Origional Article

ANTIOXIDANT ROLE OF MENTHA (MINT) AGAINST CHLOROQUINE-INDUCED OXIDATIVE STRESS IN MALE ALBINO MICE

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ABSTRACT

Background: Many drugs, in the world, have been found to induce oxidative stress when these are given at larger doses or for longer periods. Worldwide research work is being carried out to find the antioxidant role of many herbs and plants so that these can be given prophylactically to prevent oxidative damage, done by free radicals. The objective of the study was to determine the antioxidant role of mint against chloroquine induced oxidative stress.

Material and Methods: In this randomised controlled study, ninety male albino mice were divided into three groups randomly. Each of the three groups contained 30 mice. Group A was labelled as a control group and Group B and C were labelled as experimental groups. Chloroquine (970 mg/kg of body weight) was given orally to the mice of group B, on the 9th day of the experiment. The mice of group C were given an ethanolic extract of mint consecutively for the initial eight days then they were given chloroquine, at the dose of 970 mg/kg of body weight, on the ninth day. The ethanolic extract of mint was then continued to be given from day 10 to day 16 of the experiment. Blood samples of the mice were obtained on the 17th day of experiment by intracardiac puncture technique. SPSS version 20 was used to analyze the data.

Results: A highly significant (p=0.000) decrease in serum glutathione peroxidase and a highly significant (p=0.000) increase in serum malondialdehyde was observed in mice of group B (whom chloroquine was given) as compared to those of group A. Group C mice, to whom ethanolic extract of mint was given before and after the administration of chloroquine, showed a highly significant decrease and highly significant increase in serum levels of malondialdehyde and glutathione peroxidase respectively.

Conclusion: Chloroquine, when given at a dose higher than the therapeutic dose in mice, can induce oxidative stress. Mint has antioxidant potential against chloroquine induced oxidative stress.

Key Words: Chloroquine, mentha, oxidative stress, antioxidant.

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INTRODUCTION

Drugs, when given at higher doses or for longer duration, can generate free radicals, which can induce oxidative stress. Hence, the drugs are responsible for many side effects due to their oxidative potential.

For example, it has been reported that 50 % of liver failure cases in the world occur due to drug-induced hepatotoxicity.[1] It is estimated that more than 1000 drugs are responsible for acute liver failure.[2]

Previous research has revealed that hepatotoxicity is induced by various drugs due to oxidative stress induction.[3]

The radicals, generated by the drugs, can attack the lipids of the cell membranes.[4] When polyunsaturated fatty acids are degraded, certain secondary metabolites are formed, such as malondialdehyde.[5] The raised serum levels of malondialdehyde are indicative of oxidative stress induction.[6] Chloroquine is also one of the drugs which has the potential to induce oxidative stress if a dose higher than the therapeutic dose is taken.[7] Chloroquine, if given at a dose higher than the therapeutic dose, can induce hepatotoxicity due to the

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induction of oxidative stress.[8] The enzymatic and non-enzymatic antioxidants, in the body, prevent oxidative stress.[9] These antioxidants can scavenge free radicals. Excess formation of free radicals leads to excessive utilisation of antioxidants.[10] As a result, in oxidative stress, the serum levels of antioxidants are markedly reduced.[11]

Researchers have been going around the world to find different herbs and plants which have antioxidant potential.[12] Mint (mentha) is one such plant, which is being studied for its antioxidant potential.[13] It has been found that mint is a rich source of antioxidants.[14] Flavonoids and polyphenols are present in abundance in the mint. These antioxidants activate and make new antioxidant enzymes in the body. These antioxidants also scavenge free radicals. 25 different species of mint have been discovered in the world. In Pakistan, mentha arvensis is one of the most widely used species.

The aim of this study is to access antioxidant role of mentha (mint) against chloroquine-induced oxidative stress in male albino mice.

MATERIAL AND METHODS

Ninety male albino mice were included in this randomized controlled study. The mice were bought from the University of Veterinary and Animal Sciences, Lahore. Mice were selected by non-probability consecutive sampling method. Mice were then divided into three groups randomly, with each group containing 30 mice. Group A was labelled as a control group. Group B was the experimental group, in which each mouse was given chloroquine orally, at the dose of 970 mg/kg of body weight on the 9th day of the experiment. Group C was also an experimental group in which ethanolic extract of the mint was given from day 1 of the experiment to day 8 of the experiment. Then, chloroquine at the dose of 970 mg/kg of body weight was given on day 9. The ethanolic extract of mint was continued to be given to the mice of that group from day 10 to day 16 of the experiment. Blood samples of the mice were obtained on the 17th day of the experiment by intracardiac sampling technique. Data was analysed by SPSS version 20.

RESULTS

When results were compared by one way ANOVA test among groups A, B and C, it was seen that highly significant differences in the serum values of malondialdehyde and glutathione peroxidase existed among the three groups (Table1).

