WORLD JOURNAL OF PHARMACY AND PHARMACEUTICAL SCIENCES
SJIF Impact Factor 7.421
Volume 8, Issue 4, 169-183 Research Article ISSN 2278 – 4357

ASSESSMENT OF ANTIOXIDANT STATUS IN RELATION TO MICROSOMAL EPOXIDE HYDROLASE GENE POLYMORPHISMS AMONG WOOD WORKERS

Rokaya H. Shalaby*[1], Mona M. Taha[2] and Ghada E. Mamoon[1]

[1]Professor of Molecular Biology and Cytogenetics in Zoology Department, Faculty of women for Arts, Science and Education, Ain Shams University, Asmaa Fahmy street, Heliopolis, Cairo, Egypt.

[2]Zoology Department, Faculty of women for Arts, Science and Education, Ain Shams University, Asmaa Fahmy street, Heliopolis, Cairo, Egypt.

[3]Environmental and Occupational Medicine Department, Environmental Research Division, National Research Centre, Dokki, Giza, Egypt.

Article Received on
13 Feb. 2019,

Revised on 07 March 2019,
Accepted on 27 March 2019

DOI: 10.20959/wjpps20194-13477

***Corresponding Author**
Dr. Rokaya H. Shalaby
Professor of Molecular Biology and Cytogenetics in Zoology Department, Faculty of women for Arts, Science and Education, Ain Shams University, Asmaa Fahmy street, Heliopolis, Cairo, Egypt.

ABSTRACT

Occupational exposure to wood dust may be associated with different harmful health effects in workers employed in the wood industry. These harmful effects may be a result of inducing oxidative stress which is causally related to inflammations. We aimed to evaluate the associations between EPHX1polymorphisms and change in antioxidant status (SOD, CAT and GPx) among wood dust exposed workers. Subjects &Methods: EPHX1 genotyping in exon 3 and exon 4 polymorphisms was carried out by PCR-RFLP. Biochemical assessment of altered antioxidant enzyme activities illustrated role of oxidative stress (OS) that may lead to depletion in antioxidant status. SOD, glutathione peroxidase (GPx) and catalase (CAT) (as radical-scavenging enzymes) were analysed in 50 exposed workers who were classified into four subgroups according to ages and duration of exposure (5,10,15 and 20 years of exposure) and 50 control subjects classified according to age into 4 subgroups. Also, the production of MIP-2 upon dust exposure among these workers was assessed. Results: Significant reduction in enzymatic antioxidants SOD and CAT levels and non significant reduction in GPx levels, as well as significant rise in serum MIP-2 levels in different duration of exposure among exposed

workers compared to healthy control ones. Also, significant difference in genotype frequency of EPHX polymorphisms in exon 3 or 4 in different duration of exposure.Conclusion: SOD and CAT levels and not GPx can reflect the antioxidant status in wood workers while genotype frequency of EPHX1 gene polymorphisms at exon 3 and 4 can indicate genetic damage in those workers.

KEYWORDS: Wood dust, Antioxidant status, MIP-2, EPHX Gene polymorphisms.

INTRODUCTION

In furniture manufacture, releasing wood dust was shown as machines are utilized for cutting or shaping wood materials. Wood workers could be exposed to hardwood or softwood dust (during their work activities), dust from natural wood or wood-based composites, pure wood dust or wood dust which contain adhesives, paints as well as other chemicals.[1]

Wood dust represents complex mixture; the species of tree determined its chemical composition which is composed mainly of cellulose, polyoses, and lignin, containing a large and numerous substances with lower relative molecular mass.[2]

Wood dust generated distinct ROS (superoxide anion, and hydrogen peroxide) by selectively inhibiting the enzymatic activity of superoxide dismutase or glutathione peroxidase and catalase enzymes.[3]

MIP-2 is one of C-X-C chemokine that possesses in vitro chemotactic activity in neutrophil, in addition to mitogenic activity for epithelial cells. It is a heparin binding protein, (of approximately 6 kDa), that is secreted by mouse macrophage cell line when stimulated with lipopolysaccharide.[4] Various types of cell participate in MIP-2 production such macrophages, monocytes, epithelial cells, and hepatocytes, Detection of MIP-2 were found to be as a part of inflammatory stimuli response.[5]

Microsomal EPHX1 function was to activate or detoxify carcinogenic substance as polycyclic aromatic hydrocarbons and aromatic amines[6], it plays an essential role in the metabolism of epoxide intermediate produced from cigarettes which is highly reactive[7] and other epoxides that are converted to more stable diols.[8]

Chromosome 1 (1q42.12) carries human EPHX1 gene which contains 9 exons.[8] Two common polymorphisms were reported in the coding region in EPHX1 gene that resulted in

variation in enzyme activity. One conferring to decreased activity in case of one mutation while increased activity was found in another. Transition of T **(**thymine) to C (cytosine) in exon 3 lead to alteration tyrosine amino acid (Tyr) 113 into histidine (His), that change reduces enzyme activity nearby 50% (slow allele). Another transition of A (adenine) into G (guanine) in exon 4 also results in substitution of histidine (His) amino acid 139 into arginine amino acid (Arg), that substitution increased enzyme activity by 25 (fast allele).[9]

Extremely slow/slow activity genotype was shown with reduction in EPHX1 enzyme activity of more than 50%. That may result imbalance in oxidant/antioxidant within the body.[10]

