



Original Research Article

Effect of Izol contaminated feed on the heamatological profile of albino rats

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A B S T R A C T

Keywords

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Izol is a disinfectant and the active ingredient is cresol. The effect of its oral administration, in varying concentrations, on the hematological profile and body weight of adolescent male albino rats were investigated for 28 days. Twenty-four rats weighing between 55 and 145g were randomly separated into four groups of six rats each and fed water *ad libitum*. Groups 1, 2, and 3 rats received 1 ml (0.8%), 2 ml (1.7%), and 3 ml (2.5%) of Izol per 123.12 g of feed, respectively. Group 4 served as a control and was not given Izol; rather, 123.12 g of their feed was mixed with 119 ml of water. Blood for haematological analyses were collected into EDTA bottles from the ocular region of the eyes using heparinised capillary tube. The parameters assayed included percentage packed cell volume % (PCV), erythrocyte sedimentation rate (ESR), haemoglobin (Hb) concentration, total white blood cell (WBC) count, red blood cell (RBC) count, platelet count (PC), and differential WBC count. The mean cell haemoglobin concentration (MCHC), mean cell haemoglobin (MCH), and mean cell volume (MCV) were also calculated. PCV increased significantly ($P < 0.05$) for groups 2 and 3 rats given 2 ml (1.7%) and 3 ml (2.5%) of Izol respectively, compared with control. RBC count decreased significantly ($P < 0.05$) for group I rats given 1 ml (0.8%) of Izol and increased significantly ($P < 0.05$) for group 3 given 3 ml (2.5%) of Izol. MCV increased significantly ($P < 0.05$) for all the groups compared with control. While lymphocytes increased significantly ($p < 0.05$) for Group 3 rats given 3 ml (2.5%) of Izol, neutrophil decreased significantly ($P < 0.05$) for the same group compared with control. Hb decreased significantly ($P < 0.05$), for groups 1 and 2 given 1 ml (0.8%) and 2 ml (1.7%) of Izol respectively, compared with control. MCHC and MCH recorded significant ($p < 0.05$) decreases for all groups compared with control. Platelet count and ESR showed no significant ($p > 0.05$) changes in all groups compared with control. The results indicated that short-term ingestion of low Izol concentration may, in addition to causing marked increase in weight, stimulate erythropoiesis and precipitate macrocytic hypochromic anaemia and may not evoke cytotoxicity in adolescent male albino rats.

Introduction

Disinfectants are germicides which are too corrosive or toxic to be applied to tissues, but which are suitable for application to inanimate objects (Roper, 1998).

Disinfectants are continually used to disinfect bathing water, hospital equipments (e.g. thermometers, stethoscopes, dialysis equipment,

hemodialysers, endoscopes, blood culture bottles, etc.). They are also used in cleaning wounds, laboratory benches, hospitals, wheel chairs, floors, walls, furnishings, lavatories; incorporated into formulations and soaps; in washing fabrics, etc (Bruce, 1998).

Cresol solutions are common disinfectants sold, perhaps most famously, under the trade names Izol, Lysol and Izal. Human exposure occurs through discharge of some quantities/volumes, by the manufacturers, into water bodies. In Nigeria similarly, some rural habitants use cresol solutions to disinfect their wells after the accumulated clay deposits have been removed and then allow the well for some days before the water can be drunk. Furthermore, human exposure may occur in facilities which manufacture, process or use cresols.

Approximately 126,000 — 300,000 individuals are exposed to cresols at workplace in the USA. The Largest sub-group is formed by mechanics (app. 148000) exposed to cresol-containing cleaning compounds — e.g. when cleaning automobile carburettors (US-EPA, 1986). Other sources of human exposure include absorption through the intact skin, through the food chain (e.g. eating contaminated fish or probably through herbicides), inhalation from exhaust of vehicles powered with petroleum-based fuels, etc. Cresols are organic compounds which are methylphenols. That is, they have a methyl group substituted onto the benzene ring of a phenol molecule. They are widely occurring natural and manufactured group of aromatic organic compounds which are categorized as phenols (sometimes called phenolics).

Depending on the temperature, cresols can be solid or liquid because they have

melting points not far from room temperature. Like other types of phenols, they are slowly oxidized by long exposure to air. They are colourless in pure form but could be yellowish, brownish-yellow or pinkish liquid when contaminated with impurities.

Cresol has three isomers, ortho-cresol (o-cresol), meta-cresol (m-cresol), and para-cresol (p-cresol). These may occur separately or as a mixture. The term tricresol is used to describe a mixture of o, m-and p.cresols (HSDB, 1995).

