Coll. Antropol. **31** (2007) Suppl. 1: 13–16
Original scientific paper

The Mechanisms of UV Radiation in the Development of Malignant Melanoma

Mirna Šitum[1], Marija Buljan[1], Suzana Ožanić Bulić[1] and Dubravka Šimić[2]

[1] Department of Dermatology and Venereology, University Hospital »Sestre Milosrdnice«, Zagreb, Croatia
[2] Division of Dermatology and Venereology, University Hospital »Mostar«, Mostar, Bosnia and Hercegovina

A B S T R A C T

The sunlight was one of the first agents recognized to be carcinogenic for humans. There is convincing evidence from epidemiologic studies that exposure to solar radiation is the major cause of cutaneous melanoma in light-pigmented populations and plays a role in the increasing incidence of this malignancy. The molecular mechanisms by which UV radiation exerts its varied effects are not completely understood, however, it is considered that UVA and UVB are equally critical players in melanoma formation. Whereas UVA can indirectly damage DNA through the formation of reactive oxygen radicals, UVB can directly damage DNA causing the apoptosis of keratinocytes by forming the sunburn cells. Besides action through mutations in critical regulatory genes, UV radiation may promote cancer through indirect mechanisms, e.g. immunosuppression and dysregulation of growth factors. The carcinogenic process probably involves multiple sequential steps, some, but not all of which involve alterations in DNA structure.

***Key words**: melanoma, UV radiation, photoimmunology, oncogene*

Introduction

Exposure to sunlight is a major factor in induction of skin cancers including malignant melanoma, especially in Caucasian population. Melanoma is a malignant tumour arising from neural crest-derived melanocytes, pigmented cells presented normally in the epidermis and sometimes in the dermis. During the past century, changes in clothing styles, recreational activities, longevity, and other aspects of lifestyle have resulted in increased exposure to sunlight[1]. Particularly, intermittent, recreational exposure to UV component of sunlight associated with sunburns plays an important role in melanoma formation[2]. Excessive exposure of fair-skinned individuals to UV radiation, mainly natural sunlight presents an environmental risk factor. Another major risk factor for development of melanoma is genetic background of a patient. The genetic markers include three major genes: CDKN2A gene on chromosome 9, CDK4 gene on chromosome 12 and gene on chromosome 1[3]. However, in contrast to non-melanoma skin cancers, in which there are distinctive UVB-induced mutations in the p53 gene, the exact mechanisms and wavelengths by which sunlight induces, promotes or contributes to the development of melanoma have not been yet identified[2]. UVB (280–320 nm) is absorbed in nucleic acids and has been considered the aetiological wavelength for melanoma[4]. Nevertheless, recent epidemiological studies have suggested that UVA exposure increases the risk of melanoma even though this is still debated[5]. The hypothesis that UVA is important in the aetiology of melanoma is also supported by epidemiological studies, in which exposure to UVA-emitting sun beds was related to an increased melanoma risk[6]. Several retrospective studies have assessed the risk of melanoma in relation to sunscreen use[7]. One mechanism by which sunscreens may lead to an increased melanoma risk is that their use may allow prolonged intensive sun exposures, which may increase the melanoma risk.

UV Radiation and Malignant Melanoma

UVA and UVB are equally critical players in melanoma formation. However, data are conflicting on the specific roles of UVA and UVB, which differ in their mo-

Received for publication October 1, 2006

lecular and biological ways of action. The predominant component of sunlight is UVA which penetrates deeper into skin than UVB due to longer wavelengths (Table 1). Whereas UVA can indirectly damage DNA through the formation of reactive oxygen radicals, UVB can directly damage DNA causing the apoptosis of keratinocytes by forming the sunburn cells[8]. Besides action through mutations in critical regulatory genes, UV radiation may promote cancer through indirect mechanisms, e.g. immunosuppression and dysregulation of growth factors[9]. Overexpression in the skin of three growth factors for melanocytes; basic fibroblast growth factor (bFGF), stem cell factor (SCF) and endothelin (ET)-3, together with UVB radiation, led to melanoma *in vivo*[9]. In contrast to melanoma cells which produce different cytokines and growth factors with paracrine and autocrine effects[10], melanocytes are dependent on the growth factor expression by neighbour cells, e.g. keratinocytes and fibroblasts[11]. An imbalance between stimulatory and inhibitory factors provoked by exogenous stimuli, e.g. UV, could activate the melanocytes and drive them towards transformation by uncontrolled proliferation and migration[9]. UV radiation is, at the same time, a powerful stimulus to melanin production and a destructive agent on the melanocytes[1]. UV radiation can also serve as a tumour promoter after an initiating dose of a chemical carcinogen. Thus UV radiation may play a variety of roles during cutaneous carcinogenesis.

