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(RESEARCH ARTICLE)

Analysis of lipid peroxidation in the head hair of an urban population

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Abstract

Human head hair is considered a repository of information on metabolic changes, toxic exposures and diseases. Hair analysis has been investigated for environmental exposure to toxic principles such as heavy metals and is being considered as a simple and non-invasive tool for biomonitoring capable of complementing clinical investigation using biological samples such as blood, sputum and urine. Hair follicles contain lipids which are peroxidizable under oxidative stress occasioned by excessive reactive oxygen species. Thiobarbituric acid reactive substances (TBARS) are products of lipid peroxidation. This work aimed to provide data on TBARS concentrations in human head hair samples as a tool for assessing environmental exposure. Head hair samples were collected from apparently healthy female and male volunteers aged between 10 and 50 years (middle age). The samples were subjected to alkaline digestion, and spectrophotometric technique was used to assay for TBARS as a marker of oxidative stress. Significantly higher levels of TBARS were found among the female than the male volunteers. The teens and the adults (30 – 40 years) showed higher levels than the middle-aged class. There was no strong association of age with TBARS level. The results support previous study reports that gender is a predictor of hair lipid peroxidation.

Keywords: Hair; Oxidative Stress; Lipid Peroxidation; Thiobarbituric Acid Reactive Substances

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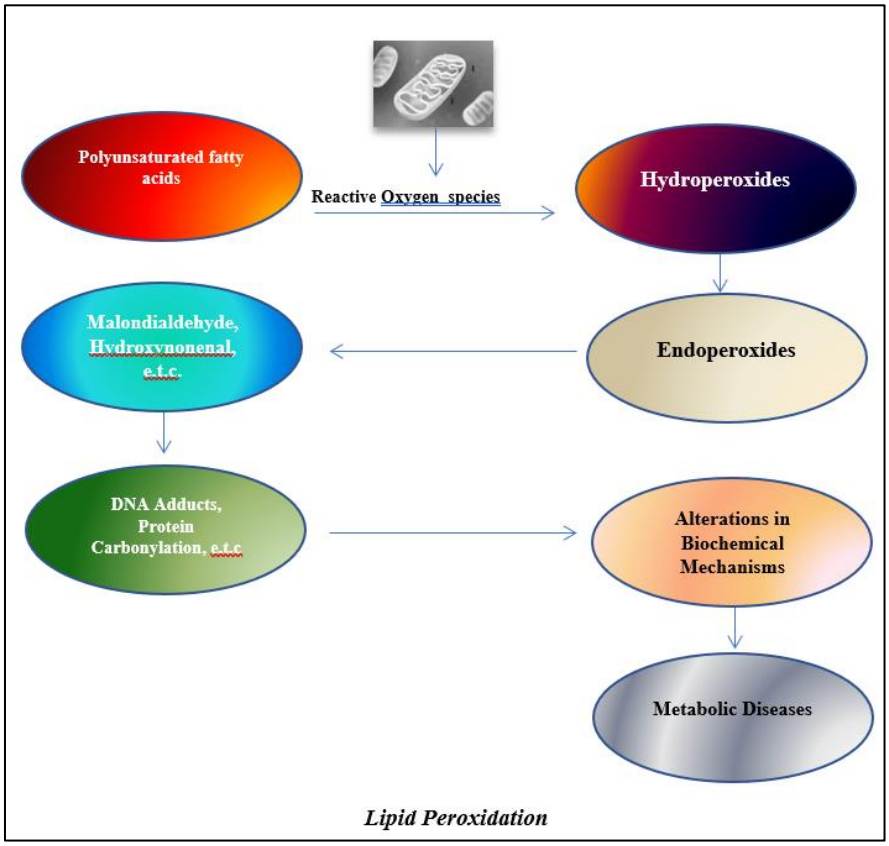
Graphical Abstract