PATIENTS AND METHODS

This study was performed on 50 workers exposed to wood dust (workers) from a factory for furniture manufacture in Cairo governorate. Also, 50 healthy individuals not occupationally exposed to wood dust were recruited as controls. The workers and controls subjects were matched for age, socioeconomic status, and dietary habits. All participants filled a questionnaire including personal, medical and detailed environmental and occupational histories. Both groups were divided into four subgroups according to ages and workers subgroups were classified according to duration of exposure to wood dust (5, 10, 15 and 20 years). First subgroup includes twelve of workers exposed to wood dust for 5 years, their age range were (20 – 25 years), second subgroup contains thirteen of workers exposed to wood dust for 10 years and their age were (25 -30 years), third subgroup includes twelve of workers exposed to wood dust for 15 years, their age range were (35- 45 years) and fourth subgroup includes twelve of workers exposed to wood dust for 20 years, their age range were (40 – 55 years). Blood samples were collected from each subjects and divided into an EDTA tube for assessment of EPHX gene polymorphisms and also for separating plasma for catalase determination and washing packed RBCs for determination of SOD and GPx enzyme activity, other dry tube for separating serum for MIP-2 estimation.

PCR restriction fragment length polymorphism analysis of mEPHX gene

Genomic DNA was isolated from total blood cells using Qiagen Kit commercial, Germany. Genomic DNA (20 ng) was amplified by polymerase chain reaction (PCR) according to (Hasset et al., 1994) [11]. The PCR was performed in a volume of 50 µL reaction mixture containing PCR buffer (1·5 mmol/L magnesium chloride, 100 ng primers, 4% dimethyl sulphoxide, 200 mmol/L of each dNTP, and 1 unit of *Taq* polymerase [Promega]). The PCR

conditions consisted of an initial single cycle of 10 minutes at 94°C, followed by 40 cycles of 94°C for 30 seconds, 56°C for 20 seconds and 72°C for 30 seconds.

Two separate PCR assays are used to detect the two mutations. The assay for the exon-3 variant uses the primer pair (5′-GATCGATAAGTTCCGTTTCACC-3′), and (5′-ATCCTT-AGTCTTGAAGTGAGGAT-3′). For exon-4 variant, primer pairs was (5′-ACATCCACTTCATCCACGT-3′) and (5′-ATGCCTCTGAGAAGCCAT-3). The PCR product of exon 3 was digested with 10 U EcoR V restriction enzyme overnight at 37°C, while PCR product for exon 4 was digested with Rsa I restriction enzyme. Then, PCR products for either exon were separated by electrophoresis through a 3% agarose gel, stained with ethidium bromide and transilluminated with ultraviolet light. For exon 3, the sizes of the restriction fragments of PCR product were 162 bps for the homozygote mutation genotype (Hist-Hist), 140 and 20 bps for the (Tyr-Tyr) wild type homozygote and 162, 140 and 20 bps for heterozygotes (Tyr-Hist).

For exon 4, the sizes of the restriction fragments of PCR product were 210 bps for the wild type homozygote (Hist-Hist), 164 and 64 bps for the (Arg-Arg) homozygote mutation genotype and 210, 164 and 46 bps for heterozygotes (Hist-Arg).

MIP-2 ELISA

Determination of serum MIP-2 concentration was done by enzyme-linked immunosorbent assay (ELISA) using commercially available kits (R&D Systems). It was performed according to the manufacturer's protocol.

Enzymatic biomarkers

1- Catalase activity: Plasma catalase activity was estimated in plasma using end point colorimerty according to Aebi 1984.[12]
2- SOD activity: Superoxide dismutase (SOD) activity was determined colorimetrically according to Nishikimi et al., 1972.[13]
3- GPx activity: Glutathione peroxidase (GPx) was determined using colorimetric method according to Plaglia and Valentine 1967 [14].

Statistical analysis

The collected data and the laboratory results were computerized. Statistical analysis was done through Statistical Package for Social Science (SPSS) software computer program version 20.

The quantitative results were expressed as means ± standard deviation (SD) and qualitative results as number (No.) and percent (%). Independent t-test, Pearson's $\chi 2$, and Analysis of variant (ANOVA) with the post-hoc test least significant differences (LSD) were used in the analysis of the results. Level of significance was adjusted at $p \leq 0.05$.

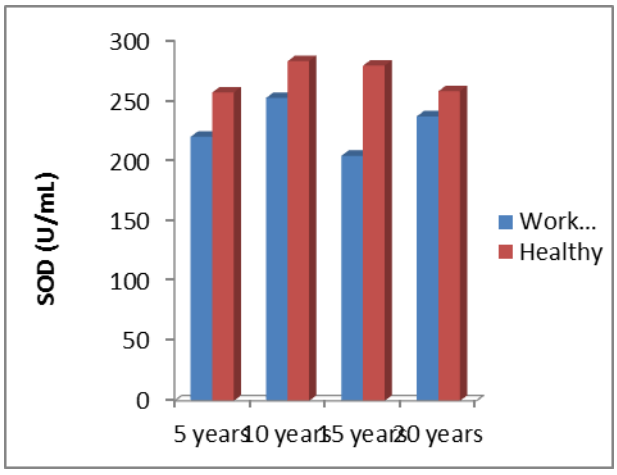
RESULTS

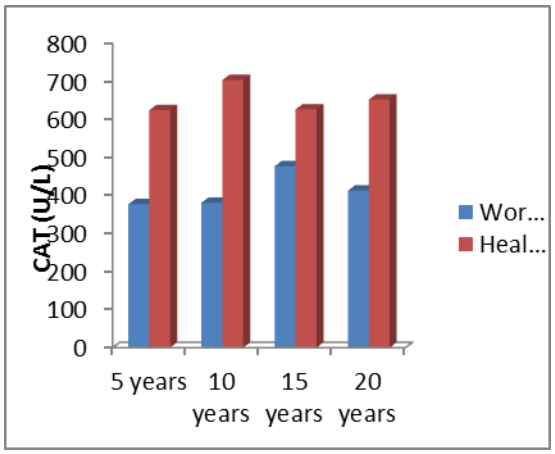
Table (1): Levels of (SOD, CAT, and GPx) enzymes activities among exposed workers and healthy groups for different duration exposure.

