Profile of Monoamine Oxidase Activity Levels in Alcohol and Tobacco Addicted Humans

Rajesh N G*, Rafik U S, Sachin L P, Archana D J

School of Life Sciences, Swami Ramanand Teerth Marathwada University, Nanded 431 606, India.

ABSTRACT

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Monoamine oxidase (MAO) activity levels have been described to be associated with the human behavioral aspects such as depressions and other neurological problems. In present study the MAO activity in the plasma of alcohol and tobacco addicted individuals were studied to check it's effect on their MAO activity. The results obtained from above study shows that, the plasma MAO activity is less in alcohol (164.78 ± 1.93 U/ml) and tobacco addicted (193.86 ± 2.97 U/ml) individuals as compared to normal individuals (453.08 ± 2.83 U/ml). This may be happens due to the effect of alcohol and tobacco on the cofactors of enzymatic reactions.

Key words: addiction, depression, monoamine oxidase,

INTRODUCTION

Amine oxidases are ubiquitous enzymes found in both microorganisms and higher organisms. Among the various types of amine oxidase, the mitochondrial flavoenzyme monoamine oxidase (MAO, EC 1.4.3.4.) is of special interest for neuropsychiatry.[1] MAO is involved in the biodegradation of aromatic monoamines, including classical neurotransmitters such as serotonin[2], noradrenalin[3], and dopamine[4]. They appear to play a central role in several psychiatric and neurological disorders. Moreover they also function as a scavengers of various other amines e.g. tyramine, octopamine, tryptamine and also able to oxidize a wide varity of primary, secondary and tertiary amines of different chemical structure.[5] MAO is a flavin-adenine dinucleotide- containing enzyme located on the mitochondrial outer membrane[6] and in human platelets.[7] It occurs in two catalitically active forms, MAO-A found primarily in catecholaminergic neurons and MAO-B localized in serotonergic neurons and in glial cells.[8] The levels of MAO activity have been strongly related with depressive and non depressive states of human being. As a part of antidepressant treatment, the prescription of MAO inhibitors antidepressant drugs is well known.

Depression refers to a wide range of mental health problems characterized by the absence of positive effect (loss of interest and enjoyment in ordinary things and experiences), low mood and a range of associated emotional, cognitive, physical and behavioral symptoms[9] It is estimate that 21% of the world population is affected by this disorder and according to the prediction of World Health Organization; it will be the second leading cause of death by the year 2020.[10] To overcome this neurological disorder variety of antidepressant MAO inhibitors such as Amitriptyline, Clomipramine, Fluoxetine, Fluvoxamine, Tranylcypromine, and Phenelzine are currently available in the pharmaceutical market.[11] These drugs decreases the activity of MAO by inhibibiting its catalytic activity.[12] Apart from the addictions, the chewing of tobacco or drinking of alcohol has been commonly observed as a transient rescue to get rid from the depressive state or day to day tensions.

Cigarette smoking is common among persons with alcohol dependence or abuse with as many as 80% of persons who are alcohol dependent also being smokers. Not only is smoking common in persons with heavy alcohol consumption, but also nicotine dependence appears more severe in smokers with a history of alcohol dependence.[13] It is reported that the habits of alcohol consumption[14, 15] and smoking[16, 17, 18, 19] are associated with changes in catecholamine metabolism and MAO activity.

In present study the levels of MAO activity in randomly selected alcohol and tobacco addicted human volunteers has been described. An attempt has been made to correlate the MAO activity in addicted and normal human beings.

Address for Correspondence:

Rajesh N G, *School of Life Sciences, Swami Ramanand Teerth Marathwada University, Nanded 431 606, India.*

E-mail: rngacche@rediffmail.com

MATERIALS AND METHODS

Benzyl amine, semicarbazide, dinitro phenyl hydrazine, was obtained from s. d. Fine Chemicals Ltd. Mumbai, DPPH (2, 2-diphenyl-1-picryl hydrazine) was procured from Sigma-Aldrich Co. (St. Louis MO, USA). The tobacco, alcohol addicted and normal volunteers were selected randomly at Nanded city (MS) in the age group of 30-50. The blood samples were collected in sample bottle with the help of registered medical practitioner.

Isolation of Platelet MAO

The isolation of platelet MAO is carried out as per the described method[20] with slight modification. Briefly 5 ml of blood sample was collected in a plastic tube containing 2 ml of 0.129 M sodium citrate as anticoagulant. Platelet rich plasma (PRP) was centrifuged at 200 g for 30 min. PRP was centrifuged at 27,000 g for 10 min. and platelet pellet was resuspended in 1 ml of 0.3 M sucrose and membranes were disrupted by repeated freezing and thawing. This extract was used as a source of MAO for further studies.

