

Original Article

Effects of Changing Sleep Habits on the Metabolic and Hematological Marker: A Pilot Study

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Abstract

Objective: The aim of this intervention study was to investigate how changes of sleep habits affect hematological and biochemical markers in healthy adults.

Methods: Five groups of 10 participants (50 in total) were involved. One group was considered the control group while the others were asked to modify their sleeping habits in terms of bedtime (sleep at 10 pm), lighting, rock or relaxing music. Blood markers were studied at the beginning and at the end of seven consecutive days.

Results: This survey showed an increase in Potassium ($p=0,006$) and Creatine Kinase ($p=0,023$) in the group of participants that changed their bedtime habits. Alkaline Phosphatase ($p=0.039$) increased in those who slept with the presence of light. Lymphocytes ($p=0.008$), Glucose ($p<0.001$) and CRP ($p=0.013$) decreased after the end of this intervention. Sleeping with relaxing music led to a decrease in Albumin ($p: 0.018$) and CRP ($p<0.001$) while sleeping with to rock music increased levels of Triglycerides ($p=0.004$), Bilirubin ($p=0.020$) and Phosphorus ($p=0.023$) and decreased Uric Acid ($p=0.019$), Lactate Dehydrogenase ($p=0.013$) and Creatine Kinase ($p=0.044$). From the participants who changed their bedtime habits the 50% reported wakefulness and morning fatigue, nightmares, drowsiness and lack of appetite. All the participants who slept with light reported intermittent sleep, difficulty in waking up and morning fatigue. The participants who fell asleep listening to relaxing or rock music reported intermittent sleep, nervousness and appetite changes.

Conclusion: Negative factors that affect the quality of sleep can affect the biochemical and hematological markers and contribute to the outbreak or deterioration of illnesses.

Keywords: Sleep, Lighting, Music, Metabolic Diseases, Circadian Clock

Introduction

Sleep is a daily, necessary, complex and important biological process for all people, regardless of age, gender, or race. Its absence or possible disorders are associated with high risk of morbidity and mortality (AlDabal & BaHamam 2011). Sleep plays a significant role in the control of brain function and the human body, while the reduction of sleeping hours at night can possibly lead to serious endocrine and metabolic disorders affecting human physiology. Sleep deprivation and disorders have significant negative short-term and long-term health effects (Medic et al., 2017) contributing to the development of pathological conditions such as diabetes, hypertension, cardiovascular disorders, obesity and psychological disorders (Johnson et al., 2018). Sleep is a repetitive physical state that is regulated by endogenous circadian rhythms. These are periodical biological rhythms with a similar repetitive pattern of 24 hours which can be adapted to external stimuli such as light (Pavlova & Latreille 2019). Circadian rhythms are controlled by Melatonin (MEL) which is a hormone that is synthesized in the pituitary gland and regulates sleep cycle (Lee et al., 2019)

There are two phases of sleep, Non-Rapid Eye Movement sleep (NREM) which is divided into four stages and Rapid Eye Movement sleep (REM). Almost 4 or 5 NREM and REM sleep cycles occur during night sleep and alternate in a cyclical fashion (Altevogt & Colten 2006). In addition, sleep is actively involved in biological processes that are characterized by the secretion of hormones that are responsible for regulating metabolism. Among those hormones are growth hormone (GH), cortisol, melatonin (MEL), leptin (LEP) and ghrelin. GH hormone begins to be secreted a few hours after the onset of sleep, especially during slow-wave sleep and is affected by frequent awakenings. The regulation of daytime and nighttime cortisol secretion depends on the time of onset, duration and quality of sleep (Leproult & Van Cauter 2010). The synthesis and release of MEL from the pituitary gland is stimulated by darkness and suppressed by light, indicating its dependence on the circadian rhythm (Cipolla-Neto et al., 2014). Finally, Leptin secretion indicates maximum values of circadian rhythm between midnight to early morning hours and minimum values in the afternoon while sleep deprivation has been found to increase Ghrelin levels (Knutson et al., 2007)

