Original Article

ASSESSMENT OF OXIDATIVE STRESS AND ANTIOXIDANT STATUS IN NEWLY ADMITTED HEALTHY UNDERGRADUATE STUDENTS IN NNAMDI AZIKIWE UNIVERSITY AWKA, NIGERIA

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DOI: 10.47750/pnr.2022.13.S01.165

Abstract

Background: The economic and social development of any country is highly dependent on health. National initiative to curb the dangers of student's ill health and social wellbeing in Nigeria is still lacking. The study is aimed to investigate the oxidative stress and antioxidant status of newly admitted undergraduate students who fell sick within 12 months period of stay in Nnamdi Azikiwe University, Awka, Nigeria. **Materials and Methods:** This is a longitudinal prospective study involving 530 (male=239, female=291) apparently healthy newly admitted 100 level students (control) and 354 (male-169, female=185) students who fell sick within 12 months period of stay in school (test). The test group was group into A (malaria, n=112), B (typhoid, n-61), C (malaria/typhoid, n-103), D (pneumonia, n=31) and E (peptic ulcer disease, n=47). Serum concentrations of malondealdehyde (MDA), nitric oxide (NO), total antioxidant capacity (TAC), catalase (CAT) and glutathione (GSH) were determined. **Results:** The results showed significantly increased levels of MDA, NO with decreased TAC, GSH and CAT in sick undergraduate students especially in malaria, typhoid, malaria/typhoid and peptic ulcer group compared with apparently healthy participants (P<.05 respectively). Same pattern was observed in male than in female sick participants (P<.05 respectively). **Conclusion:** The alteration in pro-oxidant markers is accompanied with imbalance in the antioxidant defense status resulting in oxidative stress. Males were more susceptible to oxidative insult than females. This shows that oxidative stress may have contributed to the pathogenesis and severity of different diseases observed which may cause irreversible cell damage if not managed. A national intervention for student's health and social wellbeing is strongly advocated.

Keywords: Pro-oxidants, anti-oxidants, pattern, oxidative stress, undergraduate students, Nigeria.

INTRODUCTION

Health is essential for social and economic development of any country [1]. National health policies that take into cognizance the intervention against the menace of student's ill health in Nigeria is limited. Environmental and social amenities in the Nigerian Universities are very poor. Several studies have demonstrated the detrimental effects of reactive oxygen species and oxidative stress in various disease conditions including both acute and chronic diseases [2, 3, 4]. Other reports also documented some positive relationship between daily activity stress, toxic effect of reactive oxygen species and immune functions [5] and people deficient in antioxidants are more susceptible to severe bacterial, parasitic and fungal infections [6]. Free radicals are produced in the body in both physiological and pathological conditions [7]. This free radical are normally controlled by antioxidant defense system that includes intracellular enzymes such as; glutathione peroxidase, superoxide dismutase and low molecular weight antioxidant such as vitamin C and vitamin E. Oxidative stress always occur when there is excess production of free radical more than the body antioxidant system can curb. This causes damage to the biomolecules such as protein, lipids and nucleic acid [7] and often results in sickness. Among students in Nigeria, high level of stress either from examinations, poor feeding habits, environmental factors such as pollution, poor sanitation infections can cause health problem. Living in close or

over populated quarters also poses a health risk and increase a student chances of contracting disease [8, 9]. Also, home sickness is common among university students especially the first year students. Most university students consume road side already prepared or ready to eat food and snacks such as dough nuts, meat pie, chin chin, and soft drinks. More so, the excessive and continuous stress among university undergraduate students and the somatic events associated with it has effect that go beyond mere triggering of disease to affecting their quality of life [10]. The present study therefore aimed at evaluating the oxidative stress marker and antioxidant status of both healthy and sick undergraduate students of Nnamdi Azikiwe University Awka, Anambra State, Nigeria.

MATERIALS AND METHODS

Study Design

This is a longitudinal prospective study designed to assess the oxidative stress and antioxidant status of newly admitted undergraduate students in Nnamdi Azikiwe University Awka, Anambra State, Nigeria within the 12 months period of stay in school.

