The effects UV light exposure on hair zinc concentration in vitro

UV ışığa maruziyetin saç çinko konsantrasyonu üzerine *in vitro* etkileri

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ABSTRACT

Purposes

Low hair zinc level has been shown to be a good indicator of mild to moderate zinc deficiency. Ultraviolet (UV) radiation has mutating effects on cell DNA and induces inflammatory factors in cells. The aim of this study was to observe the in vitro effect of UV exposure, on zinc concentration of hair samples.

Material and Method

Hair samples of 72 patients were included into the study of whom the clinician suspected of zinc deficiency. Hair samples were divided into three group and treated with a phototherapy device (Davlin USA model 3 series PC/SP 311/48) and exposed to either 16,5 j/L UVA, or 4910 mj UVB or both UVA and UVB together, for 8 times within 24 hour intervals. Zinc measurements of hair sample pairs (UV exposed or not) were analyzed concurrently by atomic absorption spectrophotometry (AAS).

Results

Hair samples of 72 patients (female 45, male 27) were included into the study. Mean age of patients was 6.1±3.6 years (min-max 1.0-15 years). Zinc concentration of hair was 120.9±82.0 µg/gr before UV exposure and 109.2±71.0 µg/gr after UV exposure (p=0.002). Decrease in hair zinc was not associated with gender and was significant in group both UVA and UVB exposed (p=0.01).

Conclusion

Hair zinc level was significantly reduced in UV exposed samples in concurrently analyzed before and after UV exposed hair samples. The most decrease was with UVA+UVB exposure which is also present in the sunlight. Further investigations with *in vivo* studies also lightening the mechanism of zinc decrease in hair are needed.

Keywords: hair, UVA, UVB, zinc

ÖZET

Amaç

Düşük saç çinko seviyesinin hafif-orta çinko eksikliğinin iyi bir belirteci olduğu gösterilmiştir. Ultraviole (UV) radyasyon hücre DNA'sı üzerine mutasyona neden olur ve hücrelerde inflamatuar faktörleri indükler. Bu çalışmanın amacı, UV ışığa maruziyetin saç çinko konsantrasyonu üzerine in vitro etkilerini gözlemektir.

Gereç ve Yöntem

Klinisyenin çinko eksikliğinden şüphe ettiği 72 hastaya ait saç örnekleri çalışmaya dahil edildi. Saç örnekleri 3 gruba ayrıldı ve fototerapi cihazı ile işlem gördü (Davlin USA model 3 series PC/SP 311/48). Saç örnekleri 24 saatlik aralıklarla 8 kere, 16,5 j/L UVA, 4910 mj UVB ve UVA+UVB ye maruz bırakıldı. Saç örneği çiftlerinde (UV'ye maruz kalan ve

kalmayan) çinko ölçümleri eş zamanlı olarak atomik absorpsiyon spektrofotometresi (AAS) ile yapıldı.

Bulgular

72 hastaya ait saç örneği (45 kadın, 27 erkek) çalışmaya dahil edildi. Hastaların yaş ortalaması 6.1±3.6 (min-max 1.0-15 yaş) idi. Saç çinko konsantrasyonları UV maruziyetinden önce 120.9±82.0 µg/gr, UV maruziyetinden sonra 109.2±71.0 µg/gr (p=0.002) idi. Saç çinko konsantrasyonundaki azalma cinsiyetten bağımsız idi ve UVA ve UVB'ye birlikte maruz kalan grupta anlamlı idi (p=0.01).

Sonuç

UV maruziyeti öncesi ve sonrası eşzamanlı analiz edilen saç örneklerinden UV'ye maruz kalan örneklerde saç çinko düzeyi anlamlı şekilde azalmıştı. En fazla azalma, güneş ışığında da olduğu gibi UVA+UVB maruziyetinde görüldü. Saç çinko düzeyindeki azalmanın mekanizmasını aydınlatacak ileri in vivo çalışmalara ihtiyaç vardır

Anahtar kelimeler: Saç, UVA, UVB, çinko

INTRODUCTION

Zinc is an essential micronutrient for human health and has numerous structural and biochemical roles. Zinc is the intrinsic metal component or activating cofactor for more than 70 important enzyme systems, including carbonic anhydrase, the alkaline phosphatases, dehydrogenates, and carboxypeptidases. It is involved in the regulation of nucleoproteins and the activity of various inflammatory cells and plays a role in growth, tissue repair and wound healing, carbohydrate tolerance, and synthesis of testicular hormones (1-5).

