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Antioxidant capacity total in non-melanoma skin cancer and its relationship with food consumption of antioxidant nutrients

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Abstract

The non-melanoma skin cancer is the most common cancer and accounts for more than half of the diagnoses of cancer, and basal cell carcinoma (BCC), the most frequent cutaneous neoplasm, corresponding to 70-80% of cutaneous tumors. Oxidative stress is an important trigger for skin carcinogenesis. Thus, it is important to evaluate oxidative stress, in order to discern effective therapeutic strategies able to stop it or attenuate it, thereby prevent the installation of non-melanoma skin cancer. Cross-sectional study with controls, involving 84 individuals of both sexes aged between 38-84 years, divided into two groups: control group of healthy people(n = 24) and the case group included individuals who presented non-melanoma skin and they have undergoing surgery (n = 60). The blood samples of the individuals were obtained for evaluation of biomarkers of oxidative stress (F2-isoprostane, nitrite, thiobarbituric acid reactive substances (TBARS) and total antioxidant capacity). The usual dietary intake and nutritional status of the subjects were evaluated. The significance level for this study was 5%. Patients in the case group had higher serum concentrations of biomarkers of oxidative stress, F2-isoprostane concentrations were significantly higher compared to controls. The results showed high rates of overweight and obesity in the case and control groups. The dietary concentrations of antioxidant minerals zinc, copper and selenium in the case group were significantly lower compared to controls. The correlation between markers of oxidative stress and dietary concentrations of antioxidant nutrients showed the influence of food intake of vitamins A and E in reducing oxidative stress, since these nutrients behave as important antioxidants, acting as sweepers of RL, by removing of the body the negative

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ANTIOXIDANTE TOTAL CAPACIDAD EN CÁNCER DE PIEL NO MELANOMA Y SU RELACIÓN CON EL CONSUMO DE ALIMENTOS DE LOS NUTRIENTES ANTIOXIDANTES

Resumen

El cáncer de piel no melanoma es el cáncer más común y representa más de la mitad de los diagnósticos de cáncer, y el carcinoma de células basales (BCC), la neoplasia cutánea más frecuente, representando el 70-80% de los tumores cutáneos. El estrés oxidativo es un disparador importante en la carcinogénesis de la piel. Por lo tanto, es importante para evaluar el estrés oxidativo, con el fin de prever y estrategias terapéuticas eficaces capaces de detener o mitigar ella, para evitar de este modo la instalación de cáncer de piel no melanoma. Estudio transversal con los controles, con la participación de 84 sujetos de ambos sexos con edades comprendidas entre 38 a 84 años, divididos en dos grupos: grupo control de sujetos sanos (n = 24) personas y el grupo de casos incluyeron los individuos que presentaron para el cáncer de piel no melanoma tiene someterse a la cirugía (n = 60). Las muestras de sangre de los sujetos fueron obtenidos para la evaluación de los biomarcadores de estrés oxidativo (F2-isoprostano, nitritos, sustancias reactivas al ácido tiobarbitúrico (TBARS) y capacidad antioxidante total). Se evaluó la ingesta dietética habitual y el estado nutricional de los sujetos. El nivel de significación para este estudio fue de 5%. Los pacientes en el grupo de casos tenían mayores concentraciones séricas de biomarcadores de estrés oxidativo, las concentraciones de F2-isoprostano fueron significativamente mayor en comparación con los controles. Los resultados mostraron altas tasas de sobrepeso y obesidad en los grupos de casos y controles. Las concentraciones dietéticas de antioxidante minerales de zinc, cobre y selenio en el grupo de casos fueron significativamente más bajos en comparación con los controles. La correlación entre los marcadores de estrés oxidativo y las concentraciones dietéticas de nutrientes antioxidantes destacó la influencia de la ingesta de alimentos de vitaminas A y E en la reducción del estrés oxidativo, ya que estos nutrientes se comportan como antioxidantes importantes, actuando como barrenderos RL, el cuerpo se deshaga de estos efectos negativos sobre el equilibrio redox de la piel.

effects on the redox balance of the skin. We emphasize the importance of adopting healthy eating habits that optimize the consumption of antioxidant nutrients as a strategy to prevent oxidative damage to the skin.