Sample Population/Participants Recruitment

Five hundred and seventy (570) newly admitted apparently healthy undergraduate student of Nnamdi Azikiwe University, Awka, between the ages of 17 – 21 years were conveniently recruited for this study. A baseline sample was collected from all the volunteered participants during their medical examination and testing before registration and this was regarded as control. The participants were followed up while they continue their activities until some of them started to report back to the clinic for various sicknesses. A total number of three hundred and fifty four participant (354) which includes 185 females and 169 males reported sick. At the point of sickness, the second samples were collected from the participants and this was regarded as the test group.

From the provisional diagnosis by the attending clinician, out of the three hundred and fifty four (354) participants that reported back at the NAU Medical Centre with sickness, one hundred and twelve (112) participants had malaria and served as (Group A), sixty one (61) participants had typhoid and served as (Group B). One hundred and three (103) had typhoid and malaria which served as (Group C). Forty seven (47) of them had Pneumonia and regarded as (Group D) while thirty one (31) had peptic ulcer and grouped as E.

Sample Collection

At each stage of the sampling, ten (10 ml) of blood sample was consecutively and aseptically collected by venepuncture from each of the participant at their point of first registration and during sickness at Nnamdi Azikiwe University Medical Center, Awka, The blood sample collected at every stage was dispensed into heparin citrate container and stored at -80ºC using small aliquot until assayed for oxidative stress parameters.

Study Site

This study was carried out at Medical Center, Nnamdi Azkiwe University, Awka.

Inclusion Criteria

In the baseline stage or the recruitment stage, the participants for this study were apparently healthy male and female undergraduate students between the ages of 17 – 21 years. They were newly admitted into year one (100 level) undergraduate course program and they willingly gave their consent to participate in this study. Also, in the second stage, only the participants who reported sick and consulted the clinicians at NAU medical center, Awka were included.

Exclusion Criteria

Participants who were on drugs such as antimalarial, antibiotics and supplements of any kind were excluded. Those with chronic medical condition such as hypertension etc. were excluded. Also, participants who are drug addicts, smokers, strict vegetarians, those outside the age ranges and any other person who did not meet the inclusion criteria were excluded from the study.

Anthropometric Analysis

Weight and height measurements were taken from each participant at the first and second stage of blood sample collections. Body Mass Index (BMI) was calculated using weight in kilogram divided by height in meter square [11].

Laboratory for the study

The oxidative stress markers were analyzed at Chemical Pathology Laboratory, Medical laboratory Science Department, NAU, Okofia, Nnewi Campus.

DETERMINATION OF MALONDEALDEHYDE

Malondealdehyde was determined using colorimetric method as described by Janero. [12].

Principle:

MDA in the catabolite of lipid peroxide can react with thiobarbituric acid (TBA) and produce red compound, which has a maximum absorption peak at 570 nm

DETERMINATION OF NITRIC OXIDE

Nitric oxide was determined using colorimetric method as described by Schmidt et al. [13].

Principle

The nitrate in the sample is reduced to nitrite by nitrate reductase enzyme. The total nitrite is detected with Griess reagent as a colored azo dye product and absorbance is measured at 540 nm.

DETERMINATION OF GLUTATHIONE

Glutathione was determined using colorimetric method as described by Ellman. [14].

Principle:

The assay involves carefully optimized enzymatic recycling method using glutathione reductase and Ellman's reagent (DTNB). Glutathione reductase reduces GSSG to GSH. DTNB (5-5'- dithiobis [2-nitrobenzoic acid]) reacts with GSH to form yellow color chromophore, 5-thionitrobenzoic acid (TNB).

DETERMINATION OF CATALASE

Catalase was determined using colorimetric method as described by Fossati et al. [15] was used.

Principle:

Catalase reacts with a known quantity of H2O2. The reaction is stopped after exactly one minute with catalase inhibitor. In the presence of peroxidase (HRP), remaining H2O2 reacts with 3,5-Dichloro -2-hydroxybenzene sulfonic acid (DHBS) and 4-aminophenazone (AAP) to form a chromophore with a color intensity inversely proportional to the amount of catalase in the original sample.

DETERMINATION OF TOTAL ANTIOXIDANT CAPACITY

Total Antioxidant Status was determined using colorimetric method as described by Koracevic et al. [16].

