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Mindfulness Meditation Promotes Reduction of Metabolic Stress and Antioxidant Defense in Hajjis

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Abstract

It is well established in the literature that mindfulness meditation helps in reducing metabolic and psychological stress, thus improving well-being. The present study aims at investigating this beneficial effect of mindfulness meditation in the context of the Islamic Pilgrimage (El-Hajj), through an oxidative and metabolic approach. Our experimental group includes 30 volunteer subjects who regularly practiced mindfulness meditation through Islamic prayer. The control group includes 30 subjects who do not practice prayer. Those who practice prayer (hajjis) are sampled one week prior to their departure to the pilgrimage and one week after their return from it. Stress parameters include Malondialdehyde (MDA), Catalase (CAT), and Cortisol. Metabolic parameters include control of fasting blood glucose, triglycerides, total cholesterol (TC), high-density lipoprotein (HDL), and low-density lipoprotein (LDL). Our results show a significant reduction in plasma cortisol levels ($p=0.014$) in the hajjis after return from the pilgrimage. This decrease is associated with a decrease in lipid peroxidation levels ($p = 0.082$) and a significant increase in antioxidant defense by catalase ($p = 0.0466$).

We suggest that mindfulness meditation through Islamic pilgrimage and prayer accounts for a downward modulation of plasma cortisol, MDA and the activation of antioxidant defense by catalase in hajjis after pilgrimage.

Key words: Meditation, Islamic prayer, Cortisol, Stress, Hajj, Malondialdehyde, Catalase.

1. Introduction

Increasing evidence is being uncovered on the practice of mindfulness meditation (MM) and its beneficial effects in terms of physical and mental health. Several authors report that MM appears to help in the regulation of emotions and mood disorders, such as anxiety, via a reduction in stress levels [1][2][3]. These findings are consequential since the stress factor is involved in the emergence and consequences of many pathologies [4][5]**.** Its origins can be traced to, among other things, an excessive production of free radicals and epigenetic modifications triggered by environmental and/or emotional variations[6]. The reduction of such free radicals relies in part on an enzymatic defense system such as catalase (CAT), superoxide dismutase (SOD), glutathione reductase (GPx), and paraxonase [7][8][9]. These are antioxidant enzymes that play an important role in reducing oxidative stress and are the basis for scavenging reactive oxygen species (ROS). Thus, antioxidants play an important role in the regulation of cellular signals [9]. To date, the most widely used marker for oxidative stress is malonaldehyde (MDA). MDA is indeed the main marker of lipid damage and oxidative stress induced by free radicals [10].

Catalase is a tetrameric enzyme that catalyzes the breakdown of hydrogen peroxide (H_2O_2) into oxygen and water. It is found in virtually all aerobic cells and is virtually responsible for protecting cells from the toxic effects of hydrogen peroxide [11]. A deficit in erythrocyte catalase thus causes an increase in the concentration of hydrogen peroxide [12]. In humans, it is involved in the antioxidant defense of several physiological and pathological conditions [13]. Antioxidant defenses are important in protecting healthy tissues and in preventing excessive activation of nuclear factor-kappa B (NF-κB) by the oxidative cellular environment during inflammation [14].This transcription factor is indeed the target of reactive oxygen species (ROS) and nitrogenous species (RNS) [15].

The stress response is governed by the stress system, which is located in both the central and peripheral nervous systems. Cortisol, a glucocorticoid hormone secreted by the adrenal cortex, is one of the main peripheral effectors of the stress system [16]. This hormone is regulated by the cortisol production axis via the hypothalamic-pituitary-adrenal axis (HHC) [17]**.** It has been shown that meditation practice is associated with a better antioxidant response; on the contrary, psychosocial stress is linked to an oxidative state [18].

