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**Research Article**

# THE EFFECT OF LOCALLY MANUFACTURE POTS ON LEVELS OF ALP &LDH ENZYMES

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**ABSTRACT**

The aim of this study is to assess the effect of some toxic elements released from locally made cooking pots made of soft drink cans (i.e. Pepsi cans=Tins=) and Atmonia pots on ALP & LDH. Ninety rats (average body weight 160 -220 gms) were divided into three groups (30 male per group) as follow: Group one (G1) represented control (without treatment), Group two (G2) and Group three (G3) received (Atmonia, Pepsi can) fed into 3gms/day of cooked food using feeding sucker plastic bottle for three months, we sacrificed 10 rats from each group monthly to estimate the level of ALP & LDH enzymes .The results showed that some heavy metals had led to a significant increase ($P< 0.01$) in ALP and LDH levels.



## INTRODUCTION

Metals and alloys are used as food contact materials, mainly in processing equipments, containers and household utensils but also in foils for wrapping food stuffs , In the Sudan some metals are reused locally through certain processes for manufacturing cooking pots ,the locally made cooking pots contain some alloys that are dangerous for human life, certain heavy metals are nutritionally essential for a healthy life, small quantities of these heavy metals are referred to as the trace elements e.g., iron and copper, manganese, and zinc, these elements (or some form of them) are commonly found naturally in foodstuffs (in fruits and vegetables) and in commercially available soft drinks (IOSHIC 1999). Many toxic elements could be released from cooking pots into the cooked food if they are not lined with some alloys, exposure to these metallic element in form of cooked materials is usually through oral intake, these elements had been considered toxic if their total amounts exceed the recommended exposure limits or bring about an unacceptable change in the composition of the foodstuffs (Abou-Arab, A.A- K 2001). Many workers have discussed toxicity of heavy metals and their relationship with some diseases, Glanze (1996) reported that there are 35 metals that concern humans because of occupational or residential exposure Twenty-three of these are the heavy elements or "heavy metals": antimony, arsenic, bismuth, cadmium, cerium, chromium, cobalt, copper, gallium, gold, iron, lead, manganese, mercury, nickel, platinum, silver, tellurium, thallium, tin, uranium, vanadium, and zinc. He also stated that small amounts of these elements are common in our environment and diet and are actually necessary for good health but large amounts of any of them may cause acute or chronic toxicity (poisoning). Similarly, the International Occupational Safety and Health Information Centre (IOSHIC, 1999) reported that heavy metal toxicity can result in damaged or reduced mental and central nervous system function, lower energy levels, damage to blood composition, lungs, kidneys, liver, and other vital organs. Long-term exposure may result in slowly progressing physical, muscular, and neurological degenerative processes that mimic Alzheimer's disease,

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muscular dystrophy, and multiple sclerosis, Allergies are not uncommon and repeated long-term contact with some metals or their compounds may even cause cancer, for some heavy metals, toxic levels can be just above the background concentrations naturally found in nature (Laurens 2009).

### *Objectives*

To assess the effect of some toxic elements released from locally made cooking pots made of soft drink cans (i.e. Pepsi cans=Tins=) and Atmonia pots on ALP & LDH levels.

## MATERIAL AND METHODS

### *Experimental Animals*

Ninety adult males Wistar rats weighting about (160-220 gram each) were allocated for the experiments. They were then divided into 3 groups of 30 rats each. All groups of rats were kept under standard conditions (Temperature, light, humidity). Rats were fed with standard chow and free tap water when they will be out of metabolic cage. Then, rats were fed with food cooked into two types of locally cooking pots. These were Atomonia and Pepsi can pots. They were known as "Cooking Halla". All test animals were used after the time of adaptation under laboratory conditions. Group one used as control and received standard chow, group two received 3 gm/rat/day of food cooked in atmonia pots, group three received 3 gms/rat/day of food cooked in pepsi cans pots in addition to standard chow. Rats ware fed for three months on a food made out of millet flour. Millet flour which prepared by addition of millet flour to cool water and kept for two hours for fermentation. Then the fermented food was cooked in the test pots (Atmonia, Pepsi cans) on a calm fire. Each pot received 9 Kgs of fermented homogenous millet dissolved in tap water for serial cooking's. Each cooked food from the two cooking pots collected separately in sterilized clean trays and kept until dry. The dried food was milled until became fine and kept under laboratory conditions until use. Every one rat of the two test group (Atmonia, Pepsi can) was fed into 3gms/day of cooked food using feeding sucker plastic bottle. The control rats ware fed into normal feeding chow as described above. All test and control rats were kept under laboratory conditions.

