REVIEW ARTICLE

The Glutathione S-Transferasee (GSTT and GSTM) Genotyping and Alcohol Level in Drunks

Mona N. Al-Terehi[1*], Usama S. Altimari[2], Abed J. Kadhim[2], Aamal M. Kadhum[3]

[1]*College of Science, University of Babylon, Hillah, Iraq*
[2]*Al-Nisour University College, Baghdad, Iraq*
[3]*Medical Laboratories Techniques Department, Al-Mustaqbal University College, Iraq*

Received: 17th August, 2021; Revised: 07th September, 2021; Accepted: 19th November, 2021; Available Online: 25th December, 2021

ABSTRACT

Alcoholism is most the important problem is spreading among the young in the Iraqi population, studies found the health and social harmful effects of alcohol thus the present study deal with The Glutathioe S-Transferase in alcohol use disorder, the GSTT and GSTM genotyping was used in the current study, the current finding showed GSTT+GSTT was present in low percent in drunks than the control group in non-significant differences (p = 0.156). the GSTT found in high percent's in drunks and control with null genotyping observed in non-significant differences (p = 0.801), the GSTM found in low percent in the drunks than control and null genotyping was higher in drunks than control in significant differences (p = 0.009). and finally the GSTT null GSTM and GSTM null GSTT were significant differences (p = 0.021), The levels of alcohol was detected according to GST genotyping, the high level of alcohol observed in GSTT+GSTM null genotyping (96.25 ± 22.58) while low percent observed (62.00 ± 2.00) in GSTM null GSTT, all differences were non-significant, it can be concluded that the GSTM null genotyping was strong association with alcohol use disorder, and the null genotyping of both GSTs genes have higher levels of alcohol than other genotyping.

Keywords: Glutathione S-Transferase, GSTT, GSTM, Genotyping, Alcohol Level, Drinking Habbit

International Journal of Pharmaceutical Quality Assurance (2021); DOI: 10.25258/ijpqa.12.4.19

How to cite this article: Al-Terehi MN, Altimari US, Kadhim AJ, Kadhum AM. The Glutathione S-Transferasee (GSTT and GSTM) Genotyping and Alcohol Level in Drunks. International Journal of Pharmaceutical Quality Assurance. 2021;12(4):331-333.

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

The glutathione S-Transferases (GSTs) are family of soluble enzymes included diverse types of detoxification member's work to reduce conjugated of glutathione and reduction reactions. The substrates of the GSTs are different toxic molecules like carcinogens and electrophiles, after conjugation of these molecules they are more easily to excreted into urine or bile.[1]

The GST of human included classes Alpha, Theta, Mu and Omega with multiple members of each class, its encoded by paralogous genes clusters on separated chromosomes, Alpha on chromosome 6p12,[3,4] Mu on chromosome 1p13,[4,5] Theta on chromosome 22q11.2,[6,7] and Omega on chromosome 10q24.3,[8] while the reverse transcribed pseudo-gene of the Omega class, on chromosome 3,[8] the GST common polymorphisms in the populations of the genes is a deletion in GST M1-1 and GST T1-1 in human.[9] Investigations suggested that these deletions may be happened by homologous recombination events, at least one of the neighboring GST genes didn't intact In both cases, the homozygous for the null allele exhibit a clear phenotype with respect to glutathione conjugation of specific substrates, the deletion of GSTM1 Homozygous removes GST activity with respect to conjugation of the characteristic GST M1-1 substrate trans-stilbene oxide (TSO), as measured in lymphocyte homogenates.[10-12] The GSTs genotyping may have different susceptibilities to environmental exposures, the deletion of the GSTM1 gene can be increased cancer risk by environmental exposure while the presence of the intact GSTM1 gene would be protective for carcinogen-derived DNA adduct formation and cytogenetic damage.[13]

METHODOLOGY

About 40 drunks have age ranged (21–59 years) and BMI ranged (22–30 kg/m^2) and 29 non-drunk individuals have age ranged (20–34 years) and BMI ranged (19–35 kg/m^2), blood samples were collected from each contributor with written consent, blood stored at –20°C till DNA extraction, serum used to detect alcohol level for drunks.