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Hacemos hincapié en la importancia de adoptar hábitos de alimentación saludables que optimizan el consumo de nutrientes antioxidantes como estrategia para prevenir el daño oxidativo de la piel.

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Palabras clave: *Neoplasias de la piel. El estrés oxidativo. F2-isoprostanos. Los antioxidantes. Biomarcadores.*

Introduction

The National Cancer Institute (NCI) estimates about 580 000 new cancer cases for 2014 and the most incident cancers in the Brazilian population will be: non-melanoma skin cancer (182 000), prostate (69 000); breast (57 000); colon and rectum (33 000), lung (27 000) and stomach (20 000)[1].

The non-melanoma skin cancer (NMSC) is the most common cancer and accounts for more than half of the diagnoses of cancer[1]. Risk factors include: clear skin sensitive to ultraviolet radiation, excessive exposure to sun, weather conditions (tropical climate and climate at very high altitudes), the personal and family history for the disease[1,2,3]. Basal cell carcinoma (BCC) is the most common skin cancer, accounting for 70-80% of skin tumors[3]. Environmental factors have a strong influence on the tumorigenic process, and this is the reason of the occurrence of the variations in their incidence in different geographical locations and ethnicities[4,5].

The skin is the largest body organ and it is an important link between the environment and the body whose function is to protect it from physical, chemical and microbiological damaging agents which can alter the skin structure and function, by inducing the formation of reactive oxygen species (ROS) and nitrogen (RNS).

These species are essential to maintain cellular homeostasis and survival, when at equilibrium between formation and removal. However, when there are marked changes in the balance, a pro-oxidant state is generated, thus leading to oxidative stress.

Ultraviolet radiation (UVR) from sunlight is a potent environmental risk factor in the pathogenesis of skin cancer[6,7]. Reactive oxygen species and nitrogen produced by UV radiation are associated with both the initiation, promotion, and with the progression of carcinogenesis[6,7]. They are capable of inducing the production of transcription factors such as activator protein 1 (AP-1) and nuclear factor kappa B (NF-KB). The NFkβ activated, in turn, stimulates the production of pro-inflammatory cytokines, such as tumor necrosis factor α (TNF α) and interleukin-1β (IL-1 β), and it may play an important role in redox regulation.

ROS induce lipid peroxidation (LPO)· which is responsible for: changes in membrane permeability, destroying the cell membrane structure, failure of the mechanisms of exchange of metabolites, and in an extreme condition, cell death[9,10]. During the LPO, the enzyme cyclooxygenase catalyzes the production of prostaglandins, which in combination with nitric oxide causes: inflammation, benign tumors, solar keratosis (SK), which can progress to squamous cell carcinoma (SCC), probably because the cells inflammatory produce reactive oxygen species, thereby increasing oxidative damage to DNA and the immune system[10,11].

The objective of this article is to evaluate the markers of oxidative stress in people who had non-melanoma skin cancer in healthy people and in this regard and its association with habitual food intake relative to antioxidant nutrients, in order to contribute to consolidate new therapeutic formats and to program an effective preventive approach that aims at adopting healthy eating habits that ensure a satisfactory intake of antioxidant nutrients.

Material and methods

Cross-sectional study with controls, involving 84 individuals between 38-84 years. The individuals were divided into two groups: cases (previously diagnosed with non-melanoma skin cancer, n = 60) and control (people who have never had a non-melanoma skin cancer, n = 24). People with non-melanoma skin cancer were recruited among patients who were attended at the Dermatology Clinic of the HGV, in 2011, whose results of the biopsy revealed a diagnosis of basal cell carcinoma (BCC) or squamous cell (SCC).

