Original Article

Enteral intake of Aluminum Sulphate in acidic medium enhances absorption and alters tissue content of other trace elements in male albino rats

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Summary:

Background: Aluminium (Al) intoxication was recognised as a causal agent in patients undergoing haemodialysis and then was linked to Alzheimer's disease. Nevertheless, environmental pollution with Al is mainly via drinking water, particularly when acidic rain falls on Al containing soil and bedrock. In addition, aluminium utensils are widely used for cooking. Therefore, an increasing concern is emerging for the role of pH in the absorption of aluminium from ingested food.

Materials and methods: The objective of this study was to establish the effect of acidification with1 ml/dL of concentrated acetic acid on the absorption of aluminium from a 5 mM aluminium sulphate solution taken orally in albino rats. Levels of Al, Ca, Cu, and Fe in plasma. brain, kidney, and liver were measured by atomic absorption.

Results: The results indicate that acidification of Al salt solution with acetic acid enhances its absorption. Increased plasma Al level ($p<0.05$) was associated with increased deposition in all tissues and a reduced overall body mass ($p<0.05$) and mass of cerebral hemispheres ($p<0.01$) relative to the controls. The increased plasma levels of Al correlated positively with increased Al deposited in the kidney ($r=0.790$) liver ($r = 0.967$), and brain ($r=0.955$) despite the blood brain barrier. Increased Al also correlated negatively with levels of Ca, Cu, and Fe in all tissues except in brain in which there was a positive correlation with Ca deposition.

Conclusion: The study shows an increased absorption and deposition of Al in the tissues from rats ingesting acidified Al solution with acetic acid.

Key words: Aluminum absorption, acetic acid, brain atrophy.

Introduction:

Initially aluminium (Al) intoxication was recognised as a causal agent in patients undergoing haemodialysis and then was linked to Alzheimer's disease (1). Nevertheless.environmental pollution with Al is mainly via drinking water particularly when acidic rain *falls on* Al-containing soil and bedrock (2). Further routes of pollution by this trace element may come from the use of aluminium salts in preservatives, colouring agents (3) or the use of Al-containing antacids, vaccines phosphate binding and dialysate fluid (4).Fortunately, the bio-availability of Al from drinking water is as low as .0.3% (5). Still plasma Al could be elevated when Al intake coincides with some dietary factors such as citric acid (6) whereas phosphorus and silicone reduce its absorption (7), most cooking utensils contain Al and generally the prepared foods are acidic nature. Therefore, the initiation of a project to investigate the effects of the free enteral intake of a large dosage of aluminium sulphate solution with acetic acid on the absorption of Al and on the possible toxic effect on the brain is justified.

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Materials and Methods:

Al absorption was studied in mature male albino rats weighing 83.5 ± 4.35 g The animals, which were divided into 3 groups of 10 animals in each, were placed in plastic cages with an ample supply of rats pellets. The control group was supplied with regular lap water while in the treated groups 5 mM of $Al_2(SO_4)_3 . 18H_2O$ / L was given to groups (Al + O) and *(Al +A).* The later group received an additional 1 ml of concentrated acetic acid /dL. The usage of a large dosage of Al salt was lo overcome its low absorption and availability.

The Al-containing solution was made more palatable by adding glucose 5 gm/ dL. The addition of the acid was intended to study the effect of acidic medium on Al absorption of enteral intake of Al-containing solution. Throughout the 12 weeks of the project, the volume of fluid intake was measured every other day and the body weight was determined twice-weekly. On termination 3-5 ml of blood was obtained by cardiac puncture while the animal was under ether anaesthesia. The animals were sacrificed to obtain the brain, kidneys and liver, as well as plasma for trace element analysis (Al. ('a. Cu, and Fe). The left cerebral hemisphere was dissected gently and weighed accurately for comparison. Tissue specimens were digested in concentrated acid mixture

of H_2So_4 and UNO_3, of 1:1 ratio. Levels of Al, Ca, Cu, and Fe in plasma, brain, kidney, and liver were measured by atomic absorption spectroscopy. All data obtained were tabulated and expressed in terms of X ± SD, and the differences were assessed using student t-test. Linear regression analysis was calculated to find out the type of relation that exists between elevated Al deposited in tissue and the stale of other trace elements contained in the tissue specimens under investigation. The study was conducted according to national guidelines for animal usage in research.

