



Study of Glutathione S Transferases and Malondialdehyde Levels in Male Smokers from Vidharbha Region, India

**Ranjit S. Ambad^{1*}, Suryakant Nagtilak², Ankita Kondalkar¹
and Ashish Anjankar³**

¹Department of Biochemistry, Datta Meghe Medical College, Shalinitai Meghe Hospital and Research Centre, Nagpur (Datta Meghe Institute of Medical Sciences, Sawangi (Meghe), Wardha), India.

²Department of Biochemistry, NAMO Medical Education and Research Institute Silvassa DNH, India.

³Department of Biochemistry, Jawaharlal Nehru Medical College, Datta Meghe Institute of Medical Sciences, Sawangi (Meghe), Wardha, India.

Authors' contributions

This work was carried out in collaboration among all authors. Author RSA conceptualization of research paper, literature search, writing manuscript, data collection and analysis of the results, writing and editing of manuscript. Authors SN, AK, AA literature search, writing manuscript, data collection and analysis of the results. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JPRI/2021/v33i36B31964

Editor(s):

(1) Dr. S. Prabhu, Sri Venkateswara College of Engineering, India.

Reviewers:

(1) Sangeetha Arullappan, Universiti Tunku Abdul Rahman (UTAR), Malaysia.

(2) Mohammad Daud Ali, Mohammed Al-Mana College for Medical Sciences, Saudi Arabia.

Complete Peer review History: <https://www.sdiarticle4.com/review-history/71077>

Original Research Article

**Received 02 May 2021
Accepted 09 July 2021
Published 12 July 2021**

ABSTRACT

Introduction: Free radicals are compounds having two unpaired electrons. Free radicals oxidize macromolecule in the body, including proteins, nucleic acids, and lipids.

The body uses antioxidants to fight free radicals. Antioxidants may come from the diet or be generated internally (endogenous). Endogenous antioxidants include superoxide dismutase, glutathione (GSH), catalase, and glutathione peroxidase.

Aim: To study of glutathione s transferases and malondialdehyde (MDA) levels in male smokers from Vidharbha region attending Datta Meghe Medical College

Materials and Methods: A minimum sample size of 80 was used with 40 smoking males and 40 non-smoking males. This was a cross-sectional analytical observational analysis conducted in the

*Corresponding author: E-mail: ambad.sawan@gmail.com;

medical study.

Result: The mean GSH concentrations of the smoking and non-smoking research participants were 1.69 ± 0.97 mol/L and 2.20 ± 1.40 mol/L, respectively, when plasma GSH concentrations were measured. Smokers had lower plasma GSH concentrations than non-smokers, but the disparity was not statistically significant ($p=0.1885$) suggests that smoking has no discernible effect on GSH levels.

Conclusion: Smoking tends to reduce plasma GSH while rising plasma MDA levels in men. Owing to a loss of antioxidant potential and a higher oxidative load, smokers are more susceptible to oxidant stress. Young smokers, in particular, should quit smoking as soon as possible before developing health complications in order to reap the full benefits of quitting.

Keywords: MDA; GSH; Deoxyguanosine; oxidative stress and Antioxidant.

1. INTRODUCTION

Free radicals are compounds having two unpaired electrons form. Free radicals oxidize any macromolecule in the body, including proteins, nucleic acids, and lipids. Malondialdehyde (MDA) and deoxyguanosine P are carbonyl compounds formed when proteins, nucleic acids, and lipids are oxidized by free radicals. [1,2] Our body uses antioxidants to remove free radicals. Antioxidants may be endogenous. Endogenous antioxidants include superoxide dismutase, glutathione, catalase, and glutathione peroxidase. GSH is one antioxidant that is often examined to see how a rise in free radicals affects the body. Glutathione has the molecular formula $C_{10}H_{17}N_3O_6S$ and its molecular weight is 307.3235 g/mol. Glutathione is an antioxidant that is made up of the amino acids glutamate, cysteine, and glycine. Glutathione can be found in any cell in the body, but the liver has the highest concentration. The cysteine thiol (SH) group is responsible for glutathione's biological activity and acts as a proton donor. Toxic agents are introduced into the body by smoking. Inhaled cigarette smoke contains toxic chemicals that are harmful to one's skin. Smoking remains a public health concern worldwide, especially in developed countries like Indonesia. In Indonesia, cigarette use is slowly increasing [3]

From 34.2% in 2007 to 36.2% in 2013, Indonesia's smoking rate rose. According to the World Health Organization (year), if the Indonesian government does not introduce a strict tobacco control policy, the number of smokers in the country could increase by 45 %by 2025. (2). Looking at After China, the United States, Russia, and Japan, Indonesia is ranked sixth in the world in terms of tobacco consumption. According to the results of a survey undertaken by the Ministry of Health of the

