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VI Borekar

Ph.D. Scholar, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

AP Somkuwar

Professor & Head, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

AR Sawarkar

Assistant Professor, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

RP Limsay

Assistant Professor, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

SA Dubey

Assistant Professor, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

SA Umap

Assistant Professor, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

Corresponding Author:**VI Borekar**

Ph.D. Scholar, Department of Veterinary Pharmacology & Toxicology, Nagpur Veterinary College, Seminary Hills, Nagpur, Maharashtra, India

Effect on haematology by *Zingiber officinale* on lead induced toxicity in broilers

VI Borekar, AP Somkuwar, AR Sawarkar, RP Limsay, SA Dubey and SA Umap

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Abstract

The present research investigation was carried out to study effect of 50% ethanolic rhizome extract of *Z. officinale* on physical parameters on Lead induced toxicity in broiler birds. 140 broiler chicks were divided into 7 groups each comprising of 20 birds. The toxicity was induced by administration of lead acetate @ 200 mg/kg feed from T₂ to T₇ and T₁ as normal control. T₃ & T₄ was given AA @ 200 mg/kg feed & Ext. @ 200 mg/kg b.w, respectively. T₅ was given AA @ 200 mg/kg feed & Ext. @ 200 mg/kg b.w. T₆ was given Ext. @ 300 mg/kg b.w. b.w, respectively. At the end of experiment, it was observed that, *Zingiber officinale* rhizomes extract @ 200 and 300 mg/kg bw alone (T₄ and T₆) and with L-ascorbic acid (T₅ and T₇) has restored the normal values of RBC, WBC, Haemoglobin concentration and PCV% which illustrated the protective effects of *Z. officinale* against lead toxicity induced in broiler birds.

Keywords: Medicinal plants, *Zingiber officinale*, haematology, lead toxicity, broiler birds

1. Introduction

Lead (Pb) exposure is a major public health problem; therefore it has been paid attention by researchers in probing further into its toxicity. Lead is used extensively in building materials, pigments, to glaze ceramics, water pipes and glass, paints, dyes, artificial jewelry, cosmetics, protective coatings, acid storage batteries and also as gasoline additives. So it is considered to be one of the major environmental pollutants and has been incriminated as a cause of accidental poisoning in domestic animals and birds more than any other substances (NRC, 1984) [1]. In poultry, lead (Pb) produces acute and chronic poisoning and induces a broad range of physiological, biochemical and behavioral dysfunctions. The main source of metals in chickens arises from contaminated poultry feeds and water. In view of the fact that poultry feed has been reported to be affected due to the use of heavy metals contained feed additives in poultry feed production system (Islam *et al.*, 2007) [2].

Lead blood level measurement is one of the most important indicators to lead poisoning (Tietz, 1999) [3]. Commonly, lead poisoning cause anaemia (both hemolytic and hemorrhagic), the mechanism of anaemia caused by lead briefly occurred by two ways: first, a shortened erythrocyte lifespan and impairment of heme synthesis, lead causes increased concentration of protoporphyrin by inhibiting heme synthetase, the enzyme which combine protoporphyrin and iron to form heme. Second, lead causes inhibition of the enzyme δ -aminolevulinic acid dehydratase (ALA-D) resulting in a failure of utilization of δ -aminolevulinic acid which is excreted in increased quantities in urine (Blood and Rodostits, 1989; Murray *et al.*, 2006) [4, 5].

Ginger, the rhizomes of the plant *Zingiber officinale* (Family *Zingiberaceae*), is arguably one of the most widely used culinary agent and spice in the world (Baliga *et al.*, 2012) [6]. Phytochemical studies have shown that the unique culinary and medicinal properties of ginger are due to the presence of phytochemicals like zingerone, shogaols, gingerols, pardols, β -phellandrene, curcumene, cineole, geranyl acetate, terphineol, terpenes, borneol, geraniol, limonene, β -elemene, zingiberol, linalool, α -zingiberene, β -sesquiphellandrene, β -bisabolene, zingiberenol and α -farnesene (Palatty *et al.*, 2013) [7]. Therefore, the present research work was planned to investigate the protective activity of *Zingiber officinale* along with L-ascorbic acid against lead acetate induced toxicity in broiler birds with reference haematological parameters.

2. Materials and Methods

2.1 Preparation of 50% Ethanol cold extract

The plant material i.e. roots of plant *Zingiber officinale* was collected from the local market of Nagpur and were dried at room temperature. 500 gram of powder of dried rhizome of *Zingiber officinale* was mixed with 2000 ml of 50% Ethanol in Stoppard flask and allowed to stand at room temperature for 72 hrs with frequent agitation until the soluble matter get dissolved. After 72 hrs, the mixture was filtered through muslin cloth, so as to remove the insoluble material. The filtrates were again filtered through filter paper and then poured in clean and already weighed petri plate and allowed for complete evaporation at room temperature and finally stored in desiccators in cool and dry place.