TABLE 1
WAVELENHGHTS OF THE NON-IONIZING RADIATION

Non-ionizing radiation	(nm)
UVC	<290
UVB	290–320
UVA	320–400
UVA2	320–340
UVA1	340–400
Visible	400–700
Infra-red	>700

From: Rigel S. D., R. J. Friedman, L. M. Dzubow, D. S. Reintgen, B. C. J. Ystryn, R. Marks: Cancer of the skin. (Elsevier Inc., 2005).

Immunology

Incomplete or complete regression of melanoma, occurrence of vitiligo-like depigmentation and halo nevi, as well as higher rate of melanoma in immunosuppressed patients point to the fact that melanoma is an immunogenic tumour[12]. The molecular characterization of melanoma antigens recognized by autologous T-cells or antibodies was a scientific breakthrough[13]. The major melanoma antigens include CDKN2A mutated antigen, Mage-1,-3 and NY-ESO-1 that belong to the shared tumour specific antigens and differentiation antigens like tyrosinase, gp100, MelanA/MART-1. Their expression can be followed *in situ* at the protein level using monoclonal antibodies. These proteins are processed inside the cell and presented on the melanoma cell surface as MHC/peptide complexes. CD8+ cytotoxic T-cells recognize these antigens, and after appropriate activation, kill such tumour cells in MHC-dependent manner through release of cytotoxic granules (e.g. perforin and granzyme B) or activation of FAS/TNF pathways[2]. CD8+ T-cells are believed to be the major effector cells for an anti-melanoma-specific immune response, but CD4+ T-cells, as well as antibodies, also play a critical role. Since melanoma is an immunogenic tumour, a variety of immune escape mechanisms may be found in the advanced tumours, e.g. loss of tumour-specific antigens, loss of MHC class I molecules as well as secretion of immuno-inhibitory cytokines such as IL-10 and TGF-beta[14].

Genetics

p53 is a widely studied tumour suppressor gene that has the ability to trigger cell-cycle arrest and apoptosis in response to diverse stress stimuli. Under cellular stress, such as hypoxia, DNA damage, ionizing radiation, exposure to anticancer drugs and oncogene activation, the p53 protein is rapidly accumulated in cells[15–18]. Depending on the extent of DNA damage, p53 can either bring about cell cycle arrest in an effort to repair the damage, or induce apoptosis when the damage is too severe[19,20]. Apoptosis presents self-protective mechanism to eliminate unnecessary or severely damaged cells. More than half of all human tumours contain p53 mutations, and most of the mutations are located in the DNA-binding domain[21]. The p53 tumour suppressor gene is inactivated by point mutations in approximately 50% of all human tumours. Such a high mutation frequency indicates a strong selection for loss of normal p53 function during tumorigenesis[22].

The most promising molecule in stabilising the p53 protein is CP-31398. After CP-31398 treatment, an increase in the stability of p53 protein results in its active conformation which correlates to apoptotic response. However, CP-31398 was not able to revert all mutant p53 proteins to wild-type conformation and to induce apoptosis in all mutant p53 cell lines. The ability of CP-31398 to induce apoptosis through the intrinsic mitochondrial pathway may well depend on the mutational status of p53. Mutant p53 reactivation by small molecules has an evident potential for the discovery of efficient and specific anti-cancer drugs[23].

Molecular Mechanisms

The classic model of carcinogenesis includes four stages: initiation/induction, promotion, pre-malignant progression and malignant conversion (Table 2). Initiation is the result of genetic damage caused by exogenous agent (e.g. UV radiation) that alters cellular proliferative controls and/or the differentiation pathway. Promotion

TABLE 2
STAGES OF CARCINOGENESIS

Stages of carcinogenesis
Initiation – induction
Promotion
Premalignant conversion
Malignant progression

From: Rigel S. D., R. J. Friedman, L. M. Dzubow, D. S. Reintgen, B. C. J. Ystryn, R. Marks: Cancer of the skin. (Elsevier Inc., 2005).

involves the expansion of the initiated cell population or clone and is thought to be due to epigenetic effects. Progression and conversion are marked by cells with a high level of genetic instability, chromosomal abnormalities, surface substance expression and oncogene activity[24]. The effects of non-ionizing radiations on human cells rely on complex cellular interactions. Once radiation is absorbed the molecule is raised to an excited state. In an effort to dissipate the absorbed energy and return to the resting state the energy can be converted to chemical change which in turn results in biologic alterations.