Republic of Indonesia in 2011, non-university students account for 31% of smokers in Indonesia, and private university students in Sumatra account for nearly 75% of smokers. The age group aged 18to 24 years in the United States has the largest prevalence of smokers. [4]

Toxic compounds in tobacco smoke, such as tar, nicotine, lead, and carbon monoxide, are considered to be harmful to one's health. The use of smoking raises blood lead levels. [5]

Furthermore, smoking can impair lung capacity and increase the risk of chronic obstructive pulmonary disease and skeletal muscle weakness over time. A research on laboratory animals found that tobacco smoke intensified the death of pulmonary alveolar cells. Free radicals may be induced by the gaseous and particulate components of tobacco smoke. Free radicals were found in smokers' periodontal tissues, according to the findings of a report. Another study found that exposure to nicotine, whether electronic or traditional, would disrupt rat spermatogenesis by increasing free radical levels. Since free radicals are highly reactive molecules, they can readily react with other substances.

A rise in oxidants (free radicals) and/or a reduction in antioxidants in the body is referred to as oxidative stress. The MDA concentration is one of the most commonly used markers to assess oxidative stress. MDA is the product of free radicals peroxidizing lipids. A rise in MDA levels in the body means an increase in free radicals. Aside from MDA, glutathione is another marker that is commonly used to assess oxidative stress (GSH). GSH is an endogenous antioxidant that aids in the cleavage of free radicals. The level of GSH reduces under oxidative stress. In addition to MDA and GSH, oxidative stress markers include superoxide dismutase (SOD), glutathione peroxidase (GPx),

catalase, and carbonyl compounds. Smoking has been related to increased aging, cell death, and disease via the process of oxidative stress. There is a propensity for promoter of free radicals to rise and antioxidants to decrease in smokers. However, these experiments included people of all ages, from infants to the elderly. In a study by Koubaa et al. [6] MDA concentrations were shown to be lower in smokers than in non-smokers. In a study of thirty healthy adult subjects, no significant variations in GSH and MDA concentrations were seen in smokers (15 smokers and 15 non-smokers). Latest study on the relationship between smoking and MDA and GSH concentrations in smokers was agreed to be conducted due to the large number of smokers in Indonesia, especially among university students, and the conflicting findings of previous studies. [7]

Since disturbances in glutathione synthesis and metabolism cause impaired glutathione activity as an antioxidant, glutathione levels in the body become a significant factor to consider. 8 The amount of malondialdehyde released as a result of lipid peroxidation is used to determine antioxidant activity. Malondialdehyde has the chemical formula $C_3H_4O_2$ and is a volatile molecule. According to Suparno et al. 9 high levels of plasma MDA, an oxidative stress marker, may trigger DNA disorders. Thus the study aims to evaluate glutathione s transferases and malondialdehyde levels in male smokers from vidharbha region

2. MATERIALS AND METHODS

Study comprising randomly selected smokers (≤ 10 cigarette per day aged 25 to 49 yrs)

The participants' willingness to participate in the study was recorded by signing informed consent that included their name, age, gender, smoking frequency, and daily cigarette consumption. This was a cross-sectional analytical observational analysis conducted in Dept of Biochemistry, Datta Meghe Medical College and Shalinitai Meghe Hospital and Research Center.

Study Period: September 2020 to January 2021.

2.1 Sample Collection

A 3-mL syringe was used to extract a 2 mL blood sample from each (non-fasting) study participant. The blood samples were centrifuged at 5000 rpm for 2 minutes to extract plasma, which was then

collected at -20°C before being used to measure GSH and MDA levels. The Biochemistry and Molecular Biology Laboratories of Datta Meghe Medical College's Faculty of Medicine and Shalinitai Meghe Hospital and Research Centre. assessed plasma GSH and MDA concentrations. Using the Sigma GSH and MDA assay kits, the levels of plasma GSH and MDA is calculated biochemically. The Sigma GSH assay kit was used to biochemically test plasma GSH concentrations. GSH and MDA levels were assayed by readymade quality control assayed kits manufactured by M/S- chemicals, USA. Below are the criteria for the sampling.

- **Inclusion criteria:**

- a. The participants in the survey were all smokers between the ages of 25 to 49.
- b. The only condition for participation was that the men were in good health.

- **Exclusion Criteria:**

- a. Males who were sick or with a diagnosis of a metabolic disease were number one.
- b. Such as diabetes mellitus, heart disease, and kidney disease.

3. Results

Smoking survey participants had an average age of 20.0 ± 2.03 years, a mean smoking length of 6.0 ± 1.0 years, and an average daily cigarette consumption of 10.0 ± 3.34 cigarettes. Table 1 shows the features of the subjects.