The eligibility criteria for the case and control groups were: more than 20 years old; no pre-existing disease, including: melanoma, insulin dependent diabetes mellitus, severe heart disease (with hemodynamic repercussions or cianosantes), liver dysfunction (jaundice or cirrhosis), renal failure (requiring dialysis), HIV +; no use of chemotherapy or radiation in the previous six months; no severe psychiatric illness that would limit the ability of understanding and acceptance to participate by donating the material, no consume of supplementation of vitamins and minerals. The specific inclusion criterion for control group was: no diagnosis of BCC / SCC and for the case group was the previously diagnosed BCC / SCC whose skin lesion has already undergone excision. The exclusion criterion was not accepting participate. The study was approved by the Ethics Committee of the Federal University of Piauí, according to certificate number presentation for ethical consideration (CAAE) 0055.0045.000-10.

Assessment of nutritional status

Weight, height, triceps skinfold (TSF), arm circumference (AC), arm muscle circumference (MAMC) were the nutritional assessment parameters used. The simplified normal range according Jellife (1966)[12] was used to interpret the results. For the classification of EN compared to the percentage of AC, TSF and MAMC values of Blackburn, GL and Bistrian, BR were used (1977)[13] and for the elderly, the values of AC, TSF and MAMC were compared to the 50th percentile according to sex and age recommended in National Health and Nutrition Examination Survey - NHANES III (Kuczmarski et al, 2000)[14].

For classification of nutritional status according to body mass index (BMI) to less than 60 years age group, were adopted benchmarks set by the World Health Organization (2000)[15] and for the age group over 60 years, the benchmark established by the Pan American Health Organization (PAHO, 2001)[16] was adopted.

Assessment of usual dietary intake of antioxidant nutrients

The individuals filled a FFQ (Food Frequency Questionnaire) validated by Crispin and Ribeiro and Panato[17]. The estimate of habitual food intake was made from the methodology developed by Sichieri and Everhardt (1998)[18], in a validation study. Diet composition was assessed by NutWin 2.5 software. The amounts of dietary intake of the antioxidant nutrients were adjusted by energy, according to the method residual nutrient by Stampfer and Willett (1986)[19]. It was used as a reference the estimated average requirement (EAR) defined for antioxidant nutrients.

Determination of the concentrations of total antioxidant capacity

The total antioxidant capacity of plasma collected with citrate was determined using the commercial Antioxidant Assay Kit (Cayman, USA). The assay is based on the ability of the sample antioxidant to inhibit the oxidation of ABTS® (2,2-azino-di [3-ethylbenzthiazoline sulfonate]) to the ABTS® • + metmyoglobin. The amount of produced ABTS® • + can be monitored by absorbance at 750 or 405 nm.

Statistical Analysis

For statistical analysis a database was constructed, by using the Statistical Package for the Social Sciences - SPPS version 15.0 (2007). The results were displayed as mean and standard deviation. Statistical analysis initially involved the application of the Kolmogorov-Smirnov test to verify normal distribution of the quantitative variables. For comparison of means, ANOVA Student's t test or Mann-Whitney test was used as appropriate; the correlation between variables was obtained by Spearman correlation. In all tests, it was adopted $p < 0.05$ considered statistically significant.

Results

The mean and standard deviation for age and anthropometric parameters used to assess nutritional status of the subjects in the case group and control values are shown in table I There was a statistically significant difference in age between the groups.

The mean values of BMI and TSF suggest overweight and accumulation of fat mass respectively in both the control group and a study group, and the mean values of nutritional indices and AC and MAMC suggest normal the concentration of lean and fat mass in the

Table I
Anthropometric parameters according to the case and control groups

Variables	*Control (n = 24)*	*Case (n = 60)*	*Value of p*
Age (years)	56.7 ± 11.6	62.7 ± 14.2	**0.0492**
Weight (kg)	66.4 ± 13.1	63.9 ± 10.1	0.6300
Height (m)	1.6 ± 0.1	1.6 ± 0.9	0.4750
BMI (Kg/m^2)	27.3 ± 3.5	26.3 ± 3.7	0.2710
TSF (mm)	20.2 ± 6.8	18.4 ± 7.8	0.1750
AC (cm)	30.9 ± 3.1	30.0 ± 4.5	0.1910
MAMC (cm)	24.6 ± 3.0	24.5 ± 4.0	0.7100

Legend: BMI - Body Mass Index; TSF - Triceps Skin Fold; AC - Arm Circumference; MAMC - Circumference Muscular Arm. Mann-Whitney test, $p > 0.05$ difference is not significant.