Table 1. Comparison of serum malondialdehyde and glutathione peroxidase, among groups A, B and C, by one way ANOVA.

Parameters	Group A (n= 30)	Group B (n= 30)	Group C (n=30)	p-value
Serum malondialdehyde (ng/ml)	0.12±.037	0.22±.12	0.18±.05	0.00
Serum glutathione peroxidase (mg/dl)	1.03±.013	0.79±.12	0.94±.17	0.00

Values are presented as mean ± SD

*p< 0.00 highly significant.

Highly significantly raised serum levels of malondialdehyde were observed in mice of group B, in which a single oral dose of chloroquine was given. While highly significant decline in the serum levels of glutathione peroxidase was observed in the same mice. (Table 2) A highly significant decrease in serum malondialdehyde was observed in mice of group C, in which ethanolic extract of mint was given. Highly significantly raised serum levels of glutathione peroxidase were observed in group C mice. (Table 2)

Table 2: Comparison of serum malondialdehyde and glutathione peroxidase by post hoc Tukey test among the groups A, B and C.

Group comparisons	Serum malondialdehyde (ng/ml)	Serum glutathione peroxidase (ng/dl)
Group B versus Group A	0.00	0.00
Group Versus Group B	0.00	0.00

Values are presented as mean ± SD

DISCUSSION

The comparison of the results of serum malondialdehyde and glutathione peroxidase between group A and group B revealed a highly significant increase in the level of serum malondialdehyde, while a highly significant decrease in the level of serum glutathione peroxidase in group B as compared to that in group A. The highly significant elevated levels of serum malondialdehyde are indicative of lipid peroxidation because malondialdehyde is a secondary metabolite which is produced during the lipid peroxidation process. This shows that oxidative stress had been induced by chloroquine.

The serum levels of glutathione peroxidase were declined as a result of their excessive utilisation in scavenging the free radicals, which were produced in the lipid peroxidation process. These findings are consistent with those other studies.[15] For induction of hepatotoxicity in female Wister rats, they used chloroquine. The chloroquine was given at the dose of 970 mg/kg of body weight orally. Hydroperoxides and thiobarbituric acid reactive substances were found to be raised significantly. Their significance showed that a lipid peroxidation process had occurred. The other finding of their study was the decline in serum glutathione peroxidase level, which occurred due to the excessive utilisation of glutathione peroxidase in scavenging free radicals. Hence, it was proved that chloroquine, at the dose of 970 mg/kg, had induced oxidative stress in male albino mice.

Previous research has indicated that the ethanolic extract of mint contained an excessive amount of different antioxidants such as phenolic acids and flavonoids. Hence, in the current study, ethanolic extract of mint was used to see the antioxidant effect of mint.

The comparison of serum malondialdehyde and serum glutathione peroxidase between group C and group B revealed a highly significant decrease in malondialdehyde and a highly significant increase of glutathione peroxidase in group C mice as compared to those in group B mice. Reduced serum levels of malondialdehyde, a secondary metabolite of the lipid peroxidation process, showed a decline in the lipid peroxidation process by the ethanolic extract of mint. On the other hand, the raised serum glutathione peroxidase level showed that the ethanolic extract of mint had antioxidant potential. Hence, these findings indicate that mint (mentha) possessed antioxidant effects. Research work done showed that mentha arvensis possessed an antioxidant role. Their study revealed that the phenolic acids, which were antioxidants in nature, had hydroxyl groups which were capable of scavenging free radicals.[16-18] The antioxidants also could convert Fe^{+3} into Fe^{+2}, hence inhibiting the non-enzymatic lipid peroxidation process.

Similar findings were obtained by Wani et al, (2018).[19] Their results also showed that the ethanolic extract of mentha arvensis possessed the highest concentration of flavonoids and phenolic acids. These antioxidants could lose electrons due to which they were able to stop free radical chain reactions. The phenolic acids inhibited the conversion of Fe^{+3} into Fe^{+2}. Hence, these ended the non-enzymatic lipid peroxidation process.

Polyphenols are antioxidants, which are found in mentha arvensis in abundance. Their antioxidant actions include iron chelation and inhibition of xanthine

oxidase and NADPH oxidase, enzymes which generate reactive oxygen species. These inhibit lipoxygenase and cyclooxygenase and increase the formation of antioxidant enzymes.

CONCLUSION

Chloroquine, given at a dose higher than the therapeutic dose, induce oxidative stress. Mint possesses antioxidant potential against chloroquine induced oxidative stress.