Principle:

The standardized solution of Fe-EDTA complex react with hydrogen peroxide by Fenton-type reaction, leading to the formation of hydroxyl radical (*OH). These reactive oxygen species degrade benzoate, resulting in the release of TBA Reactive Substances (TBARS) and the inhibition of color development defines as anti oxidant activity (AOA).

Statistical Analysis

The data obtained was statistically analyzed using Statistical Package for Social Science (SPSS) version 21. ANOVA was used to determine the difference between the different sicknesses which include; malaria, typhoid, malaria/typhoid, pneumonia and peptic ulcer. Student independent t-test was used to determine the difference between test and control.

RESULTS

Anthropometric parameters

The result showed the comparative analysis of anthropometric characteristics between the sick (test) and apparently healthy subjects (control). In all the variables which include; Age, BMI, SP and DBP, the difference between the subjects were not statistically significant (p> .05) (Table 1).

Table 1. Anthropometric data of test and control groups

Group	n	Age(years)	BMI	SBP(mmHg)	DBP(mmHg)
Group 1	354	18.74±1.18	21.85±3.29	115.71±4.98	73.71±4.87
Group 2	530	18.74±1.18	21.16±3.16	115.71±4.98	73.85±4.90

T- value	0.000	0.391	0.000	-0.173
P- value	1.000	0.696	1.000	0.863

BMI=body mass index, SBP=systolic blood pressure, DBP=diastolic blood pressure.

Oxidative Stress Parameters in Test and in Control Group

The mean MDA (nmol/L) and NO (μmol/L) were significantly higher in the test group (2.29±1.43 and 56.60±14.91) when compared with control group (1.80±0.88 and 45.43±13.63) while the mean TAC (μmol/L), GSH (mg/dl) and CAT (iμ/ml) were significantly lower in the test group (809.20±244.34, 18.94±1.71 and 26.86±7.06) when compared with control group (1091.11± 304.83, 22.05± 3.62 and 39.04± 7.06) ($p < .05$ respectively) (Table 2).

Table 2. Pro-oxidant and Anti-oxidant markers in Test (Sickness) and Control (apparently healthy participants) (mean±SD)

Group	n	MDA(nmol/L)	TAC(μmol/L)	GSH(mg/dl)	NO(μmol/L)	CAT(iμ/ml)
Group 1(Test)	354	2.29±1.43	809.20±244.34	18.94±1.71	56.60±13.63	26.86±7.06
Group2(control)	530	1.80±0.88	1091.11±304.83	22.05±3.62	45.43±14.91	39.04±7.06
T-value		2.435	-6.080	-6.409	-4.625	-7.260
P-value		0.016*	0.000*	0.000*	0.000*	0.000*

MDA=malondealdehyde, TAS=total antioxidant status, GSH=glutathione, NO=nitric oxide, CAT=catalase, *=the mean level is significant at p≤.05.

Oxidative Stress Parameters in different Sickness

The MDA (nmol/L) was significantly higher in students with malaria, typhoid, mala/typhoid and peptic ulcer when compared with students with pneumonia and control group (P<.05 respectively).

The mean NO (μmol/L) was significantly higher in students with malaria, mal/typhoid and peptic ulcer when compared with control group (P< .05 respectively) but was not significantly different between the test groups (P> .05).

However, the mean TAC (μmol/L) was significantly lower in students with mala, mal/typhoid, pneumonia and peptic ulcer when compared with control group (P<.05 respectively). Additionally, TAC (μmol/L) was significantly lower in students with mala/typhoid and pneumonia when compared with students with malaria, typhoid and peptic ulcer disease (P<.05 respectively).

The mean GSH (mg/dl) was also significantly lower in all the test groups when compared with control group (P<0.05 respectively) but was not significantly different when compared between the test groups (P>.05).

Furthermore, CAT (iμ/ml) was significantly lower in all the test groups when compared with control group (P<.05 respectively) but was not significantly different when compared between the test groups (P>.05) (Table 3).