El-Hajj, (or *The Pilgrimage* in arabic) is the Islamic pilgrimage to Mecca and its associated holy sites. It is one of the Five Pillars of Islam (alongside the five daily prayers) and is performed in the 12th month of the Islamic (lunar) calendar. During the days of the pilgrimage, pilgrims perform identical rituals, consisting in a series of religious and symbolic rites, following the footsteps of the Prophets Abraham and Muhammad. The pilgrimage often represents the culmination of years of preparation, both spiritual, physical and logistical. During the pilgrimage, the behavior of pilgrims should be characterized by piety and morality, abstention from all temptations, tolerance in relations with others, and avoidance of conflicts. According to Islam, every able-bodied Muslim should undertake the pilgrimage once in his or her life [19]. The Islamic prayer being also a form of meditation, we investigate its contribution to the reduction of the parameters of stress, both oxidative and psychological, in the particular context of the Islamic pilgrimage. More specifically, we are interested in identifying the relationship between oxidative stress and mindfulness meditation via an evaluation of some metabolic markers of oxidative status in Algerian praying pilgrims before and after their pilgrimage. These markers concern malondialdehyde (MDA) and catalase (CAT).

2. Methods

2.1. Participants

60 participants are chosen for this study and divided into two groups: a first group of 30 pilgrims who report practicing the five daily Islamic prayers (hajjis), and a second group of 30 healthy controls who do not pray. The mean age of the first group is 58.75±6.75 years while that of the controls is 30.24±9.24 years. All volunteers were recruited at the public health establishment of the Wilaya (state) of BLIDA, in the center-north of Algeria. The study has been approved by the Ethics Committee, Theme of Scientific Research in Health (ATRSS). All patients signed an informed consent form for this study.

2.2. Procedure

Blood samples for biochemical analyses are taken from the group of hajjis one week before departure for the pilgrimage and one week after their return from it. The control group are sampled only once. We exclude from our study subjects who are on corticosteroids. The characteristics of participants are presented in Table 1.

During the pilgrimage, hajjis perform identical rituals [20]. Upon arrival in Mecca, each hajji makes seven circumambulations (Tawaf) around the Kaaba (the building towards which Islamics pray). He or she then leaves for the plain of Arafat, a few kilometers east of Mecca, where the pilgrimate culminates in the "Day of Standing". Hajjis make overnight stops at Mina on the way to Arafat, and at Muzdaliffah on the way back. They stop on the way back at Jamarat to stone pillars which symbolize effigies of Satan. After a farewell Tawaf, hajjis leaves Mecca. Prayers must be performed five times a day as usual, consisting of specific movements, recitations and invocations.

Table 1: Epidemiological Characteristics of Pilgrims and Control Groups

	Pilgrims N=30	Controls N=30
Men n(%)	19(63.33%)	15(50%)
Female n(%)	11(36.66%)	15(50%)
Sex-ratio	19/11	1
Age (years)	58,75±6.75	30,24±9.24

*Age, is expressed as mean ± standard deviation; the categorical variables gender male (M) and female (F) are expressed as numbers and percentages (%).

2.3. Biochemical analysis

2.3.1. Metabolic parameters

Venous blood samples are collected at 8 am after overnight fasting (12 hours). Serum levels of glucose (Gly), triglycerides (TG), total cholesterol (TC), high-density lipoprotein (HDL), and low-density lipoprotein (LDL) are analyzed by spectrophotometric methods using the COBAS automatic analyzer. These variables are expressed in g/L.

2.3.2. Oxidative stress parameters

Cortisol levels are determined in plasma and quantified by the automated VIDAS Enzyme Linked Fluorescent Assay. The measurement of Malondialdehyde (MDA) by thiobarbituric acid (TBA) allows the quantification of lipid peroxidation which is the major marker of oxidative stress. This procedure is carried out by spectrophotometry and determined by a standardized assay method [21]. The reaction mixture consists of 100 µl of plasma, 200 µl of 20% thiobarbituric acid and 600 µl of 1% orthophosphoric acid. The concentration of TBARS is calculated by using the molar extinction coefficient ($1.56\times105M^{-1}$ cm^{-1}). It is expressed in µmol/l.

Catalase (CAT; EC 1.11.1.6) activity is analyzed using the method described by Claiborne (1985). The reaction mixture (1000µl) consists of 20µl of serum from each sample, 50 mM potassium phosphate (pH 7.0), and a 19 mM H_2O_2. The reaction is initiated by the addition of H_2O_2, and absorbance changes are measured at 240 nm (25°C) for 30s. The CAT activity is expressed as the unit that is defined as µmol of H_2O_2 consumed per min per mg of protein.