All rats were fed into food consists of protein (meat) to avoid cannibalism. At end of each experiment Wistar rats were anesthetized after 12 hours fasting. Whole blood samples were collected from the hepatic portal veins at two tubes. Bloods of the first tubes were left for 15 minutes at room temperature then centrifuged at 4000 rpm for 20 minutes for separation of the serum which were kept in plastic vials at 26±3°C until analysis. For ethical considerations, the collection was done from Wistar rats and with the permission of Department of Scientific Research, Khartoum University.

### *Collection of Blood Samples*

Blood samples were collected by cardiac puncture and allowed to clot for 2 hours at room temperature, followed by centrifugation at 3500 rpm for 10 minutes to obtain the serum.

### *Estimation of Plasma ALP & LDH*

Serum ALP & LDH levels were measured by the Hitach 902 Analyzer by using commercial kits (Bio System, S.A, Costa Brava30, Barcelona, Spain 2011)

## RESULTS AND DISCUSSION

**Table 1** of means of ALP from Wistar rats after feeding on feed Cocked in cocking pots made of Atmonia and Pepsi cans compared with normal range and control after one month.

| Treatment | G1 (1) | G1(2) | G1(3) | G1(4) | G1(5) | Total | Mean |
|---|---|---|---|---|---|---|---|
| Atmonia | 280.08 | 243.23 | 229.90 | 218.85 | 211.48 | 1183.54 | 236.71** |
| Pepsi cans | 108.71 | 184.76 | 170.90 | 129.90 | 114.24 | 708.51 | 141.70** |
| Normal reading | 146.00 | 146.00 | 146.00 | 146.00 | 146.00 | 730.00 | 63.15 |
| Control | 58.69 | 45.14 | 70.94 | 70.02 | 70.94 | 315.73 | 146.00 |
| Grand total | | | | | | 2937.78 | |
| Grand mean | | | | | | | 145.00 |

The results are expressed as Mean (n = 10) per treatment and respective control groups. Levels of significance values were considered to be statistically significant.

CV% = 15.8%
SE± = 13.6

**Table 2** of means of ALP from Wistar rats after feeding on feed Cocked in cocking pots made of Atmonia and Pepsi cans compared with normal range and control after two month

| Treatment | G1 (1) | G1(2) | G1(3) | G1(4) | G1(5) | Total | Mean |
|---|---|---|---|---|---|---|---|
| Atmonia | 330.08 | 293.23 | 279.90 | 268.85 | 261.48 | 1433.54 | 286.71** |
| Pepsi cans | 158.71 | 220.90 | 234.76 | 179.9 0 | 164.24 | 958.51 | 191.70** |
| Normal reading | 146.00 | 146.00 | 146.00 | 146.00 | 146.00 | 730.00 | 146.00 |
| Control | 58.69 | 45.14 | 70.94 | 70.02 | 70.94 | 315.73 | 63.15 |
| Grand total | | | | | | 3437.78 | |
| Grand mean | | | | | | | 145.00 |

The results are expressed as Mean (n = 10) per treatment and respective control groups. Levels of significance values was, $^{**}p<0.01$ considered to be statistically significant.