Cresols are used as antiseptics, disinfectants, and parasiticides in veterinary medicine. Cresols are also used in the manufacture of synthetic resins tricresyl phosphate, salicylaldehyde, coumarin, and herbicides. Cresol serve as components of degreasing compounds in textile scouring and paint brush cleaners as well as fumigants in photographic developers explosives.

Most exposures to cresols are at very low levels that are not harmful. When cresols are breathed, ingested or applied to the skin at high levels, they can be very harmful.

Breathing high level of cresols for a short time results in irritation of the nose and throats. Aside from these effects, very little is known about the effects of breathing cresols, for example, at lower level over longer times.

Ingesting high level results in kidney problems, mouth and throat abdominal pain, vomiting, and various effects on the blood and nervous system. Short-term and long-term studies with animals have similar effects from exposure to cresols. No human or animal studies have shown

harmful effects from cresols on the ability to have children. It is not known what the effects are at long-term ingestion or skin contact with low levels of cresols.

Consequent upon these, it is necessary to evaluate the possible haematological effects of cresol (IZOL) on humans using albino rat as animal model.

Materials and Methods

Experimental Animals

Twenty-four (24) adolescent male albino rats were used for the experiment. They were collected from a breeder in Nsukka, Enugu State of Nigeria. The rats were allowed to acclimatize for seven days before the experiment commenced. They were fed growers marsh water *ad libitum* in the course of acclimatization and experimentation.

Experimental Design

Twenty-four (24) adolescent male albino rats were randomly divided into four groups of six rats per group. The group consists of groups 1, 2, 3 and the control group. Each group was further subdivided into three replicate groups of two rats per cage. The rats in group 1, 2 and 3 received 1 ml (0.8%), 2 ml (1.7%), and 3 ml (2.5%) of Izol per 123.12 g of feed respectively. The fourth group, control, was not given Izol; their feed was only mixed with 119 ml of water per 123.12 g of feed. These treatments were administered to the rats on daily basis for 28 days. Their weights were determined before starting the treatment, which is after the 7 days of acclimatization, and on weekly basis in the course of the 2 days of treatment.

In the first fourteen days of the treatment, blood was collected from three rats in each of the groups, that is, one rat from each replicate. In the next fourteen days, blood was also collected from the remaining three rats in each of the groups. The blood was analyzed for the required hematological parameters.

Weight Determination

The weights of the rats were determined with a weighing balance. An empty dry container was weighed and the weight recorded. Then, the rats were put singly into the container and the weight recorded as well. The weight of each rat therefore is: Weight of container containing rat - weight of container.

Sample Collection

Using heparinised capillary tubes, blood samples were collected from the eyes. The blood was bleed directly into labeled EDTA sample bottles containing appropriate quantity of EDTA anticoagulant. The bottles were tightly stoppered and immediately rocked to properly mix the blood and the anticoagulant thereby preventing clotting.

Haematological studies

Differential Leukocyte Count

Thin films of each of the samples were made on a clean glass slide. This involved wiping the slide clean with tissue paper, placing a small drop of blood at the centre of the lower part of the slide, and spreading the blood forward with a spreader which touches the blood (allowing the blood to run along its edge) at an angle of 45° to the slide (Inyang *et al.*, 2006). The film was air-dried and

covered with *Leishman* stain. After 2 minutes, the stain was diluted with distilled water at the ratio of 1:2 and the slide was rocked gently to ensure proper mixing of the stain and water.

It was allowed to stand for 10 more minutes before washing off with distilled water. The back of the slide was cleaned with tissue paper and the slide was allowed to air-dry. A drop of oil immersion was placed on the stained film and covered with a cover slip. It was then observed under the microscope with 10x objectives. 100 leukocytes were counted and differentiated into neutrophils, eosinophils, basophils, lymphocytes, and monocytes.

Packed Cell Volume (PCV)

The microhaematocrit method was adopted (Baker and Silverton, 1985). About $\frac{3}{4}$ of a capillary tube was filled with the anticoagulated blood and one end of the tube sealed with flame from a candle light (Maurice, 1973). This was centrifuged at 10,000 rpm for 5 minutes in a microhaematocrit centrifuge. The capillary tube was removed from the centrifuge and the PCV read with the haematocrit chart (Maurice, 1973).

Erythrocyte Sedimentation Rate (ESR)

The Westergren's method was adopted. 1.6 ml of EDTA anticoagulated blood sample was added to, and properly mixed with 0.4 ml of trisodium citrate in a small container. With the aid of a pipette filler, the Westergren's pipette was filled to the '0 mark' with this sample. The pipette was fixed vertically to the Westergren's stand and the set-up was allowed to stand undisturbed for 1 hour. At exactly one hour, the level of the red cells (mm), ESR, was read.