2.4. Statistical analysis

Results for the quantitative variables are expressed as mean ± standard deviation, while qualitative variables are expressed as percentages (%). "N" denotes the number in each group.

Fisher's homogeneity test is used to check for the distribution of quantitative variables. The unpaired t-test is used to compare the means of continuous variables with normal distribution between two groups. The Mann-Whitney U test is used to compare the means of continuous variables that are not normally distributed. One-way analysis of variance (ANOVA) Tukey test is used to compare continuous variables between the "pilgrims before pilgrimage", "pilgrims after pilgrimage" and "control" groups in the case where the variables are normally distributed. In contrast, the Kruskal-Wallis ANOVA test is used for continuous data that are abnormally distributed. The chosen level of statistical significance is a p-value of less than or equal to 0.05. The statistical analysis is performed using GraphPad version 6.01 (Software, San Diego, Californie, USA)

3. Results

3.1. Epidemiological characteristics of participants

The epidemiological characteristics of participants are represented in Table1. Our study shows a predominance of males in the hajjis' group with a sex ratio of 19/11, while in the control group, the sex ratio is 1. The control group is younger than the pilgrims' group.

3.2. Results of metabolic variables

Statistical analysis of metabolic variables and oxidative stress variables in the hajjis' group before and after pilgrimage in comparison with the control group is summarized in Table 2. Our results show no significant difference or abnormal changes for metabolic variables between hajjis and the control group, namely Gly, TG, CT, HDL, LD).

On the other hand, we found a significant decrease in cortisol levels in the hajjis' group before pilgrimage versus controls (p=0.0319) and a highly significant decrease after pilgrimage in comparison with the controls group (p< 0.0001) (Table2). If we compare the groups according to the period before and after pilgrimage, we find that pilgrimage had a significant effect in encouraging the decrease of cortisol levels (p= 0.0144) (Table2)

Table 2: Statistical analysis of metabolic variables and oxidative stress variables in the pilgrims' group, before and after pilgrimage in comparison with the control group

	Pilgrims		**Controls**	**P-value**			
	Befor N=30	After N=30	N=30	B vs A	B vs C	A vs C	ANOVA
Cortisol (ng/ml)	136.7 ± 7.23	114.3 ± 5.14	160.4 ± 7.98	0.0144*	0.0319*	< 0.0001***	< 0.0001***
Gly (g/l)	1,01±0,08	0,94±0,03	0,88±0,19	0,9367	0,2722	0,1445	0,2944
TG (g/l)	1,18±0,27	1,05±0,24	1,11±0,20	0,5782	0,6805	0,3254	0,5978

CT (g/l)	1,68±0,09	1,59±0,06	1,66±0,07	0,4743	0,7257	0,6657	0,7649
HDL (g/l)	0,43±0,13	0,40±0,03	0,46±0,02	0,8792	0,2587	0,1039	0,2377
LDL (g/l)	1,04±0,10	1,17±0,06	0,98±0,07	0,2257	0,6146	0,0592	0,1632
MDA (µmol/ml)	1,30±0,13	1,95±0,32	3,24±0,46	0,082	0,0289*	0,0023**	0,0054# #
Catalase (U/min)	1,20±0,10	1,95±0,32	1,85±0,20	0,0466*	0,0442*	0,7949	0,0743

Results are expressed as mean±standard deviation. B: Before (pilgrims before pilgrimage). A: After (pilgrims after pilgrimage). vs: versus; C: Control group; n= number; Gly: Glucose; TG: Triglycerides; TC: Total cholesterol; HDL: High-density lipoprotein; LDL: Low-density lipoprotein (LDL); MDA: Malondialdehyde; (##) : very significant for the one way ANOVA test; (*) : significant; (**) very significant.