Many factors contribute to sleep disorders including a variety of pathological conditions as well as factors related to lifestyle or environmental conditions (Medic et al., 2017). The latter can promote circadian rhythm disorders through various mechanisms. Artificial light during sleep can deregulate the circadian clock by suppressing melatonin secretion that consequently affects the onset of sleep (Johnson et al., 2018, Tahkamo et al., 2019). The use of light at night is considered a potential disorder as organisms are not normally prepared to deal with light signals at night. Light is converted into electrical signals by retinal receptors and transmitted through the optic nerves to the brain that are important for vision, autonomic nervous system function and the circadian clock. Exposure to light at night has been associated with an increased risk of obesity, diabetes and dyslipidemia (Opperhuizen et al., 2019) Auditory stimuli can lead to objective and adjective effects on sleep and cause metabolic and endocrine disorders such as increased adrenalin, noradrenalin and cortisol production, increased heart rate and blood pressure. During the night, the auditory stimuli increase the first stage of NREM sleep, reduce REM sleep at the same time and therefore affects the normal sleeping stages and the subjective quality of sleep (Muzet 2007)

The aim of this survey was to investigate the effect of a change in sleep habits on the hematological and biochemical markers.

Materials and Methods

Participants: A total of 50 adult volunteers participated in this intervention study. Age under 18 years old as well as cases of seriously ill participants (pathological or accidental cause of illness) were exclusion criteria for the present study. The participants were divided into five groups as follows: a) a group that changed its bedtime habits, specifically people who were asked to sleep at 10 pm, b) a group who slept with the light on (40 WAT), c) a group that was listening to relaxing music during sleep, d) a group that listened to rock music during sleep and e) a control group. The participants with music intervention were asked to listen to the same music (a particular web link was indicated). Volume was set to 40% or S2422Hz., The participants' demographic characteristics and sleep disorders that occurred during the study were recorded.

Hematological / Biochemical Markers: On the first and last day of the intervention study, fasting blood samples were collected for analysis. An analysis of hematological markers (Red Blood Cells - RBC, White Blood Cells - WBC, Hematocrit - HCT, Hemoglobin - HGB, Platelets - PLT) was performed, as well as an analysis of biochemical markers such as Fasting Glucose (FBS), renal function markers such as Urea (UREA), Creatinine (CREA) and Uric Acid (URIC), CRP as an inflammation marker of lipid profile, including Total Cholesterol (TC), Low Density Lipoprotein (LDL), High Density Lipoprotein (HDL) and Triglycerides (TGs), liver function markers such as Total Bilirubin (TBIL) and Direct Bilirubin (DBIL), Total Proteins (TPs) and Albumin (ALB), Transaminases (Serum Glutamic Oxaloacetic Transaminase - SGOT and Serum Glutamic Pyruvic Transaminase SGPT), Gamma-Glutamyl Transferase (γ GT), Alkaline Phosphatase (ALP), Lactate Dehydrogenase (LDH), electrolytes such as potassium (K), sodium (Na), calcium (Ca), magnesium (Mg), phosphorus (P) and enzymes such as Creatine Kinase (CK) and MB isoenzyme. Fasting blood sampling was performed at the same time for all participants, before and after the intervention. Analysis was performed using automatic analyzers Advia 2120i (Siemens Healthcare GmbH, Germany) and Olympus Au2700 (Olympus Europa SE & Co. KG, Hamburg, Germany).

Code of ethical conduct: This research study complies with the fundamental principles of ethical behavior which govern the conduct of research. The research protocol was also approved by the Ethics Committee of the University of Peloponnese and from the Panarkadian Hospital of Tripolis (1035, 15107, 23650 and ΓΝ/ΕΞ/2368-1/04/05/2015201). Participants were informed about the objectives and procedures of the study. A signed consent was obtained before the start of the survey.