The mean GSH concentrations of the smoking and non-smoking study participants were 1.69 ± 0.97 mol/L and 2.20 ± 1.40 mol/L, respectively, when plasma GSH concentrations were measured (Table 2). Smokers had lower plasma GSH concentrations than non-smokers, but the disparity was statistically non-significant ($p=0.1885$). This suggests that smoking has no discernible effect on GSH levels. The study group had a mean MDA concentration of 2.80 ± 2.11 nmol/L, while the non-smoking study subjects had a mean MDA concentration of 1.46 ± 1.28 nmol/L, according to the results. As a result, smokers had higher plasma MDA concentrations than non-smokers, but the disparity was not statistically important ($P = 0.0200$) (Table 2).

Table 1. Smokers and nonsmokers have different features

Variable	Smokers(n=40)	Non-smokers(n=40)
Age(years)	20.00 ±2.03	21.00 ± 0.82
Duration of smoking years	6.00 ± 1.00	--
Number of cigarettes per day	10.00 ± 3.34	--

Table 2. Smokers and nonsmokers have different GSH and MDA concentrations

Variable	Smokers n =40	Non-smokers n=40	P-value
GSH(µmol/L)	1.69 ±0.97	2.20±1.40	P = 0.1885
MDA(µmol/L)	2.80± 2.11	1.46 ±1.28	P = 0.0200

4. DISCUSSION

The age group in study group were 25to 49 years old and had been smoking for an average of 5 years. In terms of average smoking length, it can be calculated that the research participants began smoking when they were 13to 17 years old, i.e. when they were teenagers (in junior or senior high school). The causes that cause these teenagers to start smoking at such a young age include social influence, self-actualization, a labile temperament, and a high level of curiosity. The results of this study matched those of Reimondos et al. [10] and Subandrate et al. [11] reported that the average age of smoking initiation was 13to 15 years. Our smokers were habitual or routine smokers who smoked an average of nine cigarettes a day. This is in line with previous studies, which showed that smokers consumed an average of 10to 11 cigarettes a day, or one pack.

Toxic agents are introduced into the body by smoking. Over 2000 chemicals enter the lungs as gaseous or particulate matter as you smoke. Toxic chemicals in cigarette smoke, such as tar, nicotine, lead, carbon monoxide, nitrogen monoxide, benzopyrene, and nitrosamines, are metabolized in the liver and produce oxidants, or free radicals [12] Long-term and continuous smoking causes toxic compounds to accumulate in the bloodstream, resulting in an uptick in free radicals including hydroxyl and superoxide radicals. When free radicals increase, oxidant markers like MDA rise, whereas antioxidant markers, level of GSH, glutathione peroxidase, catalase, and superoxide dismutase diminished. [13]

In this analysis, smokers had lower mean GSH concentrations than non-smokers, but the disparity was not statistically important. According to the findings of Abdul-Rasheed et al. [14] smokers' glutathione concentrations are not

significantly lower than non-smokers' glutathione concentrations. In another analysis, however, GSH concentrations were observed to be lower in smokers as compare to non-smokers. Free radicals such as hydroxyl radicals, hydrogen peroxide, and superoxide anions may have increased as a result of the accumulation of cigarette smoke oxidants. Endogenous antioxidants like GSH become more competitive when free radicals increase, lowering their plasma concentrations. GSH is a tripeptide with a sulfhydryl group that can cleave free radicals. GSH may specifically quench free radicals or cleave hydroxyl radicals and superoxide anions by acting as a cofactor of glutathione peroxidase, glutathione transhydrogenase, and superoxide dismutase. Present study reported that smokers had higher mean MDA levels than non-smokers, but the difference was not statistically significant. In an analysis performed by Erguder et al. [15] similar non-significant findings were found, who reported no significant variation in serum MDA concentration between smokers and non-smokers. In contrast, a survey of 22-25 year-old Indian students documented a significant increase in serum MDA concentration in smokers relative to non-smokers. This may have been possible due to oxidative stress induced by the build-up of toxic compounds and superoxide radicals caused by long-term and frequent tobacco use. MDA is the end product of a series of free radical-mediated lipid peroxidation reactions. MDA concentrations that rise mean that the cells are under oxidative stress. [16-20]

5. CONCLUSION

The smoking tends to reduce plasma GSH while rising plasma MDA levels in smokers. Owing to a loss of antioxidant potential and a higher oxidative load, smokers are more susceptible to oxidant stress. Young smokers, in particular, should quit smoking as soon as possible before developing health complications in order to reap

the full benefits of quitting. Present study supports hypothesis that any form of tobacco is dangerous to health.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT AND ETHICAL APPROVAL

As per international standard or university standard guideline participant consent and ethical approval has been collected and preserved by the authors.

