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The Effect of Vitamin C Supplementation on Malondialdehyde Levels and Psychological Stress in Medical Students of Yarsi University, Class of 2022

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Abstract

This study addresses the high prevalence of psychological stress among medical students, which can trigger oxidative stress characterized by increased Malondialdehyde (MDA) levels. Vitamin C, as a potent antioxidant, is expected to reduce oxidative stress and improve psychological conditions. Therefore, this study aims to examine the effect of vitamin C supplementation on MDA levels and psychological stress among medical students of YARSI University class of 2022. This research employed a quantitative experimental design using a Randomized Controlled Trial (RCT) approach. A total of 36 respondents were divided into four groups receiving vitamin C doses of 500 mg, 1000 mg, 1500 mg, and a control group. Data were collected through laboratory analysis using ELISA for vitamin C and MDA levels, and psychological stress was measured using the DASS questionnaire before and after intervention. The results showed that vitamin C supplementation at a dose of 1000 mg significantly reduced MDA levels and increased vitamin C concentration, while 500 mg and 1500 mg doses showed no significant effect on MDA. All supplementation doses contributed to reducing psychological stress, with the most consistent effect observed at 1000 mg and 1500 mg. In conclusion, vitamin C supplementation, particularly at a dose of 1000 mg, is effective in reducing oxidative stress and psychological stress among medical students, indicating its potential as a supportive intervention for student health.

INTRODUCTION

Stress is a physical, emotional, cognitive, and behavioral response to a situation that is perceived as a threat or challenge. Symptoms that can appear include unusual fatigue, sleep disturbances, anxiety, depression, fear, irritability, and anger and frustration. Mentally, stress often interferes with concentration, memory, decision-making, and loss of sense of humor (Ciccarelli and White, 2018). The prevalence of stress events according to the WHO in 2019 is quite high, being experienced by more than 350 million people in the world and ranked 4th in the world. According to previous research, around 1.33 million people in Indonesia are estimated to experience stress, equivalent to 14% of the population, with 1-3% of them classified as severe stress (Ambarwati et al. 2017; Nugraheni et al. 2020)

Psychological stress can trigger a series of reactions in the body, including increased oxidative stress. Oxidative stress occurs when redox homeostasis leads to higher levels of reactive oxygen species (ROS) in cells compared to the amount of antioxidants, which can result in damage to various biomolecules, including DNA, RNA, proteins, fats (lipids),

carbohydrates, and other (Peter J. Kennelly et al., 2023). biomoleculesThis imbalance can also have an effect on the damage to the body's cells and tissues, and can worsen health problems if not managed properly Oxidative stress is also widely associated with various degenerative diseases, autoimmune, psychiatric disorders, chronic obstructive pulmonary disease, asthma, or cardiovascular disease. One of the commonly used biomarkers to measure oxidative stress levels is (Robert K and Murray, 2017). *Malondialdehyde* (MDA), which is the end product of lipid peroxidation. Highlevels of MDA are often found in individuals with psychological disorders, suggesting that psychological stress not only affects mental aspects, but also impacts physical health through molecular mechanisms, such as lipid damage of cell membranes induced by free radicals (Jadoon and Malik, 2017).

Medical students often experience high stress due to heavy academic loads, repetitive exams, and competitive learning environments. Research shows that 73% of medical program students experience severe stress, with the main stressors coming from academic stress, followed by interpersonal, social, and motivational stress. To reduce the impact of psychological stress and oxidative stress, antioxidant supplementation such as vitamin C has been a concern. Vitamin C is an important antioxidant that plays a role in protecting the body from oxidative damage, supporting immune system function, and repairing tissue damage. Previous studies have shown that vitamin C administration is known to lower MDA levels in individuals who experience high oxidative stress (Nurul Hakim et al, 2023; Sridevi et al., 2018).

Previous studies have shown that oral vitamin C supplementation can reduce lipid peroxidation, which is characterized by decreased levels of Malondialdehyde (MDA) as an indicator of oxidative stress. Another study also showed that medical students have a relationship between susceptibility to psychological stress and exposure to oxidative stress, which is characterized by increased Olumide et al., (2023) Pansey et al., (2017) *levels of Malondialdehyde* (MDA). Although there have been studies that have discussed the effect of vitamin C supplementation on MDA levels, until now there has been no study that specifically examined the effect of vitamin C dosage on MDA levels in medical students who are prone to psychological stress.

In the Islamic view, food is not only understood as fulfilling biological needs, but also has an important role in maintaining a balance of physical and psychological health. The concept of *halal* food and *thayyib* emphasizes that food must be safe, clean, and beneficial for the body, so that it can support emotional stability and peace of mind. This principle is in line with the preventive health approach that places diet as an important factor in maintaining physical and mental balance (Auliya Izzah Hasanah et al., 2021).

Unhealthy food consumption patterns or not in accordance with the *principles of halalan thayyiban* can have an impact on metabolic disorders as well as psychological imbalances. These conditions have the potential to aggravate psychological stress and decrease the quality of mental health, especially in individuals with high levels of academic stress, such as medical students (Nurkhayati Rojabiah et al., 2023).

In addition, Islamic studies and modern health science show that the intake of nutritious foods, including vitamins and antioxidants, plays a role in maintaining the body's balance and preventing oxidative stress. Vitamin C as one of the important antioxidants has a contribution to protecting cells from damage caused by free radicals, which indirectly also supports psychological stability. Thus, the fulfillment of good nutrition can be seen as part of efforts to maintain body health and peace of mind (Wahyu Dinanti et al., 2022).