DNA Extraction and Genotyping Detection

DNA was extracted using the favorgene extraction kit for frozen blood, amplification of GSTM and GSTT conducted

**Author for Correspondence: monanajah1981@gmail.com*

using the oligos mentioned by Moasser *et al*., (2014),[14] at annealing temperature 60ºC for 30 seconds in separating tubes, PCR products were visualized using agaros gel electrophoresis, 1% Agaros, 75 V, 0.5 X TBE buffer, for 40 minutes.

Data Analysis

The genotyping detection GSTT+GSTM, GSTT, GSTM, and null genotyping, frequency of genotyping were dependent, significant analyzed via Odd ratio and CI 95%, ANOVA one way also used, the $p < 0.05$ used for significant detection.

RESULTS AND DISCUSSION

The current findings were clarified to assessment the genotyping of GST in drunks and the level of alcohol levels, the genotyping was observed in the present study included GSTT+GSTM, GSTT, GSTM, GSTT null GSTM and GSTM null GSTT, the GSTT has 312bp and the GSTM has 215 bp, the null genotype mean that the sites didn't amplify and this deal with Al-Musawi *et al*., (2020)[15] (Figure 1).

The genotyping distribution in drunks and control group shows that the GSTT+GSTT was present in lower percent (57.5%) of drunks than the control group (75.9%), 10% of drunks were null genotyping of GSTT+GSTT and it has not observed in the control group in non-significant differences (p 0.156). The GSTT found in high percent's in drunks and control (85%, 82.74%) respectively, and the null genotyping observed in (15%) of drunks and (17.24%) of control in non-significant differences (p 0.801), the GSTM found in low percent in the drunks (62.5%) than control (93.1%) and null genotyping was high in drunks (37.5%) than control (6.89%) in significant differences (p 0.009). And finally, the GSTT null GSTM and GSTM null GSTT were significant differences (p 0.021) in study groups which were (27%) in drunks and (6.89%) in control for a GSTT null GSTM while (5%) and (17.25%) of GSTM null GSTT for drunks and control group respectively (Table 1).

The level of alcohol was detected according to GST genotyping, the elevated level of alcohol observed in GSTT+GSTM null genotyping (96.25 ± 22.58) while low percent observed (62.00 ± 2.00) in GSTM null GSTT, all differences were non-significant (Table 2).

Alcohol use disorder or (Alcoholism) is a complex disorder trigger via environmental and genetic factors. The genetic factors of alcoholism were resulted from the action of multiple or interacting genes. The detection of these genes is an interest in scientific field and has used some strategies, the current study deal with GST genotyping because of the great important in oxidative stress detoxification, Alcohol has been found to be induced free radicals formation and lipid peroxidation which can lead to DNA damage.[16] The present results show a strong association between GSTT and GSTT null genotyping with alcohol abuse in drunks, also the high level of alcohol was found in GSTT+GSTM null genotyping,

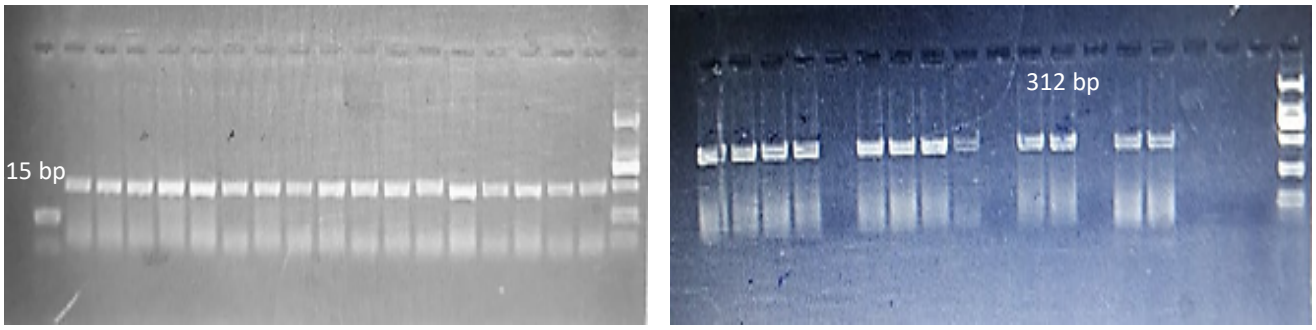

Figure 1: Agaros gel electrophoresis of GSTT and GSTM genotyping in drunks, 215 bp of GSTM and 312 bp of GSTT, 75 V, 40 min, 1% agaros with ethidum bromide staining