	5 years		10 years		15 years		20 years		P value
	Workers	Healthy	Workers	Healthy	Workers	Healthy	Workers	Healthy	
SOD (U/mL)	219.91± 64.546	256.75 ± 62.24	252.23 ± 50.89	282.85 ± 44.84	204.07 ± 56.77	279.15 ± 54.38	236.91± 39.29	257.83± 42.85	<0 .003*
CAT (U/L)	376.92 ± 133.64	623.67 ±123.43	380.73 ± 124.23	702.93 ± 70.32	204.07 ± 56.77	626.08 ± 91.42	412.50± 121.07	651.42 ±102.96	<0 .003*
GPx (mU/mL)	231.33 ± 62.66	240.42 ± 34.19	222.07 ± 48.46	267.38 ± 62.31	225.86 ± 80.30	277.15 ± 92.52	258.83 ±62.50	264.83 ± 39.86	0.211

SOD: Superoxide Dismutase, CAT: Catalase, GPx: Glutathione peroxidase **: highly significant

In present study the levels of (SOD, CAT) antioxidants enzymes demonstrated significant reduction ($P < 0.003$ and 0.003 respectively) where mean levels of (SOD, CAT) were (219.91± 64.546 U/mL, 376.92 ± 133.64 U/L), (252.23 ± 50.89 U/mL, 380.73 ± 124.23 U/L), (204.07 ± 56.77 U/mL, 204.07 ± 56.77 U/L), (236.91± 39.29 U/mL, 412.50±121.07 U/L respectively) among workers exposed to wood dust for different duration exposure(5,10,15, and 20years respectively) when comparing with healthy groups as results represented in table (1) & fig (1).





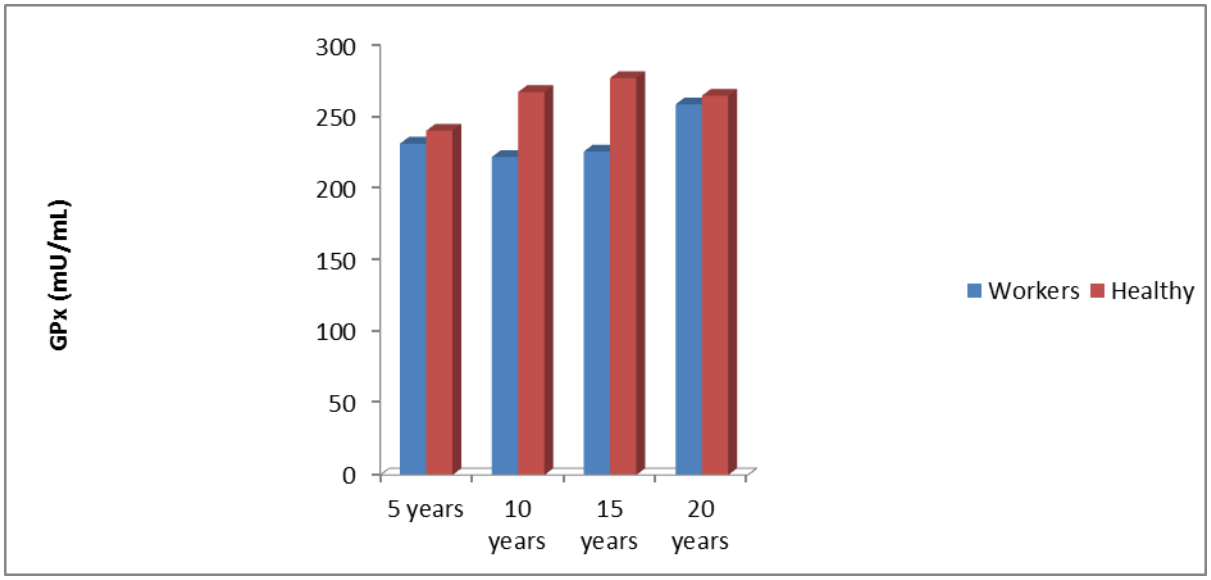


Fig (1): Levels of SOD, CAT and GPx in workers and healthy groups in different duration exposure.

Table (2): Levels of macrophage inflammatory protein-2 among exposed workers and healthy control groups for different duration exposure.