Trace elements in hair, in particular zinc, are being widely investigated (6-9). Low hair zinc level has been shown to be a good indicator of mild to moderate zinc deficiency (10-12). The

unique property of hair is that it can provide a history of the mineral status of an individual. Minerals once incorporated into the hair are no longer in any dynamic equilibrium with the rest of the body. Thus the concentration of minerals in hair at a given distance from the scalp should reflect the mineral concentration in an individual at a particular and specific earlier time. It has been shown that zinc levels in the hair reflect zinc status well and individuals known to have a zinc deficiency have significantly lower zinc levels than normal values (13). Human hair is a convenient, readily available sample and Atomic Absorption Spectrophotometry (AAS) is the sensitive, accurate and relatively interference free technique for the determination of zinc in hair.

Ultraviolet radiation (both UVA and UVB light) from the sun exerts a number of effects on scalp skin and hair. Below the surface UV radiation has mutating effects on cell DNA and induces inflammatory factors in cells. The oxidative damage caused by UV light alters the amino acids like especially tryptophan, cystine, tyrosine, and histidine in hair shaft proteins, degrades moisturizing lipids, and breaks down the melanin pigments that help protect hair from UV light (14-16). Free radicals can be generated during UV irradiation, X-ray or gamma radiation are usually the products of reactions catalyzed by metals. At high concentrations of free radicals, they may be important mediators of damage to cell structures, including lipids and membranes, proteins and nucleic acids, when oxidative stress occurs (17).

As the physiological function of zinc ion include influence on redox state, enzyme activity, gene transcription, energetic metabolism, cell cycle, cell migration and invasivity, apoptosis and proliferation, the disbalance in the levels of zinc causes interference in these systems, and has important consequences (3,18).

The aim of this study was to observe the in vitro effect of UV exposure, on zinc concentration of hair samples.

MATERIAL AND METHOD

Hair samples of 72 patients were included into the study who admitted to pediatrics or dermatology clinics with compliance of growth retardation or dermatitis of whom the clinician suspected of zinc deficiency.

Hair was used as a biopsy material reflecting the elemental status of the body. Hair samples were collected from the suboccipital area (the base of the back of skull), by cutting with stainless steel scissors. The proximal ends of the samples of hair were usually within 2 cm of the scalp, approximately 0.3 g, were used for the analysis.

Hair samples were divided into three group and treated with a phototherapy device (Davlin USA model 3 series PC/SP 311/48). Samples were then exposed to either 16,5 j/L UVA, or 4910 mj UVB or both UVA and UVB together, for 8 times within 24 hour intervals.

In each of these samples the procedures described below were applied before zinc analysis.

Acetone-water wash: Hair samples were placed in 100 ml beakers, covered with and agitated for 10 minutes with a mechanical shaker. The water was then decanted and replaced with acetone (Merck).The samples were washed three times with acetone each being agitated for a period of 10 minutes and subsequently rinsed with bidistilled-deionized water. After being washed the hair samples were dried at 100°C for 4 hours followed by cooling to room temperature and finally reweighed on an analytical balance.

Wet digestion: Hair samples were placed in 10 ml pyrex tubes and left to digest in 1 ml reagent grade nitric acid (Merck 925) overnight. All samples were covered with watch glasses and refluxed at 100°C for about 1 and half hours until a clear residue of remained. After cooling for one hour the digested samples were decanted into 25 ml volumetric flasks and brought to volume with added rinsings of bidistiled-deiodinized water.

Zinc measurements of hair sample pairs (UV exposed or not) were analyzed concurrently by atomic absorption spectrometry (AAS), using a Thermo Scientific™ iCE™ 3000 Series. The zinc hollow cathode lamp current was 5 mA and the wave length was 213.9 nm, slit width 0.5 nm. Standard solutions were run before analysis and for every 10 test samples for verifying the assay accuracy and maintaining the quality of the standard solutions. Results were expressed as µg/gr hair. Coefficient of variance was r^2=0,9996 with a stock standard concentration 1000 µg/mL. Reproducibility was 3.1%, lower limit of linearity was 0.0277 µg/mL.