Statistical analysis: A descriptive analysis of the sample (patients and control subjects) was conducted. Quantitative variables are presented as mean \pm standard deviation (SD) for normally distributed variables, and as median otherwise. In order to investigate whether there is a statistically significant correlation a) of the biochemical indicators before and after the intervention, independently of the intervention group the t-test was applied in pairs comparing two mean values and b) of the biochemical indicators before and after the intervention depending on the

intervention group applied the non-parametric Wilcoxon test to compare the median values in pair observations. In order to investigate whether there is a statistically significant difference in the values of biochemical indicators between the different intervention groups and compared to the control group, after the intervention, a one-way analysis of variance (ANOVA) was performed. The statistical analysis was performed using IBM SPSS Statistics 24 (SPSS, Chicago, IL, USA), while the significance level was set at 5%.

Results

Demographic data: A 84% of the participants in the intervention were women and 16% men with mean age $31,7 \pm 13,5$. 86% were born and live in the province. 66% of participants are married or in a relationship. Regarding the employment situation, 37.5% of the sample declare themselves unemployed and 39.6% are employed, 64% of the sample are students. The majority of the sample (84.4%) has an annual family income of up to 30,000 euros. The average value of the Body Mass Index (BMI, kg / m²) is 24.1 (6.0) kg / m². Demographic characteristics per group are depicted in each table.

Hematological / Biochemical Markers: Tables 1 summarize the results of the application of the t-test in pairs in order to study their mean difference before and after the intervention, per intervention group. Regarding the group which concerns the bedtime changing habits (Table 1), it appears that there is a statistically significant increase in the mean value of K and CK_{MB} (p-value <0.05). The study also reveals, in the group who sleep with the light on, that there is a statistically significant increase in the mean of ALP (p-value < 0.05) as well as a statistically significant reduction in the mean value of LY (Lymphocytes), CRP and GLUC (p-value < 0.05) (Table 1). Regarding the group who slept listening to relaxing music, statistical analysis also shows a statistical significant reduction in the mean value of ALB and CRP, (p-value <0.05) (table 1). Furthermore, concerning the participant who slept listening rock music it appears that there is a statistically significant decrease in the mean of URIC, LDH, CK and increase in the mean of TRIG, total bilirubin, and phosphorus (p-value <0.05). (Table 1). As far as the control group is concerned, a statistically significant increase only in the mean value of MCH was reported between two consecutive measurements. (Table 1). Finally, a statistical analysis was carried out comparing the

difference in the values of the biochemical and hematological indicators before and after the intervention (Table 2). The results confirm a statistically significant difference in the mean value of MPV, Glucose, Triglycerides, ALP and

CRP (p-value <0.05). The problems that were encountered by the participants during the study are listed in Table 3, and entail intermittent sleep, difficulty in waking up, morning fatigue and drowsiness throughout the next day.

Table 1. Results of the biochemical markers before and after the intervention (difference=value after intervention – value before the intervention).. SD: Standard Deviation. * statistically significant difference 5%. T- test was realized