Table II
Dietary concentrations averages and standard deviations of the consumption of antioxidant nutrients in the diet to the case and control groups

Parameters	*Control (n = 24)* *Average ± SD*	*Case (n = 60)* *Average ± SD*	*EAR#*	*p*
Vitamin A (µg)	380.9 ± 197.1	558.0 ± 570.0	625	**0.0010**
Vitamin C (mg)	162.4 ± 19.3	57.9 ± 50.6	75	0.2220
Vitamin E (mg)	6.1 ± 14.2	9.6 ± 7.02	12	**0.0010**
Copper (µg)	1040.0 ± 1009.0	800.0 ± 400.0	700	**0.0330**
Zinc (mg)	10.1 ± 12.6	6.5 ± 3.6	9.4	**0.0310**
Selenium (µg)	52.2 ± 44.5	76.2 ± 38.8	45	**0.0280**

Legend: Sd = Standard Deviation; Estimate average application EAR different for sexes (vitamin A 625 µg/d males and 500 µg/d female. Zinc 9,4 mg/d male 6, 8 mg/d female. Vitamin C 75 mg male and 60 mg/d female). Mann-Whitney test significant difference ($p < 0.05$). Dietary nutrient concentrations adjusted for eneragy.

two groups. The values of anthropometric data did not differ between groups.

Table II shows the estimated dietary concentrations of antioxidant nutrients from case and control groups. It was possible to detect a statistically significant difference in the habitual dietary intake of vitamin A, E and the minerals zinc, copper and selenium between groups.

Figure 1 shows the comparison of serum concentrations of total antioxidant capacity of the control group (1.8 ± 0.4 mMol/L) and case group (2.3 ± 1.1 mMol/L).

The analysis of simple linear correlation between CAOT and dietary concentrations of antioxidant nutrients in control and case groups are in table III.

The association between CAOT and dietary concentrations of antioxidant nutrients in the control group showed negative weak ($r = -0.488$) and significant correlation ($p = 0.015$) between CAOT and Vitamin A.

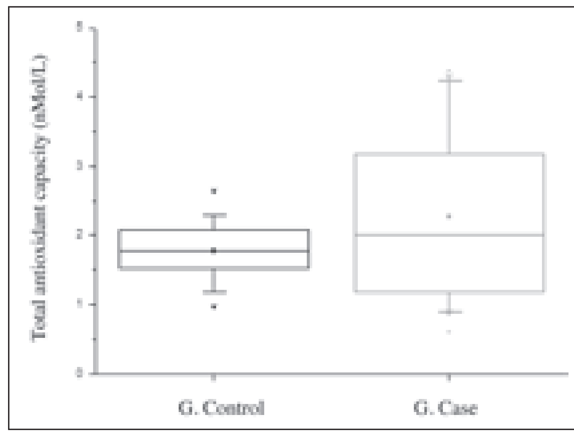

Fig. 1.—Antioxidant Capacity Total control groups (n = 24) and case group (n = 60). Legend: ANOVA test for repeated measures. There was no statistically significant difference (p = 0.8767).

Discussion

The main environmental risk factor for NMSC is exposure to ultraviolet radiation, which can cause excessive amounts of reactive oxygen species and nitrogen species, contributing to the installation of oxidative stress[6,7], which has a major effect on cutaneous immune system, inducing a state of location[6,7,8] immunosuppression, compromising the antitumor defense system.

