–186.
33. PASSERON, T., T. NAMIKI, H.J. PASSERON, P.E. LE, V.J. HEARING, Forskolin protects keratinocytes from UVB-induced apoptosis and increases DNA repair independent of its effects on melanogenesis, *J. Invest. Dermatol*., 2009, **129**, 162–166.
34. PLEASANCE, E.D., R.K. CHEETHAM, P.J. STEPHENS, D.J. MCBRIDE, S.J. HUMPHRAY, C.D. GREENMAN, *et al.*, A comprehensive catalogue of somatic mutations from a human cancer genome, *Nature*, 2010, **463**, 191–196.
35. RAIMONDI, S., F. SERA, S. GANDINI, S. IODICE, S. CAINI, P. MAISONNEUVE, M.C. FARGNOLI, MC1R variants, melanoma and red hair color phenotype: a meta-analysis, *Int. J. Cancer.*, 2008, **122**, 2753–2760.

36. RIGEL, D.S., Cutaneous ultraviolet exposure and its relationship to the development of skin cancer, *J. Am. Acad. Dermatol.*, 2008, **58**, S129–S132.
37. SCOTT, M.C., K. WAKAMATSU, S. ITO, A.L. KADEKARO, N. KOBAYASHI, J. GRODEN, R. KAVANAGH, T. TAKAKUWA, V. VIRADOR, V.J. HEARING, Z.A. ABDEL-MALEK, Human melanocortin 1 receptor variants, receptor function and melanocyte response to UV radiation, *J. Cell. Sci.*, 2002, **115**, 2349–2355.
38. SMITH, A.G., N. LUK, R.A. NEWTON, D.W. ROBERTS, R.A. STURM, G.E. MUSCAT, Melanocortin-1 receptor signaling markedly induces the expression of the NR4A nuclear receptor subgroup in melanocytic cells, *J. Biol. Chem.*, 2008, **283**, 12564–12570.
39. SIMON, J.D., D.N. PELES, The red and the black, *Acc. Chem. Res*., 2010, **43**, 1452-1460.
40. SNELLMAN, E., C.T. JANSEN, K. LESZCZYNSKI, R. VISURI, T. MILAN, K. JOKELA, Ultraviolet erythema sensitivity in anamnestic (I-IV) and phototested (1-4) Caucasian skin phototypes: the need for a new classification system, *Photochem. Photobiol*., 1995, **62**, 769–772.
41. STAMATAS, G.N., B.Z. ZMUDZKA, N. KOLLIAS, J.Z. BEER, Non-invasive measurements of skin pigmentation in situ, *Pigment. Cell. Res*., 2004, **17**, 618–626.
42. SUTHERLAND, J.C., K.P. GRIFFIN, Absorption spectrum of DNA for wavelengths greater than 300 nm, *Radiat. Res*., 1981, **86**, 399–409.
43. TADOKORO, T., N. KOBAYASHI, B.Z. ZMUDZKA, S. ITO, K. WAKAMATSU, Y. YAMAGUCHI, K.S. KOROSSY, S.A. MILLER, J.Z. BEER, V.J. HEARING, UV-induced DNA damage and melanin content in human skin differing in racial/ethnic origin, *FASEB J.*, 2003, **17**, 1177–1179.
44. VINCENSI, M.R., M. D'ISCHIA, A. NAPOLITANO, E.M. PROCACCINI, G. RICCIO, G. MONFRECOLA, P. SANTOIANNI, G. PROTA, Phaeomelanin versus eumelanin as a chemical indicator of ultraviolet sensitivity in fair-skinned subjects at high risk for melanoma: a pilot study, *Melanoma Res*., 1998, **67**, 293–299.
45. WEATHERALL, I.L., B.D. COOMBS, Skin color measurements in terms of CIELAB color space values, *J. Invest. Dermatol.*, 1992, **99**, 468–473.
46. WEINSTOCK, M.A., Assessment of sun sensitivity by questionnaire: validity of items and formulation of a prediction rule, *J. Clin. Epidemiol.*, 1992, **45**, 547–552.
47. WESTERHOF, W., O. ESTEVEZ-USCANGA, J. MEENS, A. KAMMEYER, M. DUROCQ, I. CARIO, The relation between constitutional skin color and photosensitivity estimated from UV-induced erythema and pigmentation dose-response curves, *J. Invest. Dermatol.*, 1990, **94**, 812–816.
48. WULF, H.C., P.A. PHILIPSEN, M.H. RAVNBAK, Minimal erythema dose and minimal melanogenesis dose relate better to objectively measured skin type than to Fitzpatricks skin type, *Photodermatol. Photoimmunol. Photomed*., 2010, **26**, 280–284.
49. YOUNG, A.R., C.A. CHADWICK, G.I. HARRISON, O. NIKAIDO, J. RAMSDEN, C.S. POTTEN, The similarity of action spectra for thymine dimers in human epidermis and erythema suggests that DNA is the chromophore for erythema, *J. Invest. Dermatol*., 1998, **11**, 982–988.
50. *** Project: The impact of climatic and environmental factors on personal ultraviolet radiation exposure and human health, www.icepure.eu