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EFFECTS OF ALUMINUM SUPPLEMENTATION ON GROWTH PERFORMANCE, SOME HAEMATOLOGICAL, BIOCHEMICAL AND IMMUNOLOGICAL INDICES OF MALE RABBITS

(With 4 Tables)

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تأثير الأمداد بالألومنيوم في الغذاء على النمو وبعض مكونات الدم وبعض التغيرات البيوكيميائية والمناعية في ذكور الأرانب

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أجريت هذه الدراسة على عدد ثمانية واربعون من ذكور الأرانب النيوزيلندية عند عمر الفطام (٥ أسابيع) وذلك بهدف تقييم التأثيرات السلبية لأضافة عنصر الألومنيوم في الغذاء علي أداء النمو والمظاهر الهيماتولوجية والمناعية للدم وكذلك بعض المكونات البيوكيميائية للبلازما ومدى تراكم الألومنيرم فى بعض أعضاء الجسم. قسمت الحيوانات الي اربعة مجموعات متساوية (١٢ في كل مجموعة) بمتوسط وزن ٤٨٨ جرام. غذيت المجموعة الأولي علي عليقة متزنة دون اضافة الألومنيوم وأعتبرت مجموعة قياسية بينما غذيت كل من المجموعات الثانية والثالثة والرابعة علي علائق زودت بثلاث مستويات من عنصر الألومنيوم (١٠٠٠، ٢٠٠٠، ٣٠٠٠ جزء في المليون علي التوالي). أظهرت النتائج أن حيوانات المجموعة الثانية والثالثة والرابعة سجلت قيم أقل معنويا فى متوسطات وزن الجسم النهائى ومعدل الزيادة اليومية فى الوزن مقارنة بالمجموعة القياسية. سجلت المجموعة الرابعة قيم أقل معنويا فى متوسطات معدل استهلاك الغذاء يوميا مقارنة بالمجموعة القياسية. سجلت حيوانات المجموعة الثانية والثالثة والرابعة قيم أعلى معنويا فى متوسطات معدل كفاءة التحويل الغذائى مقارنة بالمجموعة القياسية بما يعنى زيادة معدل استهلاك الغذاء بالنسبة لمعدل الزيادة اليومية فى الوزن. أظهرت المجموعة الثالثة والرابعة انخفاض معنوى فى تركيز هيموجلوبين الدم، عدد كرات الدم الحمراء والنسبة المئوية لمكونات الدم الخلوية مقارنة بالمجموعة القياسية. أظهرت المجموعة الثالثة والرابعة زيادة معنوية فى العدد الكلى لكرات الدم البيضاء مصحوبا بزيادة فى نسبة الكرات المتعادلة والكرات حمضية الصبغ وانخفاض فى نسبة الكرات الليمفاوية مقارنة بالمجموعة القياسية. كما لوحظ انخفاض

معـــنوى فى محتوى البلازما من البروتين الكلي والجلوبيولين فى المجموعة الرابعة مقارنة. كما لوحظ انخفاض معنوى فى محتوى البلازما من الجلوكوز وتركيز هرمون الغدة الدرقية (T_3) فـــى المجموعات الثانية والثالثة والرابعة مقارنة بالمجموعة القياسية. هذا وقد أظهرت المجموعـــة الثالثة والرابعة ارتفاع معنوى فى تركيز الكوليسترول فى البلازما بينما أظهرت المجموعات الثانية والثالثة والرابعة زيادة معنوية فى محتوى البلازما من عنصر الألومنيوم مقارنـــة. أظهـــرت المجموعة الثالثة والرابعة زيادة معنوية فى تراكم الألومنيوم فى أنسجة العضـــلات والكبد بينما زاد تراكم الألومنيوم معنويا فى أنسجة الكلية والمخ فى المجموعات الثانـــية والثالـــثة والرابعة مقارنة. نستنتج من هذه الدراسة أن احتواء علائق الأرانب النامية على مستويات عالية من عنصر الألومنيوم (خاصة ٢٠٠٠ ، ٣٠٠٠ جزء فى المليون) أدى الى انخفاض معنوى فى الأداء الأنتاجى وفى كفاءة الأنشطة الحيوية لها نتيجة التأثير السلبى لعنصر الألومنيوم على تفاعلاتها ووظائفها الفسيولوجية.