1. Introduction

Human hair is a store of biochemical and environmental information over time. The analysis of human hair provides information on metabolic changes, environmental exposures, genetic predispositions, diseases and hormonal imbalances [1, 2, 3, 4, 5]. Its ease of accessibility, durability and as a store of biochemical information for an extended period of time makes it a sample of choice over any other biological samples such as blood and urine for forensic investigations. Hair is produced by the hair follicles that are rooted below the skin surface [6]. These hair follicles are open to stressors such as oxidants which interfere with hair cycle including anagen, catagen and telogen [7] leading to hair loss as a consequence of oxidative stress. The redox imbalance following over-production of reactive species relative to the antioxidant cascades culminates into the oxidative stress. The phenomenon is known to be a mechanism behind the etiology of certain non-communicable diseases such as diabetes, cancer and cardiovascular disease [8, 9, 10] as well as the 'pathological' loss of hair [11, 12]. Lipids peroxidation occurs following reactive oxygen species interaction with lipids in the cell membranes leading to modification of their permeability characteristics. Lipid peroxidation is one of the known indices of oxidative stress. The process also occurs in human hair follicles because of their contents of unsaturated lipids [11]. Aside as a predictor of environmental exposure, the hair analysis may provide an indirect screening test for pathological disorders in the body. Analysis of lipid peroxidation in the hair is considered simple and non-invasive tool for biomonitoring of cellular state of health or overall wellbeing of the human body [13]. Dearth of data from the hair analysis with respect to lipid peroxidation informed our decision to undergo the study.

2. Materials and Methods

2.1. Sample preparation and analysis

Each head hair (colored black) sample was obtained from the nape close to the scalp of the male and female apparently healthy volunteers, resident permanently in Ogbomoso town (8°09'48.2"N; 4°15'59.1"E), Oyo State Nigeria. The samples were carefully wrapped in polyethylene bags, labeled and their particulars such as hair color and hair treatment method were recorded. A total number of 72 samples of head hair from 49 males and 23 females were obtained from the volunteers aged 10 to 50 years. Each sample was minced and washed with acetone several times until a clear solution was obtained and finally rinsed in deionized water in a pyrex beaker. The sample was dried in an electric oven

at 105° C for 5 mins. The treated samples were stored in the desiccator at room temperatures (35 – 36 ^{0}C). Known weight (0.02 – 0.05 g) of the hair sample was digested with 2 ml 2 M NaOH solution and heated to 80^{0}C on a heating mantle for about 20 mins to form a solution. The digested samples were each centrifuged at 3500x*g* for 10 mins and spectrophotometric determination of thiobarbituric acid-reactive substances as an index of lipid peroxidation were carried out according to the method developed by Sheu *et al*. [13]. Briefly, color development was done by adding 1.5 ml 9% H_3PO_4 to 1.5 ml of the digest and mixed thoroughly. A 0.5 ml 30 mM thiobarbituric acid (TBA) was then added to the digest mixture, vortexed and placed in boiling water bath for 1 hr thereafter cooled under running water. The contents were mixed with 2.5 ml butan-1- ol and extracted by centrifuging at 1000x*g* for 20 mins. The butanolic extract was subjected to spectrophotometry at 534 nm. A sample blank contained 2.5 ml of butanol. The extinction coefficient was taken as 2.58 x 10^4 M^{-1} cm^{-1}.

2.2. Statistical analysis

Data were processed using GraphPad Prism software version 9.0 Results of the analysis were expressed as means ± SD. They were analyzed by 2-way ANOVA; $p < 0.05$ was taken for statistically significant difference. Correlation coefficient was carried out between age and TBARS concentration.