Results:

Aluminium salt ingesting animals showed normal locomotion with no noticeable ill signs. Nevertheless, rats ingesting the acidified Al salt solution with 1 ml of acetic acid gained less weight ($p<0.05$) with clear decrease in brain weight ($p<0.01$), and in daily fluid intake ($p<0.05$) than those ingesting the non-acidified Al salt solution (Fig 1). Analysis of plasma and tissues Al levels gave evidence indicating that enteral fluid intake 5mM of Al sulpate with acetic acid enhanced the absorption of Al (table1). 'The progressive rise in plasma Al levels of the control, (Al + 0) and then (Al + A) groups correlated positively with the magnitudes of Al deposited in the brain, kidney, and liver (Fig 2). Furthermore, the increased Al deposited in these tissues altered their content of Ca, Cu, and Fe. In the brain, the pooled data of Al levels of the three groups showed positive correlation with Ca levels (r=0.841). but it was negative with Cu (r= -0.806) and Fe (r= -0.842) (Fig3). Similarly, the pooled data of Al levels in the kidney and liver showed negative correlation with their Ca, Cu, and be levels. All correlations were highly significant ($p<0.001$).

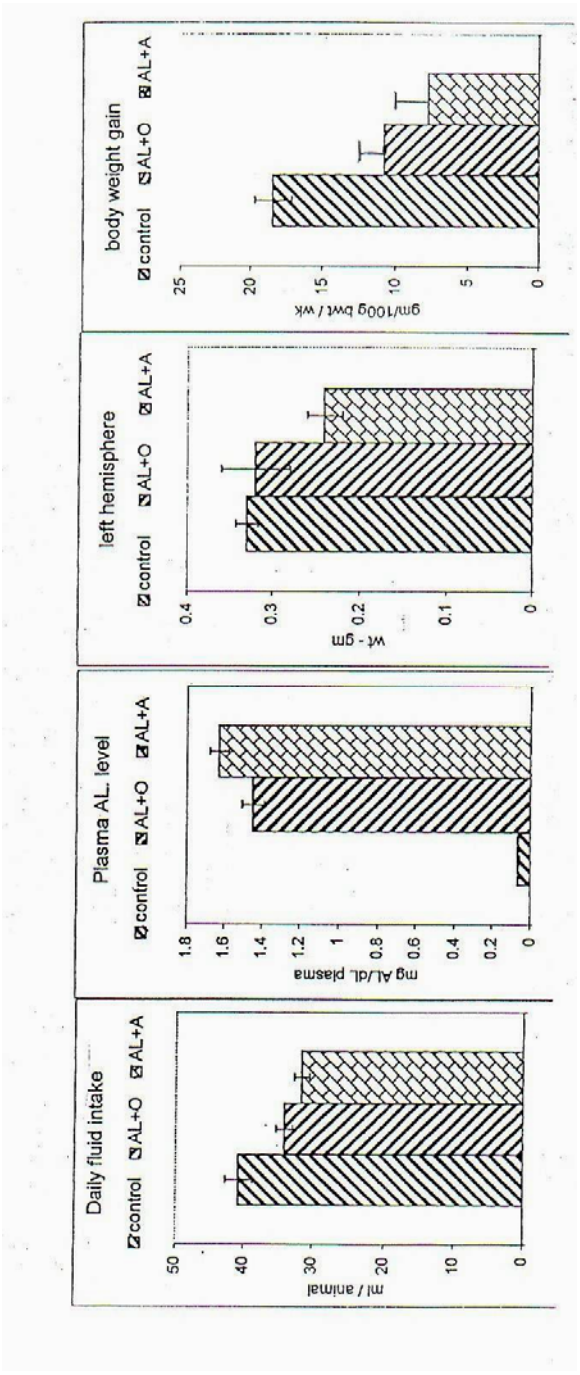

Fig.1 shows the effect of eternal fluid intake of 5 M m of aluminium sulphate with (A1+A) or without (A1+0) acetic acid on daily fluid intake, plasma. A1 level and weights of the left hemisphere and the body gain, $(\overline{X} \pm SD)$.

Table 1: Trace elements contents in plasma (mg dL^{-1}) and in tissues (ppm of wet wt) in rats ingesting 5mM A1 sulphate with (A1+A) or without (A1+0) of 1 ml of acetic acid expressed in term of $(\overline{X} \pm SD)$.