Table 1: The GST genotyping (GSTT and GSTM) in study groups

Genotyping	*Drunks*	*Control*	*Odd ratio (CI95%)*	*Sig*
GSTT+GSTT				
Present	23(57.5%)	22(75.9%)	8.6170 (0.4384 to 169.385)	0.156
Null	4(10%)	0		
GSTT				
Present	34(85%)	24(82.75%)	0.8471 (0.2316 to 3.0981)	0.8019
null	6(15%)	5(17.24%)		
GSTM				
Present	25(62.5%)	27(93.10%)	8.1000 (1.6809 to 39.0325)	0.0091
Null	15(37.5%)	2(6.89%)		
GSTT null GSTM	11(27%)	2(6.89%)	13.7500 (1.4831 to 127.4794)	0.0211
GSTM null GSTT	2(5%)	5(17.25%)		

Table 2: The alcohol level in drunks according to GST genotyping

Genotyping	*Alcohol level mg/cm^3*	*Sig*
GSTT+GSTM	75.26 ± 9.89	0.419
Null	96.25 ± 22.58	
GSTT	72.41 ± 7.160	0.502
Null	84.83 ± 16.01	
GSTM	74.20 ± 9.11	0.988
Null	74.40 ± 8.76	
GSTT null GSTM	66.45 ± 8.21	0.828
GSTM null GSTT	62.00 ± 2.00	

Independent t test

the GSTs are contributed to the metabolism of different types of toxic molecules like potential carcinogenic compounds, such as organic epoxides, peroxides, steroids and aromatic amino/nitro compounds.[17] Helzlsouser *et al*., (1998)[18] reviewed the isozymes of GST like GSTT1 and GSTM1, which have overlapping substrate specificity, however, the GSTs can interact with alcohol or its contaminants in disease like breast cancer development. For instance the missed of the GST genes can be decreased the ability to conjugate the products of lipid peroxidation, cytotoxic compounds and free radicals producing through the metabolism of alcohol.[19] The present results deal with other evidence that found the null genotyping of the GST was associated with other disease in alcohol consumes patients.[20,21] The present study has many limitations like the difficulty of sample and data collection from drunks. The current investigates concluded that the GSTM null genotyping was strong association with alcohol use disorder, and the null genotyping of both GSTs genes have higher levels of alcohol than other genotyping.