UVB causes mutations and immunosuppressive effects essential to photo-carcinogenesis. UVB-induced DNA damage leads to modifications in oncogene and tumour suppressor gene expression, the most important event in tumour initiation. UVB primarily affects the epidermis causing disruption in DNA and the formation of pyrimidine dimmers. In addition, UVB effects the production of reactive oxygen species (e.g. hydrogen peroxide, superoxide anions and singlet oxygens). These induce single strand breaks in DNA[24]. UVB and UVA radiation both exert immunomodulatory effects. In addition, UV radiation may have other effects on the local environment in which the tumours develop, such as increased vascularity and production of growth factors that can contribute to tumour growth and progression. The molecular mechanisms by which UV radiation exerts its varied effects are not completely understood. The carcinogenic process probably involves multiple sequential steps, some, but not all of which involve alterations in DNA structure, e.g. mutations.

Prevention

The overall annual incidence and mortality rate of melanoma is constantly rising in recent decades in all parts of the world, with the highest rates in Australia. Therefore the role of primary and secondary prevention becomes crucial in reducing the lifetime risk of malignant melanoma as well as other non-melanoma skin cancers. While secondary prevention with early detection is the most effective strategy for those who sustained unprotected sun exposure in youth, primary prevention by effective sun protection throughout life for those at risk to develop skin cancer may reduce the incidence of skin cancer. However, according to the published clinical data, there is no direct relationship between dose of UV radiation and melanoma induction.

The protective role of sunscreens has been controversial according to some epidemiological studies[2]. The interpretation of these studies must be done with great caution, because persons with a history of sunburn are more likely to use sunscreens, so the higher risk may be misinterpreted. One of the studies has indicated that UVA sunscreen protection may be crucial in preventing the efferent immune response suppression important in tumour formation. It is reasonable to suppose that the improvement in performance of modern sunscreens will lead to a worthwhile benefit as a preventive agent against melanoma, although these benefits may not be seen for several decades[25].

While speaking of prevention on *in vitro* basis, two lipid soluble antioxidans, -tocopherol and -carotene, have been shown to prevent UV-mediated oxidative stress in keratinocytes and fibroblasts[26,27]. -tocopherol reduces cell proliferation, probably by affecting protein kinase C dephosphorylation, which in turn reduces DNA-binding capacity of the AP-1 transcription factor[28,29]. -tocopherol protection presents probably a combined effect of a decreased proliferation rate, providing time for extended repair, and reduced level of oxidative stress resulting in diminished apoptosis signalling. These effects suggest a potent and broad UV protective capacity of -tocopherol affecting several cellular functions[4]. On the other hand, -carotene is protecting primary cultures of melanocytes from UVA or UVB-induced oxidative stress, which is less protective compared to the action of -tocopherol. The balance between proliferation and apoptosis might, in a longer perspective, have implications for the risk of transformation and subsequent development of melanoma. This is the basis for understanding the protective effect of -tocopherol against UV-induced melanocyte damage[4].

In conclusion, the incidence of melanoma has increased dramatically in the past 40 years. The reason for this increase is uncertain but may involve increased recreational sun exposure, especially early in life[30]. Routine use of sunscreens, use of protective clothing, and avoiding intense midday ultraviolet exposure should be recommended. The patient should be educated in the clinical features of melanoma and advised to report any growth or other change in a pigmented lesion. Hopefully this would rise awareness in general population and change the outcome of the disease in the future.

