

ORIGINAL ARTICLE

Assessment of The Total Oxidant Levels In People With Different Lifestyle In Al-Diwaniyah

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Abstract

Background Reactive oxygen species (ROS) are formed as a result of different metabolic reactions that occur normally in the human body, these free radicals harm the cell DNA, and other cellular structures and represent a waste product that must be eliminated from the body. Lipid peroxidation is one of the major outcomes of free radical-mediated injury that directly damages membranes and generates a number of secondary products including aldehydes such as MDA, which is the most abundant individual aldehyde, resulting from lipid peroxidation.

Objective The purpose of this study is to Evaluate the total oxidant levels among people with different social classes in the Al-Diwaniya population, Iraq. The possible relationship among serum of SOD, MDA and CRP in patients with obesity to identify the difference in the levels of total antioxidants with people having different lifestyles.

Materials and Method Samples were obtained from the Department of Medical Laboratories and Internal Medicine Patients registered with the clinic at Diwaniyah General Hospital Between February 1 and April 1, 2021. Blood samples were used to analyze biochemical properties of serum, such as SOD and MDA. It was evaluated in patients using ELISA technology. Statistical analysis was carried out using GraphPad Prism 9.2.0.

Results Our study in MDA (P-Value <0.0001) concentrations compared to Medium Work and Easy Work are significant. The results in SOD in the mean values between Hard Work and Medium Work (P-Value = 0.2279) is non-significant. The results in CRP in the mean values between Hard Work and Medium Work (P-Value <0.0001) are significant.

Conclusion The level of Free Radical (Oxidative Stress) was elevated in the hard work as compared with medium work and easy work. The level of SOD, MDA and CRP elevated in hard work as compared with medium work and easy work.

Keywords: Total antioxidants; Oxidative stress; Obesity; CRP.

1 Introduction

Oxygen species that are highly reactive (ROS) and formed as a result of different metabolic reactions that occur normally in the human body, these free radicals harm the cell DNA, and other cellular structures and represent a waste product that must be eliminated from the body [1]. The formation of ROS and cellular oxidative damage may be a component of the developmental toxicological processes of drugs and lifestyle factors. Oxidative stress occurs when ROS levels are very high. ROS production overwhelms antioxidant defenses, it results in cellular macromolecule oxidative damage [2]. Attempts are made to provide an updated basis for future studies on the toxic effect of such pollutants, in particular the notion of increased risk for developmental toxicity due to combined and cumulative exposure to a variety of environmental pollutants, on oxidative stress in the mechanism of developmental toxicity [3]. In any case, residential thinks about the impacts of different ways of life designs on ROS have been insufficiently conducted, considering subjects were constrained to certain populaces. Subsequently, this ponders inspected the impacts of the way of life, eating propensities, maladies, and other components within Al-Diwaniya province [4]. Plasma viscosity is important in treating infection, autoimmune diseases, and malignancies, since it is routinely used in primary care for both diagnosing and monitoring of inflammatory illnesses, is one of the most frequently used inflammatory markers. Overall, the rate of testing for inflammatory markers such as C reactive protein (CRP) has increased linearly over last 15 years [5]. While there is significant variation between General practitioner (GP) practices for test results that look for inflammatory markers, when it comes to the rates and frequency of abnormal outcomes, the inflammatory marker testing rate and prevalence are quite stable [6]. Malondialdehyde (MDA) is employed in medical research as a biomarker for oxidative stress. The synthesis of numerous active substances occurs as a result of lipid peroxidation, which causes cellular damage. A vital biomarker for hypertension, diabetes, atherosclerosis, heart failure, and cancer has been bio monitored in both in-vivo and in-vitro research. In patients who suffer from multiple types of diseases, such as lung cancer, complicated regional pain syndrome, and glaucoma, higher levels of MDA are detected [7]. It appears that the MDA assay, which has been found to be rather accurate, may also be used to discover oxidative stress in other disease pathologies. In this review, we'll focus on the ability of MDA to accurately and reliably Evaluate diverse medical conditions [8].

