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ORIGINAL ARTICLE

Decrement of the Purkinje Cells Diameter after Oral Intake of Lithium in Albino Rats

TAZEEN KOHARI[1], FARAH MALIK[2,], AFTAB AHMAD[3]
[1]*Associate Professor of Anatomy, Islam Medical and Dental College, Pasrur Road, Sialkot*
[2]*Assistant Professor of Anatomy, Liaquat College of Medicine and Dentistry, Jauhar Karachi*
[3] *Associate Professor of Anatomy, M. Islam Medical and Dental College, Gujranwala.*
Correspondance to Dr. Tazeen Kohari, E-mail: tazeenk67@gmail.com. Phone: 0323-2967849

ABSTRACT

Background: The histology of Cerebellar gray matter consists of a middle Purkinje cells layer with flask shaped Purkinje cells. The field of Neurology has documented that different organic compounds and metals are lethal to the excitatory Purkinje Neurons. Researches have proved Lithium to be hazardous to nervous tissue and especially Cerebellum For the past sixty years Lithium is the favorable drug for treatment of Bipolar Disorder.
Aim: To Analyse and record the changes of decrement of the size of Purkinje cell Diameter after chronic Lithium ingestion.
Methods: Sixteen albino rats were selected and were treated with lithium for a period of fifteen days and the data for changes in Purkinje cells Diameter was observed.
Results: The Observations of Our study showed highly significantly decreased diameter of the Purinje cells in Group B (Lithium Carbonate) animals as compared to Group A Animals which were on Lab Diet
Conclusion: The Morphometric Data proved that Lithium Carbonate is Toxic to Purkinje cells, and it educated our Population to use Lithium with caution.
Keywords: Purkinje cell Diameter, Gray matter, Hazardous

INTRODUCTION

The basic plan of cerebellar cortex is the same in vertebrates; thin layer of cerebellar gray matter covers the white matter. The cortex contains the afferents of Climbing and Mossy fibers and five varieties of neurons which are Granule, Stellate, Basket, Golgi and Purkinje cells. Purkinje cells are large and sole output of the cerebellar cortex[1].

The human Purkinje cell layer consists of large pyramidal shaped cells arranged along the upper margin of granular layer, have vesicular nucleus and deeply staining Nucleolus. Human cerebellar purkinje cells are 50µm transversely and 50-100 µm longitudinal their somata is 30-35 µm vertical[2,3,4]. Neural tissue (cerebellum) is damaged by metals such as, Copper, Cadmium[5] and other soft metals like Lithium[6].

Lithium the alkali metal is still the most popular drug for mood disorders, but due to reported deleterious effects of this drug on cerebellar tissue there has been decreased rate of prescribing this drug[7]

There is a deficient literature of atrophy of cerebellar cortex, as a result of Lithium carbonate ingestion. Our research is proof of the degenerative changes of the antimaniac drug (Lithium Carbonate) on the Cerebellar Cortex.

The objective of the study was to analyse and record the changes of decrement of the size of Purkinje cell Diameter after chronic Lithium ingestion.

METHODS

For this study sixteen rats weighing 155-165 grams were selected and divided into two groups. For the purpose of research work these animals were retrieved from Charles River Breeding Laboratories, Brooklyn, Massachusetts, USA. After cross breeding they were kept in the animals at Animal House of Basic Medical Sciences Institute, JPMC Karachi. They were put under observation for one week prior to the experiment. Group A comprised of eight animals on Lab diet for fifteen days and Group B was given Lithium carbonate[8] 20mg/kg for 15 days.

On fifteenth day the animals were sacrificed, brain was removed and the cerebellum was separated from the rest of the brain and fixed in formaldehyde[9] for 24 hours.

The cerebellar tissue was dehydrated by passing through ascending grades of alcohol obtained on glass slides four micron thick sections were collected for staining with haematoxylin and eosin[10].

The changes of the Purkinje cells diameter were observed under light microscope. Micrometry was done for Purkinje Cells Diameter in horizontal axis with ocular reticule. The Ocular micrometer was placed in the right eye and the reticule was adjusted in the left. The divisions of Ocular micrometer were coincided with stage micrometer scale. Calibrations of Ocular counting reticule with stage micrometer scale under 40 X were recorded The micrometry was carried out in selected fields of the tissue. The data was subjected to statistical analysis by using SPSS 2007 version-16.

A statistical difference between means and experimental data was carried out by student 'T' test.
Statistical analysis: Statistical analysis Purkinje Cells Diameter of major group-B (Lithium carbonate treated) shows a highly significant decrease of the Purkinje Cell Diameter at fifteenth day.

RESULTS

On histology and micrometry of H&E stained sections of the Purkinje Cells Diameter was Observed and recorded. The Observations showed highly significantly decreased purkinje cells diameter due to apoptosis in lithium treated group B animals as compared to Control Group.

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Group-A: In rodents of Group A on Lab diet the mean values of the Diameter of Purkinje cells was 14.4±0.51µm on fifteenth day.
Group B: The mean values of the Diameter of Purkinje cells in Group B animals who ingested lithium carbonate was 8.0±0.45µm on fifteenth day.
P Value in Group-A (Lab Diet): A highly significantly increased (P< .001) in the mean values of the diameter of Cerebellar purkinje cells was observed in Group A as compared to the Group B animals.
P Value in Group B (Lithiumcarbonate Treated Group): Animals of Group B showed a highly significantly decreased (P<.001) in the mean values of the Diameter of Purkinje neurons as compared to Group A Rodents.

Table1: Comparison of Mean values of the Diameter (µm) of Cerebellar Purkinje Neurons in Various Groups of Rats

Groups	n	4th Week	
		Mean	SEM
A normal diet	8	14.4 µm	0.51
B Normal Diet +Lithium Carbonate	8	8.0µm	0.45

Table 2: P Values of the diameter (in microns) of cerebellar purkinje neurons among various groups of Albino Rats

Groups	Days	P value
A	15 Days	P<0.001***
B	15 Days	P<0.001***

DISCUSSION

Cerebellar purkinje cells are flask shaped or pyramidal shaped cells in Purkinje cell layer they are excitatory and inhibitory neurons[11].

Lithium carbonate efficacy is well known in depressive disorders but Amer and Karam found out that Lithium use caused Cerebellar degeneration due to neuronal loss. They in their study proved that LI2CO3 caused release of Caspaces -3 which is an Apoptotic marker and resulted in Purkinje cells pyknosis and Karyolysis leading to decrease in Purkinje cell diameter which resulted in neuronal degeneration[12,13]. The same observations of the decreased Purkinje cells Diameter was observed in our study, this may be due to the fact that lithium causes release of reactive oxygen species which leads to Apoptosis, Nuclear Fragmentation and Chromatin crumpling which showed shrunken Purkinje cells with irregular cell margins. The decreased diameter of purkinje cells most probably resulted in cell death which was evident by the increased distance between neurons.

The same results of shrunken purkinje cells and eosinophilic stained, crumpled nucleus was reported byLossi[14,15]. They in their study had documented that Lithium caused Purkinje cell axonal injury leading to the ischemic damage to purkinje nucleus resulting in shrunken diameter of the neuron My results of this study are in agreement with the above this may be due to the reason that Lithium use increases the concentration of Caspases 3[15], which caused chromatin condensation, nuclear apoptosis[6], DNA fragmentation and nuclear envelope collapse along with irregular shaped soma due to these mechanisms I found decreased diameter of purkinje neurons which were visualized under high magnification and was reported.

CONCLUSION

Our study is a marker for Neurologists and psychiatrists to prescribe Lithium in patients with caution.
Conflict of interest: Nil

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