NUTRITION

Comparison of Prooxidant-Antioxidant Balance between the Hospitalized Patients and the Healthy Subjects

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ABSTRACT

Objective: **In this study we aimed to evaluate the prooxidant-antioxidant balance (PAB) in the hospitalized patients in comparison with healthy subjects.**

Setting: **This cross sectional study was done from 2010 to 2012 in Mashhad University of Medical Sciences teaching hospitals.**

Methods: **The PAB assay, which is used in this study, measures the prooxidant burden and the antioxidant capacity simultaneously in one assay, thereby calculating a measure of redox status. One hundred seventy four patients who were admitted to the hospital, and 171 age and sex-matched healthy subjects were recruited to the study as case and control groups respectively. Anthropometric characteristics, biochemical parameters, prooxidant-antioxidant balance and serum high-sensitivity C-reactive protein (hs-CRP) were assessed in both groups. Statistical analysis was then preformed on the data.**

Results: **PAB values were significantly higher in case group as compared with the control group ($P < 0.05$). No significant correlation was observed in age, body mass index and hip circumference with serum PAB values PAB values in either group. Also serum levels of the albumin, pre-albumin, and hs-CRP showed no significant correlation with PAB values in the patient group ($P < 0.05$).**

Conclusion: **Being assessed by PAB assay, hospitalized patients demonstrated significant higher prooxidant antioxidant balance, as compared with the healthy subjects.**

KEY WORDS

pro oxidants, antioxidants, oxidative stresses, hospitalized adolescents

INTRODUCTION

Oxidative stress is defined as an imbalance between the pro-oxidants and antioxidants in favor of the pro-oxidants (Cherubini et al., 2005). Pro-oxidants such as reactive oxygen species (ROS) and reactive nitrogen species (RNS) are highly reactive molecules that may be derived either from metabolic processes or external sources (Alamdari et al., 2007, Johansen et al., 2005). They can potentially react with the body's own molecules such as proteins, DNA, and lipids and damage them (Griffiths and Lunec, 2001). ROS include free radicals such as superoxide ($\cdot O^2$-), hydroxyl ($\cdot OH$), peroxyl ($\cdot HRO^2$), hydroperoxyl ($\cdot HRO^2$-) and non-radical species like hydrogen peroxide (H_2O_2) and hydrochlorous acid (HOCl). RNS include free radicals such as nitric oxide ($\cdot NO$) and nitrogen dioxide ($\cdot NO_2$-), and non-radical species such as peroxynitrite (ONOO-), nitrous oxide (HNO_2) and alkyl peroxynitrates (RONOO) (Johansen, Harris *et al*., 2005).

Several studies have provided an insight into the role of oxidative stress in the pathogenesis of several conditions including atherogenesis, ischemic cerebral injury, hepatic damage, liver fibrosis, hypertension and cardiovascular disease (CVD) (Kelly *et al*., 2008, Pietrangelo, 1996, Rodrigo *et al*., 2008, Tara *et al*., 2010, Parizadeh *et al*., 2011, Ferrari, 2001) as well as some physiologic stresses like pregnancy (Boskabadi *et al*., 2013). Therefore, not only controlling oxidative stress especially in hospitalized patients could be a part of disease treatment and preventing complications, but also it seems that prooxidant antioxidant balance (PAB) values could be used as a prognostic factor for morbidity and mortality and monitoring the efficacy of treatment by estimating the extent of oxidative stress. In some previous studies, amounts of some oxidants and antioxidants in hospitalized patients have been investigated and remarkable results have been obtained (Powers *et al*., 2008, Mishra *et al*., 2005).

For the evaluation of the PAB, the determination of both oxidant and antioxidant status is often necessary. Although various methods have been proposed, but measuring the total oxidants and antioxidants are laborious, expensive, time consuming, and often imprecise.

The PAB assay, designed previously by Alamdari et al., is a simple, rapid, and inexpensive method that can measure the prooxidant burden and the antioxidant capacity simultaneously in one assay (Alamdari, Paletas *et al*., 2007).

Regarding limited data concerning pro-oxidant antioxidant balance levels in patients who have been admitted to hospital, also to assess the validity of PAB assay method, in this study we aimed to compare the prooxidant-antioxidant balance values of hospitalized patients with values for healthy subjects and assess whether there is a correlation between PAB values and biochemical parameters.

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Table 1. Patients and their diagnoses in different wards of the hospital

Wards (number of patients)	Diagnosis	Number of patients
Internal (52)	Esophageal cancer	20
	Icterus	32
Cardiac (31)	Myocardial Infarction	31
Burn Unit (36)	Burning injury	36
ICU (30)	Myasthenia	6
	Cerebrovascular Accidents	5
	Thrombosis	4
	Convulsion	3
	Subarachnoid Hemorrhage	3
	Sinusoidal Thrombosis	2
	Guillain barré syndrome	2
	Intracranial Hemorrhage	1
	Decreased level of consciousness	1
	Myelitis transversa	1
	Encephalopathy	2
Surgery (25)	Orthopedic	11
	Thoracic surgery	14

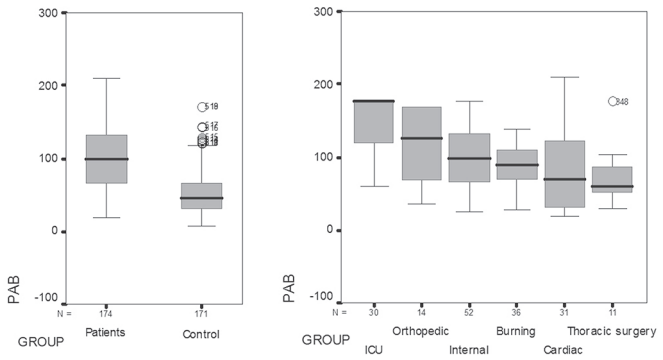

Figure 1. Distribution of PAB values of the patients versus controls and patients in different wards.