2.2 Experimental animals

The Institutional Animal Ethics Committee (IAEC) (CPCSEA Reg. No. 244/GO/ReBi/S/2000/CPCSEA Dated 01.08.2000) approved the experimental protocol. The experimental protocol met the national guidelines as per the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India. The present research work was carried out 140 Broiler chicks were purchased from M/s. Atharvaraj Hatcheries, Nagpur. Broiler chicks were kept under standard managerial conditions as per the norms of BIS. All the birds were maintained on deep litter system at Poultry Farm, Department of Poultry Sciences, Nagpur Veterinary College, Nagpur and were fed on commercial diet and provided free access of drinking water during experimental period. On arrival, all chicks were weighed and randomly distributed in to seven treatment groups viz, T₁, T₂, T₃, T₄, T₅, T₆, and T₇ of 20 chicks in each group. Brooding with respective treatment group was carried out with electric hover brooders and continued until 14th day of age. All the Broiler chicks were fed on commercial feed purchased from M/s Supreme Agrovet Industries, Nagpur and provided free access of drinking water during experiment.

2.3 Experimental protocol

Broiler chicks (140) were divided into 7 groups T₁, T₂, T₃, T₄, T₅, T₆, and T₇ each group comprising of 20 birds. The lead

toxicity was induced by administration of lead acetate @ 200 mg/kg (Ibitoye *et al.*, 2011)^[8] of feed for 30 days. Group T₁ served as normal control, Group T₂ served as lead acetate control. Group T₃ and T₄ were received L-Ascorbic acid @ 200 mg/kg feed and plant extract @ 200 mg/kg b.w along with Lead acetate, respectively. Group T₅ was received L-Ascorbic acid @ 200 mg/kg feed and plant extract @ 200 mg/kg b.w along with Lead acetate. Group T₆ was received plant extract @ 300 mg/kg b.w along with Lead acetate. Group T₇ was received L-Ascorbic acid @ 200 mg/kg feed and plant extract @ 300 mg/kg b.w along with Lead acetate. The experiment was conducted for period of thirty days.

After 30th day of experiment, prior to sacrifice, blood samples were collected aseptically from wing vein in glass vials containing 1% Ethylene Diamine Tetra Acetic Acid (EDTA) for hematological estimation. Hematological studies such as hemoglobin concentration (Hb), using Sahli's Method (acid hematin), total RBC count, total WBC count and Packed Cell Volume (PCV) by micro tube method as mentioned by Benjamin (1985)^[9].

3. Results

From the results showed in table 1, it was observed that the extract @ 200 and 300 mg/kg bw alone (T₄ and T₆) and with L-ascorbic acid (T₅ and T₇) has significantly restored the altered values of RBC, WBC, haemoglobin concentration and PCV% in lead intoxicated broiler birds. Extract @ 200 and 300 mg/kg bw (T₄ & T₆) alone and with L-ascorbic acid (T₅ & T₇) showed significant increased RBC values toward normal (T₁) when compared to lead control group (T₂). Group T₄, treated with extract@ 200 mg/kg bw showed lowest increase in WBC as compared to other treatments groups, however it was also significantly higher than that of lead control group. Extract @ 200 & 300 mg/kg bw with L- ascorbic acid (T₅ and T₇) showed more significant increase in haemoglobin values towards the normal than extract @ 200 & 300 mg/kg bw alone (T₄ and T₆) when compared with lead treated control group (T₂). Similarly, extract @ 200 & 300 mg/kg bw with L-ascorbic acid (T₅ and T₇) found significant increase in PCV% towards the normal than extract @ 200 & 300 mg/kg bw alone (T₄ and T₆) when compared with lead control group (T₂).

Table 1: Effect of *Zingiber officinale* rhizomes extract on haematology on lead induced toxicity in broiler