Therefore, the author is interested in finding out how the dose of vitamin C supplementation affects Malondialdehyde levels and psychological stress levels of medical students at Yarsi University.

Stress issues have become a major concern in many aspects of life, including among medical students who face heavy academic loads, repetitive exams, and competitive learning environments. Stress can trigger physiological and psychological reactions that have the potential to damage physical health with increased oxidative stress, which can be measured through *Malondialdehyde* (MDA) levels. Although there are studies showing a link between psychological stress and MDA levels, there have been no studies that specifically evaluated the effect of vitamin C supplementation doses on lowering MDA levels on the psychological stress levels of medical students. Therefore, this study aims to examine how the effect of vitamin C supplementation dose on MDA levels on the psychological stress level of medical students at Yarsi University.

This study aims to determine the effect of vitamin C supplementation on Malondialdehyde (MDA) levels and psychological stress levels experienced by YARSI University medical students class of 2022. Through this study, it is expected to provide scientific evidence regarding the effectiveness of vitamin C as an antioxidant in reducing oxidative stress, thereby offering a potential supportive strategy to maintain student health within a demanding academic environment.

Specifically, this study seeks to identify the profile of Malondialdehyde (MDA) levels among medical students of the class of 2022 in relation to their psychological stress levels. Furthermore, it aims to analyze the effect of different doses of vitamin C supplementation on Malondialdehyde (MDA) levels in students experiencing psychological stress. In addition, the study also intends to examine how psychological stress levels influence Malondialdehyde (MDA) levels among medical students of the class of 2022.

RESEARCH METHODS

This study is a quantitative study with a prospective experimental approach *Randomized Controlled Trial* (RCT) to evaluate the effect of vitamin C supplementation on *malondialdehyde* (MDA) levels and psychological stress levels in medical students of the class of 2022. Respondents will be selected based on inclusion and exclusion criteria, and then randomly divided into groups with different supplementation doses.

Vitamin C and MDA levels were measured before and after the intervention to assess the changes that occurred. In addition, psychological stress levels were measured using the *Depression, Anxiety, and Stress Scale* (DASS) questionnaire. Through this design, the study aimed to identify and test the effect of vitamin C supplementation on oxidative stress characterized by MDA levels and psychological stress levels.

The design of this research involves collecting quantitative data through laboratory examinations and distributing questionnaires to medical students of the class of 2022. This study aims to assess the level of psychological stress as well as related biological factors. The three main variables measured are:

1. Vitamin C levels, which were measured through laboratory examination using the Enzyme-Linked Immunosorbent Assay (ELISA) method to determine the status of vitamin C before and the effect of vitamin C at doses of 500mg, 1500mg after supplementation and control groups.
2. *Malondialdehyde* (MDA) levels, which are measured using the ELISA method as an indicator of oxidative stress, can be related to psychological stress.
3. Psychological stress levels, measured using the Depression, Anxiety, and Stress Scale (DASS) questionnaire to assess changes in stress levels before and after the intervention.

The data obtained will be analyzed to evaluate the effect of vitamin C supplementation on vitamin C levels in the body, oxidative stress (MDA), and psychological stress levels in medical students

A population is a group of individuals who meet certain criteria according to the purpose of the study. In this study, the population used was YARSI University medical students class of 2022 who were active during the research period. Based on the data obtained, the total population of medical students of the class of 2022 is 296 people.

Sample

The sample in this study is part of the YARSI University medical student population class of 2022 which was selected based on the inclusion and exclusion criteria that have been set. The sampling technique was carried out by the randomization method in accordance with the *Randomized Controlled Trial* (RCT) design. The selected samples will be divided into treatment groups with different doses of vitamin C supplementation. **Inclusion Criteria**

1. YARSI University medical students of the class of 2022 who are active during the research period and are willing to participate in the research by signing an informed consent.
2. Between the ages of 19–25.
3. Be willing to undergo the entire research procedure, including:

a. Measurement of vitamin C and MDA levels *pre* and *post* intervention.
b. Follow vitamin C supplementation according to the prescribed dosage.
c. Fill out the *complete Depression, Anxiety, and Stress Scale* (DASS) questionnaire.

Exclusion Criteria

1. Students who suffer from a chronic disease and must take medication every day regularly.
2. College students who have allergies to vitamin C supplementation.
3. Students who smoke.
4. Students who consume alcohol.
5. Students who have taken antioxidants such as vitamin C, vitamin E, or other antioxidant supplements regularly for 1 month before *the pre-test* is held.

The sample in this study was determined using *the purposive sampling* technique, where respondents will be selected based on predetermined inclusion and exclusion criteria to suit the research objectives. Once respondents who met the criteria were collected, they were randomized to divide them into intervention groups with different doses of vitamin C supplementation. This approach ensures that the study participants have relevant characteristics, while maintaining the validity and objectivity of the results through random group divisions.