Hematological / Biochemical Markers	Group 1 (Bedtime changing habits) Males 20% Females 80% Average age: 28.9 Years Old		Group 2 (Night lighting during sleep) Males 30% Females 70% Average age: 28.4 Years Old		Group 3 (Sleeping with relaxing music) Males 10% Females 90% Average age: 33.2 Years Old.		Group 4 (Sleeping with rock music) Males 20% Females 80% Average age: 23.11 Years Old		Group 5 (Control) Males 10% Females 90% Average age: 3 Years Old	
	Mean Difference (SD)	p-value	Mean Difference (SD)	p-value	Mean Difference (SD)	p-value	Average Difference (SD)	p-value	Average Difference (SD)	p-value
White Blood Cells (k/µl)	0.12 (1.29)	0.775	-0.8 (1.3)	0.086	-0.05 (1.55)	0.915	0.67 (1.8)	0.270	0 (0.05)	0.819
Lymphocytes (k/µl)	-0.12 (0.51)	0.479	-0.2 (0.18)	0.008*	0 (0.45)	0.989	0.31 (1.43)	0.511	0.01 (0.04)	0.324
Monocytes (k/µl)	-0.04 (0.07)	0.104	-0.02 (0.06)	0.436	0.05 (0.07)	0.071	0.01 (0.14)	0.823	0 (0.02)	0.619
Neutrophils (k/µl)	0.32 (1.52)	0.523	-0.63 (1.29)	0.158	-0.14 (1.3)	0.737	0.61 (1.03)	0.094	-0.02 (0.06)	0.305
Red Blood Cells (M/µl)	0.02 (0.15)	0.717	-0.04 (0.21)	0.509	-0.02 (0.25)	0.800	0.09 (0.21)	0.215	-0.03 (0.11)	0.462
Hemoglobin (g/dl)	0.05 (0.45)	0.734	-0.12 (0.61)	0.550	-0.04 (0.67)	0.854	0.23 (0.63)	0.280	0.05 (0.17)	0.381
Hematocrit (%)	-0.04 (1.47)	0.934	-0.26 (1.78)	0.655	-0.53 (2.7)	0.550	1.16 (2.07)	0.111	-0.11 (0.21)	0.129
Mean Corpuscular Volume (fl)	-0.13 (0.95)	0.677	0.39 (1.37)	0.392	0.68 (1.03)	0.067	0.55 (1.85)	0.372	0.1 (0.23)	0.204
Mean Corpuscular Hemoglobin (pg)	0 (0.22)	>0.999	0.06 (0.25)	0.460	0.1 (0.3)	0.316	-0.05 (0.76)	0.841	0.19 (0.19)	0.012*
Mean Corpuscular Hemoglobin Concentration (g/dl)	0.05 (0.55)	0.782	-0.05 (0.68)	0.821	0.77 (3.17)	0.462	-0.33 (0.86)	0.255	0.11 (0.25)	0.200
Red Blood Cell distribution Width (%)	0.12 (0.27)	0.187	-0.05 (0.28)	0.581	0.26 (1.21)	0.513	0.11 (0.41)	0.423	0.02 (0.25)	0.767
Platelets (k/µl)	-2.9 (15.17)	0.560	10.5 (29.9)	0.296	-5.6 (30.27)	0.573	-7.5 (45.73)	0.617	-0.3 (1.06)	0.394
Mean Platelet Volume (fl)	0.28 (0.44)	0.076	-0.23 (0.35)	0.069	-0.36 (0.59)	0.084	-0.15 (0.66)	0.489	7.98 (25.41)	0.347
Plateletcrit (%)	0.003 (0.01)	0.411	0 (0.04)	0.806	-0.02 (0.04)	0.169	0 (0.03)	0.662	0 (0.01)	0.223
Platelet Volume Distribution Width (fl)	0 (0.42)	>0.999	-0.52 (0.75)	0.055	-0.31 (1.42)	0.506	-0.11 (0.75)	0.668	-0.11 (0.33)	0.336
Glucose (mg/dL)	0.5 (7.18)	0.831	-16.2 (7.57)	<0.001*	-2.6 (13.55)	0.559	0.9 (12.59)	0.826	-1.3 (3.62)	0.286
Urea (mg/dL)	-0.8 (5.47)	0.655	-2.2 (9.22)	0.470	1.5 (5.34)	0.397	-0.6 (8.24)	0.823	-0.2 (1.4)	0.662