	5 years		10 years		15 years		20 years		P value
	Worker	Healthy	Worker	Healthy	Workers	Healthy	Workers	Healthy	
MIP-2 (ng/L)	183.82 ±11.05	174.58 ±19.17	223.92 ±24.86	190.31 ± 16.88	327.76± 55.43	215.15 ± 24.17	638.25± 219.45	451.33 ± 102.51	P< 0.001

MIP-2: macrophages inflammatory protein

Levels of macrophage inflammatory protein recorded significant elevation (P<0.001), where mean levels of MIP-2 were (183.82 ±11.05ng/L, 223.92 ±24.86ng/L, 327.76± 55.43ng/L, and 638.25± 219.45ng/L, respectively) among workers exposed to wood dust for different duration exposure (5, 10, 15, and 20 years, respectively) when compared with healthy control groups, where mean levels were(174.58 ±19.17ng/L, 190.31 ± 16.88ng/L, 215.15 ± 24.17ng/L, 638.25± 219.45ng/L respectively), results were represented in table (2)&fig(2).

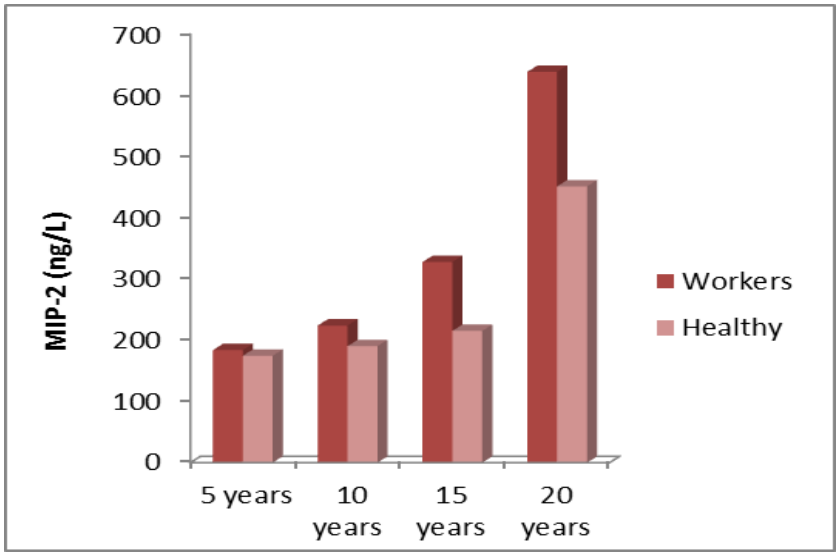


Fig (2): Levels of macrophage inflammatory protein-2 in different duration among exposed workers and healthy group.

Table (3): Genotype frequency of EPXH1 polymorphism in Exon 3 (Try113His) among exposed workers and healthy control groups for different duration exposure.

	5 years		10 years		15 years		20 years		P value
	Workers	Healthy	Workers	Healthy	Workers	Healthy	Workers	Healthy	
Exon 3: Tyr-Tyr	3 (25.00%)	7 (58.34%)	4 (30.77%)	7 (53.84%)	3 (23.08%)	11 (84.61%)	5 (41.67%)	6 (50.00%)	
Tyr-Hist	6 (50.00%)	4 (33.33%)	4 (30.77%)	5 (38.46%)	5 (38.46%)	1 (7.69%)	3 (25.00%)	6 (50.00%)	0.047*
Hist-Hist	3 (25.00%)	1 (8.33%)	5 (38.46%)	1 (7.70%)	5 (38.46%)	1 (7.69%)	4 (33.33%)	0 (0.00%)	

In the present study genotype frequency of EPXH1 polymorphism exon 3 illustrated significant difference (P< 0.047) in exposed workers different duration exposure (5,10,15, 20, respectively) comparison with healthy control groups, where Tyr-Tyr genotype was (25.0%,30.77%, 23.08%, 41.67%, respectively) in exposed workers and (58.34%, 53.84% ,84.61%, 50.00%, respectively),while Tyr-Hist was (50.00%, 30.77%, 38.46%, 25.00%, respectively) in exposed workers and (33.33%, 38.46%, 7.69% ,50.00%, respectively) in addition, homozygose mutant genotype Hist-Hist was (25.0%, 38.46%, 38.46%, 33.33% , respectively) in exposed workers and (8.33%, 7.70%, 7.69%, 0.00% respectively),table (3)& fig (3).

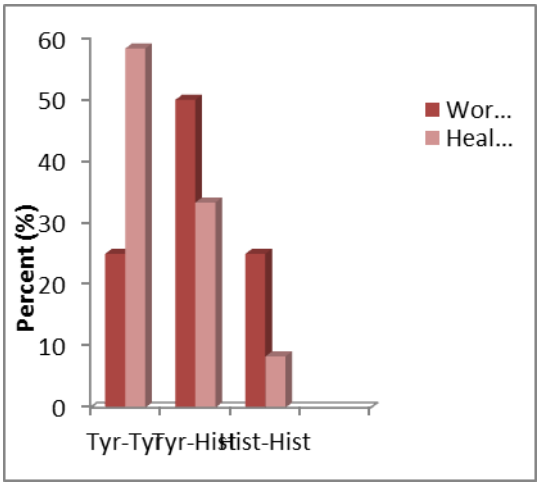


Fig (3a): Microsomal epoxide hydrolase genotypes (exon 3) at 5 years duration among exposed workers and healthy group.