Table III
Analysis of simple linear correlation between total antioxidant capacity and dietary concentrations of antioxidant nutrients of case and control groups

CAOT	*Dietary concentrations of antioxidant nutrients*					
	Vit A	*Vit C*	*Vit And*	*Selenium*	*Copper*	*Zinc*
G. Control	**r = -0.488**	r = 0.267	r = -0.206	r = 0.146	r = 0.206	r = -0.160
	p = 0.015	p = 0.226	p = 0.334	p = 0.15	p = 0.334	p = 0.455
G. Case	r = -0.064	r = -0.002	r = -0.192	r = -0.046	r = -0.110	r = -0.040
	p = 0.654	p = 0.988	p = 0.145	p = 0.732	p = 0.406	p = 0.765

Legend: CAOT - Total Antioxidant Capacity; Vitamin A; Vitamin C; Vitamin E. Spearman Correlation.

Researches directed to mechanisms for restoring the redox balance of the skin as a prevention or treatment for NMSC strategy have been encouraged and the use of antioxidants is emerging as a promising alternative[20]. There is more and more evidence that antioxidants promote rapid repair of oxidative damage (through sequestration of reactive oxygen species in the lipid membrane, eg.), helping to reduce the production of free radicals, lipoperoxidation, promoting the repair DNA damage and protein and thus reducing oxidative damage to the skin[21].

This study showed a predominance of females in 19 (79.2%) of the controls and 39 (65%) of cases, similar to the results of Mantese et al.[22], Becher et al.[23] Spindola et al.[24], which also performed studies in patients with NMSC. This result is due to an earlier search of health services by women, making them more aware of their own bodies and yet they constitute the target audience for several educational campaigns against cancer, alerting them about the problem, its risk factors and possible manifestations[23].

The average age was statistically higher in the case group compared to control (p = 0.0492). With advancing age there is an increase in ROS and RNS, depression of the immune system and reduction of DNA repair mechanisms, beyond the effects of cumulative sun exposure over a lifetime[22], thereby increasing vulnerability to NMSC.

The mean values of anthropometric indices AC, TSF and MAMC suggest accumulation of fat mass and lean mass. The classification of nutritional status showed 51.7% in normal weight and overweight / obesity in 48.3% of cases by BMI, signaling to the low nutritional impact caused by NMSC, since it has a high cure rate and effective treatment with the removal of skin lesion. The NMSC does not cause digestive and absorptive disorders that compromise the nutritional status of their bearers, which favorably affects the mental and emotional conditions, providing welfare, better appetite and greater freedom in the selection of food, which can lead to increased body weight.

These current results are aligned with Tartari et al.[25], Azevedo and Bosco[26], who assessed the nutritional status of patients with cancer by BMI and found high levels of eutrophication and / or overweight.

The mean BMI of the cases was not according to official recommendations contained in the global perspective on food, nutrition and cancer prevention[27] report, what could be a factor that would contribute to the generation of oxidative stress.

The average age of the group was 62.7 ± 14.2 years, phase whose physiological changes due to aging can interfere with nutritional status, such as: decreased basal metabolism, redistribution of body mass, changes in digestive function, changes in sensory perception and decreased sensitivity to thirst[28]. Overweight and obesity also find support in a positive energy balance associated with physical inactivity. When calorie intake exceeds energy expenditure will increase accumulation of adipose tissue and weight, too. The greater the amount of energy, the more intense intense the energy metabolism, which produces to potentially harmful reactive molecules to cellular structures[29].

According to Farinatti[29], the lower caloric intake tends to attenuate the process of cell damage as you get older, with reduced lipid peroxidation, lower accumulation of oxidized proteins and oxidative DNA damage.

In respect to serum from CAOT, we can suppose that, in situations of oxidative stress evidenced, as noted in NMSC, there is mobilization of antioxidants to neutralize the oxidative burst· suggesting an increase in antioxidant defense mechanisms along the time of exposure to oxidative stress. The endogenous antioxidant defense system is stimulated by oxidative stress, which, in turn, causes a metabolic defense response[32], thus explaining the increased concentrations of CAOT in cases compared to controls.

According to Yet et al. opinion[32], in situations of increased production of reactive species, there is a compensatory increase in the activity of antioxidant defense system in the body of patients; and their study detected increased activity of the antioxidant system in women with breast cancer, that we also observed when comparing the control group with the case group, although not statistically significant.