Uric acid (mg/dL)	-0.41 (0.61)	0.064	0.26 (0.78)	0.322	0.11 (1.28)	0.791	-0.56 (0.62)	0.019*	0.09 (0.14)	0.081
Cholesterol (mg/dL)	-3 (11.46)	0.429	1.6 (12.88)	0.704	1.3 (10.85)	0.714	-1.8 (20.85)	0.791	-0.4 (2.91)	0.674
High-Density Lipoproteins HDL (mg/dL)	1.7 (4.16)	0.229	5.5 (10.48)	0.131	0.7 (4.16)	0.608	2.4 (4.58)	0.132	0.4 (1.07)	0.269
Low-Density Lipoprotein LDL (mg/dL)	-5.6 (11.18)	0.148	-3 (6.63)	0.186	-3.8 (11.06)	0.306	-8.6 (19.37)	0.194	-1.9 (0.99)	<0.190
Triglycerides (mg/dL)	-2.4 (17.12)	0.668	-3.9 (14.43)	0.415	10 (18.36)	0.119	21.2 (17.38)	0.004*	-0.7 (1.83)	0.257
Creatinine (mg/dL)	-0.04 (0.07)	0.120	0 (0.07)	0.963	0 (0.07)	0.963	-0.03 (0.07)	0.221	-0.01 (0.02)	0.129
Total Bilirubin (mg/dL)	0.08 (0.29)	0.437	0.13 (0.4)	0.349	-0.06 (0.27)	0.483	0.1 (0.11)	0.020*	0.01 (0.06)	0.588
Direct Bilirubin (mg/dL)	0.02 (0.13)	0.700	0.04 (0.14)	0.385	-0.05 (0.09)	0.123	0.02 (0.05)	0.354	-0.01 (0.01)	0.132
Total Protein (g/dL)	-0.04 (0.28)	0.662	0.06 (0.33)	0.576	-0.15 (0.22)	0.062	0 (0.41)	>0.999	-0.04 (0.12)	0.309
Albumin (g/dL)	-0.03 (0.18)	0.616	-0.28 (0.8)	0.299	-0.15 (0.16)	0.018*	-0.12 (0.38)	0.343	-0.12 (0.1)	0.015
Calcium (mg/dL)	0.16 (0.57)	0.396	0.16 (0.41)	0.250	0.07 (0.54)	0.691	-0.19 (0.33)	0.106	-0.03 (0.11)	0.394
Phosphorus (mg/dL)	0.05 (0.33)	0.641	0.06 (0.39)	0.663	0.36 (0.59)	0.085	0.21 (0.24)	0.023*	-0.02 (0.14)	0.662
Magnesium (mg/dL)	0.02 (0.15)	0.629	-0.03 (0.07)	0.202	0.01 (0.17)	0.872	0.04 (0.1)	0.232	-0.06 (0.11)	0.111
Potassium (mEq/L)	0.29 (0.26)	0.006*	0.15 (0.31)	0.160	0.25 (0.48)	0.137	0.06 (0.4)	0.649	-0.03 (0.13)	0.468
Sodium (mEq/L)	0.8 (1.81)	0.196	-0.9 (1.79)	0.147	-1.1 (2.13)	0.137	-0.5 (1.84)	0.413	-0.1 (1.29)	0.811
Serum Glutamic-Oxaloacetic Transaminase (IU/L)	-0.3 (2.21)	0.678	0.7 (4.27)	0.617	-0.7 (1.77)	0.242	0.9 (5.67)	0.627	0.2 (1.03)	0.555
Serum Glutamic Pyruvic Transaminase (IU/L)	-0.4 (1.96)	0.534	1.1 (3.07)	0.287	0.5 (2.99)	0.610	1 (5.56)	0.583	0.3 (1.34)	0.496
Gamma-glutamyl transferase (IU/L)	-1 (1.49)	0.063	-0.3 (1.77)	0.604	-2.1 (3.75)	0.111	-1.5 (4.93)	0.361	-0.8 (0.92)	0.085
Alkaline phosphatase (U/L)	0.5 (3.5)	0.662	3.7 (4.83)	0.039*	-0.6 (3.78)	0.627	-1.2 (5.29)	0.491	-0.1 (1.29)	0.811
Lactate dehydrogenase (U/L)	13.7 (29.63)	0.178	-23.3 (47.86)	0.158	-1.7 (18.89)	0.782	-31.2 (32.1)	0.013*	0.7 (3.16)	0.502
Creatine Kinase (U/L)	-4.6 (26.25)	0.593	1.5 (20.92)	0.826	-23.1 (40.27)	0.103	-46 (62.01)	0.044*	-7.1 (19.76)	0.285
Creatine Kinase Isoenzyme CKMB (ng/ml)	2.7 (3.13)	0.023*	0.5 (2.76)	0.581	0.3 (2.98)	0.758	-0.1 (0.88)	0.726	-0.33 (0.71)	0.195
C-reactive protein (mg/L)	-0.06 (0.13)	0.170	-0.19 (0.2)	0.013*	-0.24 (0.13)	<0.001*	0 (0.15)	>0.999	0 (0.04)	0.829

