Original Article

Fruitful Clinical Results of L-Arginine Supplementation in Illnesses of Various Etiologies

Saleem Ahmed Bhutto[1], Muhammad Muqeem Mangi[2], Abdullah Khilji[1], Zulfiqar Ali Bhatti[1], Rehana Guddi Siddiqui[3] and Mir Khater Ali Talpur[1]

ABSTRACT

Objective: To evaluate pros and cons of L-Arginine supplementation as protective adjuvant against lithium hepatitis in rats.

Study Design: Experimental Study

Place and Duration of Study: This study was conducted at the Basic Medical Sciences Institute, Jinnah Post Graduate Medical Centre, Karachi, Sindh from October 2012 to March 2013.

Materials and Methods: Sixty adult rats were selected and divided into four groups A, B, C and D with further division into three Subgroups based on periods of treatment 2, 6 and 12 weeks respectively. A was control group, B was on lithium, C was on both lithium and Arginine and group D was on Arginine alone. The initial and final body and absolute and relative liver weights of rats were recorded after scarification at the end of each treatment period. Livers were cut into 3um thick sections stained with hematoxylin and eosin for histological & morphometrical examination

Results: The findings in groups A and D were normal while in group B animals body and liver weights were increased with morphological changes.

Group C animals exhibited significant changes in body and liver weights in comparison with B animals with least inflammation and morphological changes due to L-Arginine co-administration with lithium

Conclusion: It is suggested that lithium induced hepatic-toxicity could be attenuated with L-Arginine.

Key Words: lithium, L-Arginine, liver, anti-oxidant, anti-platelet

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INTRODUCTION

One of the anti-psychotic drugs used is lithium carbonate. It is the drug of choice for bipolar disorder routinely used in clinical practice. There is little margin between toxic and safety dose [2]. Lithium reduces the synthesis of cyclic adenosine monophosphate (cAMP) and inhibits the influx of calcium ions by limiting its channel opening [3].

Lithium affects the transport of mono or divalent cations throughout the whole body. It cannot maintain an electrical gradient across biological membranes therefore disturbs the action potential in the brain and the organs.

[1.] Department of Anatomy, Khairpur Medical College Khairpur Mir's.

[2.] Department of Physiology, Suleman Roshan Medical College Tando Adam Sindh.

[3.] Department of Preventive Medicine, GMMC, Sukkur.

Correspondence: Saleem Ahmed Bhutto, Assistant Professor of Anatomy, Khairpur Medical College Khairpur Mir's.
Contact No: 03093624879
Email: saleem.bhutto@khprkmc.edu.pk

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Lithium replaces sodium and Potassium for the production of single action potential therefore hampers the transmission processes in the brain. It disturbs the biological clock of brain by disturbing the glycogen synthase kinase 3-beta. Lithium inhibits inositol phosphatase and other important enzymes in the normal recycling of membrane phosphoinositides and G-proteins are made to be uncoupled from their receptors [4, 2].

Lithium produces toxic effects on neuromuscular, cardiovascular, gastrointestinal and renal tissues [5, 2]. Lithium administration significantly reduces the activities of anti-oxidant enzymes in liver, e.g. superoxide dismutase (SOD), glutathione peroxidase (GPx). It also decreases the concentration of malonyl dialdehyde (MDA), abnormally raises lipids e.g. cholesterol, triglycerides, phospholipids, and fatty acids in liver tissues with parallel decline in ATP. Lithium damages the DNA and biological membranes [6, 7].

Basically cytotoxic effects of lithium in the liver tissue are manifested by the disturbance of Nitric Oxide metabolism.[8]

Nitric oxide, in the body and Liver is synthesized by L-arginine a semi-essential amino acid[9]. Hypercholesterolemia, diabetes Mellitus and vascular endothelial

dysfunction can be corrected by Arginine via Nitric Oxide metabolism [10].

Keeping in view the above facts, this study was designed to observe the effects of lithium on rat liver with protective role of Arginine and this was done by:

- Observing the morphometric and histological effects of lithium alone and lithium and Arginine combined on rat liver under light microscope.
- Statistical analysis of observations.

MATERIALS AND METHODS

Sixty healthy adult albino rats of Massachusetts breed of either sex, of 90-120 days of age, weighing 200-250 grams were selected for this experimental study. The standard protocol of healthy living and diurnal rhythm of the body and mind to day & Night variation cycle was observed appropriately. All animals were divided into four groups; A, B and C. Each group was further divided into three subgroups e.g. A1, A2 & A3, B1, B2 & B3 and C1, C2 & C3. This subdivision is based on duration of treatment and that period is 2, 6 and 12 weeks respectively. The drugs used in this study were Lithium Carbonate (as "neurolith"-Adamjie Pharma) and L-arginine as "Arginine" (as General Nutrition Corporation- Pittsburg USA).