Excess weight recorded by BMI may contribute to the increase in isoprostane concentrations; this association finds support in the accumulation of adipose tissue, which leads to increased production of ROS and RNS, oxidative stress and concentrations of their markers. The age could also contribute to the pro-oxidant status in the case group, because as we get older, the total amount of antioxidants in the plasma begins to decrease but the amount of ROS and RNA increases[34].

Assessment of habitual food intake concerning to antioxidant nutrients in the control group showed that the average vitamin C and the minerals copper, zinc and selenium dietary concentrations exceeded the EAR carrying, with great probability of adequacy of intake of these nutrients. In the case group, only the average intake of selenium and copper exceeded the EAR, the average vitamin A, C and E and the mineral zinc dietary concentrations were lower than the values of EAR, therefore not reaching the recommended nutritional coverage, showing great probability of inadequate intake of these nutrients.

The findings of this study suggest to inadequate zinc intake in the study group, which is identified by an irregular and insufficient intake of protein sources, represented by food: beef, fish and eggs and still no reports of intake of whole grains, cereals integrals, nuts walnuts, almonds, wheat germ, nuts, pistachios, liver and other foods rich in this nutrient.

The probable deficit of vitamins C, A and E can be identified by an irregular and even occasional consumption of foods such as orange juice, cashew, cherry, pumpkin, orange, papaya, melon, mango, strawberry, liver, liver oil cod, carrots, sunflower oil, almonds, vegetable oils, seeds, almonds and others.

It is noteworthy that the classification of food consumption pattern concerning the antioxidant nutrients allowed to establish the predominance of quantitative deficit of intake of all nutrients evaluated in the case group, revealing conducive to installation deficiencies tables, disabling the body to enjoy the effects played by these antioxidant nutrients.

The correlation found in this study between CAOT and dietary concentrations of antioxidant nutrients in the control group suggests that dietary intake of vitamins A could induce a compensatory mechanism to oxidative stress, slowing the production of free radicals and slowing the depletion of antioxidant defenses, contributing to lower serum concentrations of CAOT in this group. And the results of the same correlation in the case group, highlighted the influence of the consumption of food sources of vitamin A in reducing oxidative stress, since the nutrient in focus behaves as an important antioxidant, acting as sweeper RL, freeing the body of these negative effects on the redox balance of the skin.

A proper, balanced diet, rich in antioxidants and nutrients acts as a preventive mechanism in generating RL, hence it can be inferred that the individuals who had NMSC, included in this study, did not adopt positive strategies capable to protect the skin.

The probability of nutrients inadequacy in focus, even not acting alone in the propensity to skin cancer, may contribute to the depletion of endogenous antioxidant mechanisms, generating an imbalance oxidant / antioxidant, which can lead to oxidative stress. If people continue the current food consumption, this will cause serious nutritional compromise, thus disabling the body to meet the demands antioxidative, now increased, due to previously diagnosed NMSC.

In face of the action of these nutrients in the prevention of oxidative stress and inflammatory stress, it becomes imperative to adopt eating habits that ensure adequate intake of food sources of these nutrients, as a preventive strategy to oxidative damage in the population studied.

Conclusion

The assessment of nutritional status showed high prevalence of overweight and obesity and excess fat and lean mass, reflecting the low impact of the disease on nutritional status. The maintenance of BMI in excess of the levels recommended by the World Cancer Research Foundation[27] proves to be able to explain the propensity for disease progression.

This fact suggests the need for nutritional counseling of these individuals, since adiposity may precipitate the pro-oxidant, increasing the chances of disease recurrence and cause other nutritional and metabolic disorders.

It was possible to confirm the previous diagnosis of NMSC led to the pro-oxidant status in the case group, probably due to the late effects of UVR, driven largely by the generation of a chronic oxidative stress, a chronic increase in free radicals, chronic inflammatory stress and depletion of antioxidant defenses. Factors such as being overweight, advanced age, striking sun exposure, alcohol consumption and smoking may also have contributed to the pro-oxidant status checked.