Table 2. Comparative results of the correlation of biochemical markers between intervention groups. before and after the intervention. Bonferroni test was realized

Hematological / Biochemical Markers	Intervention Group (Average Value SD)				p-value
	Group 1	Group 2	Group 3	Group 4	
Lymphocytes (k/μl)	-0.12 (0.51)	-0.20 (0.18)	0.002 (0.45)	0.31 (1.43)	0.513

Hemoglobin (g/dl)	0.05 (0.45)	-0.12 (0.61)	-0.04 (0.67)	0.23 (0.63)	0.597
Hematocrit (%)	-0.04 (1.47)	-0.26 (1.78)	-0.53 (2.7)	1.16 (2.07)	0.285
Mean Corpuscular Hemoglobin Concentration (g/dl)	0.05 (0.55)	-0.05 (0.68)	0.77 (3.17)	-0.33 (0.86)	0.724
Platelets (k/ μ l)	-2.9 (15.2)	10.5 (29.9)	-5.6 (30.3)	0.3 (1.06)	0.594
Mean Platelet Volume (fl)	0.28 (0.44)	-0.23 (0.35)	-0.36 (0.59)	-0.15 (0.66)	0.050*
<i>Bonferroni</i>	0.010 group 1/3				
Plateletcrit (%)	0.003 (0.01)	0.003 (0.04)	-0.02 (0.04)	0 (0.03)	0.405
Platelet Volume Distribution Width (fl)	0.00 (0.42)	-0.52 (0.75)	-0.31 (1.42)	-0.11 (0.75)	0.605
Glucose (mg/dL)	0.50 (7.18)	-16.2 (7.57)	-2.6 (13.54)	0.9 (12.59)	0.003*
<i>Bonferroni</i>	0.001 group1/2. 0.008 group2/3. 0.003 group 2/4				
Triglycerides (mg/dL)	-2.4 (17.1)	-3.9 (14.4)	10.0 (18.36)	21.2 (17.38)	0.006*
<i>Bonferroni</i>	0.021 group1/4; 0.012 group2/4				
Calcium (mg/dL)	0.16 (0.57)	0.16 (0.41)	0.07 (0.54)	-0.19 (0.33)	0.315
Potassium (mEq/L)	0.29 (0.26)	0.15 (0.31)	0.25 (0.49)	0.06 (0.4)	0.522
Sodium (mEq/L)	0.80 (1.81)	-0.90 (1.79)	-1.10 (2.13)	-0.5 (1.84)	0.128
Serum Glutamic Pyruvic Transaminase (IU/L)	-0.40 (1.96)	1.10 (3.07)	0.50 (2.99)	1 (5.56)	0.7866
Gamma-glutamyl transferase (IU/L)	-1.00 (1.49)	-0.30 (1.77)	-2.10 (3.76)	0.80 (0.29)	0.347
Alkaline phosphatase (U/L)	0.50 (3.50)	3.7 (4.83)	-0.60 (3.78)	-1.2 (5.29)	0.079
Creatine Kinase (U/L)	-4.6 (26.2)	1.5 (20.92)	-23.1 (40.27)	-46 (19.76)	0.056
C-reactive protein (mg/L)	-0.06 (0.13)	-0.19 (0.20)	-0.24 (0.13)	0.003 (0.15)	0.004*
<i>Bonferroni</i>	0.052 group 2/4; 0.007 group 3/4				