Table 3. Serum mean±SD of Oxidative Stress Parameters in Different Sickness

Group	n	MDA(nmol/L)	TAC(μmol/L)	GSH(mg/dl)	NO(μmol/L)	CAT(iμ/ml)
Group A(Mal)	112	2.15±1.27	787.42±253.61	19.39±2.00	59.00±11.67	27.87±7.81

Group B(Typ)	61	2.17±1.72	850.16±153.07	18.59±1.81	48.92±13.64	23.99±5.16
Group C(Mal/Typ)	103	2.33±1.46	256.50±60.45	18.59±1.53	58.46±13.64	27.07±6.16
Group D (Pneu)	31	1.80±1.31	241.64±108.06	19.31±0.63	47.92±22.79	25.07±7.23
Group E (PUD)	47	2.24±1.33	809.22±32.26	18.64±0.95	60.12±10.99	26.53±6.48
Group F (Contrl)	530	1.80±0.88	1091.11±304.83	22.05±3.62	45.43±14.91	39.04±6.43
F-value		4.748	7.648	8.314	5.702	10.642
P-value		0.007*	0.000*	0.000*	0.000*	0.000*
A/B		0.272	0.476	0.193	0.036*	0.129
A/C		0.676	0.000*	0.126	0.893	0.909
A/D		0.020*	0.000*	0.918	0.090	0.419
A/E		0.590	0.835	0.299	0.842	0.657
A/F		0.002*	0.000*	0.000*	0.000*	0.000*
B/C		0.489	0.000*	0.998	0.065	0.187
B/D		0.007*	0.000*	0.443	0.889	0.781
B/E		0.736	0.733	0.958	0.089	0.464
B/F		0.020*	0.009*	0.000*	0.448	0.000*
C/D		0.001*	0.182	0.414	0.121	0.479
C/E		0.819	0.728	0.956	0.780	0.731
C/F		0.001*	0.001*	0.000*	0.001*	0.001*
D/E		0.002*	0.372	0.507	0.122	0.726
D/F		0.997	0.002*	0.040*	0.705	0.003*
E/F		0.005*	0.012*	0.003*	0.010*	0.002*

A=malaria, B=typhoid, C- malaria and typhoid, D=pneumonia, E= peptic ulcer disease, MDA=malondealdehyde, TAS=total antioxidant status, GSH=glutathione, NO=nitric oxide, CAT=catalase, /=vs, *=the mean level is significant at 0.05 level.

Gender Distribution of Oxidative Stress parameters in the test group

The mean levels of MDA (nmol/L) and NO (μmol/L) were significantly higher in male sick undergraduate students (2.18±1.59, 54.33±13.92) when compared with their female counterparts (2.39±1.39, 54.33±13.30) ($p<.05$ respectively). Conversely, the

mean TAC (µmol/L), GSH (mg/dl)and CAT (iµ/ml) were significantly lower in male sick undergraduate students (785.76±236.96, 18.79±2.09, 29.01±7.60) when compared with their corresponding females (824.25±251.82, 19.13±1.34, 25.16±7.14) (p<.05 respectively) Table 4.

Table 4. Gender distribution of oxidative stress parameters among test group

Group	n	MDA(nmol/L)	TAC(µmol/L)	GSH(mg/dl)	NO(µmol/L)	CAT(iµ/ml)
Group A (male)	169	2.39±1.59	785.76±236.96	18.79±1.34	58.40±13.92	25.16±6.60
Group B (female)	185	2.18±1.39	824.25±251.82	19.13±2.09	54.33±13.30	29.01±7.14
T-value		-3.598	-3.652	3.044	2.295	0.598
P-value		0.012*	0.002*	0.006*	0.018*	0.022*

MDA=malondealdehyde, TAS=total antioxidant status, GSH=glutathione, NO=nitric oxide, CAT=catalase, *=the mean level is significant at 0.05 level.

DISCUSSION

The imbalance between oxidant and antioxidant is a resultant of oxidative stress and depressed body defense mechanism. Antioxidant enzymes play a significant role in the body by depressing the harmful activies of free radicals. Measurement of these antioxidants is therefore an indirect and non-invasive test that could be used to assess the induction of oxidative stress in a cell.