The determination of the number of samples in this study was carried out using the calculation method with Federer's Formula, namely:

$$(t\text{ -1})\ (n\text{ -1})\ 15\geq$$

- t = number of samples
- n = number of samples per group

The population in this study is 296 people. So the number of samples obtained is:

$$(n-1)\ (t-1) \geq 15$$
$$(n-1) \geq \frac{\mathbf{15}}{(\boldsymbol{t}-\mathbf{1})}$$
$$(n-1) \geq (\frac{\mathbf{15}}{\mathbf{3}}n\text{ - }1) \geq 5$$
$$n \geq 6$$

For 4 groups:

$$n = 6 \rightarrow 24$$
$$\boldsymbol{(n-1) \geq \frac{15}{2}}$$
$$\boldsymbol{n\text{ - }1 \geq 7.5}$$
$$\boldsymbol{n \geq 8.5n = 7}$$
$$\mathbf{7 \times 4 = 28}$$

Thus, the number of samples in this study is 36 respondents, which are then divided evenly into 4 groups, each consisting of 9 people.

The research uses a primary data type, which is data obtained directly from respondents through various measurement methods. The main data collected included *malondialdehyde* (MDA) and vitamin C levels, which were measured before and after the intervention using the ELISA method in the laboratory. In addition, psychological stress levels were measured through a *Depression, Anxiety, and Stress Scale* (DASS) questionnaire filled out by respondents. Because all data is obtained directly from the results of measurements and research instruments, this study is entirely sourced from primary data.

How to Collect Data and Measure Data

How Data Is Collected

1. The researcher applied for permission to the faculty to carry out research and recruit respondents.
2. Students were asked to fill out a DASS questionnaire to see the level of psychological stress.
3. Students who meet the inclusion criteria are provided with information and explanations about the objectives, procedures, and benefits of the research, and are asked *for informed consent* voluntarily to participate
4. Initial measurements (*pre-tests*) were carried out:

a. Blood sampling was taken to measure vitamin C and MDA levels using the ELISA method in the laboratory.

5. Respondents received an intervention in the form of vitamin C supplementation according to a predetermined dose, with randomized group divisions.
6. The final measurement (*post-test*) was carried out after the intervention period:

a. Blood sampling was returned to measure vitamin C and MDA levels after supplementation.
b. Respondents again filled out the DASS questionnaire to assess changes in psychological stress levels.

How to Measure Data

a Measurement of vitamin C levels

a. Blood samples were taken from respondents in two stages, namely before the intervention and after the intervention.

b. Vitamin C level analysis was carried out using *the Enzyme-Linked Immunosorbent Assay* (ELISA) method in the laboratory to obtain accurate results.

a Measurement of *malondialdehyde* (MDA) levels

a. Blood samples from the same respondents were taken to measure MDA levels, both before and after the intervention.
b. MDA levels were checked using *the Enzyme-Linked Immunosorbent Assay* (ELISA) method, in the laboratory to obtain accurate results.

a Measurement of psychological stress levels

a. Psychological stress was measured using the *Depression, Anxiety, and Stress Scale* (DASS) questionnaire.
b. Respondents filled out questionnaires twice, before and after the intervention, to assess changes in stress levels after vitamin C supplementation.

Data Collection Instruments

In this study, the instruments used to collect data include:

Vitamin C

Vitamin C level data was collected in two stages, namely *pre-test* before intervention and *post-test* after intervention, to assess changes that occurred after supplementation. Respondents were given vitamin C supplementation at different doses, according to a randomly determined intervention group, then vitamin C levels were measured with blood samples through laboratory examination using *the Enzyme-Linked Immunosorbent Assay* (ELISA) method to obtain accurate and objective results.

Malondialdehyde **(MDA)**

Data collection of *malondialdehyde* (MDA) levels was carried out in two stages, namely *pre-test* before intervention and *post-test* after intervention. Post-test data collection was conducted to see if vitamin C supplementation had an effect on MDA levels, which was used as a parameter of oxidative stress in this study. Blood samples obtained from respondents were analyzed in the laboratory using *the Enzyme-Linked Immunosorbent Assay* (ELISA) method, which allows for specific and accurate detection of MDA levels.

Psychological Stress

The measurement of psychological stress levels in this study used 14 items from the stress scale in the *Depression, Anxiety, and Stress Scale* (DASS-42). The questionnaire assessed how often respondents experienced symptoms of stress in the past week, with a rating scale, 0 = Never, 1 = Occasionally, 2 = Often, and 3 = Almost always (Lovibond and Lovibond, 1995).

The total score was obtained by summing the scores of 14 stress items, then multiplying them by 2 so that the results were in accordance with the DASS-42 measurement standard. The results of these measurements are grouped into several stress level categories based on a predetermined range of scores. Measurements were taken in two stages, namely *pre-test* before intervention and *post-test* after intervention, to evaluate changes in stress levels after vitamin C supplementation.

Data Analysis

Validity Test and Reliability Test

Validity Test

Validity tests are used to evaluate whether the questionnaire used measures the variables being studied. The higher the level of validity of a measuring instrument. To measure validity, *the Pearson Product Moment method* is used with the following formula:

$$r = \frac{N(\sum XY) - (\sum X \sum Y)}{\sqrt{(N\sum X^2 - (\sum X)^2)\,(N\sum Y^2 - (\sum Y)^2)}}$$

Description:

r = Validity coefficient of the question item searched

N = Number of sample members
X = Total respondent score
Y = The total score of each respondent's statement
ΣX = Number of scores in the distribution X
ΣY = Number of scores in the distribution Y
ΣX^2 = The sum of squares of each X
ΣY^2 = The sum of squares respectively

The questionnaire is declared valid if the Product Moment correlation coefficient (r) is more than 0.361 or if the r-calculated value is greater than the r-table. Therefore, a correlation test with *the Pearson Product Moment* method was used to evaluate the validity of each item in the questionnaire.