Group 1: bedtime changing habits Group 2: night lighting during sleep Group 3: sleeping with relaxing music Group 4: sleeping with rock music SD: Standard Deviation *statistically significant difference 5%

Table 3. Problems reported during the intervention

	Group 1 n (%)	Group 2 n (%)	Group 3 n (%)	Group 4 n (%)
Difficulty with the onset of sleep	2 (20%)	3 (30%)	5 (50%)	2 (20%)
Intermittent sleep	5 (50%)	10 (100%)	4 (40%)	6 (60%)
Nightmares	4 (40%)	0 (0%)	1 (10%)	0 (0%)
Dreams	2 (20%)	0 (0%)	2 (20%)	3 (30%)
Difficulty in wakening up-Morning fatigue	5 (50%)	7 (70%)	3 (30%)	3 (30%)
Drowsiness in the next day	4 (40%)	8 (80%)	6 (60%)	4 (40%)
Hunger	2 (20%)	1 (10%)	1 (10%)	4 (40%)
Anorexia	4 (40%)	1 (10%)	0 (0%)	0 (0%)
Nervousness				
Stress	3 (30%)	5 (50%)	4 (40%)	0 (0%)

Group 1: bedtime changing habits Group 2: night lighting during sleep Group 3: sleeping with relaxing music Group 4: sleeping with rock music

Discussion

The results of the present study show that even a short-term change in sleeping habits can affect hematological and biochemical markers.

Sleep is an integral part of the well-being and normal functioning of the body and it is a condition whose attributes can be identified and modified to contribute to achieving optimal health-related conditions.

The present survey shows a statistically significant reduction in Glucose levels after the end of the intervention as far as the group that sleep with the presence of light is concerned. Though the highest Glucose levels were found in people who were sleeping by listening to rock music. Although music therapy has been shown to have positive health outcomes, it appears that the type of music played during sleep plays an important role in this effect. Surveys have shown that enjoyment and distraction depending on the kind of music could inhibit sleep causing sleep disorders (Dickson and Schubert 2019). Sleep has a causal relation with the homeostasis glucose regulation and appetite control, thus reduced sleep contributes to a high obesity risk and type II diabetes. (Briaçon-Marjollet et al 2015)

Regardless of the results of our research, recent studies reveal significantly higher levels of glucose and insulin regarding the sleeping group who slept with the lights on, thus indicating glucose intolerance and sensitivity, Albreiki et al (2017). Gil-Lozano et al. performed a control trial study to prove light exposure during sleep led to sleeping disorders and high insulin resistance (Gil-Lozano et al 2016). A recent study in rats, performed by Arasteh et al., showed that the continuous light exposure during night sleep led to high glucose levels without having any significant effects on serum cholesterol and insulin levels (Arasteh et al 2010). Regarding an experimental study performed by Opperhuizen et al. where male glucose tolerant rats were exposed for 2 hours to night light, it was found that exposure to night light had acute adverse effects on glucose metabolism, leading to acute intolerance of beta cells (Opperhuizen et al. 2017). However, it is worth mentioning that rodents are nocturnal mammals that present reverse activity and reverse resting cycles compared to humans (Albreiki et al 2017).