The present study observed significantly higher levels of MDA and NO with lower levels of TAS, GSH and CAT in the sick participants when compared with the apparently healthy participants. This reveals sickness induced oxidative stress. Previous studies have noted the negative influences of oxidative stress in lifestyle diseases [17, 18]. Many intrinsic and extrinsic factors adversely affect the body homeostasis molecularly thereby, causing stress as well as ill health and immunosuppression in the affected individuals and exposing them to various types of pathogens. The intrinsic factors include exam stress and malnutrition [8, 19, 20] while, the extrinsic sources of oxidative attack which can affect the students include radiation (both ionizing and non-ionizing), atmospheric pollutants, biological and chemical toxins, toxic gasses and oxidizing disinfectants [21]. Foreign microbes invading the body due to most times unhygienic environments of some students hostels and campuses, routine ingested foods with low nutrient value can lead to the production of tissue or cell-damaging oxidants thereby disturbing immune responses and causing oxidative stress [22, 23]. It has been noted that the high intake of fast foods, processed foods, meat and other unhealthy life styles can induce oxidative stress [21]. During oxidative stress, the released ROS might overwhelm the antioxidant defense mechanism mediated by GSH, CAT, and TAS amongst others, thereby reducing their level. MDA and total anti-oxidant status (TAS) values have been employed as qualitative indicators for the oxidant-antioxidant balance in human homeostasis as well as biomarkers for cell membrane degeneration, stresses or diseases induced by free radicals following increased membrane lipid peroxidation [24, 23]. MDA can enhance the depletion of glutathione, increase proinflammatory cytokines, and accelerate the collagen deposition by stellate cells, thereby increasing oxidative stress with induction of cell or tissue damage [25]. Antioxidants enzymes such as superoxide dismutase (SOD) catalyzes the oxidation and reduction of superoxide radicals to hydrogen peroxide (H2O2), CAT breaks down and inhibits H2O2 conversion to hydroxyl radical, while GPx through GSH stops the release of hydrogen peroxide [26]. Superoxide anions and hydrogen peroxide interact in a Fe2+-catalyzed redox reaction to generate hydroxyl radical which has been documented as the most harmful ROS [27, 26]. The oxidative stress in sickness as observed in the present study corroborates the findings of Tsatsakis et al. [28] and Padureanu et al. [29]. Reactive oxygen species have been associated with many disease conditions including cancer, immune disorders and cardiovascular diseases [30, 28].

All the factors responsible for the oxidative stress directly or indirectly participate in immune system defense mechanism. Any alteration leading to immunosuppression can trigger disease development. Studies have proven that individuals deficient in antioxidants are more susceptible to severe bacterial and fungal infections [6]. Reactive species are important in killing pathogens

but as a negative side effect can also injure the host tissues. This is particularly apparent during chronic inflammation, which may cause extensive tissue damage with a subsequent burst in oxidative stress [31]. The production of free radicals involves macrophages and neutrophils to combat the invading microbes. The whole of the process is performed in host cells during the activation of phagocytes or the effect of bacteria, virus, parasites, and their cell products reactivity with specific receptors. These cellular damages in general lead to altering immune response to microbes and ultimately altered susceptibility to bacterial, viral, and parasitic infections [32].

Biological stress also is a complex phenomenon involving many biochemical changes which can contribute to oxidative stress. Oxidative stress has been implicated in the causation or display of psychiatric disorder [2, 33]. Previous study has linked psychological stress to adverse effects of ROS, leading to oxidative stress [5]. The anxiety or distress experienced by students anticipating examinations will also increase oxidative stress. Reports showed that reactive oxygen and nitrogen species such as hydrogen peroxide, superoxide are produced in the body in both physiological and pathological conditions [7, 34, 35]. In physiological conditions, they increase as regulatory mechanisms, intercellular signalling species, or as bactericidal agents. Their production is continuously regulated by the antioxidant defense systems though oxidative cell damages still occur [7, 35].

Furthermore, increased levels of MDA, NO and decreased TAS, GSH and CAT in male than in female sick participants in this study revealed higher oxidative stress in male than in female. This may be attributed to the antioxidant properties of estrogen, gender differences in NADPH-oxidase activity and other mechanisms [36, 37, 38]. Barp et al. [39] also reported a higher oxidative stress in male rat than the female. Another study showed that in vivo biomarkers of oxidative stress were higher in young men than in women of the same age [40]. Similarly, it was observed that ROS production was higher in the vascular cells from males than in the cells from females [41]. Bilgin etal. [42] and Bhatia et al., [43] reported a greater antioxidant potential in females over males. This study indicates that there is an apparent association between gender and oxidative stress, where women seem to be less susceptible to oxidative stress. In some studies, catalase activity levels were found to be the same between genders in the brain, heart and lung but higher in the female kidney [44].