Hematological Parameters	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇
	Control Group	Lead acetate @ 200 mg/kg feed	Lead acetate @ 200 mg/kg feed + AA @ 200 mg/kg feed	Lead acetate @ 200 mg/kg feed + Ext. @ 200 mg/kg b.w.	Lead acetate @ 200 mg/kg feed + Ext. @ 200 mg/kg feed	Lead acetate @ 200 mg/kg feed + Ext. @ 300 mg/kg b.w.	Lead acetate @ 200 mg/kg feed + Ext. @ 300 mg/kg b.w. + AA @ 200 mg/kg feed
RBC (x 10 ⁶ Cells/dl)	2.62±0.03 ^{ab}	2.16±0.09 ^c	2.79±0.07 ^a	2.39±0.07 ^b	2.44±0.08 ^b	2.46±0.09 ^b	2.50±0.12 ^b
WBC (x 10 ³ Cells/dl)	25.26±1.48 ^{ab}	18.26±0.49 ^d	27.42±0.89 ^a	21.90±0.79 ^c	25.21±1.63 ^{ab}	23.71±0.40 ^{bc}	26.59±0.53 ^a
Hemoglobin (g/dl)	11.20±0.47 ^a	8.06±0.42 ^d	10.69±0.58 ^{ab}	9.36±0.39 ^c	10.89±0.36 ^{ab}	9.94±0.38 ^{bc}	10.82±0.31 ^{ab}
PCV%	31.61±1.21 ^a	22.19±1.12 ^d	30.07±1.46 ^{ab}	26.08±0.95 ^c	30.68±1.11 ^{ab}	27.85±0.96 ^{bc}	30.46±0.74 ^{ab}

Values are mean S.E. for 20 birds in each group. Values not sharing a common superscript in a column differ significantly (P<0.05)

4. Discussions

In lead toxicity, haematological parameters are considered as an indication of its intoxication to produced anaemic condition, which results from inhibition of the heme synthesizing enzymes with concurrent elevation of protoporphyrin (Lee, 1981)^[10]. In present study, reductions in the total WBC may be due to the aplastic anaemia due to increased blood poisoning by lead and also infiltration of bone marrow by lead (Ibitoye *et al.*, 2011)^[8] where lead may

affect on blood cell precursors. Decreased haemoglobin concentration and PCV% in lead treatment groups when compared with normal control may be due to the deleterious effect of lead on blood causing anaemia by shortening the lifespan of RBC and impairment of heme synthesis (Jain, 1986)^[11]. Lead also increases concentration of protoporphyrin by inhibiting heme synthetase, the enzyme which combined protoporphyrin and iron to form heme. Second, lead causes inhibition of the enzyme δ -aminolevulinic

acid dehydratase (ALA-D) resulting in a failure of utilization of δ -aminolevulinic acid which is excreted in increased quantities in urine (Blood and Rodostits, 1989; Murray *et al.*, 2006) ^[4,5] and lead reduces the process of haemosynthesis through inhibition of such enzymes (Osweiler, 1996) ^[12].

In accordance with the present findings, Khan *et al.* (2008) ^[13] reported that following lead acetate administration, there was decrease in haemoglobin and PCV. Correspondingly, Szymezak *et al.* (1983) ^[14] also observed the hemoglobin level reduction. Kamruzzaman (2006) ^[15] reported that lead administration significantly decreased RBC, WBC and haemoglobin content in rats. McDonald (1988) ^[16] reported that a hypo chromic, regenerative anaemia was observed in some affected birds with lead. Marques *et al.* (2006) ^[17], who also reported that significant decrease in RBC, haematocrit in lead exposed mice. Gurer *et al.* (1998) ^[18] also described hypochromic anaemia due to lead toxicity.

In the present finding, extract of *Zingiber officinale* rhizomes @ 200 and 300 mg/kg bw alone and with L-ascorbic acid has significantly increased RBC, WBC, Haemoglobin concentration and PCV% which illustrated the protective effects of *Z. officinale* extract and ascorbic acid on dietary lead exposure as it affects on the haematology of the birds. Studies regarding the protective effect of *Z. officinale* on hematology in metal toxicity are not much mentioned. The improvement in haematological parameters by ethanolic extract of *Z. officinale* which were declined by lead toxicity may be due its antioxidant, radical scavenging, hepato & renal protective properties. However, the findings of present work are mostly in agreement with previous investigation as, Duke and Ajensu (1985) ^[19], Famurewa *et al.*, (2011) ^[20] and Jakes and Susan (2007) ^[21] who reported the use of ginger in condition like rheumatism and bleeding rashes. The mineral element like iron present in the ginger are may facilitates the formation of blood cells and reducing deficiency of causing anemia (Umoh *et al.*, 2014) ^[22]. Duke and Ajensu (1985) ^[19] also recommended use of decocted rhizome of ginger in treating pulmonary haemorrhage and haemostasis. Ascorbic acid (AA) is a well-known antioxidant vitamin involved in several biochemical processes in biological systems. Vij *et al.*, (1998) ^[23] also reported significant protective effect of Ascorbic acid against toxic effect of lead on heme synthesis and drug metabolism.

4. Conclusion

Form this investigation it can be concluded that, extract of *Zingiber officinale* rhizomes @ 200 and 300 mg/kg body weight alone and with L-ascorbic acid has possesses remarkable protective effects against lead induced toxicity in broiler birds by restoring the normal value of the haematological parameters.

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