MATERIALS AND METHODS

Subjects

After obtaining approval from the Ethics Committee of Mashhad University of Medical Sciences, this cross sectional study was performed on 174 patients who were admitted to six wards of Ghaem Hospital (Mashhad, Iran) including internal, cardiac, intensive care unit (ICU), orthopedic, thoracic surgery and burn unit. Subjects who were consuming anti-inflammatory drugs except low doses of aspirin or a statin were excluded from the study. One hundred seventy one age- and sex-matched healthy subjects were recruited as the control group. Exclusion criteria for the controls were receiving any drugs and any known acute or chronic disease. Demographic and clinical data were obtained by direct interviewing with patients and their family.

Anthropometric measurements

Weight, height, body mass index (BMI), waist circumference, and hip circumference were measured for all subjects. Blood pressure was measured twice while the patients were seated and rested for at least 30 minutes using a standard mercury sphygmomanometer. Patients were considered smokers if they smoked ≥ 1 cigarette/day at the time of admission or in the preceding 12 months (Bassey, 1986). Total body fat, lean body mass and body water indices were investigated using a bio impedance analyzer (Body stat 1500 MDD, England) for hospitalized and healthy subjects.

Table 2. Comparison of Clinical and Biochemical Characteristics of the Patients and Controls

Characteristics	patients (n = 174)	Control (n = 171)	P value
Female (%)	45.4% (79/174)	27.5% (47/171)	< 0.001
Age (year)	47.10 ± 1.50	54.71 ± 0.73	< 0.001
Smoker (%)	19.0% (33/174)	7.6% (13/171)	< 0.001
SBP (mmHg)	113.8 ± 0.17	123.2 ± 0.11	< 0.001
DBP (mmHg)	70.6 ± 0.12	76.8 ± 0.09	< 0.001
Weight (Kg)	63.19 ± 1.12	74.18 ± 0.94	< 0.001
Height (cm)	163.24 ± 0.60	164.86 ± 0.63	0.37
BMI (kg/m^2)	23.64 ± 0.42	27.35 ± 0.32	< 0.001
WC(cm)	86.45 ± 1.33	97.72 ± 1.49	< 0.001
HC(cm)	100.55 ± 7.52	103.36 ± 1.26	< 0.001

SBP: systolic blood pressure; DBP: Diastolic blood pressure; BMI: body mass index; WC: waist circumference; HC: hip circumference. Values are expressed as mean ± SEM. Between groups comparisons were assessed by t-test, chi-square test or Mann-Whitney test (P < 0.001).

Blood Sampling

Blood samples were collected from each subject. Collected serum was centrifuged at 2500 rpm for 15 minutes at room temperature and was collected after centrifuging at 2500 rpm for 15 minutes at room temperature and was stored at -8℃ prior to analysis.

Biochemical analysis

Stored frozen serum samples were used for measuring levels of pre-albumin, albumin and total protein.

Prooxidant-antioxidant balance assay

Prooxidant-antioxidant balance was determined according to a previously described method by Alamdari *et al*. (Alamdari, Paletas *et al*., 2007). This method uses TMB (3,3 5,5 tetramethylbenzidine) and TMB cation to measure the balance of oxidants and antioxidants simultaneously in one experiment by using two different kinds of reactions; one enzymatic reaction where the chromogen TMB is oxidized to a color cation by peroxides; and a chemical reaction where the TMB cation is reduced to a colorless compound by antioxidants. In this method the mixture of hydrogen peroxide and uric acid has been chosen as representatives of oxidants and antioxidants, respectively (Alamdari, Paletas *et al*., 2007).

Inflammatory parameter measurement

Serum high-sensitivity C-reactive protein was measured by a polyethylene glycol (PEG)-enhanced immunoturbidimetry method with an Alycon analyzer (ABBOTT, Chicago, IL, USA).

Statistical analysis

Data analysis was carried out using Statistical Package for the Social Sciences (SPSS, Apache Software Foundation, and Chicago, IL, USA, 18th release). All data were checked for normality and presented as mean ± standard error of the mean (SEM) or, in the case of non-normally distributed data, as median with the 25th to 75th inter-quartile ranges. General linear model was used to adjust confounding variables in two groups. Comparison between groups was performed using t-test for parametric data and Mann-Whitney test for nonparametric data. Correlations among serum biomarkers and anthropometric parameters with PAB were evaluated using standard two-tailed bivariate correlation