Trace elements	AL	Ca	Cu	Fe
		Plasma mg dL^{-1}		
Control	0.064 ± 0.005 ***	9.96 ± 0.32 -	0.068 ± 0.002 *	0.96 ± 0.061 -
AL + O	1.48 ± 0.08 *	8.93 ± 0.373 -	0.083 ± 0.004 -	0.982 ± 0.073 -
AL + A	1.63 ± 0.078	8.92 ± 0.467	0.087 ± 0.003	0.89 ± 0.037
		Brain - ppm		
Control	2.29 ± 0.13 ***	415,03 ± 10.51 **	8.19 ± 0.19 -	104.28 ± 6.15 *
AL + O	29.59 ± 0.97 ***	590.37 ± 10.75 ***	7.11 ± 0.06 *	95.51 ± 2.36 ***
AL + A	44.57 ± 0.97	1479.0 ± 22.1	5.66 ± 0.04	16.67 ± 0.78
		Kidney-ppm		
Control	2.06 ± 0.02 ***	548.07 ± 6.52 **	27.18 ± 0.98 **	286.28 ± 13.06 **
AL + O	25.47 ± 1.19 ***	489.97 ± 9.19 **	20.85 ± 0.58 *	218.33 ± 12.9 *
AL + A	53.17 ± 1.03	443.6 ± 7.42	17.5 ± 0.45	197.05 ± 5.27
		Liver -ppm		
Control	0.95 ± 0.014 ***	305.53 ± 10.13 ***	6.56 ± 0.21 -	241.34 ± 8.46 -
AL + O	9.79 ± 0.89 **	190.25 ± 8.31	6.02 ± 0.28 -	238.75 ± 6.28 **
AL + A	12.99 ± 1.02	15101 ± 3.05	5.34 ± 0.19	197.61 ± 2.59

- *- means not significant*
- ** means $p < 0.05$*
- *** means $p < 0.01$*
- **** means $p < 0.001$*

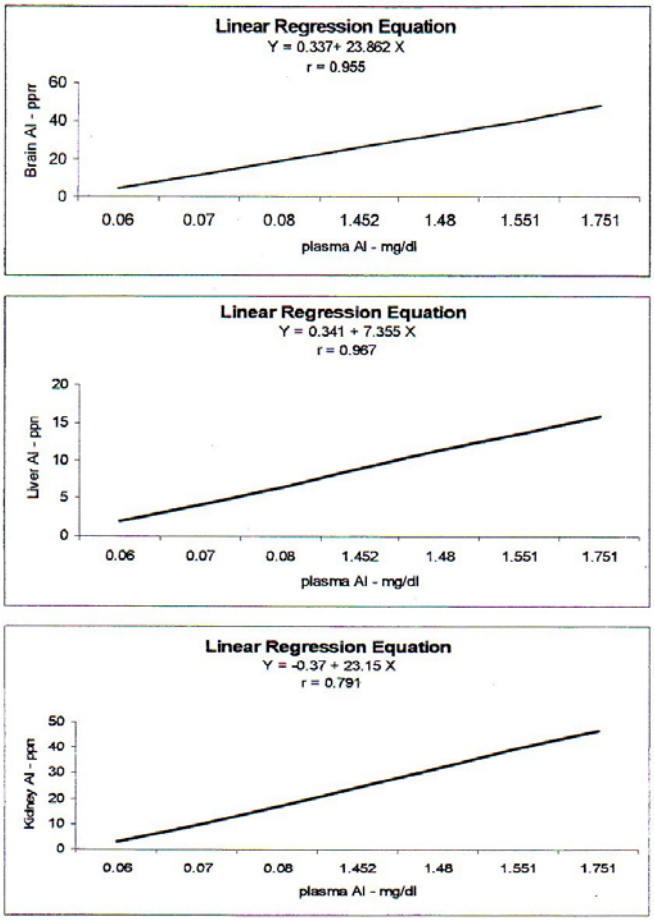

Fig.2 shows linear regression Curves of plasma A1 and tissue A1 contents.

Discussion:

The enteral intake of Al salt with acetic acid enhanced significantly the absorption as evidenced in elevated plasma levels and increased deposition in all organs studied. This confirmed early findings where citric acid in lemon juice as another weak organic acid increased the bio-availability of aluminum. The mechanism of citrate and other organic acids enhancement of 'AI absorption are para-cellularly by opening the tight junctions that are present between mucosal cells rather than intra-cellularly (6).