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AUTHOR CONTRIBUTIONS

SK: Data collection, analysis, writing

REFERENCES

1. Zaky HS, Gad AM, Nemr E, Hassan W, Abd El-Raouf OM, Ali AA. Modulatory effects of some natural products on hepatotoxicity induced by a combination of sodium valproate and paracetamol in rats. J Biochem Mol Toxicol 2018 Jul;32(7):e22162. https://doi.org/10.1002/jbt.22162.
2. Iorga A, Dara L, Kaplowitz N. Drug-induced liver injury: cascade of events leading to cell death, apoptosis or necrosis. Int J Mol Sci. 2017 May 9;18(5):1018. https://doi.org/10.3390/ijms18051018.
3. Njoku DB. Drug-induced hepatotoxicity: metabolic, genetic and immunological basis. Int J Mol Sci 2014 Apr 22;15(4):6990-7003.https://doi.org/10.3390/ijms15046990.
4. Singh D, Cho WC, Upadhyay G. Drug-induced liver toxicity and prevention by herbal antioxidants: an overview. : Front Physiol. 2016 Jan 26;6:363. https://doi.org/10.3389/fphys.2015.00363.
5. Gentile F, Arcaro A, Pizzimenti S, Daga M, Cetrangolo GP, Dianzani C, Lepore A, Graf M, Ames PR, Barrera G. DNA damage by lipid peroxidation products: Implications in cancer, inflammation and autoimmunity. AIMS genetics. 2017 Aug;4(02):103-37. doi:10.3934/genet.2017.2.103.
6. Tsikas D. Assessment of lipid peroxidation by measuring malondialdehyde (MDA) and relatives in biological samples: Analytical and biological challenges. Analytical biochemistry. 2017 May 1;524:13-30. https://doi.org/10.1016/j.ab.2016.10.021.
7. Dattani JJ, Rajput DK, Moid N, Highland HN, George LB, Desai KR. Ameliorative effect of curcumin on hepatotoxicity induced by chloroquine phosphate. Environ Toxicol Pharmacol. 2010 Sep 1;30(2):103-9. https://doi.org/10.1016/j.etap.2010.04.001.
8. Ugo Nwanjo H. Aqueous extract of Tridax procumbens leaves: Effect on Lipid peroxidative stress and antioxidant status in chloroquine-induced hepatotoxicity in rats. J Herbs Spices Med Plants. 2008 Dec 15;14(3-4):154-65. https://doi.org/10.1080/10496470802598719.
9. Haida Z, Hakiman M. A comprehensive review on the determination of enzymatic assay and nonenzymatic antioxidant activities. Food Sci Nutr. 2019 May;7(5):1555-63. https://doi.org/10.1002/fsn3.1012.
10. Pari L, Amali DR. Protective role of tetrahydrocurcumin (THC) an active principle of turmeric on chloroquine induced hepatotoxicity in rats. J Pharm Sci. 2005 Apr 30;8(1):115-23.
11. Gheita TA, Kenawy SA. Measurement of malondialdehyde, glutathione, and glutathione peroxidase in SLE patients. Systemic lupus erythematosus: Methods and protocols. 2014:193-9. https://doi.org/10.1007/978-1-4939-0326-9_14.
12. Manfo FP, Nantia EA, Kuete V. Hepatotoxicity and hepatoprotective effects of African medicinal plants.

Toxicological Survey of African Medicinal Plants. 2014 Jan 1:323-55. https://doi.org/10.1016/B978-0-12-800018-2.00011-X.
13. Rajesh K, Swamy V, Shivakumar S, Inamdar JV, Kurnool NA. Hepatoprotective and antioxidant activity of ethanol extract of Mentha arvensis leaves against carbon tetrachloride induced hepatic damage in rats. International Journal of PharmTech Research. 2013;5:426-30.
14. Patil K, Mall A. Hepatoprotective activity of Mentha arvensis Linn. leaves against CCl4 induced liver damage in rats. Asian Pacific Journal of Tropical Disease. 2012 Jan 1;2:S223-6. https://doi.org/10.1016/S2222-1808(12)60156-X.
15. Yahfoufi N, Alsadi N, Jambi M, Matar C. The immunomodulatory and anti inflammatory role of polyphenols. Nutrients. 2018 Nov 2;10(11):1618. https://doi.org/10.3390/nu10111618.
16. Dar MA, Masoodi MH, Mir MA. Phytochemical screening and antioxidant potential of root of Mentha arvensis L. from Kashmir region. IJPSR. 2014;5(4):1572-80.
17. Park YJ, Baskar TB, Yeo SK, Arasu MV, Al-Dhabi NA, Lim SS, Park SU. Composition of volatile compounds and in vitro antimicrobial activity of nine Mentha spp. Springerplus. 2016 Sep 21;5(1):1628. https://doi.org/10.1186/s40064-016-3283-1.
18. Ahmad N, Fazal H, Ahmad I, Abbasi BH. Free radical scavenging (DPPH) potential in nine Mentha species. Toxicol Ind Health . 2012 Feb;28(1):83-9. https://doi.org/10.1177/0748233711407238.
19. Wani S, Basir SF. Analysis of antioxidant activity, total phenolic and total flavonoid contents of Allium sativum, Mentha arvensis and Murraya koenigii. Int. J. Adv. Res. Sci and Eng. 2018;7(4):2632-46.