SUMMARY

Forty eight of weanling New Zealand White (NZW) male rabbits of 5 weeks old and averaged 488 g body weight were used to evaluate the negative effects of adding aluminum (Al) in diet on growth performance, blood haematological and immunological indices, plasma biochemical constituents and accumulation of Al in some organ tissues. Animals were divided randomly into 4 groups. The first group fed on control diet while the second, third and fourth groups were fed on the control diet mixed with Al sulfate to perform three added levels of Al, 1000 (T1), 2000 (T2) and 3000 (T3) ppm. The results showed reduction ($P<0.001$) in averages of final body weight and daily weight gain for T1, T2 and T3 groups when compared to control one. Averages of feed intake decreased ($P<0.05$) in T3 group of rabbits compared to control. T1, T2 and T3 groups of male rabbits showed lower ($P<0.001$) feed conversion efficiency than the control one. Blood Hb concentrations, RBCs count per mm^3 blood and PCV % were lowered ($P<0.01$) for T2 and T3 rabbits than control. Total count of leucocytes increased ($P<0.01$) for T2 and T3 rabbits when compared to control. Rabbits of T2 and T3 had lower ($P<0.01$) lymphocytes % and higher ($P<0.01$) neutrophils and eosinophils % than control. There were decreases ($P<0.01$) in concentrations of plasma total protein and globulin for T3 rabbits, and in plasma glucose of the treated rabbits when compared to control. Plasma cholesterol increased ($P<0.01$) for T2 and T3 rabbits compared to control. Rabbits of T1, T2 and T3 groups had lower ($P<0.01$) concentrations of thyroid hormone (T_3) than control. Plasma Al concentrations increased ($P<0.01$) for T1, T2 and T3 rabbits compared to control. Al accumulation increased ($P<0.01$) in muscles and livers of T2

and T3 rabbits compared to control while it was highly accumulated ($P<0.01$) in kidneys and brains of the treated groups. This study indicated that adding Al, especially at 2000 and 3000 ppm, adversely reduced growth performance and metabolic activities of male rabbits as a result of the negative effect of Al on their physiological reactions as shown from the obtained results.

Key Words : *Aluminum, Growth performance, Physiological reactions, Male rabbits.*

INTRODUCTION

Aluminum (Al) is an ubiquitous element, being the most prevalent metal on the earth's crust (Atienzar *et al.*, 1998). It is usually found in some drinking water, in several pharmacological preparations and in many processed animal diets (Chinoy and Patel 2001), however, it is normally found at very low levels in animal feeds (Hussein *et al.*, 1989). Al does not appear to have any role in biological function in the animal body but it could have toxic effects particularly on the brain and kidneys when present in high levels in the blood (Bishop, 1997). High levels of Al have been shown to adversely affect growth performance of male rats (Batanineh *et al.*, 1998). This negative effect of Al administration could be mediated by depressing feed intake and feed efficiency (Guoo and Ching, 1994), reducing the availability of dietary phosphorus (Hussein *et al.*, 1989), and impairment of energy metabolism (Yashino *et al.*, 1998). Indeed, Al is one of the potent substances that increase free radicals in the body and produce its damaging effects through its pro-oxidant action (Katyal *et al.*, 1997).

Recently, increased attention is being paid to Al due to its serious effects on the central nervous system, energy metabolism and blood haematology (Graves *et al.*, 1990 and WHO, 1997). In this respect, blood haematological parameters such as haemoglobin, hematocrit, erythrocytes count and plasma iron concentrations were reported to be decreased in Al-treated rats (185 mg Al/kg) as observed by Guo-ChiHung *et al* (2000). Also, some immune functions were stimulated in terms of increased number of spleenocytes and thymocytes in rats received oral treatment of Al at 500 mg/litre (Glynn *et al.*, 1999). Some works showed an increase in serum Al due to Al administration in male rats received AlCl3 at 2 % for 4 weeks (Brown and Schwartz, 1992). On the other hand, accumulation of Al in the body has been observed to induce metabolic disorders related to bone, blood and brain (Alfrey,

1989). An elevation in Al accumulation was noticed in tissues of brain and kidney in rats received Al citrate ingestion at 7 mg/100 g of BW (Rahnema and Jennings, 1999).