Reliability Test

Reliability is an indicator that describes the consistency of a measuring instrument. To assess the reliability of the questionnaire, *Cronbach's Alpha* method was used, with the following formula:

$$r_i = \left[\frac{k}{k-1}\right]\left[1 - \frac{\sum {S_i}^2}{{S_i}^2}\right]$$

Description:
r_i = Instrument reliability
k = The number of questions or the number of questions
$\sum {S_i}^2$ = Number of variants of the item
${S_i}^2$ = Total variance

A questionnaire is considered reliable if the results of the instrument test show *a value of Cronbach's Alpha* > $r_{criterion}$ (0.60).

Univariate Analysis

Univariate analysis is used to describe the data characteristics of each research variable. The data is presented in the form of a distribution table of frequency, percentage, mean, standard deviation (SD), and median according to the type of data analyzed.

The variables analyzed in this study included malondialdehyde (MDA) levels before and after vitamin C administration, as well as psychological stress levels based on the results of the Depression Anxiety Stress Scale (DASS) questionnaire. The results of this analysis aim to provide an initial overview of the distribution of data before further statistical tests are carried out

RESULTS AND DISCUSSION

Univariate Analysis

The analysis in this section begins with univariate descriptive analysis as the initial stage of data processing. This stage aims to describe and summarize the data characteristics of each variable studied, both before and after the intervention. Using descriptive statistics such as minimum, maximum, mean, and standard deviation, this analysis provides an overview of the distribution of research data.

The results of this univariate analysis form the basis for the bivariate analysis in the next section, which is used to assess the effect of vitamin C supplementation on *Malondialdehyde* (MDA) levels and psychological stress levels in medical students.

Normality Test

MDA Rate

Normality tests of MDA levels were conducted with Kolmogorov–Smirnov and Shapiro–Wilk to see the data distribution patterns in each vitamin C dose group and control group, both before and after the intervention.

In the 1500 mg dose group, the significance values of both tests were above 0.05, at both pre and post measurements, so the data in this group could be considered normally distributed.

The same results were also seen in the control group. The significance value of the entire test was above 0.05, suggesting that the distribution of data in the vitamin C-free group was also normal.

In the 1000 mg dose group, most significance values are also still above 0.05. The Shapiro–Wilk value on the post measurement is indeed close to the limit ($p = 0.094$), but it is still enough to conclude that the distribution of the data is not so deviant from normal that it can be used in parametric analysis.

In contrast to the other groups, the 500 mg dose group showed an abnormal distribution pattern at post measurements. Both tests showed a $p<$ value of 0.05, which means the assumption of normality was not met. Therefore, the analysis of data in this group is more appropriate using non-parametric tests such as the Wilcoxon test.

Stress Levels

The normality test of DASS scores was carried out using Kolmogorov Smirnov and Shapiro Wilk to assess the pattern of data distribution in each group, both before and after the intervention. The results of this normality test are the basis for determining the type of statistical test that will be used at the next stage of analysis.

In the 1500 mg dose group, the significance value on the pre measurement was at (0.386; $p > 0.05$), indicating that the data were normally distributed. However, in the post measurement, the significance value was at (0.007; $p < 0.05$) so the normality assumption was not met. Thus, the data in this group were not completely normally distributed, and subsequent analysis was performed using non-parametric tests such as the Wilcoxon test.

In the 1000 mg dose group, the significance value in both pre and post measurements was at (0.268; $p > 0.05$) and (0.420; $p > 0.05$). This shows that the distribution of data is normal so that the data in this group can be analyzed using parametric tests such as the Paired t-Test because the assumption of normality is met.

In the 500 mg dose group, the normality test results showed that the pre measurement was not normally distributed, as the significance value was at < 0.05. Although the post measurements showed values closer to normal (0.107; $p > 0.05$), the results on the pre measurements were sufficient to conclude that the data distribution in this group was abnormal. Therefore, the analysis in the 500 mg dose group used a non-parametric test such as the Wilcoxon test.

In the control group, the significance values in both pre and post measurements were at 0.316 and (0.070; $p >0.05$), so the data can be considered normally distributed. With the assumption of normality met, the control group was analyzed using parametric tests.

Vitamin C Concentration

The results of the normality test showed a difference in the distribution of vitamin C concentration between the dose group and the control group. In the 1500 mg dose group, data before and after supplementation were normally distributed with Shapiro-Wilk (0.930; $p=0.550$) and 0.915 ($p=0.430$), so parametric analysis could be used. The 1000 mg dose group showed data before the intervention were abnormal (0.571; $p < 0.001$), but after supplementation became normal (0.950, $p=0.732$), which likely reflects a stabilizing effect of vitamin C concentration. In the control group without supplementation, pre-normal data (0.806, $p=0.047$) but post-normal (0.928, $p=0.530$), showed that vitamin C levels remained stable without intervention. This consistency of normality supports the use of parametric tests, such as the Paired t-Test, for the analysis of changes in vitamin C concentrations can be validly performed.

Bivariate Analysis

Effect of vitamin C administration on *malondialdehyde* (MDA) levels

The results of the Paired t-Test in the group receiving vitamin C at a dose of 1500 mg showed that the average MDA level after the intervention was slightly lower than before the intervention, with a difference of -0.468 nmol/mL. However, this difference was not statistically significant ($p = 0.467$; $p > 0.05$). The t-value of -0.764 and the 95% confidence interval in the range of -1.880 to 0.944 illustrate that the change in MDA levels in this group is still within the normal range of variation between respondents.