Group A animals served as Control and were on normal diet alone. Group B animals were treated with Lithium carbonate. Group C animals were given both Lithium carbonate and arginine alone. The Lithium in water and the arginine in feed were administered. The dosage of lithium was 20 mg/kg body weight per day [11] and that of Arginine was 300mg/ kg body weight per day [12].

Initially all the animals were weighed and their initial body weights and final body weights were recorded before scarification after every 2, 6 and 12 weeks of treatment. Livers were carefully removed and weighed to determine absolute and relative liver weights.

Liver was cut into 3 microns thick sections and stained with H&E for morphological examination. Micrometry was done with the help of a stage micrometer scale, Ocular micrometer scale and Ocular counting graticule.

The statistical significance of the differences of various quantitative changes between Lithium Carbonate and Lithium Carbonate + L-arginine treated rats from control rats were evaluated by the students ''t'' test [13], using computer software SPSS version 16 in the windows XP2000. The differences were statistically significant if the P- value was equal to or less than 0.05.

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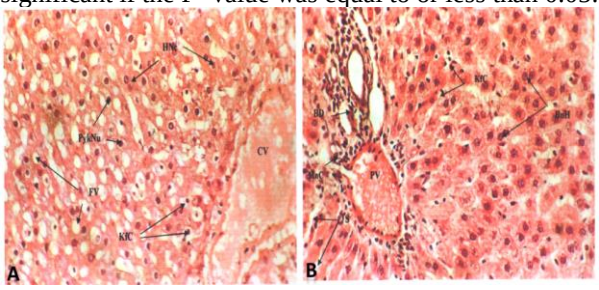

Figure No.1: Photomicrographs A & B stained with H&E depicting morphological changes.

KFC= kuffer cells, HC= Hepatic cords, CV= central vein, BD= Bile duct, BnH= Binuclear Hepatocytes, S= Sinusoids, Pykna= pyknotic nuclei, HNc= Hepatic Nuleus

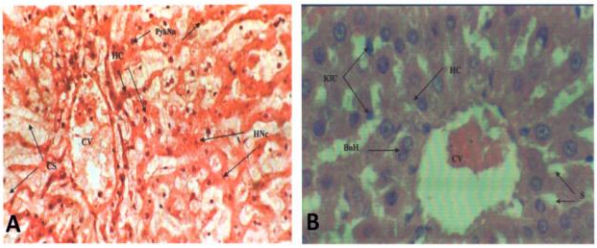

Figure No.2: Photomicrographs A & B stained with H&E revealing morpho-metrical differences of liver lobule under various magnifications

DISCUSSION

The study results revealed an increase in body weight only in group B animals. This can be explained on the basis of altered body and hepatic metabolism.

The Group C Animals showed decrease in body weight. It was comparable more or less to control group. The loss of body weight can be explained by the modulating effect of arginine on body weight [14]. who also found that the dietary arginine reduced fat mass in Zucker diabetic fatty rats.

There was an increase in Absolute & Relative Liver Weight in B group animals due to lithium toxicity. It might be due to edema, inflammatory infiltrate, pyknosis & karyorrhexis, apoptosis and necrosis and compensatory cellular hypertrophy and hyperplasia, marked venous and sinusoidal congestion, more Fatty deposits in cells in liver as suggested by [15]. Also, these observations agree with the work of [17], who described similar findings after co-administration of zinc with lithium in rats.

In C group animals the Absolute & Relative Liver Weight were decreased when compared with B group but the weight loss was not like control. It might be due to less inflammatory and pathological changes in liver, reduced cellular hyperplasia and hypertrophy, less Fatty Deposits, less sinusoidal congestion and inflammatory infiltrate. These findings match with the findings of [16].

Morphological & morphometrical changes increasingly worsened sequentially with the increase in treatment duration e.g 2, 6 and 12 weeks with lithium. As depicted by data, a full-blown inflammation, congestion and apoptosis and necrosis occurred in liver tissue with compensatory responses as suggested by [17]. These observations are also in conformity with the work of [18]. Vijaimohan studied the protective effect of Sobatum against lithium induced toxicity in rats.

Observations in Group C Animals showed the protective effect of Arginine against the Lithium toxicity. Liver morphology was restored to the near normal. Examination revealed an organized Lobular cytoarchitecture of liver as is depicted in the results of [19] who used L-arginine for liver damage in experimental acute cholestasis, an immuno-histochemical study.[20]

CONCLUSION

The present study suggests that L – Arginine supplementation as an adjuvant with treatment is beneficial and significantly attenuates hepatotoxicity of Lithium in albino rats. This further warrants research on man & animal.

Author's Contribution:

Concept & Design of Study:	Saleem Ahmed Bhutto
Drafting:	Muhammad Muqeem Mangi, Abdullah Khilji
Data Analysis:	Zulfiqar Ali Bhatti, Rehana Guddi Siddiqui, Mir Khater Ali Talpur
Revisiting Critically:	Saleem Ahmed Bhutto, Muhammad Muqeem Mangi
Final Approval of version:	Saleem Ahmed Bhutto

Conflict of Interest: The study has no conflict of interest to declare by any author.

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