3.3. Assessment of the MDA rate

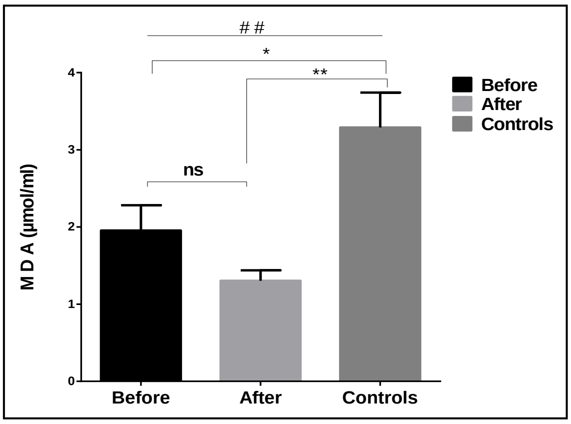

Fig1. Comparison of mean MDA values in the group of pilgrims before and after pilgrimage and the group of controls

#: Represents significance in the ANOVA test, (##): very significant, (*): significant, (**) very significant; ns: not significant

Our results show a significant increase in mean MDA levels in the control group compared to the hajji's group before pilgrimage (p=0.0289) (Figure 1). We also note a highly significant increase in MDA compared to hajjis after pilgrimage (p=0.0036) in comparison to controls.

We note a highly significant (p=0.0093) variation in MDA levels between the pre- and post-pilgrimage groups (Table 2).

Although our results show a 33% decrease in MDA levels in the hajjis' group after pilgrimage (1.95±0.32 µmol/ml) in comparison with the pre-pilgrimage period (1.30±0.13 µmol/ml), there was no significant difference between these two groups (p=0.082, ns).

3.4. Assessment of the catalase rate

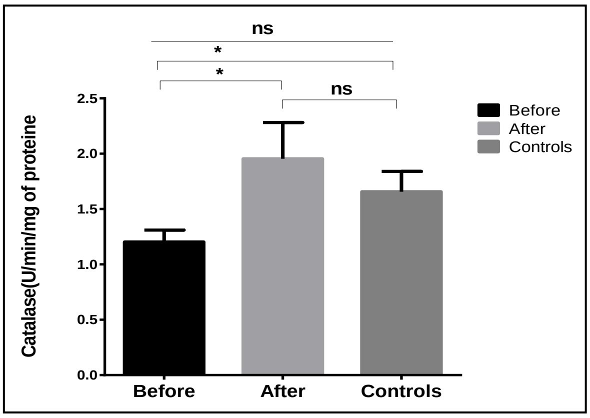

Fig2. Comparison of the mean values of catalase activity in the pre- and post-pilgrimage groups and the control group

#: Represents significance in the ANOVA test, (##): very significant, (*): significant, (**) very significant; ns: not significant

Our results show a significant increase (p=0.7949) in catalase activity in the hajjis' group post-pilgrimage compared to the control group, and a decreased without significant difference (p=0.0442) in the hajjis' group during the pre-pilgrimage period when compared to the control group.

However, there is a significant increase (p= 0.0466) in catalase activity in the post-pilgrimage hajjis' group (1.95±0.32U/min) when compared to the pre-pilgrimage hajjis' group (1.20±0.10U/min). The Tukey test shows no significant difference (p= 0.0743) for catalase activity between the three groups in our study (Table 2).

4.Discussion

To the best of our knowledge, this is the first study in the Maghreb and the Islamic world to examine the effect of pilgrimage and prayer via mindfulness meditation on markers of stress. Meditation is used to relieve mental and physical suffering[22][23][24]. It aims to find an inner balance that allows the reconnection of body and mind to maintain a healthy mind in a healthy body [25]. It is well established that subjects who do not practice mindfulness meditation are prone to constant stress [3][26]. Indeed, psychological stress develops in the long term into physiological stress via the production of free radicals, which are at the origin of several pathologies such as cancers [27][28], diabetes, cardiovascular diseases[29][30], cognitive disorders such as Parkinson's and Alzheimer's disease [31] and bipolar disorders [32].

Our study shows that the meditative characteristics of Islamic prayer during pilgrimage contributes to the reduction of oxidative stress. Indeed, we show a significant reduction of metabolic stress markers, namely MDA, Catalase and Cortisol, in pilgrims after their return from pilgrimage.