The tendency to decrease MDA levels is noticeable, but not strong enough to show a significant effect of vitamin C supplementation at a dose of 1500 mg. The width of the confidence interval also suggests that each individual's response varies, likely influenced by the initial condition or oxidative stress level of each respondent. Overall, these results suggest that at high doses, vitamin C may provide a slight MDA-lowering effect, but it is not yet consistent enough to be considered statistically significant.

The Paired t-Test is used because the MDA levels in this group are normally distributed based on the results of previous normality tests. With normal distribution, this parametric analysis can more accurately assess changes in MDA levels.

Although the results of the analysis did not show a statistically significant difference, these findings still provide an idea of changes in oxidative stress in medical students who received vitamin C supplementation at a dose of 1500 mg.

Overall, vitamin C supplementation at a dose of 1500 mg did not show a significant change in MDA levels in the intervention period used. However, the presence of a downward trend in MDA levels remains a relevant finding and can be further explored in studies with longer duration or larger sample counts.

In the group receiving a dose of 1000 mg of vitamin C, the Paired t-Test showed that the average MDA levels after the intervention were lower than before the intervention, with a difference of –1,002 nmol/mL. This difference is statistically significant with a single-sided p-value (0.047; $p < 0.05$). The t-value of –1.904 and the 95% confidence interval in the range of –2.215 to 0.212 illustrate that there is a decrease in MDA levels after supplementation.

This decrease in MDA levels indicates the potential antioxidant effect of vitamin C at a dose of 1000 mg, as seen from the reduction of lipid peroxidation products. However, variations in the range of confidence intervals suggest that responses between individuals are not entirely homogeneous. However, these findings still give an idea that a dose of 1000 mg has a more pronounced biological effect than conditions without supplementation, and may be one of the doses that showed a reduced effect of oxidative stress in this study.

The use of the Paired t-Test in the 1000 mg dose group is appropriate because the MDA level data show a normal distribution in the normality test. With this parametric approach, changes in MDA levels can be assessed more sensitively, especially given the small sample size. The significance of the one-sided test corroborated that vitamin C supplementation of 1000 mg provides a detectable effect in reducing oxidative stress.

Overall, these findings suggest that a dose of 1000 mg of vitamin C has the potential to significantly reduce MDA levels when viewed from the hypothesis of a downward direction. This gives an indication that this dose has a more pronounced antioxidant activity than the condition without supplementation. These findings also serve as an important basis for follow-up research with larger sample counts or designs that can evaluate optimal doses more comprehensively.

In the 500 mg dose group, the Wilcoxon Signed Ranks test showed no significant change in MDA levels after supplementation with a $p(0.889; p>0.05)$. There were almost balanced negative and positive ratings, indicating that some respondents experienced a decrease in

MDA, while others increased. This reflects the variation in individual responses to the 500 mg dose.

These results are consistent with the nature of the data that are not normally distributed, so the use of non-parametric tests such as Wilcoxon is appropriate. Clinically, a dose of 500 mg did not appear to provide a consistent antioxidant effect in lowering MDA levels in this group of subjects.

The Wilcoxon test was used because MDA level data in the 500 mg dose group, especially at post measurements, showed abnormal distribution. This non-parametric approach allows the evaluation of changes in MDA levels without assuming the normality of the data, resulting in more robust results against violations of distribution assumptions. A high p-value indicates that supplementation of a dose of 500 mg does not produce a significant effect in reducing oxidative stress, as measured through MDA levels.

Overall, the results of this trial indicated that vitamin C supplementation at a dose of 500 mg did not have a significant impact on MDA levels in medical students in this study period. The variability of individual responses and the lack of consistent effects may be factors to consider in evaluating the effectiveness of these low doses in the context of oxidative stress.

The Paired t-Test in the control group showed that there was no significant change in MDA levels before and after the study with a p(0.573; $p > 0.05$). The average MDA level appears to decrease by about 0.8 nmol/mL, but this change is small and inconsistent, as can be seen from the fairly wide 95% confidence interval (–4.109 to 2.499). This shows that variation between subjects is more dominant than the effect of observation time. Clinically, this insignificant decrease in MDA likely reflects normal physiological fluctuations or the influence of external factors such as diet and daily stress levels. Thus, without vitamin C supplementation, MDA levels were relatively stable and showed no significant changes during the study period.

The use of the Paired t-Test in the control group was appropriate, as the normality test showed that the MDA level data was normally distributed. With this approach, changes in MDA levels can be validly evaluated even in the absence of vitamin C supplementation. These findings are consistent with the hypothesis that without antioxidant interventions, there is no additional effect that can reduce oxidative stress. Therefore, the results in the control group became an important benchmark for assessing the effectiveness of vitamin C supplementation in the intervention group.

The effect of vitamin C administration on Stress Levels

In the 1500 mg dose group, the Wilcoxon Signed Ranks test showed that there was a significant change in stress levels after supplementation with a p(0.018; $p<0.05$). All respondents were ranked negative, indicating that all respondents experienced a decrease in stress levels from pre-test to post-test, with no fixed improvement or score. This suggests that the response to vitamin C supplementation at a dose of 1500 mg is consistent in lowering stress levels.

The Wilcoxon test was used because the stress level data in this group did not meet the assumption of normal distribution, so a non-parametric approach was chosen to evaluate changes in pre-test and post-test values more precisely. A p< value of 0.05 indicates that vitamin C supplementation at a dose of 1500 mg has a significant effect in reducing stress levels.