Statistical analysis

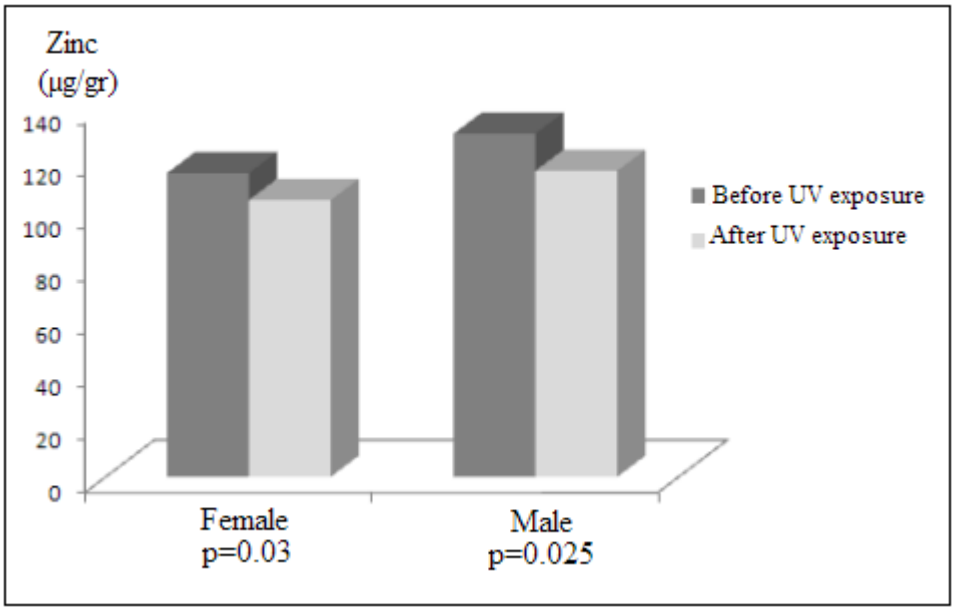
Mean±standard error of mean values of Zn of both groups in hair was calculated for each group. ANOVA analysis was used to compare both groups. The 95% confidence interval (CI) on the mean was calculated. Chi-square test was used to compare the cases out of 95% CI. All statistical analyses were performed using the statistical package SPSS version 20 (SPSS, Inc., Chicago, IL, USA).

RESULTS

Hair samples of 72 patients (female 45, male 27) were included into the study. Mean age of patients was 6.1± 3.6 years (min-max 1.0-15 years).

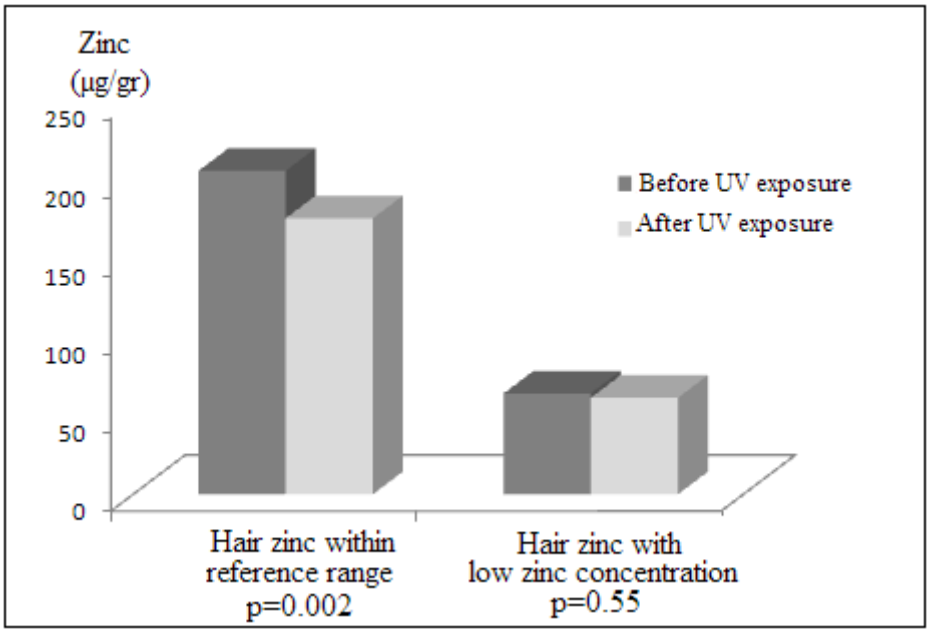
Firstly zinc concentration of hair samples were compared according to their UV exposure or not. Zinc concentration of hair was 120.9±82.0 µg/gr before UV exposure and 109.2±71.0 µg/gr after UV exposure. The difference was significant (p=0.002). Decrease in hair zinc was not associated with gender; in both male and female the decrease was significant (female p=0.03, male p=0.025, respectively) (Figure 1).