The mean dietary antioxidant nutrients concentrations of the case group pointed to high probability of inadequacy of vitamins A, C, E and the mineral zinc, not reaching the recommended nutritional coverage, favoring the pro-oxidant status verified in the cases, which greatly hinders the fight against free radicals produced.

References

1. National Cancer Institute. José Alencar Gomes da Silva, Coordination of Surveillance and Prevention of Cancer. 2014 estimates: incidence of cancer in Brazil. Rio de Janeiro: INCA, 2014. 126p.
2. Ministry of health. Brazil. Secretary of health, National Cancer Institute, Coordination of Prevention and Awareness. The situation of cancer in Brazil. Rio de Janeiro: INCA; 2006.120p.
3. Sampaio SAP, Rivitti EA. Dermatologia. 3ª ed. ampl. São Paulo: Artes Médicas, 2007.
4. Bariani RL, Nahas FX, Barbosa MVJ, Farah AB, Ferreira LM. Basal cell carcinoma: an updated epidemiological and therapeutically profile of an urban population. *Acta Cirúrgica Brasileira* 2006; 21(2).
5. Chinem VP, Miot HA. Epidemiologia do Carcinoma Basocelular. *An. Bras. Dermatol. Rio de Janeiro* 2011; 86(2): 292-305.
6. Narendhirakannan RT, Hannah MAC. Oxidative stress and skin cancer: an overview. *Ind J Clin Biochem* 2013; (apr-june) 28(2) :110-115.
7. Ichihashi M, Ueda M, Budiyanto A, Bito T, Oka M, Fukunaga M, Tsuru K, Horikawa T. UV-induced skin damage. *Toxicol* 2003; 189: 21-39.
8. Bickers DR, Athar M. Oxidative stress in the pathogenesis of skin disease. *J Invest Dermatolog* 2006;26(12):2565-75.
9. Vasconcelos SML, Goulart MOF, Moura JBF, Benfato VM, Benfato MS, Kubota LT. Reactive oxygen and nitrogen Species, antioxidants and oxidative damage markers in human blood: Main analytical methods for its determination. *Quim Nova* 2007; 30(5):1323-1338.
10. Svobodova A, Walterova D, Vostalova J. Ultraviolet light induced alteration to the skin. *Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub* 2006 Jul; 150 (1):25–38.
11. Ouhtit A, Ananthaswamy HN. A model for UV-induction of skin cancer *J. Biomed Biotechnol*. 2001; 1(1): 5–6.
12. Jellife DB. The assessment of the nutritional status of the community. *Monogr Ser World Health Organ. Geneva* 1966; 53:3-271. 271p.
13. Blackburn GL, Bistrian BR. Nutritional and metabolic assessment of the hospitalized patient. *JPEN* 1977; 1:11-22
14. Kuczmarski MF, Kuczmarski RJ, Najjar RM. Descriptive anthropometric reference data for older Americans. *J Am Diet Assoc* 2000; 100:59-66.
15. World Health Organization (WHO). Physical status: use and interpretation of anthropometry. Geneva; 2004.463p
16. Organização Pan-Americana da Saúde (OPAS). División de Promoción y Protección de la Salud (HPP). XXXVI Reunión del Comitê Asesor de Investigaciones en Salud – Encuestra Multicêntrica. Salud Bienestar y Envejecimiento (SABE) en América Latina e el Caribe. Informe preliminar. Washington, DC:OPAS; 2001. 19p.