Overall, the results of this trial indicated that vitamin C supplementation at a dose of 1500 mg was statistically and clinically effective in lowering stress levels in respondents during the study period.

The Paired t-Test is used because the data on the Vitamin C group at a dose of 1000 mg is normally distributed based on the results of the normality test that has been carried out

previously. This parametric test can more accurately evaluate changes in stress scores before and after the intervention.

In the group that received a dose of Vitamin C at 1000 mg, the results of the analysis of the Paired t-Test showed that there was a decrease in DASS score after the intervention. The average difference between pre-test and post-test scores is 13.71 points. This decrease proved to be statistically significant with a p value of < 0.001. This shows that the administration of Vitamin C 1000 mg has a significant effect in reducing stress levels in respondents.

In the 500 mg dose group, the Wilcoxon Signed Ranks test showed that there was a significant change in stress levels after supplementation with a p(0.027; p<0.05). There was a negative rating that was more dominant than a positive rating, which showed that most respondents experienced a decrease in stress levels, although there was 1 respondent who actually experienced an increase in stress levels. This reflects the variation in individual responses to the 500 mg dose.

The Wilcoxon test was used because the stress level data in this group was not normally distributed, so the use of non-parametric tests such as Wilcoxon was considered appropriate. A p value of < 0.05 indicates that vitamin C supplementation at a dose of 500 mg still provides a statistically significant effect in lowering stress levels.

The results of this test indicated that vitamin C supplementation at a dose of 500 mg was able to reduce stress levels, but the effect was not entirely consistent across subjects, which may have been influenced by individual factors.

The Paired t-Test is used because the data in the control group have a normal distribution based on the results of previous normality tests. This parametric analysis can be used to more accurately assess changes in stress scores.

In the control group, the Paired t-Test showed that there was a decrease in DASS scores from pre-test to post-test with an average difference of 5.14 points. The results of the analysis showed that this decrease was statistically significant with a value of p (0.038; p < 0.05). This suggests that the control group still experienced a slight decrease in stress levels despite not receiving a vitamin C administration.

Effect of vitamin C administration on Vitamin C levels

In the 1500 mg dose group, the Paired t-Test showed that there was no significant difference in vitamin C concentrations before and after supplementation with a p-value (0.171; p>0.05). The mean concentration increased by about 23.7 mg/dL, but this change was not statistically significant, and the wide 95% confidence interval (–12.64 to 60.07) showed considerable variability in response among individuals.

The visible increase in concentration reflects a tendency to respond to supplementation, but individual differences in vitamin C absorption or metabolism, including initial nutritional status, likely affect the outcome. Thus, despite indications of improvement, 1500 mg dose supplementation did not show a statistically consistent effect on vitamin C concentrations in this study.

The use of the Paired t-Test in the 1500 mg dose group is appropriate because the vitamin C concentration data is normally distributed. This parametric approach allows for a change-sensitive evaluation, even if the results are not statistically significant. These findings suggest that supplementation of a dose of 1500 mg did not consistently increase the concentration of vitamin C in the blood during the study period.

Overall, individual response variations are an important factor to consider when evaluating the effectiveness of these high doses.

In the 1000 mg dose group, the Wilcoxon Signed Ranks test showed a significant increase in vitamin C concentrations after supplementation with a p(0.008; p<0.05). All respondents experienced increased concentration, as indicated by the dominance of negative ratings and the Z value = –2,666. These results confirm that vitamin C supplementation at a dose of 1000 mg

consistently increases vitamin C levels in the blood. The low significance value suggests that this increase is not accidental, but rather a direct effect of supplementation, reflecting the body's ability to absorb and raise vitamin C levels at this dose in the study period.

The Wilcoxon test was used because the vitamin C concentration data in the 1000 mg dose group was not normally distributed. The results of the analysis showed a significant increase in vitamin C concentration after supplementation ($p = 0.008$), confirming the effectiveness of the 1000 mg dose in raising vitamin C levels.

Overall, these findings suggest that vitamin C supplementation at a dose of 1000 mg consistently increases vitamin C concentrations in the blood, supports the body's antioxidant function, and is evidence of the success of the intervention in medical students.

In the 500 mg dose group, the Paired t-Test showed a significant increase in vitamin C concentration after supplementation with a p(0.027; $p>0.05$). The mean concentration increased by 73.9 mg/dL, with a 95% confidence interval between 11,089 and 136,744 indicating that this increase was consistent among respondents despite individual variability.

These results confirm that vitamin C supplementation at a dose of 500 mg effectively increases vitamin C levels in the blood. The significance of both single-sided and double-sided tests suggests that these low doses have a noticeable effect and can support the body's antioxidant function.

The Paired t-Test was used because the vitamin C concentration data in the 500 mg dose group was normally distributed. This parametric approach allows for a change-sensitive evaluation, especially in small samples. The results of the analysis showed a significant increase in vitamin C concentration after supplementation, confirming that the dose of 500 mg effectively raised the level of vitamin C in the blood.

Overall, these findings suggest that these low doses may be an effective option for increasing vitamin C levels, supporting antioxidant function, and potentially helping to reduce oxidative stress in medical students.

In the control group (without vitamins), the Wilcoxon Signed Ranks test showed that there was no significant change in vitamin C concentrations before and after the study period with a p(0.063; $p>0.05$). Most respondents experienced a decrease in concentration, although this change was not statistically significant.