REFERENCES

1. Fitzpatrick's Dermatology in General Medicine (McGraw-Hill, 2003). — 2 BOLOGNIA JL, JORIZZO JL, RONALD PR: Dermatology (Mosby, 2003). — 3. PIEPKORN M, J Am Acad Dermatol, 42 (2000) 705. — 4 LARSON P, OLLINGER K, I ROSDAHL I, Br J Dermatol, 155 (2006) 292. — 5. WANG SQ, SETLOW R, BERWICK M, J Am Acad Dermatol, 44 (2001) 837. — 6. DONAWHO C, WOLF P, Curr Opin Oncol, 8 (1996) 159. — 7. WESTERDAHL J, INGVAR C, MASBACK A, OLSSON H, Int J Cancer, 87 (2000) 145. — 8. ESKANDARPOUR M, HASHEMI J, KANTER L, J Natl Cancer Inst, 95 (2003) 790. — 9. BRENNER M, DEGITZ K, BESCH R, BERKING C, Br J Dermatol, 153 (2005) 733. — 10. MORETTI S, PINZI C, SPALLANZANI A, Int J Cancer, 84 (1999) 160. — 11. HERLYN M, SHIH IM, Pigment Cell Res, 7 (1994) 81. — 12. NESTLE FO, BURG G, DUMMER R, Immunol Today, 20 (1999) 5. — 13. ROSENBERG SA, Nature, 411 (2001) 380. — 14. MARINCOLA FM, JAFFEE EM, HICKLIN DJ, FERRONE S, Adv Immunol, 74 (2000) 181. — 15. SHERR CJ, Genes Dev, 12 (1998) 2984. — 16. KO LJ, PRIVES C, Genes Dev, 10 (1996) 1054. — 17. GIACCIA AJ, KASTAN MB, Genes Dev, 12 (1998) 2973 — 18 ASKER C, WIMAN KG, SELIVANOVA G, Biochem Biophys Res Commun, 265 (1999) 1. — 19. LI G, HO VC, MITCHELL DL, Am J Pathol, 150 (1997) 1457. — 20. TRON VA, TROTTER MJ, ISHIKAWA T, J Cutan Med Surg, 3 (1998) 16. — 21. BEROUD C, SOUSSI T, Nucleic Acids Res, 26 (1998) 200. — 22. SOUSSI T, Ann MY Acad Sci, 910 (2000) 121. — 23. HO CK, LI G, Br J Dermatol, 153 (2005) 900. — 24. RIGEL SD, FRIEDMAN RJ, DZUBOW LM, REINTGEN DS, CJYSTRYN B, MARKS R: Cancer of the skin (Elsevier Inc., 2005). — 25. DIFFEY BL, Br J Dermatol 153 (2005) 378. — 26. EICHLER O, SIES H, STHL W, Photochem Photobiol, 75 (2002) 503. — 27. EPSTEIN JH, Photochem Photobiol, 25 (1977) 211. — 28. BOSCOBOINIK D, SZEWCZYK A, AZZI A, Arch Biochem Biophys, 286 (1991) 264-9. — 29. MAHONEY CW, AZZI A, Biochem Biophys Res Commun, 154 (1988) 694-7. — 30. KASPER DL, BRAUWALD E, FAUCI A, HAUSER S: Harrison's Principles of Internal Medicine (McGraw-Hill Professional, 2004).

M. Šitum

Department of Dermatology and Venerology, University Hospital »Sestre milosrdnice«, Vinogradska cesta 29, 10 000 Zagreb, Croatia
e-mail: msitum@kbsm.hr

MEHANIZMI UV ZRAČENJA U NASTANKU MALIGNOG MELANOMA

SAŽETAK

Sunčeva svjetlost jedan je od prvih agenasa prepoznatih kao humani karcinogeni čimbenik. Epidemiološke studije ukazale su na činjenicu da je sunčeva svjetlost jedan od glavnih čimbenika u nastanku malignog melanoma u bijelaca, sa značajnom ulogom u porastu incidencije tog malignog tumora. Molekularni mehanizmi kojima UZ zračenje djeluje na stanicu nisu još u potpunosti razjašnjeni, no smatra se da UVA i UVB zračenje imaju jednako značajnu ulogu u nastanku melanoma. UVA zračenje neizravno oštećuje DNA prozivodnjom kisikovih slobodnih radikala, dok UVB izravno oštećuje DNA uzrokujući apoptozu keratinocita nastankom stanica opeklina. Osim djelovanja na ključne regulacijske gene, UV zračenje može utjecati na nastanak zloćudnih tumora i drugim neizravnim mehanizmima kao npr. imunosupresijom i poremećenom regulacijom različitih faktora rasta. Karcinogeneza vjerojatno uključuje složeni kaskadni proces u kojem bitnu, ali ne i jedinu ulogu, ima promjena u strukturi DNA.