Increased MDA, NO and reduced CAT, GSH and TAS observed in malaria infected participants can result from the reactive role of the host to malaria parasite infection. This observation is in support of the work of Atiku etal. [45] and Babalola et al. [46]. Malaria has been proven to be an oxidative disease [47]. Additionally, oxidative stress, hypoxia, increased inflammation, and hepatocyte apoptosis in malaria-infected livers have been previously reported [48]. Oxidative stress is considered as the main factor in the pathogenesis of malaria and contributes to the severity of malaria related complications. The free radical productions are triggered by malaria parasite which will subsequently lead to an antioxidant defense in the host cell to halt the infection [47, 48]. The release of ROS brings about the eradication of the parasite while the increase production of these free radicals on the other hand can trigger inflammation process resulting in severe degeneration of host cells and tissues leading to serious infections [47]. Free radical production in malaria infections have been attributed to the host's hemoglobin molecule, because heme molecules is used as a source of amino acids for its own nutrition during the erythrocytic stage of the disease, resulting in the excessive release of circulating heme. The presence of Fe2+ and its oxidized groups in this process can trigger intravascular oxidative stress, thereby affecting the erythrocytes and endothelial cells and increase the interiorization of the parasite in tissues including the liver and brain [49, 50]. Previous reports noted that virulence of malaria parasite infection depends largely on the patients' antioxidant capacities, which in turn is determined by the concentrations of antioxidant micronutrient [45, 51]. Reduced glutathione (GSH) in this case play a vital role in scavenging the free radicals released as a result of malaria infection.

Some studies reported that increased Nitric oxide (NO) is involved in progression of malaria infection to cerebral malaria and other severe complications though its role is still under deliberation [52, 53, 54]. Other authors on the other hand, suggest that cerebral malaria results from a low bioavailability of NO [55, 56]. Moreover, reactive oxygen species are also generated in the mitochondria through various metabolic processes. These free radical productions are triggered by malaria parasite which will subsequently lead to an antioxidant defense in the host cell to halt the infection [57].

Decrease in the levels of antioxidants in the human body, has recently attracted close attention as it is thought to underlie the development of numerous lifestyle disease conditions. In all the oxidative stress markers evaluated in the various diseases including malaria, typhoid, malaria and typhoid, peptic ulcer and pneumonia, the results showed evidence of oxidative stress in malaria, typhoid, malaria and typhoid, peptic ulcer. Based on the finding of this study, it could be deduced that reactive oxygen species occasioned by oxidative stress contribute immensely to the destructive effect of malaria parasites. This is in agreement with the report of Vasquez et al. [47] which suggests that excessive amounts of free radicals can cause severe pathologies to host cells and tissues irrespective of the ability to clear malaria parasites. This contributes to the complexity and ambiguity in the use of oxidative stress in the validation of or underpinning of oxidative stress markers as diagnostics tools in infectious diseases. However, plasmodium has a variety of antioxidant enzymes which includes reduced production of

glutathione that allow it to survive amidst this oxidative onslaught [47, 58]. More so, NO was found to be significantly higher in the participants with malaria, mala/typhoid and peptic ulcer disease when compared with the participants with typhoid while, TAS was significantly lower in the participants with mala/typhoid and pneumonia compared to the participants with malaria, typhoid and peptic ulcer. This may be due to the role of NO in the reduction or inhibition of malaria parasite. Other studies reported similar findings in both human and non-human studies. Luckhart et al. [59] reported that NO in inhibits plasmodium development in Anapheles misquitoes while Onyeneke et al. [60] reported increase in the level of NO in malaria infection in pregnant women. Nitric oxide plays a pathophysiological role in malaria infection and as a result many diseases may be related to a high or low NO level in the body. Though, the role of nitric oxide in blood-stage malaria is still very unclear [61]. The author stated that intraerythrocytic Plasmodia are protected from reactive oxygen species by hemoglobin. While, some other researchers suggest that NO has a protective role against blood-stage malaria and claim that NO is an essential factor for malaria resolution by P. Falciparum. They reported that higher serum nitric oxide levels are toxic to the parasite [62]. These effects are vital in preventing cerebral malaria, the introduction of exogenous NO, or substances that release NO, has been accepted as supportive treatment for management of malaria, with great results in improving microcirculation, reducing brain inflammation and protecting the blood-brain barrier [63, 64] thus, decreasing oxidative stress. Exogenous NO is also indicated in the prophylaxis of lung damage of malaria patients [65]. It can be toxic especially in situations of oxidative stress, which increases free radicals and reduction of antioxidants [66]. In malaria, moderate generation of reactive oxygen species may be helpful because is an essential component of the response to Plasmodium infection by phagocytes as it contributes to the clearance of the parasites [47]. Moreover, medications such as antimalarial enhance the induction of oxidative stress since their protective oxidative killing of the parasite leads to oxidative stress [47]. It has been indicated that NO can act both positively and negatively on malaria resolution as described by several authors [48, 66, 67, 68]. The authors resolved that NO has a parasiticidal role in malaria, since high serum levels of it favor parasitemia resolution without harming the host as the gas formerly regarded as a noxious agent, is contemplated today as a way to resolve this disease, since it may directly act on the inflammatory process and indirectly enable the necessary cytokines to stimulate the immune system.