3. Results

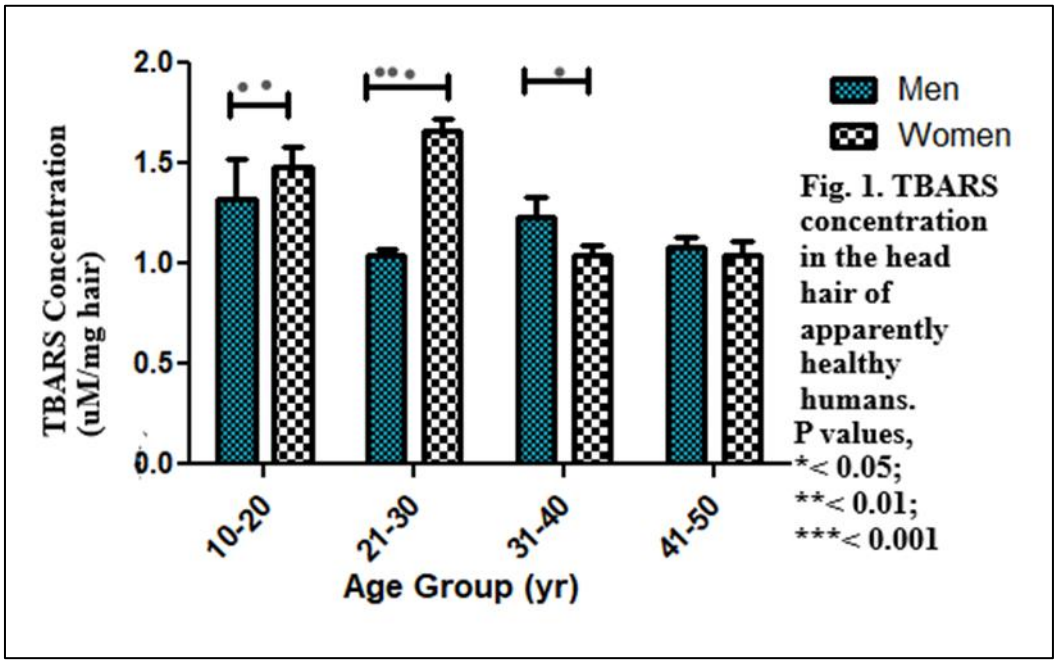


Figure 1 TBARS concentration in the head hair of apparently healthy humans**.**

The results of the analysis were shown in Figure 1. The highest value of thiobarbituric acid reactive substances (TBARS) in the head hair sample was recorded among the females age group 21 to 30 years (young adults). The females had significantly higher TBARS values than their male counterparts aged between 10 and 30 years. However, beyond 30 years old up to 40 years, the period of adulthood, the males had significantly higher TBARS. There was no significant difference in TBARS concentration between the genders at the onset of older adulthood or middle age (41-50 years). Pearson correlation coefficient between age and the genders gave – 0.5353 (r^2 = + 0.280) for male and –0.7889 (r^2 = + 0.623) for the female volunteers.

4. Discussion

TBARS are products of lipid peroxidation occasioned by excessive production of reactive oxygen species which lead to failed redox balance in the cell called oxidative stress**.** The products of lipid peroxidation namely α F2-isoprostanes, 4-hydroxynonenal (4-HNE) and malondialdehyde (MDA) are biomarkers of oxidative stress that underlie the general mechanism of some non-communicable diseases [14, 15, 16]. The increased TBARS recorded is therefore pathological. The observed differential in the TBARS levels among the genders is consistent with the widely held view that gender is

the greatest predictor of lipid peroxidation in human [17]. The highest TBARS levels recorded among the females could be ascribed to gender-related hair treatment methods among females which could have adversely affected the lipid content of hair follicles. However, this is at variance with the belief that female hormone estrogen shows antioxidant properties which male hormone testosterone does not [18]. Photosensitive lipids in human head hair medulla have been identified by Sandt and Borondics [19]. The exposure of head hair to the solar-sourced ultra-violet rays may produce light mediated conversion (photoconversion) and damage (photodamage) of the hair lipids. This may account for the higher TBARS recorded among the teens and the adults who are presumably more restless than the middle aged (≈ 50 yrs) class. But the correlation studies did not show a strong association of age with TBARS level probably supporting the reports of previous studies [20, 19, 21].

5. Conclusion

Head hair lipid peroxidation was affected by gender than by age. Further study should include larger samples and more sensitive technique than spectrophotometry should be applied.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical consideration

The authors received no financial support for the research, authorship, and/or publication of this article.

Statement of informed consent

The samples were collected from the donors with informed verbal consent.

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