Information considering the negative effects of Al supplementation on growing male rabbits and related physiological reactions are limited. The present study, therefore, was undertaken to investigate the adverse effects of three added levels of Al at 1000, 2000 and 3000 ppm in diets on growth performance, some haematological and immunological indices, some plasma biochemical constituents and residual accumulation of Al in some organ tissues of male rabbits.

MATERIALS and METHODS

The present study carried out at the farm of Animal Production Department, Faculty of Agriculture, El-Minia University. Forty eight of New Zealand White (NZW) weanling male rabbits of 5 weeks old and averaged 488 g body weight were used in this experiment until 15 weeks of age during the months from January to March. The animals were clinically healthy and proved to be free from internal and external parasites. They were randomly divided into 4 groups (12 males in each) in which the first group fed on the control diet, while the second, third and fourth groups fed on the control diet mixed with Al sulfate ($Al_2(SO_4)_3 16H_2O$) to perform three added levels Al, 1000 (T1), 2000 (T2) and 3000 (T3) ppm. Animals were fed *ad libitum* on pelleted commercial control diet containing alfalfa hay 37.5%, barley cereals 16 %, yellow corn 20 %, wheat bran 15 %, soybean meal 10 %, sugar beet molasses 1 %, calcium carbonate 0.15 %, sodium chloride 0.1 % and minerals plus vitamins mixture 0.25 %. The chemical analysis of the diet was 17.84 % crude protein, 2.37 % crude fat, 13.87 % crude fiber and 9.64 % ash. Animals were individually housed in galvanized wire cages equipped with feeders and nipple drinkers.

The data of body weights were recorded for male rabbits in each experimental group at 5, 7, 9, 11, 13 and 15 weeks of age. At the periods of 5-7, 7-9, 9-11, 11-13 and 13-15 weeks, feed intakes were recorded and daily weight gains and feed conversion efficiency were calculated. At 15 weeks of age, five males from each group were weighed and slaughtered after approximately 12 hours of fasting. Immediately after slaughtering, heparinized blood samples (5 ml) were collected from each rabbit. Whole blood samples were analyzed shortly after collection for blood hemoglobin (Hb, g/dl), packed cell volume (PCV, %), red blood

cell (RBCs, $x10^6/mm^3$) and total leucocytes count ($x10^3/mm^3$). Stained blood smears with Lieshman's stain were performed for the differential leucocytic cell counts (Dacie and Lewis, 1991). Plasma samples were obtained and stored at -20°C until assayed for biochemical analysis.

Five males from each experimental group were randomly taken for tissue Al analysis. Tissue samples were collected from muscles, livers, kidneys and brains and stored at -20°C until analysis. Plasma triiodothyronine (T_3) and thyroxine (T_4) concentrations were determined by a direct solid-phase I^{125} radioimmunoassay techniques using coat-A-count TKT_3 and TKT_4 RIA kits purchased from diagnostic products corporation (DPC, LA, CA, 90045-5597, USA). T_4/T_3 ratio was calculated. Plasma total protein, albumin, total lipids, cholesterol and glucose were measured spectrophotometrically using standard test kits supplied from Bio-Merieux Marcy-1, Etolie Charbonnieres- Les Bains, France and Bio-Analytics kits (USA). Globulin calculated mathematically by subtracting the difference between total protein and albumin. Concentrations of Al in tissues and plasma (ppm) were determined using flame atomic absorption spectrometry (Model Varian Spectr AA220) at wave length 309.3 and slit width 0.5 under flame working conditions: Lamp current: 10 mA, Fuel: acetylene, Support: Nitrous oxide).

The obtained data were analyzed by least square means analysis of variance using General Linear Models (GLM) procedure of the statistical analysis system (SAS, 1992). The model used to analyze the different traits studied for rabbits was as follows:

$Y_{ij} = \mu + T_i + e_{ij}$. Where : Y_{ij}= ij^{th} Observation, μ = Population mean; T_i = Effect of i^{th} treatments and eij= Random error. Duncan's Multiple Range test was used to detect differences between means of the experimental groups (Duncan, 1955).