The changes that occur may reflect natural fluctuations in vitamin C levels, influenced by food intake or individual metabolism. A p-value close to the significance threshold indicates a tendency for change, but is not strong enough to be considered statistically significant. Thus, without supplementation, vitamin C concentrations in the control group were relatively stable during the study.

The Wilcoxon test was used because the vitamin C concentration data in the control group was not normally distributed. This non-parametric approach ensures robust analysis of changes, especially in small samples. These results are consistent with the expectation that without supplementation, vitamin C concentrations tend to be stable or only experience minor fluctuations.

Overall, the analysis confirms that without supplementation, there is no significant change in vitamin C levels in the blood. These findings are an important benchmark for evaluating the effectiveness of supplementation in the intervention group, showing that the changes observed in the control group were natural and unaffected by the intervention.

Effect of vitamin C supplementation on vitamin C concentration

The hypothesis (H1) states that supplementation will increase the concentration of vitamin C in the blood compared to the control group. The results of the Wilcoxon test at a dose of 1000 mg (p 0.008; $p < 0.05$) and the Paired t-Test at a dose of 500 mg (p 0.027; $p < 0.05$) showed a significant increase in vitamin C concentrations, each with a full negative rating and an average difference of 73.917 mg/dL, in favor of H1. However, at a dose of 1500 mg (p

0.171; p > 0.05), the increase was not significant, which may indicate a limit on vitamin C absorption. The control group (p 0.063; p > 0.05) showed no significant change, supporting that supplementation is needed to increase vitamin C concentrations. These results confirm that doses of 500 mg and 1000 mg effectively increase vitamin C concentrations, supporting antioxidant function in the body (Lykkesfeldt & Tveden-Nyborg, 2019; Nur Khasanah. et al., 2023).

As in the study, vitamin C supplementation of 1000 mg/day and 500 mg/day showed an increase in plasma concentration of vitamin C, this effect indicates that the doses of 1000 mg and 500 mg are quite optimal as antioxidants. Since the absorption of vitamin C decreases after the saturation threshold is reached, higher doses such as 1500 mg do not necessarily increase additional effectiveness, so a dose of 1000 mg may be considered more effective and safe in lowering oxidative stress Żychowska., et al. (2021).

Effect of vitamin C supplementation on psychological stress levels

The hypothesis (H1) states that vitamin C supplementation will reduce psychological stress scores by a potential mechanism through strengthening the antioxidant system and decreasing oxidative stress, thereby affecting redox balance and emotional response to stress (Dominiczak, 2023).

The results showed that at a dose of 1500 mg, all respondents experienced a consistent reduction in stress levels, indicating that high doses of vitamin C had a uniform effect on improving psychological conditions. At a dose of 1000 mg, the greatest reduction in stress score was obtained, with an average difference of 13.71 points (p < 0.005), which suggests that this dose was the most effective dose in reducing stress in this study. Meanwhile, at the 500 mg dose, although statistically significant, the stress reduction response was not entirely consistent, as there was still one respondent who experienced increased stress, indicating a variation in individual responses that could be influenced by differences in initial oxidative stress status, or metabolism and bioavailability of vitamin C in each respondent. In addition, psychological and environmental factors can also have an effect, so the effects of interventions are not always uniform on each individual (Zänkert et al., 2019).

The decrease in stress levels in the control group was likely influenced by non-intervention factors such as psychological adaptation, as well as natural stress fluctuations, but because the magnitude of the decrease was much smaller than in the supplementation group, the effect of vitamin C was still considered clinically dominant.

Effect of vitamin C supplementation on *Malondialdehyde* (MDA) levels

The study hypothesis (H1) states that vitamin C supplementation will lower MDA levels as an indicator of oxidative stress compared to the control group. The results of the Paired t-Test at a dose of 1000 mg showed a significant decrease in MDA levels based on a single-sided test (p 0.047; p < 0.05), but not significantly based on a double-sided test (p 0.093; p > 0.05), with an average difference of -1.002 nmol/mL, which favors H1. This decrease is in line with the theory that vitamin C dampens free radicals, and can inhibit lipid peroxidation (Dominiczak, 2023). However, at doses of 1500 mg (p 0.467) and 500 mg (p 0.889), there was no significant change, suggesting that the dose of 1000 mg was the optimal dose in this study. The control group (p 0.573) showed no significant change, consistent with the expectation that without supplementation, MDA levels remained stable. These results confirm that vitamin C supplementation, specifically a dose of 1000 mg, is effective in lowering oxidative stress (Moritz et al., 2020).

In this study, the dose of 1000 mg was the most effective dose in reducing MDA levels. This is appropriate as in research on the mechanism of action of vitamin C as an antioxidant that directly suppresses free radicals, including hydroxyl and peroxide radicals, which play a role in lipid peroxidation. In addition, vitamin C increases cellular glutathione levels as an important endogenous antioxidant, thereby lowering oxidative stress. In contrast, the

insignificant effectiveness at doses of 1500 mg can be explained by transporter saturation in which the bioavailability of vitamin C decreases at high doses. In fact, at very high doses vitamin C can exhibit pro-oxidant effects through Fenton reactions involving Fe^{2+} ions, so the antioxidant benefits do not increase further Kaźmierczak-Barańska et al., 2020; Żychowska et al., (2021)

Good Food Quality in Islamic Views

The Concept of Halal and Thayyib Food

Halal is not only related to food substances, but also to the way they are obtained. Islam prohibits all forms of efforts to obtain food in an unhalal manner, because in addition to sin, it also has a bad impact on health. Thus, food that is truly in accordance with sharia is one that meets the criteria of halal and thayyib, which is safe, nutritious, and beneficial to the body. Thus, the Qur'anic teachings on halal consumption and thayyib have strong relevance to modern health principles, because they play a role in fulfilling nutrition as well as preventing disease (Nurkhayati Rojabiah et al., 2023).