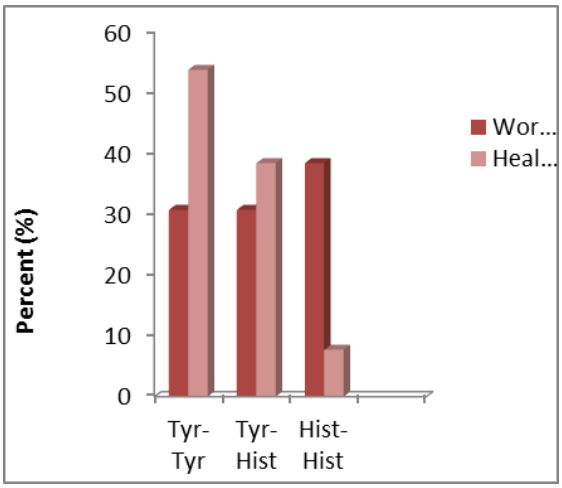


Fig (3b): Microsomal epoxide hydrolase genotypes (exon 3) at 10 years duration among exposed workers and control group

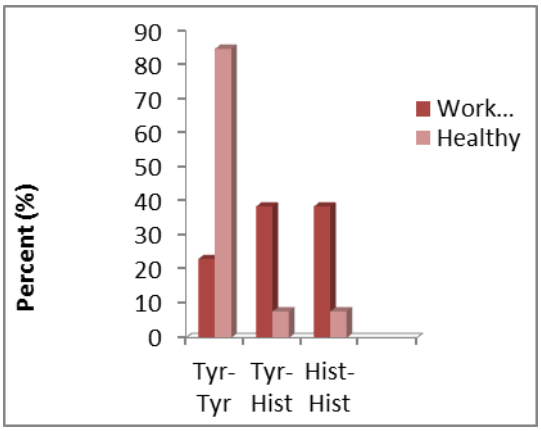


Fig (3c): Microsomal epoxide hydrolase genotypes (exon 3) at 15 years duration among exposed workers and healthy group

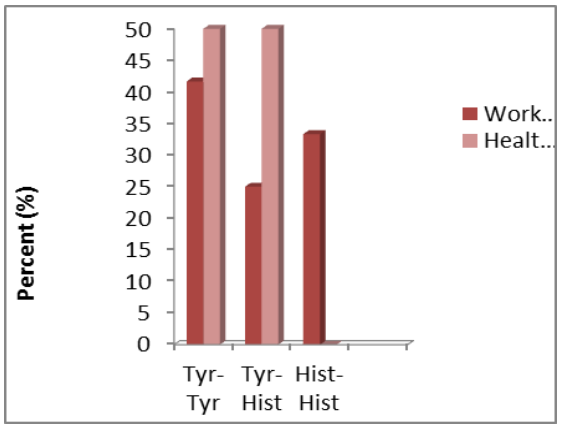


Fig (3d): Microsomal epoxide hydrolase genotypes (exon 3) at 20 years duration among exposed workers and healthy group.

Table (4): Genotype frequency of EPXH1 polymorphism in Exon 4 (Try113His) among exposed workers and healthy control groups for different duration exposure.

	5 years		10 years		15 years		20 years		P value
	Workers	Healthy	Workers	Healthy	Workers	Healthy	Workers	Healthy	
Exon 4: Hist-Hist	3 (25.00%)	8 (66.67%)	4 (30.77%)	7 (53.84%)	7 (53.86%)	11 (84.62%)	1 (8.33%)	6 (50.00%)	0.05*
Hist-Arg	6 (50.00%)	3 (25.00%)	5 (38.46%)	4 (30.76%)	2 (15.38%)	2 (15.38%)	6 (50.00%)	4 (33.33%)	
Arg-Arg	3 (25.00%)	1 (8.33%)	4 (30.77%)	2 (15.40%)	4 (30.76%)	0 (0.00%)	5 (41.67%)	2 (16.67%)	

Genotype frequency of EPHX1 polymorphism reported significant difference (P=0.05) at exon4 among exposed workers for different exposure duration (5, 10, 15, 20 years) in comparison with healthy control groups, where Hist-Hist genotype was (25.00%, 30.77%, 53.86%, 8.33%, respectively) in exposed workers and (66.67%, 53.84%, 84.62%, 50.00%, respectively) in healthy control groups, while Hist-Arg genotype was (50.00 %, 38.46%, 15.38%, 50.00%, respectively) and (25.00%, 30.76%, 15.38%, 33.33%, respectively) in healthy control groups ,in addition to Arg-Arg genotype was (25.00%, 30.77%, 30.76% and 41.67%respectively) and (8.33%, 15.40%, 0.00%, 16.67%respectively)in healthy control groups, table (4)& fig (4).

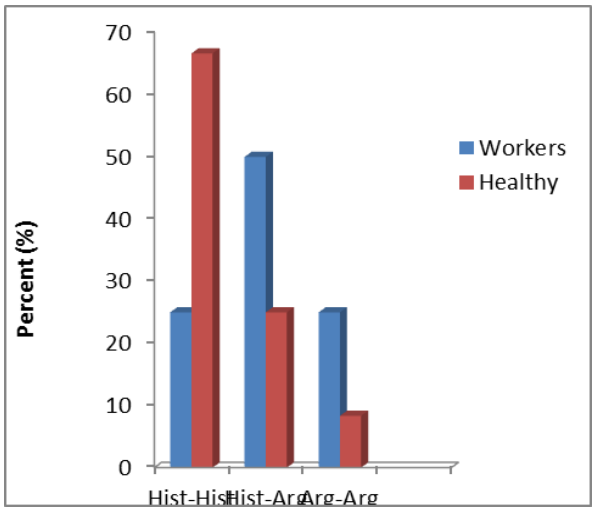


Fig (4a): Microsomal epoxide hydrolase genotypes (exon 4) at 5 yearsduration among exposed workers and healthy group.