RESULTS

The results showed significant ($P<0.001$) reductions in final body weights (FBW) at 15 weeks of age by 12.9, 23 and 28 %, and in averages of daily weight gain (DWG) from 5 to 15 weeks by 16.4, 28.9 and 36.6 % for the experimental rabbits fed T1, T2 and T3 of Al-supplemented diets respectively when compared to control (Table 1). Averages of feed intake (FI) from 5 to 15 weeks were decreased ($P<0.05$) by 8.8% for rabbits of T3 when compared to control with insignificant differences in FI due to T1 or T2 treatments. T1, T2 and T3

male rabbits showed lower (P<0.001) feed conversion efficiency by 19.4, 40.5 and 46.8% respectively than control.

Blood Hb concentrations decreased (P<0.01) in a dose-dependent manner for T2 and T3 rabbits than control with significant (P<0.01) differences between T2 and T3 rabbits (Table 2). This decrease in Hb values due to Al amounted by 17.2 and 22.2 % respectively for T2 and T3 groups when compared to control. Data showed significant (P<0.01) decreases in total RBCs count per mm^3 blood by 20.4 and 20.7 %, and in PCV % by 20.0 and 21.0 % respectively for T2 and T3 rabbit groups compared to control. Insignificant differences in RBCs and PCV values were observed between T2 and T3 rabbits. Total count of leucocytes increased (P<0.01) by 17.0 and 29.8 % respectively for T2 and T3 groups when compared to control with insignificant differences between T1 and control or between T2 and T3 ones (Table 2). Rabbits of T2 and T3 had lower (P<0.01) lymphocytes % and higher (P<0.01) neutrophils and eosinophils % than control. Meanwhile, monocyte and basophils % did not change due to Al treatments.

Plasma total protein and globulin concentrations decreased (P<0.01) respectively by 21.9 % and 22.8 % for rabbits of T3 when compared to the control (Table 3). Plasma glucose decreased (P<0.01) for Al-treated rabbits when compared to control with inignificant differences among the three treated groups. Plasma cholesterol increased (P<0.01) by 17.5 and 26.3 % for T2 and T3 rabbits when compared to control. Rabbits of T1, T2 and T3 groups had the lower (P<0.01) concentrations of thyroid hormone (T_3) by 19.6, 22.2 and 34.2 % respectively than control. Concentration of T_3 hormone was lower (P<0.01) for T3 group than T1 and T2. The T_4/T_3 ratio increased (P<0.05) for T3 when compared to control. Plasma Al concentrations increased (P<0.01) by 66.4, 84.2 and 91.1 % for T1, T2 and T3 rabbits respectively when compared to control. Al levels had no effect on plasma Al concentrations. Insignificant changes were observed in plasma concentrations of albumin, total lipids and thyroxin (T_4) due to Al treatments.

Table 4 represents the Al concentrations in tissues of muscles, livers, kidneys and brains in Al-treated rabbits vs control. Al concentrations increased (P<0.01) by 63.4 and 145.6 % in muscles, and by 161.2 and 310.8 % in livers of T2 and T3 rabbit groups respectively compared to control, while the Al was highly accumulated (P<0.01) in kidney (83.7, 148.9 and 253.4 %) and in brain (40.1, 61.3 and 111.1 %) of the three treated groups respectively. Al concentrations in livers and

kidneys increased ($P<0.01$) as the level of additive Al increased. In case of muscles and brains, Al concentrations were higher ($P<0.01$) for T3 than T1 and T2 rabbits while there were insignificant differences between T1 and T2 rabbits.

DISCUSSION

The present study showed a marked depression in growth performance of Al-fed male rabbits during growing period (5 to 15 weeks) indicating an adverse effect of added Al in diets at 1000, 2000 and 3000 ppm on growth traits in terms of FBW, DWG and FC, however, reduced FI observed only at 3000 ppm of Al. These findings are coincided with some observations that oral Al administration at 2835 ppm in male rats for 8 days led to a reduction in FI and averages of BW (Ondreicka *et al.*, 1996). This negative effect of adding Al was observed in male rats ingested solution of Al chloride along with drinking water at 1000 ppm for 12 weeks (Bataineh *et al.*, 1998) in which those animals exhibited a reduction in their averages of FI and BW due to Al administration.