2 Materials And Methods

2.1 Study population

The study population comprised from three main groups people with hard work and people with medium work and people with easy work aged 18-60 years that in Iraq, Al-Diwaniyah Governorate.

2.2 Sample size and sampling

Samples collected from the general community from specific groups divided into three groups, represented by people who work hard, others who work medium jobs, and others who work easy jobs.

2.3 Exclusion criteria

People who were excluded from this study.

1. Children in general.
2. People with cancer.
3. People with diabetes.
4. Women who are expecting a child.
5. Patients with additional chronic conditions.

3 Data collection

3.1 Questionnaire interview

A meeting interview was utilized to complete a questionnaire prepared to meet the study requirements for both cases and controls. All interviews were conducted directly face to face. The interviewer explained any questions not obvious during the survey. The questionnaire was based on current oxidative stress studies. The majority of questions were questions of yes/no that offer a dichotomous choice. The questionnaire includes questions on the study population personal profile (age, gender and work) of socio-economic data (employment, obesity and smoking families, physical activity and food) among research populations.

3.2 Body mass index

Body mass index has been determined like the body weight ratio in kg/height in square meter. Before measuring weight and height, patients were requested to remove heavy garments and shoes. For weight measurement, the medical balance was employed. The BMI=18.5-24.9 were deemed normal weight, the BMI=25.0-29.9 had been designated overweight, and the BMI person(s)30.0 were deemed obese.

3.3 Specimen collection and processing

Venous blood samples were collected from 100 people. Blood samples (5 ml each) were drawn by a well-trained nurse into glass gel tubes to separate the blood and get the serum and was left for a while with an anticoagulant. Samples were obtained by centrifugation at 4000 rpm for 10 minutes for the determination of Ferric reducing ability of power (FRAP), magnesium ion, ascorbic acid, C-reactive protein, serum superoxide dismutase (SOD) and MDA.

4 Biochemical analysis

4.1 CRP rapid quantitative test

This kit applies to the in vitro quantitative determination of Human CRP concentrations in serum, plasma and other biological fluids. Specificity: This kit recognizes Human CRP in samples. No Significant cross-reactivity or interference between Human CRP and analogues was observed.

Principle: The Finecare TM CRP quantitative test is based on the immunoassay technology of fluorescence. This test uses a sandwich approach for immunodetection. When the sample is added to the test cartridge well, CRP antibodies labeled fluorescence detectors are attached in the blood specimen to the CRP antigen on the sample pad and form immune complexes.

4.2 Determination of Serum Malondialdehyde

Malondialdehyde is measured spectrophotometrically by a spectrophotometer.

Principle: The thiobarbituric acid (TBA) test measures malonaldehyde (MDA) produced due to the oxidation of fatty acids with three or more double bonds, and it measures other TBA reactive substances such

as 2-alkenals and 2,4-alkadienals. Therefore, TBA is also referred to as TBARs (TBA reactive substances)

4.3 Human SOD1(Superoxide Dismutase 1, Soluble) ELISA Kit

Human SOD1 concentrations in serum, plasma and other biological fluids can be quantified with this ELISA kit. Human SOD1 may be detected in samples with this kit thanks to its high level of specificity. Analogs and human SOD1 had no significant cross-reactivity or interference.

Principle: This ELISA kit uses the Competitive-ELISA principle. The micro ELISA plate provided in this kit has been pre-coated with SOD1. During the reaction, SOD1 in the sample or standard competes with a fixed amount of SOD1 on the solid phase supporter for sites on the Biotinylated Detection Ab specific to SOD1. Excess conjugate and unbound sample or standard are washed away, and Avidin-Horseradish Peroxidase (HRP) conjugate are added to each micro plate well and incubated. Then a TMB substrate solution is added to each well. The enzyme-substrate reaction is terminated by the addition of stop solution and the color turns from blue to yellow. The optical density (OD) is measured spectrophotometrically at a wavelength of $450 \text{ nm} \pm 2 \text{ nm}$.