CV% = 13%
SE± = 5.9

**Table 3** means of ALP from Wistar rats after feeding on feed Cocked in cocking pots made of Atmonia and Pepsi cans compared with normal range and control after three month

| Treatment | G1 (1) | G1(2) | G1(3) | G1(4) | G1(5) | Total | Mean |
|---|---|---|---|---|---|---|---|
| Atmonia | 340.00 | 303.00 | 289.00 | 278.00 | 271.00 | 1481.00 | 296.20** |
| Pepsi cans | 168.71 | 230.00 | 244.00 | 189.00 | 174.00 | 1005.00 | 201.00** |
| Normal reading | 146.00 | 146.00 | 146.00 | 146.00 | 146.00 | 730.00 | 146.00 |
| Control | 58.69 | 45.14 | 70.94 | 70.02 | 70.94 | 315.73 | 63.15 |
| Grand total | | | | | | 3531.73 | |
| Grand mean | | | | | | | 176.58 |

The results are expressed as Mean (n = 10) per treatment and respective control groups. Levels of significance values was, $^{**}p<0.01$, considered to be statistically significant.

CV% = 79%
SE± = 88

**Table 4** of means of LDH from Wistar rats after feeding on feed cocked in cocking pots made of Atmonia and Pepsi cans after the first month

| Treatment | G1(1) | G1 (2) | G1 (3) | G1 (4) | G1 (5) | Total | Mean |
|---|---|---|---|---|---|---|---|
| Atmonia | 130.95 | 133.86 | 142.59 | 131.53 | 168.70 | 707.63 | 141.53** |
| Pepsi cans | 142.74 | 111.74 | 102.43 | 110.58 | 121.08 | 588.57 | 117.71** |
| Normal reading | 108.50 | 108.50 | 108.50 | 108.50 | 108.50 | 542.50 | 108.50 |
| Control | 81.48 | 68.70 | 76.82 | 91.95 | 95.44 | 414.39 | 82.88 |
| Total | | | | | | 2253.09 | 112.65 |

*** G1 – 10 = Replications of control.**

The results are expressed as Mean (n = 10) per treatment and respective control groups. Levels of significance values was, $^{**}p<0.01$, considered to be statistically significant.

CV% = 11%
SE+ = 7.82

**Table 5** of means of LDH from Wistar rats after feeding on feed cocked in cocking pots made of Atmonia and Pepsi cans compared with normal reading and control after two months

| Treatment | G1 (1) | G1(2) | G1(3) | G1(4) | G1(5) | Total | Mean |
|---|---|---|---|---|---|---|---|
| Atmonia | 180.42 | 199.02 | 208.36 | 171.11 | 241.96 | 1000.87 | 200.17** |
| Pepsi cans | 204.00 | 154.78 | 128.04 | 129.21 | 146.72 | 762.75 | 152.55** |
| Normal reading | 108.50 | 108.50 | 108.50 | 108.50 | 108.50 | 542.50 | 108.50 |
| Control | 81.48 | 68.70 | 76.82 | 91.95 | 95.44 | 414.39 | 82.88 |
| Total | | | | | | 2720.51 | 136.03 |

The results are expressed as Mean (n = 10) per treatment and respective control groups. Levels of significance values was, $**p<0.01$ considered to be statistically significant.

CV% = 15.85%
SE± = 13.6

**Table 6** of means of LDH from Wistar rats after feeding on feed cocked in cocking pots made of Atmonia and Pepsi cans compared with normal reading and control after three months

| Treatment | G1 (1) | G1(2) | G1(3) | G1(4) | G1(5) | Total | Mean |
|---|---|---|---|---|---|---|---|
| Atmonia | 230.42 | 249.02 | 258.36 | 221.11 | 290.96 | 1249.87 | 249.97** |
| Pepsi cans | 254.00 | 254.00 | 228.00 | 179.00 | 196.00 | 1111.00 | 222.20** |
| Normal reading | 108.50 | 108.50 | 108.50 | 108.50 | 108.50 | 542.50 | 108.50 |
| Control | 81.48 | 68.70 | 76.82 | 91.95 | 95.44 | 414.39 | 82.88 |
| Grand total | | | | | | 3317.76 | |
| Grand mean | | | | | | | 165.88 |

The results are expressed as Mean (n = 10) per treatment and respective control groups. Levels of significance values was, $**p<0.01$ considered to be statistically significant.