Haemoglobin Concentration Determination

The haemoglobin concentration was determined using the cyanmethaemoglobin method through which the haemoglobin was read using digital photoelectric colorimeter.

0.02ml of blood was added into 5ml of modified Drabkin's solution. It was allowed to stand for 4-5mins, to ensure complete conversion of haemoglobin to cyanmethaemoglobin, before taking it to the colorimeter.

Total Leukocyte (WBC) Count

With the aid of a micropipette, I added 0.02ml of blood into a Khan's test tube containing 0.38ml of Turke's fluid and mixed thoroughly. Few drops of the diluted blood sample were loaded into a charged counting chamber using a Pasteur pipette. After 2 minutes, the counting chamber was taken to a microscope and the number of white blood cells was counted using 10x objective lens.

White blood cell count is carried out in the four large corner squares of the chamber. In addition, cells lying on the lines of only two sides of each large square were counted.

The number of white blood cells per cubic millimeter of blood was obtained as follows:

Number of WBCs counted x blood dilution factor x chamber depth divided by the

Area of chamber counted = Number of WBCs counted x 50

(Blood dilution factor 20; Chamber depth = 10; Area of chamber counted = 4).

Red Blood Cell Count

With the aid of a micropipette, 0.02 ml (20 μ l) of blood was added into a Khan's test tube containing 4 ml of formol-citrate solution. It was properly mixed and few drops loaded into a charged improved Neubauer counting chamber using a Pasteur pipette. When the cells have settled out of suspension, the counting chamber was taken to a light microscope and the number of red blood cells was counted using 40x objectives. Red blood cell count is carried out in the five smaller centre squares of the chamber. Cells lying on the lines of only two sides of each square were counted. The number of red cells per cubic millimeter of blood was obtained as follows:

Number of red cells counted multiplied by 200 multiplied by 10 divided by 1/5 =
Number of red cells counted x 10,000
(200 = Dilution factor; 10 = Depth factor; 1/5 = Area counted).

Platelet Count

With the aid of a micropipette, add 0.02ml (20 μ l) of well-mixed anticoagulated blood into a Khan's test tube containing 0.38ml of ammonium oxalate diluting fluid. It was properly mixed and few drops loaded into a charged improved Neubauer counting chamber using a Pasteur pipette.

The cells were allowed 20 minutes to settle before the counting chamber was viewed under a light microscope with 40x objective lens. Platelet count is done in the same five smaller centre squares as the red blood cell count. The number of platelets per cubic millimeter of blood was determined as follows:

$$\frac{\text{No. of platelets counted} \times \text{blood dilution factor} \times \text{chamber depth}}{\text{Area of chamber counted}}$$

= Number of platelets counted x 10,000.
(Blood dilution factor = 200; chamber depth factor = 10; Area counted = 1/5)

Red Cell Indices

The red cell indices were determined by the method described by Blaxhall *et al.*, (1973).

1. MCHC (%) = $\frac{\text{Hb(g/dl)} \times 100}{\text{PCV(\%)}}$
2. MCH (Pg) = $\frac{\text{Hb(g/dl)} \times 10}{\text{RBC(mm}^3\text{)}}$
3. MCV(fl) = $\text{PCV(\%)/RBC(mm}^3\text{)}$

Statistical Analysis

Data were reported as means \pm SD (Standard Deviation) and were analyzed using one-way analysis of variance (ANOVA). The relationship between variables (groups) was assessed using Fisher's Least Significant Difference (FLSD) post HOC Tests. Both tests were done with the aid of the SPSS (Statistical Package for Social Sciences). Values of $p < 0.05$ were regarded as being significant.

Results and Discussion

The results of the effect of Izol contaminated feed on the haematological profile and body weight of adolescent male albino rats, exposed to Izol for 28 days, are presented as means \pm SD (standard deviation) in different tables as shown below.

Mean Body Weight (g)

After acclimatization, the rats recorded a mean body weight (g) of 90.00 \pm 18.17, 116.67 \pm 19.41, 105.00 \pm 22.14 and 97.50 \pm 13.32 respectively for group 1, group 2,

group 3, and control (Table 1). However, while groups 2, and 3 recorded insignificantly ($P > 0.05$) higher mean body weights compared with the control, group 1 recorded an insignificantly ($P > 0.05$) lower mean body weight compared with the control. The mean body weight (g) ranged from a minimum of 90.00 ± 18.17 , for group 1 which was given 1 ml (0.8%) of Izol, to a maximum of 116.67 ± 19.41 , for Group 2 which received 2 ml (1.7%) of Izol per 123.12 g of feed (Table 1). At the end of the experiment, the mean body weight (g) of the rats were 119.17 ± 32.77 , 154.17 ± 15.30 , 134.17 ± 21.31 , and 124.17 ± 19.34 respectively for Groups 1,2,3 and control (Table 1).