Determination of MAO activity

MAO activity measurement was performed as per the published method[21, 22] with slight modifications. In brief the reaction mixture contained 0.025 M phosphate buffer pH 7, 0.0125 M semicarbazide, 10 mM benzylamine (pH adjusted to 7), and the enzyme equivalent to 3 mg protein in a total reaction volume of 2 ml. After 30 minutes incubation at 25^0 C, 1ml of acetic acid was added and boiled for 3 min in boiling water bath followed by centrifugation. The resultant supernatant (1 ml) was mixed with equal volume of 0.05% of 2, 4-DNPH and 2.5 ml of benzene was added after 10 min incubation at room temperature. After separating the benzene layer it was mixed with equal volume of 0.1N NaOH. Alkaline layer was decanted and heated at 80^0 C for 10 min. The orange-yellow colour developed was measured at 450 nm. The enzyme activity was expressed as μM benzaldehyde semicarbazine formed/hour/3 mg protein. One unit of enzyme activity was defined as the amount of enzyme which caused an increase in absorbance of 0.001 min^{-1} at 450 nm at 25^0 C and pH 7, which corresponds to the formation of 0.01μM of product. The profile of enzyme activity in alcohol addicted, tobacco chewers and normal volunteers has been summarized in Table 1, 2, 3 respectively.

RESULTS

A random sampling study of human volunteers at Nanded city (MS) was carried out to analyze the plasma MAO activity in tobacco and alcohol addicted individuals and the same was correlated with normal individuals. The result of the present investigation Table 1-3 shows the difference of MAO activity in relation to tobacco and alcohol addictions as compared to normal individuals. The average plasma MAO activity in both alcohol (164.78 ± 1.93 U/ml) and tobacco addicted (193.86 ± 2.97 U/ml) individuals was found to be less as compared to normal volunteers (458.08 ± 2.83 U/ml).

DISCUSSION

A vast body of literature is available attributing the implications of alcohol and tobacco addictions in catecholamine metabolism and MAO activities.[14-19] It has been reported that the platelet MAO activity declines during active drinking in alcoholic patients.[23,24] Moreover the low platelet MAO activity has been considered as biochemical marker for alcoholism and decrease in MAO activity during active alcohol consumption may be concealed by a transitory increase during a certain period of withdrawal.[25] It has been suggested that the low MAO platelet activity may be a result of chronic alcohol intake and may originate from a secondary effect of alcohol on certain enzymatic cofactors such as iron and riboflavin.[26,27] This may be the possible reason for decline in MAO activity in alcohol and tobacco addicted volunteers.

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Table No 1. Profile of MAO activity (U/ml) in alcohol addicted persons

Sr.No.	Sample code	MAO activity in U/ml
1	AA1	112 ± 8.08
2	AA2	200 ± 9.16
3	AA3	146 ± 9.07
4	AA4	168 ± 9.07
5	AA5	168 ± 7.50
6	AA6	149 ± 4.50
7	AA7	187 ± 6.24
8	AA8	170 ± 6.02
9	AA9	149 ± 6.11
10	AA10	165 ± 5.00
11	AA11	171 ± 3.51
12	AA12	206 ± 5.68
13	AA13	159 ± 6.00
14	AA14	180 ± 5.56
15	AA15	175 ± 4.58
16	AA16	164 ± 4.50
17	AA17	153 ± 5.29
18	AA18	141 ±3.60
19	AA19	175 ± 4.50
20	AA20	152 ± 6.00
21	Average Activity	164.78 ± 1.93

The results shown here are the mean values of n=3 ± S.D. AA = alcohol addicted persons.

Table No 2. Profile of MAO activity (U/ml) in tobacco addicted persons

Sr.No.	Sample code	MAO activity in U/ml
1	TA1	69 ± 4.04
2	TA2	162 ± 4.72
3	TA3	253 ± 3.57
4	TA4	235 ± 5.03
5	TA5	214 ± 3.60
6	TA6	223 ± 5.13
7	TA7	184 ± 4.04
8	TA8	205 ± 5.00
9	TA9	234 ± 4.00
10	TA10	211 ± 6.00
11	TA11	176 ± 5.29
12	TA12	101 ± 4.58
13	TA13	165 ± 5.13
14	TA14	233 ± 5.00
15	TA15	164 ± 4.04
16	TA16	244 ± 3.05
17	TA17	219 ± 4.04
18	TA18	202 ± 4.50
19	TA19	163 ± 5.68
20	TA20	215 ± 5.03
21	Average Activity	193.86 ± 2.97

The results summarized are the mean values of n=3 ± S.D. TA = tobacco addicted persons,

Table No 3. Profile of MAO activity (U/ml) in Normal persons

Sr.No.	Sample code	MAO activity in U/ml
1	NL1	444 ± 4.58
2	NL2	505 ± 5.00
3	NL3	555 ± 4.50
4	NL4	457 ± 5.85
5	NL5	304 ± 4.50
6	NL6	510 ± 6.50
7	NL7	496 ± 4.72
8	NL8	520 ± 4.35
9	NL9	355 ± 4.50
10	NL10	368 ± 4.16
11	NL11	426 ± 4.16 .
12	NL12	508 ± 4.50
13	NL13	487 ± 4.00
14	NL14	512 ± 3.51
15	NL15	523 ± 4.04
16	NL16	430 ± 6.02
17	NL17	368 ± 3.60
18	NL18	546 ± 5.00
19	NL19	311 ± 4.50
20	NL20	492 ± 4.50
21	Average Activity	458.08 ± 2.83

The results obtained here are the mean values of n=3 ± S.D. NL = normal persons,

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