REFERENCES

1. Whalen R, Boyer TD. Human glutathione S-transferases. InSeminars in liver disease 1998;18(4):345-358. ©1998 by Thieme Medical Publishers, Inc..
2. Morel F, Rauch C, Coles B, Le Ferrec E, Guillouzo A. The human glutathione transferase alpha locus: genomic organization of the gene cluster and functional characterization of the genetic polymorphism in the hGSTA1 promoter. Pharmacogenetics and Genomics. 2002 Jun 1;12(4):277-286.
3. Coles BF, Kadlubar FF. Human alpha class glutathione S-transferases: genetic polymorphism, expression, and susceptibility to disease. Methods in enzymology. 2005 Jan 1;401:9-42.
4. Tan KL, Chelvanayagam G, Parker MW, Board PG. Mutagenesis of the active site of the human Theta-class glutathione transferase GSTT2-2: catalysis with different substrates involves different residues. Biochemical Journal. 1996 Oct 1;319(1):315-321.
5. Sies H, Packer L, editors. Glutathione transferases and gamma-glutamyl transpeptidases. Gulf Professional Publishing; 2005 Dec 6.
6. Coggan M, Whitbread L, Whittington A, Board P. Structure and organization of the human theta-class glutathione S-transferase and D-dopachrome tautomerase gene complex. Biochemical Journal. 1998 Sep 15;334(3):617-623.
7. Zhao Y, Marotta M, Eichler EE, Eng C, Tanaka H. Linkage disequilibrium between two high-frequency deletion polymorphisms: implications for association studies involving the glutathione-S transferase (GST) genes. PLoS genetics. 2009 May 8;5(5):e1000472.
8. Whitbread AK, Tetlow N, Eyre HJ, Sutherland GR, Board PG. Characterization of the human Omega class glutathione transferase genes and associated polymorphisms. Pharmacogenetics and Genomics. 2003 Mar 1;13(3):131-144.
9. Board PG. Biochemical genetics of glutathione-S-transferase in man. American journal of human genetics. 1981 Jan;33(1):36.
10. Seidegård J, Vorachek WR, Pero RW, Pearson WR. Hereditary differences in the expression of the human glutathione transferase active on trans-stilbene oxide are due to a gene deletion. Proceedings of the National Academy of Sciences. 1988 Oct 1;85(19):7293-7297.
11. Brockmöller J, Gross D, Kerb R, Drakoulis N, Roots I. Correlation between trans-stilbene oxide-glutathione conjugation activity and the deletion mutation in the glutathione S-transferase class mu gene detected by polymerase chain reaction. Biochemical pharmacology. 1992 Feb 4;43(3):647-650.
12. Brockmöller J, Kerb R, Drakoulis N, Nitz M, Roots I. Genotype and phenotype of glutathione S-transferase class μ isoenzymes μ and ψ in lung cancer patients and controls. Cancer research. 1993 Mar 1;53(5):1004-1011.
13. Pemble S, Schroeder KR, Spencer SR, Meyer DJ, Hallier E, Bolt HM, Ketterer B, Taylor JB. Human glutathione S-transferase theta (GSTT1): cDNA cloning and the characterization of a genetic polymorphism. Biochemical Journal. 1994 May 15;300(1):271-276.
14. Moasser E, Azarpira N, Shirazi B, Saadat M, Geramizadeh B. Genetic polymorphisms of glutathione-s-transferase M1 and T1 genes with risk of diabetic retinopathy in Iranian population. Iranian journal of basic medical sciences. 2014 May;17(5):351.
15. Al-Musawi HS, Alriyahee FA, Kamil ZS, Al-Terehi M. Genotyping of glutathione-s-transferase t1 and m1 in iraqi beh_ampersandsignccedil; et's disease. Annals of Tropical Medicine and Public Health. 2020 Aug;23:231-239.
16. Wright RM, McManaman JL, Repine JE. Alcohol-induced breast cancer: a proposed mechanism. Free Radical Biology and Medicine. 1999 Feb 1;26(3-4):348-354.
17. Taniguchi N., Higashi T., Sakamoto Y., Meister A., editors. Glutathione Centennial: Molecular Perspectives and Clinical Implications. Wiley; Chichester: 1989.
18. Helzlsouer KJ, Selmin O, Huang HY, Strickland PT, Hoffman S, Alberg AJ, Watson M, Comstock GW, Bell D. Association between glutathione S-Transferasee M1, P1, and T1 genetic polymorphisms and development of breast cancer. J Natl Cancer Inst. 1998;90:512-518.
19. Park SK, Yoo KY, Lee SJ, Kim SU, Ahn SH, Noh DY, Choe KJ, Strickland PT, Hirvonen A, Kang D. Alcohol consumption, glutathione S-transferase M1 and T1 genetic polymorphisms and breast cancer risk. Pharmacogenetics and Genomics. 2000 Jun 1;10(4):301-309.
20. Zheng T, Holford TR, Zahm SH, Owens PH, Boyle P, Zhang Y, Zhang B, Wise JP, Stephenson LP, Ali-Osman F. Glutathione S-transferase M1 and T1 genetic polymorphisms, alcohol consumption and breast cancer risk. British journal of cancer. 2003 Jan;88(1):58-62.
21. Rundle A, Tang D, Mooney L, Grumet S, Perera F. The interaction between alcohol consumption and GSTM1 genotype on polycyclic aromatic hydrocarbon-DNA adduct levels in breast tissue. Cancer Epidemiology and Prevention Biomarkers. 2003 Sep 1;12(9):911-914.