It was clear that adding Al in diet at levels 2000 and 3000 ppm decreased blood Hb, PCV and RBCs, and this decrease was dose-dependent in blood Hb, however, this negative effect of Al on hematopoiesis was insignificant at 1000 ppm of Al. This trend of decreased blood PCV values which was noticed in broiler chicks fed Al at 500, 1000 and 1500 ppm (Zein El-Dein *et al.*, 1999). Although evidence indicated that Al inhibited haemoglobin synthesis (Abreo *et al.*, 1989), the step in which Al interfered in the haemoglobin synthesis still unclear. Fulton and Jeffery (1994) noticed blood Hb and PCV to be decreased with smaller and more irregular erythrocytes in rats given Al at 50 mg/kg BW. In the same respect, these haematological indices were investigated in mice received oral Al administration at 74 or 185 mg Al/kg BW/day in relation to the formation of nitric oxide which may play an important role in the underlying causes of the anemia as reported by Guo-Chih Hung *et al.* (2000) who noticed that 185 mg Al/kg BW induced a marked increase in plasma nitric oxide and plasma Al concentrations accompanied with a decline in blood Hb, PCV and RBCs counts as well as lowered levels of plasma Fe concentrations, indicating that Al-intoxication causes an increase in nitric acid production in the plasma, which may be associated with the haematopoiesis system disturbance inducing anemia. Another possible mechanism for the

microcytic, hypochromic anemia associated with Al overload is that increased haem oxygenase activity with subsequent increased destruction of haem and/or haem protein that observed in rats received 50 mg/kg of Al lactate (Fulton and Jeffery, 1994). Taken together, the adverse effect of Al (at 2000 and 3000 ppm) on blood haematopoiesis that observed in the present study and others may reflect the trend towards depression of growth performance in high Al-treated animals.

Rabbits fed Al at 2000 and 3000 ppm exhibited higher counts of total leucocytes accompanied with a marked decrease in lymphocytes % and an increase in neutriphils %. These results partially agreed with earlier report by Abd El-Nasser *et al.* (1994) showing degenerative changes in the liver including depletion of lymphocytic elements in rabbits given oral Al chloride administration. Alterations in total leucocytes count and its differential cells for rabbits treated with Al may signify a change in their immune status reflecting their adaptability to the adverse effects of Al administration. In this regard, some immune functions were stimulated in terms of increased number of spleenocytes and thymocytes in rats received oral treatment of Al at 500 mg/litre (Glynn *et al.*, 1999).

Results of plasma assay indicated some changes in its biochemical constituents, where plasma total protein (TP) concentrations showed a decrease by 21.9 % in rabbits fed 3000 ppm Al-supplemented diet. In broiler chicks, adding Al at 1000 and 1500 ppm in diet induced decreases in plasma TP by 17.5 and 23.9 % respectively (Zein El-Dein *et al.*, 1999). This change in plasma TP, in the present study, may be associated with the decrease in plasma globulin (Glob) by 22.8 % due to Al treatment (3000 ppm). This negative effect of high Al on plasma TP and Glob in male rabbits could be mediated by a decline in protein synthesis and its secretion via altered enzyme activities as observed in mice treated with AlCl3 at 200 mg/kg BW (Chinoy and Patel 2001). The decrease in plasma glucose concentrations for Al-fed rabbits could be explained by the observation that high Al exposure affects carbohydrate metabolism (Underwood, 1977). This could be correlated with a decline in activity of phosphorylase which found in other tissues in male mice (Chinoy and Patel, 1996). This finding may explain the adverse effect of high Al administration on plasma glucose concentrations. In the present study, Al treatments did not affect plasma total lipids, but they induced increases in plasma cholesterol concentrations by 17.5 and 26.3 % in rabbits fed Al levels at 2000 and 3000 ppm respectively. In this regard, Szilagyi *et al.* (1995) observed that adding Al to the diets at 200, 1000