In the present study CRP levels decreased after the end of the intervention in regard to both the participants that slept with the light on and those who slept listening to relaxing music. The comparative study of hematological and biochemical markers between the participants that changed their bedtime habits and those who slept while listening to rock music, performed after the end of the intervention, revealed the value of this marker was at the highest levels compared to all groups. Inflammation is generally a condition that is affected by sleeping disorders. In particular, experimental studies have shown that levels of

cytokines can be affected by intermittent sleep. Cytokines are defined as peptides that are important tools for regulating inflammation. Several literature references indicate that inflammatory markers are mainly related to sleep duration and the quality of life that depends on. They are definitely not related to those parameters that are examined in the present study. Thus, Patel et al. also concluded that CRP values are increased when the total hours of night time are increased, while the reduction of night sleeping hours has not been proved to affect this marker (Patel et al. 2009). Another study that included several participants who slept only 4 hours for 10 consecutive nights, also showed the opposite results; namely high CPR levels (Haack et al 2007). It is worth mentioning that the study of Meier-Ewert et al. also proved that both total and short-term partial sleep deprivation led to increased CRP concentrations (Meier-Ewert 2004).

The present study showed an increase in triglyceride (TG) levels concerning the group that slept listening to rock music. In fact, the increase of the lipid marker value led to an indicative statistical difference between the specific intervention group and those who slept earlier than usual. Sleep fragmentation leads to a decrease in leptin concentration and eventually it functions as a biological mechanism which is responsible for short sleep duration and dyslipidemia correlation. Leptin reduces lipogenesis, increases fatty acid oxidation and hydration. As a result, the reduction of leptin levels contributes to high levels of serum triglycerides (Kaneita et al 2008, Stern et al 2016).

TGs as well as fatty acids show strong circadian fluctuations accompanied by progressively reduced levels during sleep. Sleeping activity enables the increase of lipoprotein lipase and fatty acid synthesis in fatty tissue (Briaçon-Marjollet, et al 2015). A group of researchers studied a population sample from Caucasus with diabetes and confirmed the same results. The participants, who had shorter sleeping hours than normal, also had higher TGs levels, indicating that disturbed sleep was associated with a disorder of the lipid profile which is also a risk factor of microvascular diabetes complications (Wan et al 2013). Despite of no significant differentiation of TGs in our study, regarding the participants who slept with the lights on, we should take into consideration that night lighting during sleep led to intermittent and generally disturbed sleep. A recent study of elderly people conducted by Obayashi et al.

showed that exposure to night light was related to high levels of plasma TGs (Obayashi et al 2013).

In addition to what has already been mentioned, the comparative results between the hematological and biochemical markers among the intervention groups, showed a statistically significant difference in potassium and serum phosphorus values. These results are related to the sleeping groups that changed their bedtime habits and those who slept accompanied by rock music. However, there are no literature reviews to argue or confirm our results and as a result there is a lack of comparative analysis. Despite its limitations, our study is the first to investigate and compare hematological and biochemical markers in groups of individuals who have changed their sleeping habits.

Study restrictions: The study restrictions concern the participants' small sample that couldn't possibly provide general conclusions as well as the non-uniform sampling distribution by age and gender. The comparative results of the study should also be related to variables such as age, gender, potential health problems, occupational and social conditions in order to add further scientific value to the study.

Conclusions: The present study of piloted intervention was performed in order to investigate the possible correlations of sleep disorders of different reasons with the participants' health condition. However, various aspects of modern lifestyle should also be taken into consideration, especially the increased use of digital media (smartphones, tablets, laptops), increased alcohol and caffeine consumption, intense physical activities as well as fast and stressful rhythms of life. These characteristics significantly contribute, among other things, to insufficient and poor quality of sleep. The degradation of sleep quality can possibly result in physical, mental and intellectual problems. Regarding the strong correlation between these problems and the increased rates of morbidity and mortality, further study of sleep-related disorders is imperative.

Data availability statement: The data that support the findings of this study are available from the corresponding author upon reasonable request

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