17. Crispin SP, Ribeiro RCL, Panato E. Relative validity of a food frequency questionnaire for use in adults. *Rev Nutr* 2009; 22(1): 81-95.
18. Sichieri R, Everhart JE. Validity of a Brazilian food frequency questionnaire against dietary recalls and estimated energy intake. *Rev Nutr* 1998; 18(10):1649-59.
19. Willett WC, Stampfer MJ. Total energy intake: implications for epidemiologic analyses. Am J Epidemiol 1986; 124:17-27.
20. Tatsch E, Bochi GV, Pereira RS, Kober H, Oliveira JR, Moresco RN. Effects of anticoagulants and storage temperature on blood nitrite levels. *J. Bras. Patol. Med. Lab.* 2011; 47(2): 147-150.
21. Poljsak B, Dahmane R. Free Radicals and Extrinsic Skin Aging. *Dermatol Res and Pract* 2012; 2012:135206. Published on line Feb 29, 2012. doi:10.1155/2012/135206.
22. Mantese SAO, Berbert ACLV, Gomides MDA, Rocha A. Basal cell Carcinoma - Analysis of 300 cases observed in Uberlândia-MG, Brazil. *An Bras Dermatol* 2006; 81(2):136-42.
23. Becker TCA, Baptista F, Soave JS, Weiler MA, Lacerda MAM, Alves VS, Inácio FD. Epidemiological profile of patients with skin cancer in the community of cities of the region of Campo Mourão - Parana in 2008. *Rev Saúde e Biol* 2010; 5(1): 51-58.
24. Spindola EO, Yamasato K, Tamashiro CS, Matos OMP, Figueredo SS, Pita MJ, Albuquerque LF, Duarte IB, Gunter HF, Sassioto IS, Pita MVC. Prevalence of skin neoplasia in Campo Grande, MS, 1993-2004. *Ensaios e C* 2006; 10(3): 161-170.
25. Tartari RF, Busnello FM, Nunes C H A. Nutritional Profile of Patients Submitted to Chemotherapy in a Tertiary Outpatient Clinic. *Rev Bras Cancerol* 2010; 56(1): 43-50.
26. Azevedo DC, Bosco MS. Nutritional, dietary profile and quality of life of cancer patients in chemotherapist treatment. *ConScientiae Saúde* 2001; 10(1): 23-30.
27. World Center Research Fund / American Institute for Cancer Research. Food, Nutrition, Physical Activity and the Prevention of Cancer: a Global Perspective. Washington, DC: AICR, 2007. 537p.
28. Campos MTFS, Monteiro JBR, Ornelas APRC. Factors that affect the aged people food intake e a nutrição do idoso. *Rev Nutr* 2000; 13(3):157-165.
29. Farinatti PT. Biological theories of aging: genetic and stochastic approaches. Rev Bras Med Esporte 2002;8(4):129-138. Disponível em: http://www.scielo.br/scielo.php
30. Yeh CC, Hou MF, Tsai SM, Lin SK, Hsiao JK, Huang JC, Wang LH, Wu SH, Hou LA, Ma H, Tsai LY. Superoxide anion radical, lipid peroxides and antioxidant status in the blood of patients with breast cancer. *Clin Chim Acta* 2005; 361:104-111. PubMed. PMID:16009358.
31. Kadiiska MB. Gladen BC, Baird DD, Germolec D, Graham LB, Parker CE, Nyska A, Wachsman JT, Ames BN, Basu S, Brot N, Fitzgerald GA, Floyd RA, George M, Heinecke JW, Hatch GE, Hensley K, Lawson JA, Marnett LJ, Morrow JD, Murray DM, Plastaras J, Roberts LJ 2nd, Rokach J, Shigenaga MK, Sohal RS, Sun J, Tice RR, Van Thiel DH, Wellner D, Walter PB, Tomer KB, Mason RP, Barrett JC. Biomarkers of Oxidative Stress Study II: Are oxidation products of lipids, proteins, and DNA markers of CCl_4 poisoning? *Free Radic Biol Med*. 2005;38 (6): 698-710. PubMed. PMID: 15721980
32. Yeh CC, Hou MF, Tsai SM, Lin SK, Hsiao JK, Huang JC, Wang LH, Wu SH, Hou LA, Ma H, Tsai LY. Superoxide anion radical, lipid peroxides and antioxidant status in the blood of patients with breast cancer. *Clin Chim Acta* 2005; 361:104-111. PubMed. PMID:16009358
33. Bogdan Allemann, I.; Baumann, L. Antioxidants used in skin care formulations. *Skin Therapy Lett* 2008; 13(7): 5-9.