With regards to the level of the oxidative stress markers concerning the various disease conditions, enhanced oxidative stress culminating in MDA and depletion of other antioxidants parameters was observed in malaria, typhoid, malaria/typhoid and peptic ulcer infected participants. This is inconsonance with established evidence. In typhoid infection, the causative agent, Salmonella typhi resides within the Kupffer cells in the liver [69] and it is typical of the Kupffer cells to respond to foreign particulate matter by phagocytosis and excess production of ROS could trigger hepatic injury following the decrease of endotoxin removing the potential of Kupffer cells [69]. ROS have been linked with various inflammatory gastrointestinal disorders such as gastroduodenal inflammation, ulceration, and gastric cancer [70]. Excessive levels of ROS can damage the cellular proteins [71] including cytoskeletal proteins and, ultimately, disrupt the gastrointestinal tract barrier to increase gut permeability which contributes to inflammation in a variety of gastrointestinal diseases [72]. Our study did not show significant oxidative stress in the university students with pulmonary infection. Depletion in antioxidant system has been previously linked to progressive pulmonary fibrosis [73]. Several researches have also reported that lung diseases such as asthma and chronic obstructive pulmonary disease (COPD), are linked to oxidative stress [74, 75]. Pro-oxidants have been shown to elicit inflammation through the stimulation of different kinases involving pathways and transcription factors like NF-kappa B and AP-1 [76].

CONCLUSION

The alteration in the pro-oxidant markers is accompanied by imbalance in the antioxidant defense status resulting in oxidative stress. Higher TAS, GSH, CAT and NO in females than in males showed that males are more susceptible to oxidative insult than females. This shows that oxidative stress may have contributed to the pathogenesis and severity of different disease conditions observed in the undergraduate students which may cause irreversible cell damage if not managed. A national intervention for student's health and social wellbeing is strongly advocated.

ETHICS STATEMENT

The ethical approval for this study was obtained from the board of ethics committee of Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State, Nigeria, while, the informed consent was sought and obtained prior to the study

AVAILABILITY OF DATA AND MATERIAL

The authors confirm that the data supporting the findings of this study are available within the article

COMPETING INTEREST

Authors declare no conflict of interest.

FUNDING

"This research received no external funding

AUTHORS' CONTRIBUTIONS

Conceptualization, JCA; Data curation, JCA, NRU, ACI and TU; Formal analysis, JCA, ACI, TU CEO and FAE; Investigation, JCA, NRU, ACI , TU, CEO, FAE, EGU and BCU; Methodology, JCA, CCO and NRU; Project administration, JCA; Resources, JCA, NRU, ACI, CEO, FAE and TU; Software, JCA,CCO, NRU and JEA; Supervision, CCO, NRU and JEA; Validation, CCO, NRU and JEA; Visualization, CCO, NRU and JEA; Writing – original draft, JCA, NRU and CCO; Writing – review & editing, JCA, CCO, NRU, JEA, ACI ,TU, CEO, FAE, EGU and BCU.

ACKNOWLEDGMENTS

Authors wish to acknowledge the students who voluntarily gave their consent for this study.

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