5 Results

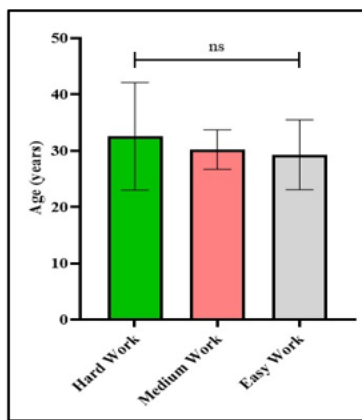
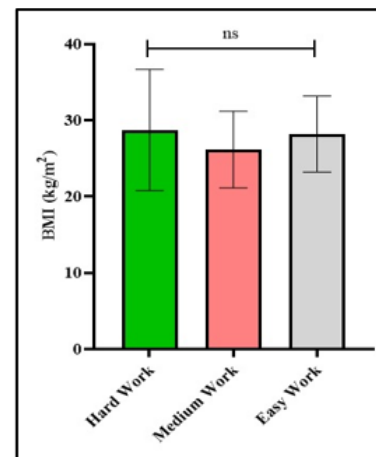
Demographic Characteristics of Hard Work, Medium Work, and Easy Work The Demographic characteristics of Hard Work (HW) with the Medium Work (MW) and Easy Work (EW) were all shown in Table 1.

The results of this study show mean age with HW elderly were (32.53 ± 9.55) years, but the age of MW younger was (30.27 ± 3.50) years and that of EW was (29.27 ± 6.19) years. There was no significant difference in mean age between HW, MW, and EW (P-Value = 0.1702). While there was an insignificant difference between MW and EW as shown in Figure 1.

Table 1: Demographic characteristics of hard work, medium work, and easy work.

Characteristic	Hard Work n=30	Medium Work n=30	Easy Work n=30	P
Age (years)				
Mean $\pm$ SD	32.53 $\pm$ 9.55	30.17 $\pm$ 3.50	29.27 $\pm$ 6.19	0.1702
Range	19-60	24-40	19-50	NS
BMI (kg/m ²)				
Mean $\pm$ SD	28.71 $\pm$ 7.93	26.14 $\pm$ 5.02	28.18 $\pm$ 4.97	0.2359
Range	17.51-64.45	19.53-38.06	18.52-35.16	NS
Gender				
Male, n (%)	23 (76.7%)	22 (73.3%)	20 (66.66 %)	0.139
Female, n (%)	7 (23.3 %)	8(26.7 %)	10 (33.33 %)	NS

*n: number of cases; SD: standard deviation; BMI: Body mass index; O: one-way ANOVA; C: chi-square test; NS: not significant at $p > 0.05$

**Figure 1:** Calculation of the Age in patients with hard work (HW) and medium work (MW) and easy work (EW).**Figure 2:** Calculation of mean body mass index (BMI (kg/m²)) in Hard Work (HW) and Medium Work (MW) and Easy Work (EW).

The body mass index (BMI) (kg/m²), with HW, was calculated at (28.71 $\pm$ 7.93) (kg/m²), that of MW was (26.14 $\pm$ 5.02) (kg/m²) and that of EW was (28.18 $\pm$ 4.97) (kg/m²) and there was insignificant difference in BMI among study groups as shown in Figure 2.

5.1 Estimation of oxidative markers concentrations (SOD, and MDA) and CRP in hard work, medium work, and easy work

The outcome of this study show increased levels of MDA (19.73 $\pm$ 4.84) μ M in hard work (HW) as compared with medium work (MW) and easy work (EW) (16.07 $\pm$ 5.71). (11.33 $\pm$ 3.52) μ M respectively; The outcomes of our study show high significant difference (P-Value < 0.0001) in the concentrations of MDA as compared with MW and EW. The results in mean values between HW and MW (P-Value = 0.0097) is significant, the mean values between HW and EW (P-Value < 0.0001) is highly significant, the mean values between MW and EW (P-Value = 0.0003) is significant. as shown in Figure 3.