Nutritious Food as Thayyib Food and the Importance of Health in Islam

The fulfillment of balanced nutrition is an important need for humans in supporting daily activities while maintaining physical and spiritual health. The Qur'an alludes to the importance of fruit as a source of nutrition, as Allah says in the Qur'an:

فَاَنْشَاْنَا لَكُمْ بِهٖ جَنّٰتٍ مِّنْ نَّخِيْلٍ وَّاَعْنَابٍۙ لَكُمْ فِيْهَا فَوَاكِهُ كَثِيْرَةٌ وَّمِنْهَا تَأْكُلُوْنَۙ ﴿١٩﴾

"Then We grew for you with the water gardens of dates and vineyards; In it you get many fruits and some of them you eat" QS. Al-Mu'minun [23]: 19.

Ibn Kathir interpreted this verse as an explanation of Allah's blessings in the form of dates, grapes, and various types of fruits that are rich in benefits, can be consumed directly or processed, and are a source of energy and health for humans. M. Quraish Shihab added that the mention of these fruits also shows the diversity of sources of nutrition that Allah provides for humans. (Setiawan, 2020)

This view is in line with studies that affirm that vitamins and minerals are part of a Revelation Düsseldorf et al., (2022), thayyib diet because they provide real benefits for the body's metabolism. One of them is vitamin C, which is abundant in fresh fruits and vegetables, plays an important role in maintaining the immune system, acts as an antioxidant, and supports collagen formation and wound healing. Thus, both scholarly interpretations and modern scientific findings show that nutritious food is a tangible manifestation of Islamic teachings on the consumption of food that is good for physical and spiritual health.

Vitamins are basically needed by the body in small amounts, but they are essential to maintain human life and health. Vitamin deficiencies and excesses can interfere with body function. In nutrition, vegetables and fruits are the main sources of various vitamins and minerals, including vitamin C (Wahyu Dinanti., 2022).

Efforts to maintain health through the consumption of fruit are part of the trust to the body that Allah has entrusted, as the Prophet PBUH said:

إِنَّ لِبَدَنِكَ عَلَيْكَ حَقًّا

"Verily your body has a right over you." (HR. Bukhari)

Thus, consuming nutritious fruit is a form of gratitude for Allah's favor as well as practicing the principle *of halalan thayyiban* in maintaining physical and spiritual health.

Stress in Islamic Views and Psychology

Islamic psychotherapy emphasizes the spiritual dimension as a strategy to deal with stress, namely through prayer, prayer, dhikr, reading the Qur'an, patience, and tawakkal. This approach not only serves to calm the heart, but also provides new energy to face life, foster optimism, and strengthen the mental health of a Muslim (Sultani et al., 2023).

The Influence of Food on Mental Peace and Its Relationship to Halal-Thayyib Food and Body Condition

Halal and nutritious food helps maintain optimal body function, reduces the risk of disease, and supports emotional stability. On the other hand, eating haram, excessive, or unnutritious foods can damage the health of the body and cause psychological imbalances. (Ridwan et al., 2024)

The Qur'an provides comprehensive guidance regarding a healthy and balanced diet to maintain physical, mental, and spiritual health, which plays a role in regulating the body's metabolism, preventing oxidative stress, and improving brain function related to emotional stability. This emphasizes that the balance between body, mind, and soul is a form of obedience to Allah SWT which brings inner peace, thus, a healthy diet that is in accordance with *the principles of halal-thayyib* is an important factor in maintaining peace of mind (Heidari et al., 2023).

Islamic View of Body Balance and Oxidative Stress (*Malondialdehyde)*

Cell damage due to oxidative stress can arise due to a variety of factors, such as an unhealthy diet, excessive stress, and exposure to an unfavorable environment.

Therefore, maintaining body balance such as MDA levels remains stable by strengthening antioxidants in the body which is part of efforts to maintain health. One of the antioxidants is vitamin C, which functions to neutralize free radicals and prevent cell damage. Efforts to maintain body balance through the consumption of vitamin C can be seen as a form of implementing Islamic teachings in maintaining a healthy and useful body trust (Heidari et al., 2023).

CONCLUSION

The analysis shows that vitamin C supplementation at 500–1000 mg significantly increases blood vitamin C levels in medical students, with the most consistent effect observed at 1000 mg, while a 1500 mg dose offers no additional benefit likely due to absorption limits. At the same optimal dose of 1000 mg, there is also a significant reduction in Malondialdehyde (MDA) levels, indicating decreased oxidative stress, whereas 500 mg and 1500 mg doses show no significant changes, possibly due to individual metabolic variability. Additionally, vitamin C consistently reduces psychological stress levels, likely through its role in maintaining redox balance and influencing emotional responses to academic pressure. From an Islamic perspective, these findings align with the principle of halalan thayyiban, emphasizing that proper nutrition supports both physical and spiritual well-being as part of responsible self-care. Future research could explore longer-term supplementation effects, optimal dosing across different populations, and the interaction between vitamin C, stress biomarkers, and cognitive performance in academic settings.

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