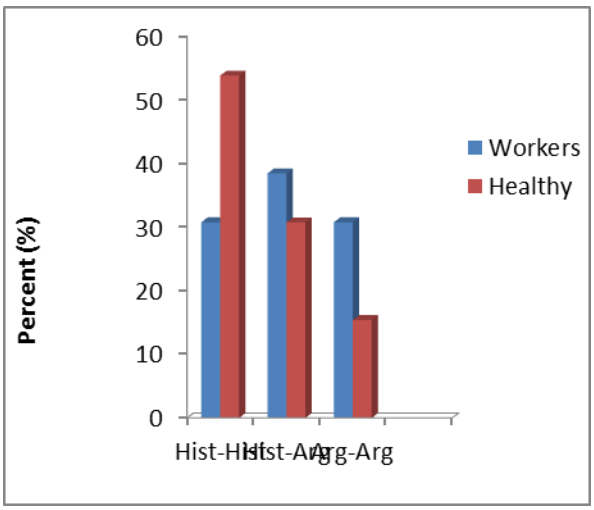


Fig (4b): Microsomal epoxide hydrolase genotypes (exon 4) at 10 years duration among exposed workers and healthy group.

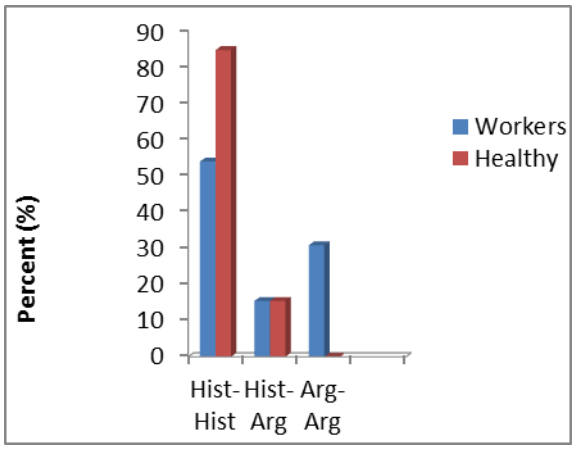


Fig (4c): Microsomal epoxide hydrolase genotypes (exon 4) at 15 years duration among exposed workers and healthy group.

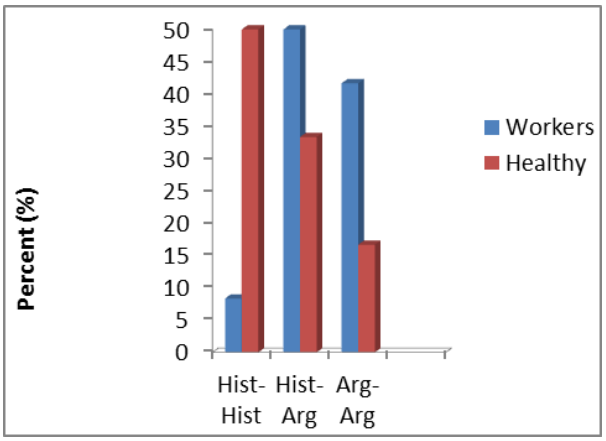
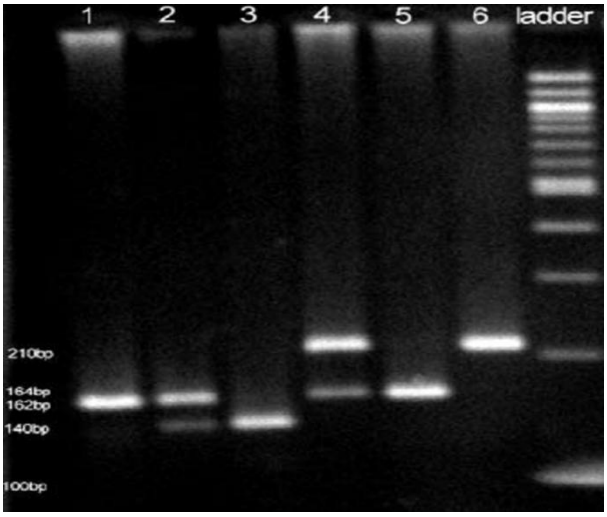


Fig (4d): Microsomal epoxide hydrolase genotypes (exon 4) at 20 years duration among exposed workers and healthy group.



1: **His/His**.
2: **Tyr/His**.
3: **Tyr/Tyr**.
4: **His/Arg**.
5: **Arg/Arg**.
6: **His/His**.
7: marker with**100 bp (ladder)**.

Fig (5): gel electrophoresis of EPHX1 gene (exon 3 and 4) by PCR-RFLP. Exon 3 in lanes 1-3, Exon 4 in lanes 4-6.

DISCUSSION

Wood workers are exposed continuously to wood dusts particles that cause a numerous harmful effects on their health [15]. Recent study mentioned that free radicals interaction with cell and macrophage leads to several disorders through occupational wood dust exposure.[16]

Present study illustrated a significant reduction in catalase activity (P<0.003) among exposed workers (Table 1), that was in agreement with Staffolani[17] who concluded that occupational exposure to wood dust promotes excessive production of hydrogen peroxide. Previous study reported that hydrogen peroxide is produced through digestion of wood dust particles by macrophages and neutrophils which undergoes respiratory burst via reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase system.[18]

Also, Aboulezz[19] concluded that reduction in catalase efficiency may be due to enzyme exhaustion trying to remove generated hydrogen peroxide during occupational exposure. Also, it may be as a result of enzyme inactivation through excessive production of ROS in mitochondria and microsomes.