and 3000 mg/kg increased serum cholesterol in a dose-dependent manner, but decreased serum triglycerides. Also, 2000 mg Al /kg diet in broiler chicks had no effect on their plasma total lipids concentrations, but reduced their plasma triglycerides (Zohouri *et al.*, 1998). The significant increases in plasma cholesterol concentrations for Al-fed rabbits are in harmony with previous study by Chinoy and Patel (2001) who showed an increase in serum cholesterol in mice treated with AlCl3 at 200 mg/kg BW. This could be explained that the hypercholesterolmic effect of Al treatment may be related to the significant decline noticed in 3β- and 17β-hydroxysteroid dehydrogenase activities, suggesting altered steroidogenesis in Al-treated animals (Chinoy and Patel, 2001). The results showed that plasma T_3 hormone concentrations were decreased for rabbits received diets supplemented with Al at 1000, 2000 and 3000 ppm accompanied with no change in thyroxine (T_4) levels. Also, the increase in T_4/T_3 ratio with Al at 3000 ppm may reflect a decline in conversion of T_4 to T_3 in Al-treated rabbits. The present results pointed out that supplemental Al appears to provide for additional increases in plasma Al concentrations, where they were elevated by 66.4, 84.2 and 91.1 % for rabbits fed 1000, 2000 and 3000 ppm of Al respectively. These results reinforce some studies showed an increase in plasma Al concentrations due to Al administration as observed in male rats received Al in drinking water at 500 mg/liter for 7-9 weeks (Glynn *et al.*, 1999).

It is likely that the significant negative response in plasma factors of male rabbits fed Al-supplemented diets may signify a case of their inactive metabolism, since plasma metabolite constituents represent a buffering state for metabolic synthesis and catabolism end products (Swenson, 1984). This interpretation may indicate that high levels of Al ingestion might negatively influence bioavailability of nutrients that required for the efficient metabolic processes as previously explained in rats received 500 and 2000 mg Al/kg diet added in the basal diet reflecting the adverse effect of high levels of Al administration in reducing growth performance of Al-fed animals (Guoo and Ching, 1994).

The appreciable increases of Al concentrations accumulated in the tissues of examined organs (liver, kidney, muscle and brain) were almost dose-dependent changes, indicating a serious effect of Al toxicity on those tissues and their physiological functions. These results on rabbits are consistent with some studies working on different species. In broiler chicks, adding Al at levels 500, 1000 and 1500 ppm in diets

resulted in an increase in Al retention within muscles (Zein El-Dein *et al.*, 1999). Also in lambs, high Al concentrations accumulated in liver and tended to increase in kidney and muscle due to adding Al at 2000 ppm to the diet (Valdivia *et al.*, 1982). In case of mice, increases in Al accumulation observed in liver tissue of male rats given 2 % Al chloride in diet (Brown and Schwarts, 1992); in liver and kidney of female rats injected with Al chloride at 10 mg/kg daily (Muller *et al.*, 1992); or given gavages at 200 and 400 mg/kg/day on gestational days 1-20 (Belles *et al.*, 2001). In tissues of brain and kidney, an increase in Al concentrations were noticed in rats received Al citrate ingestion at 7 mg/100 g of BW as reported by Rahnema and Jennings (1999). An elevation of accumulated Al in brain tissues has been reported in studies on rat received Al in drinking water at 50 or 100 mg/kg BW (Domingo *et al.*, 1996). However, other works showed insignificant change in Al concentrations in brain tissues of male lambs received 2000 ppm of Al (Valdivia *et al.*, 1982). Although it is generally believed that orally administered Al compounds are absorbed in low amounts by the body (0.1-1 %), there are many factors can enhance Al absorption in animal and presumably its accumulation in different tissues. For example, dietary compounds such as minerals can affect Al absorption and its accumulation in tissues (Deng *et al.*, 1998). Also, oral Al administration can produce significant changes in the distribution of various essential elements (Belles *et al.*, 2001). Tissue concentrations of Al can be altered by moderate changes in the diet and kidney function, since a reduction about 30 % in kidney function increased Al retention in tissues of rats received Al either by injection or oral administration as reported by Ecellbarger and Greger (1991). However, although the significant accumulation of Al in tissues has already been reported in earlier works together with the present study, the full mechanisms have not yet elucidated.

In conclusion, the current study showed that adding Al to the diets adversely reduced growth performance of male rabbits as a result of the negative effect of adding Al on their physiological reactions. The negative physiological reactions of male rabbits to Al administration were evidently pronounced at 2000 and 3000 ppm of added Al, but mostly not at 1000 ppm. So, from the economical view, high Al toxicity in rabbit rations should be taken into account to avoid its negative effects on animal physiological and productive performances.