Failure to observe neurological disorders in rats despite the accumulated Al could he due to the fact that the symptoms were too slight to notice in the animals or the dose and/or the duration of enteral intake of aluminium sulphate was not enough lo produce signs of illness. Infants in prolonged intravenous feeding of Al containing solution have exhibited impaired neurological development and, on increasing Al exposure, the impairment was associated with a reduction in the Mental Development Index (8). The reduced gain in body weight is assumed to be attributed to reduction in bone weight, since osteomalacia is a common feature of Al toxicity (9). The brain atrophy of rats which implies a significant reduction in brain weight is attributed to neural cell damage of the RNA as a result of oxidative stress and the formation of reactive oxygen species, such as superoxide and hydrogen peroxide, after being converted to short-lived highly reactive

intermediate such as hydroxyl radical (10). The reported positive correlations between plasma Al levels and that deposited in the given organs is confirmed (11), In comparing the well-known blood How to the kidneys with that of the brain relative to the magnitude of the deposited Al in these organs, one may assume that the permeability of the blood-brain barrier to Al is reasonably altered or that the blood-brain barrier has been modified to allow more Al permeate through (12, 13). The noticeable finding of this work is the alteration in the content of the trace elements Ca, Cu and be in organs in the presence of Al deposited in the brain, kidney and liver. The reason for such re-distribution is still unclear. Nevertheless, there were reports on The effects of Al presence on Ca, Cu and Fe absorption but there was no mentioned reports as yet indicating the increased Al deposition in the mentioned tissue causing redistribution of these trace elements. The reasoning of such redistribution is still lacking Distal view but still related where Grondolfi and associates were working on neuro-degerative disease in conjunction with chronic dietary deficiencies of trace elements concluded that Al altered intracellular Ca homeostatic in vitro (14). Furthermore, uraemia patients undergoing haemodialysis expressed elevated parathyroid hormone level, which normally influenced Ca absorption and its mobilization and the release of Ca into the plasma (15). This release was not evident in plasma Ca level as mostly due to effective Ca haemostatic (14). Recently, Perl and Moarems (2006) demonstrated prominent evidence that, the deposited Al in the brain was located within the neuro- fibrillory tangles of medium and large size neurons (16, 17). Nevertheless, these tangles are morphologically and biologically different from that observed in the brain of Alzheimer diseased patients (18), thus Al does not represent an etiologic cause of AD rather than it plays an active role in the pathogenesis of AD. Furthermore, that both epidemiological; and experimental findings strengthen the possibility that prolonged exposure to relatively low Al level may be neurotoxic leading to neuro-degerative disease (18, 19).

Conclusion:

The enteral intake of Al salt with acetic acid enhanced significantly the absorption as evidenced in elevated plasma levels and increased deposition in the tissues studied (brain, kidneys and liver). This increased absorption of Al was associated with brain atrophy, decrease body mass and altered tissue contents of Ca, Cu and Fe in the organs studied. Further work is encouraged to re-examine the re-distribution of these elements with prolong study and their outcome.

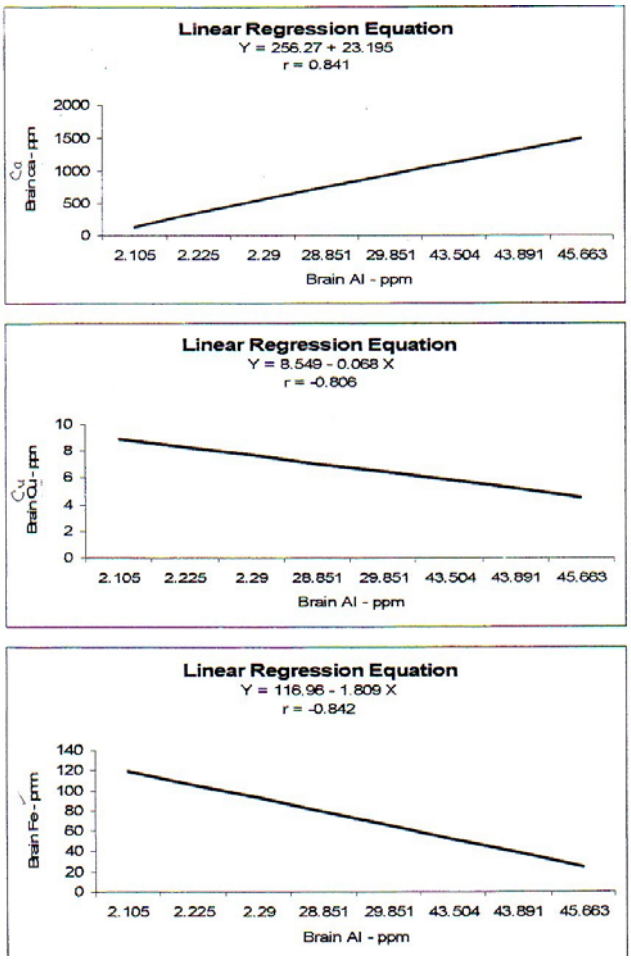

Fig. 3 shows linear regression Curves of brain A1 and brain Ca, Cu and Fe.

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