To this date, the most used marker in exploring the presence of stress remains malondyhaldehyde. It is the biomarker of choice for lipid damage during free radical-induced stress [10]. The increase in MDA in our study in the control group confirms the presence of oxidative stress. In contrast, the decrease in MDA levels in hajjis' group, particularly upon return from pilgrimage, suggests that mindfulness meditation promotes the reduction of lipid peroxidation through the decrease in MDA levels. Our results corroborate those of several studies that report a decrease in MDA levels in subjects who practice mindfulness meditation such as Tai Chi [33], Yoga [18], Zen [34], and Kouksundo meditations [35].

Stress is known to contribute to the mechanism of lipid peroxidation at the membrane level. This is a process during which oxidants such as free radicals or non-radical species attack lipids containing carbon-carbon double bonds, in particular polyunsaturated fatty acids (PUFAs). The latter have a protective effect against vascular pathologies [36]. Thus, an increase in free radicals or reactive oxygen species (ROS) can directly damage membrane lipids by promoting the increase of MDA [37]. Our results suggest that environmental, social or psychological stress is directly or indirectly involved in the mechanisms of lipid peroxidation at the membrane level by promoting the increase of ROS at the origin of lipid damage. Consequently, mindfulness meditation practiced by pilgrims plays an important role in the reduction of oxidative damage of membrane lipids such as polyunsaturated fatty acids (PUFAs), especially Omega 3 [38].

Grosso et al in 2014 used PUFAs such as omega-3 in a depressed population as an adjuvant rather than as monotherapy. Although their results revealed significant clinical efficacy, no correlation was found between the efficacy of Omega 3 and other variables such as study size, initial severity of depression, duration of the trial and patient age [39]. Also, it has been found that there is a close association between increased lipid peroxidation and psychosocial stress. This condition is usually accompanied by an increase in catecholamines and prostaglandins, whereas the practice of deep meditation such as Tai Chi, is associated with low levels of lipid peroxidation and lower sympathetic nervous system activity [40].

It has been shown in the literature that meditation is associated with a better antioxidant response, while psychosocial stress is linked to an oxidative state [18]. Our study confirms the improvement of antioxidant defense by catalase, which is an antioxidant enzyme, in praying pilgrims after pilgrimage. This increase in catalase activity levels positively affects the oxidative status of pilgrims. Our results corroborate with several studies that report a decrease in the levels of catalase activity in subjects who practice mindfulness meditation [18][41]. This confirms that high oxidative stress or low enzymatic antioxidant status are generally biological mechanisms associated with poor health outcomes and progression of a wide range of diseases [42]. Furthermore, meditation practice decreases markers of oxidative stress by increasing the transcriptional expression of antioxidant enzymes such as glutathione S-transferase, CueZneSOD (CueZn-superoxide dismutase), Mn-SOD, glutathione peroxidase (GPX) and catalase (CAT)[42] [43]. In fact, catalase plays an important role in the reduction of hydrogen peroxide to non-toxic substances [44]. On the one hand, our results suggest that the diminution of the antioxidant defense following environmental, psychological or moral stress, activates the pathway of lipid peroxidation. The latter could be due to the toxic effect of H_2O_2. On the other hand, the results of the cortisol dosage that we obtained confirm the beneficial effects of mindfulness meditation, by the reduction of the cortisol level reported in several studies [45][46]. Our results suggest that mindfulness meditation through Islamic prayer and pilgrimage decreases cortisol activation and regulates HHC. This decrease in cortisol levels is involved in oxidative damage. Furthermore, it has been shown that there is a strong correlation between oxidative damage and psychological stress, given that under conditions of psychological stress the production of mitochondrial ROS derivatives is accelerated and can cause an inflammatory state in healthy individuals [47][48][49].

5.Conclusion

There is a growing body of evidence that confirms the health benefits of meditation for body and mind. We find that mindfulness meditation during Islamic pilgrimage and prayer allows for spiritual satisfaction and reduces psychological stress. Through the results of our study, we can assume that the physical efforts and exercises during pilgrimage along with meditation through Islamic prayer activate antioxidant defense by increasing the levels of catalase enzyme activity. The latter reduces the toxic effects of hydrogen peroxide and free radicals. In addition, catalase is involved in the regulation of cortisol activity by decreasing oxidative stress, which could explain the molecular and physiological mechanisms that link meditation during pilgrimage to the praying pilgrim's well-being and health.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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