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Table 1: Effect of additive Al on growth performance of male rabbits (means ± SEM).

	Treatments					
Parameters :	Cont	T1	T2	T3	SEM	Sig.
IBW (g)	490.0	481.3	471.3	495.0	19.3	NS
FBW (g)	2114 a	1841 b	1627 c	1521 c	41.9	***
DWG (g)	23.2 a	19.4 b	16.5 c	14.7 d	0.60	***
FI (g/ day)	72.9 a	72.5 a	72.8 a	66.5 b	1.6	*
FC (g feed/g gain)	3.14 c	3.75 b	4.41 a	4.61 a	0.18	***

a,b,c,d means within the same row having different superscripts significantly different.
* (P<0.05), *** (P<0.001), NS= not significant.
IBM = Initial body weight, FBW = Final body weight, DWG = Daily weight gain,
FI = Feed intake, FC = Feed conversion.

Table 2: Effect of additive Al on haematological parameters and total leucocytes counts and its differential cell count of male rabbits (means ± SEM).

	Treatments					
Parameters :	Cont	T1	T2	T3	SEM	Sig.
Hb (g/dl)	13.88 a	13.50 a	11.50 b	10.80 c	0.20	**
RBCs ($x10^6/mm^3$)	7.06 a	6.94 a	5.62 b	5.60 b	0.08	**
PCV (%)	39.0 a	37.3 a	31.2 b	30.8 b	0.60	**
Total leucocytes ($x10^3/mm^3$)	6.24 b	6.28 b	7.30 a	8.10 a	0.25	**
Lymphocytes (%)	55.2 a	53.6 a	43.8 b	40.0 c	1.14	**
Monocytes (%)	4.2	4.4	4.8	4.6	0.31	NS
Neutrophils (%)	37.0 c	38.0 c	46.4 b	49.6 a	1.00	*
Eosinophils (%)	2.6 c	3.0 c	4.0 b	4.8 a	0.35	NS
Basophils (%)	1.0	1.0	1.0	1.0	0.00	NS

a,b,c means within the same row having different superscripts significantly different (P<0.01).
* (P<0.05), ** (P<0.01), NS= not significant.
Hb=Hemoglobin, RBCs= Red blood cells, PCV=Packed cell volume.

Table 3: Effect of additive Al on plasma metabolites and hormones of male rabbits (means ± SEM).

	Treatments					
Parameters	Cont	T1	T2	T3	SEM	Sig.
Total protein (g/dl)	7.60 a	6.38 ab	6.11 ab	5.93 b	0.43	*
Albumin (g/dl)	4.24	3.56	3.33	3.32	0.24	NS
Globulin (g/dl)	3.37 a	2.82 ab	2.78 ab	2.60 b	0.18	*
Glucose (mg/dl)	105.5 a	88.8 b	89.0 b	83.8 b	2.75	**
Total lipids (mg/dl)	291.3	288.8	285.0	281.3	5.16	NS
Cholesterol (mg/dl)	100.0 b	102.3 b	117.5 a	126.3 a	4.80	**
T_3 (ng/ml)	1.58 a	1.27 b	1.23 b	1.04 c	0.07	**
T_4 (ng/ml)	52.38	50.93	51.73	50.60	3.35	NS
T_4/T_3 ratio	33.08 b	40.14 ab	42.15 ab	49.96 a	2.74	*
Al (ppm)	6.07 b	10.10 a	11.18 a	11.60 a	0.88	**

^{a,b} means within the same row having different superscripts significantly different (P<0.01).
* (P<0.05), ** (P<0.01), NS= not significant.

Table 4: Effect of additive Al of Al accumulation in tissues of some organs in male rabbits (means ± SEM).

	Treatments				
Organs :	Cont.	T1	T2	T3	SEM
Muscle	79.7 c	106.8 bc	130.2 b	195.7 a	8.1
Liver	69.0 c	98.61 c	180.2 b	283.5 a	12.8
Kidney	52.8 d	97.0 c	131.4 b	186.6 a	8.9
Brain	79.1 c	110.9 b	127.6 b	167.3 a	6.5

^{a,b,c,d} means within the same row having different superscripts significantly different (P<0.01).