ACKNOWLEDGEMENT

Author acknowledges the immense help received from the scholars whose articles are cited and included in references of this manuscript. The author is also grateful to authors / editors / publishers of all those articles, journals, and books from where the literature for this article has been reviewed and discussed. Author also acknowledges Department of Medicine, Datta Meghe Medical College, Nagpur.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Ariyani R. Essay. Jakarta: Indonesia University; Study of detection DNA-adduct 8- hidroksi-2'-deoksiguanosin as a biomarker of cancer risk on worker at some Gas Station Jakarta; 2009.
2. Oxidative stress and antioxidant defense. Birben E, Sahiner UM, Sackesen C, Erzurum S, Kalayci O. *World Allergy Organ J.* 2012;5(1):9-19.
3. World Health Organization. WHO global report on trends in prevalence of tobacco smoking, 2015. Geneva: World Health Organisation; 2015.
4. Buran T, Sanem Gökçe Merve Kılınç, Elmas Kasap. Prevalence of Extraintestinal Manifestations of Ulcerative Colitis Patients in Turkey: Community-Based Monocentric Observational Study. *Clinical Medicine and Medical Research.* 2020;1(2):39-46. Available: <https://doi.org/10.52845/CMMR/2020v1i2a8>
5. Departemen Kesehatan RI. Riset kesehatan dasartahun. Jakarta: Badan Penelitiandan Pengembangan Departemen Kesehatan Republik Indonesia, 2013.
6. Nurbaya F, Wijayanti Y. Faktor risiko yang berhubungan dengan kadar timah hitam dalam darah. *Kemas* 2010;6:51-6.
7. Koubaa A, Triki M, Trabelsi H, et al. The effect of a 12-week moderate intensity interval training program on the antioxidant defense capability and lipid profile in men smoking cigarettes or hookah: a cohort study. *Sci World J* 2015;2015: 1-9.
8. Daniel V, Daniel K. Diabetic neuropathy: new perspectives on early diagnosis and treatments. *Journal of Current Diabetes Reports.* 2020;1(1):12-14. Available: <https://doi.org/10.52845/JCDR/2020v1i1a3>
9. Safyudin Safyudin, Subandrate Subandrate smoking tends to decrease glutathione and increase malondialdehyde levels in medical students. 2016(08).
10. Yuniastuti A. Dasar Molekuler Glutathion Dan Perannya Sebagai Antioksidan. Central Java: Universitas Negeri Semarang; 2016.
11. Suparno S, Suhartono S, Sofro MAU, Sulchan M, Tjahjono K. Kadar seng dan kadar malondialdehyde pada penderita multi drug resistant tuberculosis dan tuberkulosis sensitif. *Jurnal Gizi Indonesia.* 2018;7(1):8 DOI: 10.14710/jgi.7.1.8-14.
12. Daniel V, Daniel K. Perception of Nurses' Work in Psychiatric Clinic. *Clinical Medicine Insights.* 2020;1(1):27-33. Available: <https://doi.org/10.52845/CMI/2020v1i1a5>
13. Subandrate, Safyudin, Arifin M, et al. Kadar superoksida dismutase mahasiswa perokok di Program Studi Pendidikan Dokter Universitas Sriwijaya. *Jurnal Kedokteran Yarsi* 2015;23:76-82.
14. Reimondos A, Utomo ID, Mc Donald P, et al. Smoking and young adults in Indonesia. *Australian Demographic and Social*

- Research Institute, The Australian National University; 2012.
15. Barreiro E, Peinado VI, Galdiz JB, et al. Cigarette smoke-induced oxidative stress. *Am J Resp Crit Care Med.* 2010;182:477–88.
 16. Daniel V, Daniel K. Exercises training program: It's Effect on Muscle strength and Activity of daily living among elderly people. *Nursing and Midwifery.* 2020; 1(01):19-23.
Available:<https://doi.org/10.52845/NM/2020v1i1a5>
 17. Rajalakshmi G, Sudha G. Reduced plasma antioxidant level in smoking asthmatic. *Int Res J Pharm.* 2013;3:142-4.
 18. Abdul-Rashees OF, Al-Rubayee WT. Effects of cigarette smoking on lipid peroxidation and antioxidant status in Iraqi men at Baghdad city. *Int J Basic Appl Sci* 2013;2:47-50.
 19. Erguder IB, Ucar A, Ariturk I, et al. The effects of cigarette smoking on serum oxidant status, and cholesterol, homocysteine, folic acid, copper, and zinc levels in university students. *Turk J Med Sci.* 2009;39:513-7.
 20. Mahapatra SK, Das S, Dey SK, et al. Smoking induced oxidative stress in serum and neutrophil of the university students. *Al Ameen J Med Sci.* 2008;1:20-31.

© 2021 Ambad et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

The peer review history for this paper can be accessed here:
<https://www.sdiarticle4.com/review-history/71077>