Figure 1: The decrease in hair zinc in male (n:27) and female (n:45) after UV exposure.



The cut-off level of hair zinc was determined as 100 (µg/gr). The decrease in zinc was significant in hairs within reference range (n:38, p=0.002), however not in hairs with low zinc concentrations (n:34, p=0.55) (Figure 2).

Figure 2: The decrease in zinc in hairs within reference range (n:38) and with low zinc concentrations (n:34).



Secondly hair samples were compared separately according to their exposure to UV type. Hair zinc concentrations of samples before and after different types of UV exposure (UVA, UVB, UVA+B) are shown in Table 1. Decrease in hair zinc was significant in group both UVA and UVB exposed.

Table 1. Hair zinc concentrations of samples before and after different types of UV exposure (UVA, UVB, UVA+B) are shown.

Hair zinc (µg/gr)	**UVA (n=24)**	**UVB (n=24)**	**UVA+ UVB (n=24)**
Before UV exposure	127.3±93.9	140.9±85.1	94.3±59.3
After UV exposure	119.0±84.7	131.4±68.5	77.1±44.7
p	0.184	0.19	0.01

DISCUSSION

In fact, ultraviolet (UV) radiation is considered to be the most damaging of all environmental factors. Hair has natural protection against the sun's rays. UV rays cause major changes in the mechanical ultra-structural and sensorial properties of hair, such as change of texture, a dry appearance, increase in porosity, loss of suppleness (19,20). The UV induced damage involves deep changes in the structure of keratin caused by the photo-oxidation of amino acids, sterols and fatty acids, resulting in rupture of sulfur bridges, decomposition of lipids, decrease in melanin as well as numerous micro-molecular lesions (13-16).

To our knowledge there is no investigation about the effect of UV light on hair zinc levels or zinc measurement. In this study we firstly investigated this effect in *in vitro* conditions. Hair zinc was significantly reduced in UV exposed samples in concurrently analyzed before and after UV exposed hair samples. The most decrease was with UVA+ UVB exposure which is

also present in the sunlight. The number of samples can be the limitation factor for the statistical significance in other groups.

Zinc stabilizes cell membranes, serves as an essential cofactor for several metalloenzymes, and participates in basal cell mitosis and differentiation (21). Although zinc (II) itself has no redox capacity, it is considered a potent and important antioxidant agent (22). Its antioxidant properties are due to both the direct and indirect interference with target structures. These include induction of metallothionein expression and glutathione synthesis of oxidant production, association with cysteines (with concomitant release by other oxidants) and regulation of redox signaling. Zinc (II) is an important regulator of glutathione (GSH) synthesis. The importance of zinc in the metabolism of glutathione underscores the finding that, as zinc deficiency is accompanied by oxidant increase, many studies reveal a deficiency of glutathione under such conditions (23,24). So UV exposure of hair can trigger the activity of zinc for antioxidant regulations.

Atomic absorption spectroscopy (AAS) is an important analytical technique based upon the absorption of radiation by free atoms. Virtually all metallic elements can be directly detected with excellent accuracy, precise quantitation, and very sensitive detection limits (25). According to the principle of AAS, the analyzed sample must contain the reduced metal in the atomic vaporized state. Commonly this is done by using the heat of a flame to break the chemical bonds and form free, unexcited atoms. AAS is sensitive, accurate, specific and specific. Disadvantages however are, sometimes the inability of the flame to dissociate samples into atoms or ionization of atoms following dissociation by the flame (26). In this study the UV exposure might have lead unexpected chemical bonds of zinc, or the used flame might have show insufficiency of dissociation of atoms. So the lower measurements of zinc can be caused by damaging effect of UV which leads to abnormal chemical bounds of zinc.

However this study needs further investigations with *in vivo* studies also lightening the mechanism of zinc decrease in hair.

Compliance with Ethical Standards:

Ethical approval: The Institution Review Board of Kayseri Training and Research Hospital approved the study protocol. The study was performed in accordance with the Declaration of Helsinki.

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Conflict of interest: The authors have no conflicts of interest or financial ties to disclose.

Informed consent: Informed consent was obtained from all individual participants included in the study.

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