CV% = 13.5%
SE± = 14.2

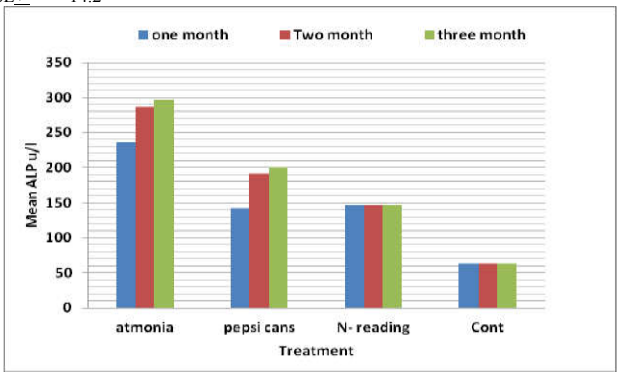


**Fig 1** Mean level of ALP/ul from Wistar rats fed on feeds cocked in cocking pots made of Atmonia and Pepsi cans in comparison with normal range and control for three months

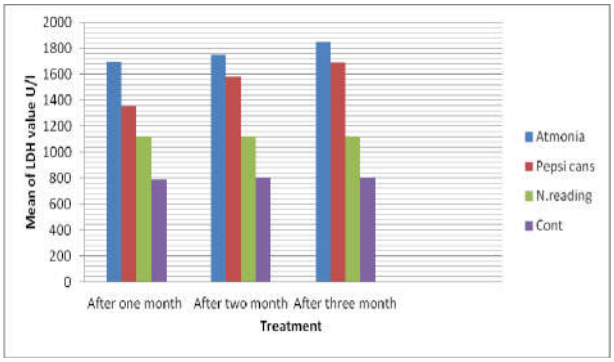


**Fig 2** Mean level of LDH U/l from Wistar rats fed on feeds cocked in cocking pots made Atmonia and Pepsi cans in comparison with normal range and control for three months

Table (1) and Fig 1 indicated that ALP for G2 and G3 were 236.71, 236.71 respectively in the first month compared with the control146.00. These results were clearly agree with Kim, (2010) who reported that heavy element such as Pb, Fe, Cu and Al were considered as the main reasons for high elevation of AST, ALT and ALP. The results obtained from this study were supported the assumption that heavy elements released in feeds cooked in cooking pots made of Atmonia and Pepsi cans were the main reasons of high elevation of liver functioning enzymes of Wistar rats such as AST, ALT and ALP. Alkaline phosphatase (ALP) an enzyme also found in the liver, bile ducts, and bones (Ki-Soo Kang, 2013). High levels of these enzymes may cause liver damage or disease, a blocked bile duct, or bone disease. Table (2, 3) and Fig 1 showed increase level of ALP in the second and third months respectively compared to their controls. This result also in line with Kim, (2010). In analysis of variance, there was significant difference between groups at P≤0.01. Table (4) and Fig 2 indicated that LDH for G2 and G3 were141.53, 117.71 respectively in the first month compared with the control 82.88. These results were in line with Paul and Giboney, (2005) reported that some toxic heavy elements were able to cause significant increase in LDH amounts in liver. Table (5, 6) and Fig 2 indicate increase level of LDH in the second and third months respectively compared to their controls. In analysis of variance, there was significant difference between groups at P≤0.01.

## CONCLUSION

Heavy elements from cooking pots made out of Atmonia & Pepsi cans such as Lead, copper, Aluminum, Ferrous and Arsenate give a significance increase (P≤ 0.01) in levels of ALP and LDH enzymes.

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