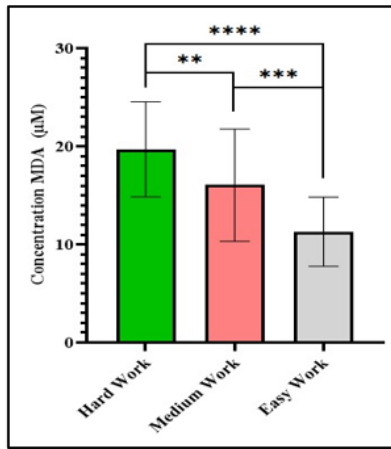


Figure 3: Calculation of oxidative stress concentrations measured as malondialdehyde (MDA (μM)) in hard work (HW) and medium work (MW) and easy work (EW).

The outcome of this study show decreased levels of superoxide dismutase (SOD) (3699.83 ± 192.50) (pg/ml) in hard work (HW) as compared with medium work (MW) and easy work (EW) (3789.33 ± 111.57), (3849.33 ± 133.06) (pg/ml) respectively; The outcomes of our study show significant difference (P-Value = 0.0018) in the concentrations of SOD as compared with MW and EW. The results in mean values between HW and MW (P-Value = 0.0316) is non-significant, the mean values between HW and EW (P-Value = 0.0017) is significant, the mean values between MW and EW (P-Value = 0.1131) is non-significant. as shown in Figure 4.

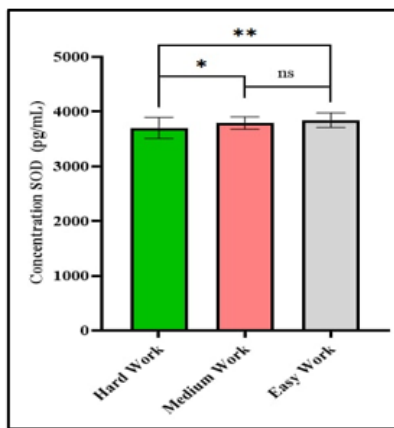


Figure 4: Calculation of Antioxidant levels by measuring the antioxidant enzyme superoxide dismutase (SOD) (pg/mL) in hard work (HW) and medium work (MW) and easy work (EW).

The outcome of this study show increased levels of CRP (9.30 ± 1.64) mg/L in hard work (HW) as compared with medium work (MW) and easy work (EW) (7.04 ± 2.02). (5.56 ± 1.70) mg/L respectively; The out-

comes of our study show a high significant difference (P-Value < 0.0001) in the concentrations of CRP. as compared with MW and EW. The results mean values between HW and MW (P-Value < 0.0001) is high significant, the mean values between HW and EW (P-Value < 0.0001) is high significant, the mean values between MW and EW (P-Value = 0.0032) is significant. as shown in Figure 5.

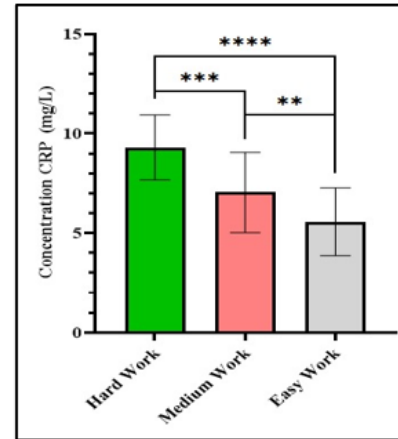


Figure 5: Calculation of inflammatory concentrations C reactive protein (CRP (mg/L)) in hard work (HW) and medium work (MW) and easy work (EW).

6 Discussion

Body mass index: Person weight in kilograms divided by their height in meters is known as their BMI. An easy and inexpensive way to determine a person's body mass index (BMI) ranges from underweight to obese [9]. There was an insignificant difference in BMI among study groups as shown in (Figure 2). All the participants in our study are classified as overweight and this consequence was agreed with the study [10] which showed that about two-thirds of persons in the United States have a BMI more than or equal to 25 kg/m^2 .