SOD enzyme transforms superoxide radicals to stable oxygen molecule (O_2) and H_2O_2.[20] Our present study mentioned that the levels of SOD enzyme activity recorded a significant reduction (P<0.003) respectively in exposed workers.That was in accordance with Gaballah[21] who found significant reduction in SOD enzyme among Egyptian wood workers. Also, recent study revealed that prolonged exposure wood dust promotes excessive releasing of •O (superoxide radicals) and H_2O_2 (hydrogen peroxide radicals) which induce chronic

oxidative stress[3], these oxidative stress represents the most toxic mechanism causing many physiological and cellular changes. This mechanism is mediated through generation of intracellular reactive oxygen species which in turn impair antioxidants enzymes activities like catalase and SOD.[22]

It was observed that most of the exposed personnel did not use facemasks or gloves. The positive genotoxicity in the present study may be due to lack of protective measures.

Also, it was shown that levels of serum MIP-2 showed increased levels with increasing duration of exposure (significant elevation ($P<0.001$)) among workers compared to healthy control group. This rise may be a result of chronic oxidative stress, or from wood dust chemical composition, or may be due to interaction between wood dust particles and macrophages. Also, these results indicated that these particles promote immune response through releasing humeral mediators (as MIP-2) which mediates the acute inflammatory response following prolonged exposure to wood dust particles.[23]

Microsomal epoxide hydrolase (mEH) is an important metabolic biotransformation enzyme that hydrolyzes epoxides, yielding trans-dihydrodiols. Such hydrolysis usually has a detoxifying effect.[24]

Current study recorded that genotype frequencies of EPHX1 in exon 3 showed significant difference ($P<0.047$) and in exon 4 ($P< 0.05$) among exposed workers and controls. According to Li[7], Tyr-Hist represents slow allele in exon 3 and Hist-Arg in exon 4 fast allele. Variation in these alleles was shown along different exposure duration as shown in table 3 and 4. Bruschweiler[25] illustrated that the wood working environment often includes simultaneous exposures to other substances (PAHs, formaldehyde, or wood preservatives. PAHs can induce mutations in human[26], resulting from oxidative DNA damage which can be induced by wood dust[21], all that may result in gene-environment interactions which can play an important role in replacement of Tyr113His in exon 3[24], these genetic polymorphisms has a significant impact on synthesis, function and activity of mEH enzyme leading to variations in individual ability in xenobiotic biotransformation and antioxidant protection[27], which in turn also may affect oxidative defenses against a number of environmental substances.[28]

So, it can be inferred that the "fast" genotype because of its highest activity during wood dust exposure can generates some intermediate products, which are efficient inducers of antioxidant depletion in exposed workers. The "intermediate" genotype mediates the above-mentioned metabolic situations and consequently, minor biological effects are expected.[29] This reflects a direct functional effect of the EPHX1 gene through its regulation of EPHX1 levels in those workers.

CONCLUSION

SOD and CAT levels and not GPx can reflect the antioxidant status in wood workers while genotype frequency of EPHX1 gene polymorphisms at exon 3 and 4 can indicate genetic damage in those workers

ACKNOWLEDGMENT

The authors express their sincere thanks to Prof Dr Amal Saad, Prof. of Environmental & Preventive Medicine, National Research Centre, for her encouragement during the study and her help in analyzing the data.

REFERENCES

1. Bislimovska D, Petrovska S, Minov J. (Respiratory Symptoms and Lung Function in Never-Smoking Male Workers Exposed to Hardwood Dust).Open access Macedonian journal of medical sciences, 2015; 500–505.
2. Farahat SA, Ibrahim YH, Abdel-Latif M N. (Nontoxicity and oxidative stress due to exposure to wood dust among carpenters). Egyptian Journal of Occupational Medicine, 2010; 83-95.
3. Antognelli C, Gambelunghe A, Talesa VN, Muzi G.(Reactive oxygen species induce apoptosis in bronchial epithelial BEAS-2B cells by inhibiting the antiglycationglyoxalase I defence: involvement of superoxide anion, hydrogen peroxide and NF-κB). Apoptosis: an international journal on programmed cell death, 2014; 102-116.
4. Driscoll KE. (TNFalpha and MIP-2: role in particle-induced inflammation and regulation by oxidative stress), 2000; Toxicol Lett., 177-183.
5. Qin CC, Liu YN, Hu Y, Yang Y, Chen Z.(Macrophage inflammatory protein-2 as mediator of inflammation in acute liver injury). World journal of gastroenterology, 2017; 3043-3052.