Obesity and being overweight may be linked to poor working circumstances, according to mounting research. Those who work long hours and in high-demand, low-control workplaces may be more likely to become obese. A fourth of the lives of medium and easy workers are spent in the workplace, and the stress and demands of their jobs may alter their food and exercise habits, which may contribute to obesity and overweight [11]. The outcome of the present study were agreed with the study [12] which there was no statistically significant association between psychosocial work characteristics and the change in body mass index, for both genders. The outcomes in our study were consistent with the studies [13]. The study found that the link between BMI, risk of obesity, and work charac-

teristics was not significantly different. Dietary, exercise and smoking changes have been linked to weight gain and may also be linked to employment circumstances [14].

C-reactive protein (CRP): Acute-phase Blood plasma contains proteins that the liver produces in response to macrophage and fat cell factors (adipocytes). It is an acute-phase hepatic protein that rises in response to macrophage and T cell production of interleukin-6.

The outcome of this study show increased levels of CRP in hard work (HW) as compared with medium work (MW) and easy work (EW). Increased levels of CRP in hard work may be due to increased physical activity. According to some researchers, the type, intensity, length and familiarity of the exercise all play a role in initiating a complicated cascade of inflammatory reactions [15]. The outcome of study were agreed with the study [16] which mentioned elevated levels of CRP in moderate work, and with the study [17] which revealed increased concentrations of CRP in intense work. The consequences of our study have disagreed with the study [18] which clarified CRP decreased among moderate workers by a large and modest amount, according to the results. People who engage in moderate physical activity have lower levels of CRP than those who are sedentary. When exercising at a high intensity, it appears that the immune system is affected. Furthermore, there is a widespread assumption that these alterations differ significantly from those following moderate exercise when they occur after hard activity [19].

Superoxide dismutase (SOD): Metalloenzymes known as SODs are enzymes that break down superoxide radicals into hydrogen peroxide and oxygen (H_2O_2), (O_2) respectively [20]. The outcome of this study show increased levels of SOD in hard work (HW) as compared with medium work (MW) and easy work (EW). The findings of our study were consistent with the study [21] which found elevated levels of SOD in people with hard work. To combat the rise in mitochondrial superoxide or other ROS sources, SOD acts as a first line of defense. The outcomes of our study were agreed with the study [22], which found an elevation in the levels of SOD six hours after the hard work and persisted for 24 hours. This study also revealed that this elevation was due to increased Extracellular SOD mRNA expression was significantly increased in skeletal muscle after severe exercise, as was aortic EC-SOD mRNA expression. SOD mRNA expression in skeletal muscle and the aorta can be increased by a single bout of exercise, according to these findings, which could explain in part why exercise is so helpful [23]. Organs with the highest amounts of SOD include

the kidney and lungs. Recent research suggests that endurance exercise increases the expression of SOD in our largest organ, the skeletal muscle, leading to higher levels of SOD in other organs peripheral to the skeletal muscle. Exercising can activate SOD's humoral function, which could be used to treat a wide range of diseases [24, 25].

Malonylaldehyde (MDA): Oxidative stress is characterized by the presence of MDA, an organic, highly reactive molecule that occurs naturally. Lipid peroxidation is responsible for its formation. Malondialdehyde is formed when reactive oxygen species destroy polyunsaturated lipids. The outcome of this study show increased levels of MDA in hard work (HW) as compared with medium work (MW) and easy work. These results were agreed with the result of another study [26] which showed that Moderate to hard-intensity work can increase reactive oxygen species (ROS), free radicals in the body, which is characterized by an increase in malondialdehyde (MDA).

7 Conclusions and Recommendation

1. The level of Free Radical (Oxidative Stress) where elevated in the hard work as compared with medium work and easy work.
2. The level of antioxidants where elevated in the easy work as compared hard work and medium work.
3. The level of MDA and CRP elevated in hard work as compared with medium work and easy work.

Conflict of Interest: No conflicts of interest exist between the authors and the publication of this work.

Ethical consideration: The ethical committee approved the study at Çankiri Karatekin University, Çankiri, Turkey.

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