6. Xu X, Hua H, Fan B, Sun Q, Guo X, Zhang J.(EPHX1 rs2234922 polymorphism and lung cancer susceptibility in Asian populations: a meta-analysis). Journal of thoracic disease, 2015; 1125–1129.
7. Li H, Fu WP, Hong ZH. (Microsomal epoxide hydrolase gene polymorphisms and risk of chronic obstructive pulmonary disease: A comprehensive meta-analysis). Oncology letters, 2012; 1022-1030.
8. Václavíková R, Hughes DJ, Souček P.(Microsomal epoxide hydrolase 1 (EPHX1): Gene, structure, function, and role in human disease). Gene, 2015; 1-8.
9. Liu HQ, Zhang CP, Zhang CZ, Liu XC, Liu ZJ.(Influence of two common polymorphisms in the EPHX1 gene on warfarin maintenance dosage: a meta-analysis). BioMed research international, 2015; 1-12.
10. Zhang YJ, Gan RY, Li S, Zhou Y, Li AN, Xu DP, Li HB.(Antioxidant Phytochemicals for the Prevention and Treatment of Chronic Diseases), 2015; 21138-21156.
11. Hassett C, Robinson KB, Beck NB, Omiecinski CJ. (The human microsomal epoxide hydrolase gene (EPHX1): complete nucleotide sequence and structural characterization). Genomics, 1994; 433–442.
12. Aebi H. (Catalase in vitro). Method Enzymol, 1984; 105: 121–126.
13. Nishikimi M, Appaji N, Yagi K. (The occurrence of superoxide anion in the reaction of reduced phenazinemethosulfate and molecular oxygen). BiochemBiophys Res Commun, 1972; 849-854.
14. Plaglia DE, Valentine WN. (Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase). J Lab Clin Med., 1967; 158-169.
15. Szewczyńska M, Pośniak M. (Assessment of occupational exposure to wood dust in the Polish furniture industry). MEDYCYNA PRACY, 2017; 45-60.
16. Radi R. (Oxygen radicals, nitric oxide, and peroxynitrite: Redox pathways in molecular medicine). Proceedings of the National Academy of Science of United States of America, 2018; 5839-5848.
17. Staffolani S, Manzella N, Strafella E, Nocchi L, Bracci M, Ciarapica V, Amati M, Rubini C, Re M, Pugnaloni A, Pasquini E, Tarchini P, Valentino M, Tomasetti M, Santarelli L.(Wood dust exposure induces cell transformation through EGFR-mediated OGG1 inhibition).Mutagenesis, 2015; 487-97.
18. Suryakar A.N, Katkam R.V, Dhadke V.N, Bhogade R.B. (A study of oxidative stress in cotton industry workers from Solapur city). Biomedical Research, 2010; 260-264.

19. Aboul Ezz HS, Khadrawy YA, Mourad IM.(The effect of bisphenol A on some oxidative stress parameters and acetylcholinesterase activity in the heart of male albino rats).Cytotechnology, 2015; 145–155.
20. Niu R, Han H, Zhang Y, Wang J, Zhang J, Yin W, Yin X, Sun Z, Wang J.(Changes in Liver Antioxidant Status of Offspring Mice Induced by Maternal Fluoride Exposure during Gestation and Lactation). Biological trace element research, 2016; 172-178.
21. Gaballah IF, Helal SF, Mourad BH.(Early detection of lung cancer potential among Egyptian wood workers).International Journal of Occupational and Environmental Health, 2018; 120-127.
22. Suliman Y AO, Ali D, Alarifi S, Harrath AH, Mansour L, Alwasel SH.(Evaluation of cytotoxic, oxidative stress, proinflammatory and genotoxic effect of silver nanoparticles in human lung epithelial cells). Environmental toxicology, 2015; 149-160.
23. Aztatzi-AguilarOG, Valdés-ArzateA, Debray-GarcíaY, Calderón-ArandaES, Uribe-Ramirez M, Acosta-SaavedraL, GonsebattME, Maciel-RuizJA, PetrosyanP, Mugica-AlvarezV, Gutiérrez-RuizMC, Gómez-QuirozLE, Osornio-VargasA, FroinesJ, KleinmanMT, De Vizcaya-RuizA.(Exposure to ambient particulate matter induces oxidative stress in lung and aorta in a size- and time-dependent manner in rats). SAGE journal, 2018; 1-15.
24. Tan X, Wang YY, Chen XY, Xian L, Guo JJ, Liang GB, Chen MW.(Quantitative assessment of the effects of the EPHX1 Tyr113His polymorphism on lung and breast cancer).Genetics and molecular research, 2014; 7437-7446**.**
25. Bruschweiler ED, Danuser B, Huynh CK, Wild P, Schupfer P, Vernez D, Boiteux P, Hopf NB.(Generation of polycyclic aromatic hydrocarbons (PAHs) during woodworking operations). Frontiers in oncology, 2012; 1-9.
26. Sivonova MK, Dobrota D, Matakova T, Dusenka R, Grobarcikova S, Habala V, Salagovic J, Tajtakova M, Pidanicova A, Valansky L, Lachvacs L, Kliment J Jr, Nagy V, Kliment J. (Microsomal epoxide hydrolase polymorphisms, cigarette smoking and prostate cancer risk in the Slovak population). Neoplasma, 2012; 79-84.
27. Dubovskaya L.V, Rybina T.M, Bakakina Y.S, Kardash O.F, Denisevich N.P, Volotovski I.D. (Genetic Predisposition to Chronic Dust Bronchitisamong Potash Miners). Journal of Medical and Biological Science Research, 2015; 55-61.
28. Yang X, Wang Y, Wang G.(Quantitative assessment of the influence of EPHX1 gene polymorphisms and cancer risk: a meta-analysis with 94,213 subjects). Journal of experimental & clinical cancer research, 2014; 1-17.

29. Bukvic N, Lovreglio P, Fanelli M, Susca FC, Ballini A, Lastella P, Foà V, Fustinoni S, Soleo L, Guanti G.(Influence of some detoxification enzyme polymorphisms on cytogenetic biomarkers between individuals exposed to very low doses of 1, 3-butadiene).Journal of occupational and environmental medicine, 2009; 811-821.