Generally, all the groups recorded steady increases in mean body weight all through the experiment, except for few exceptions- the weight(g) of the control decreased from 100.00 ± 13.78 , in day 7 of treatment, to 93.33 ± 51.34 , in day 14 (Table 1). Similarly, while group 2 maintained the same weight (124.17g) in day 14 and 21, the weight (g) of group 1, and 3 decreased from 101.67 ± 19.15 , and 119.17 ± 22.23 to 95.83 ± 20.10 and 107.50 ± 27.34 in day 14 and 21 respectively (Table 1). The significant increase in weight in each group is recorded as percentage (%), and showed that all the groups including the control appreciated in weight (Table 2). The order of increase is group 1 (32.41%) > group 2 (32.14%) > group 3 (27.78%) > control (27.35%), (Table 2). Mean within the same column and in each treatment days are not significantly different ($P > 0.05$) compared with control.

Mean Packed Cell Volume PCV (%)

At day 14, the mean PCV value of group one rats was the same when compared

with the control. Group 2 rats recorded a lower mean PCV value when compared with the control, though this decrease was not significant ($P > 0.05$). Group 3 rats recorded a significant increase ($P < 0.05$) in mean PCV value when compared with the control. The mean PCV (%) \pm SD ranged from a minimum of 33.00 ± 2.00 , recorded for group 2 rats given 2ml (1.7%) of IZOL, to a maximum of 44.00 ± 1.0 recorded for group 3 rats given 3ml (2.5%) of IZOL per 123.12g of feed (Table 3). At day 28, group 1 rats recorded a decreased PCV value when compared with the control though this decrease was not significant ($P > 0.05$). Group 2 and 3 rats recorded significant increases ($P < 0.05$) in their PCV values when compared with the control.

Mean Erythrocyte Sedimentation Rate (ESR-mm) of adolescent male albino rates exposed to Izol

At day 14, group 1 and 3 rats recorded a lower mean ESR values compared with the control, though these decreases were not significant ($P > 0.05$). Group 2 recorded a higher mean ESR value which was also not significant ($P > 0.05$) when compared with control. The mean ESRs (mm) \pm SI ranged from a minimum of 1.33 ± 0.6 , recorded for group 3 rats given 3ml (2.5%) of IZOL, to a maximum of 2.00 ± 0.5 , recorded for groups 2 rats given 2ml (1.7%) of IZOL per 123.12g of feed (Table 4). At day 28, group 2 rats recorded the same mean ESR value with control. Group 3 rats recorded a lower value when compared with control, though this decrease was not significant ($P > 0.05$). Group 1 recorded a higher value compared with control, though this increase was not significant ($P > 0.05$). It is worthy to note that the higher the ESR value, the more probable the organism will be anaemic.

Mean Haemoglobin Hb (g/dl)

At day 14, mean Hb of group 1 rats decreased, though this decrease was not significant ($P>0.05$) compared with the control. Group 2, and 3 rats recorded increased mean Hb values, though these increases were not also significant ($P>0.05$). The mean Hb (g/dl) \pm SD ranged from a minimum of 9.30 ± 1.20 , recorded for group 1 rats given 1ml (0.8%) of IZOL, to a maximum of 12.10 ± 1.10 recorded for group 3 rats given 3ml (2.5%) of IZOL per 123.12g of feed (Table 5). At day 28, the mean Hb value of all the groups decreased when compared with the control. However, these decreases were significant ($P<0.05$) for group 1 and 2, it was not significant ($P>0.05$) for group 3. The mean Hb (g/dl) \pm SD ranged from a minimum of 9.77 ± 0.95 , recorded for group 1 rats given 1ml of (0.8%) of IZOL per 123.12g of feed, to a maximum of 12.10 ± 1.10 , recorded for the control rats that were not exposed to IZOL (Table 5).

Mean Total White Blood Cell (WBC) Count ($10^3/\text{mm}^3$)

At day 14, the mean WBC count decreased for group 2, and 3 rats, though these decreases were not significant ($P>0.05$) compared with the control. Conversely, group 1 rats recorded a significantly ($P<0.05$) higher mean WBC count compared with control. The mean WBC ($10^3/\text{mm}^3$) \pm SD ranged from a minimum of 5600.00 ± 400.00 , for group 2 rats given 2ml (1.7%) of IZOL, to a maximum of 12300.00 ± 1350.31 , for group 1 rats given 1ml (0.8%) of IZOL per 123.12g of feed (Table 6). At day 28, the mean WBC count decreased for groups 1, and 3 rats, though these decreases were not significant ($P>0.05$) compared with the control.

Mean Red Blood Cell (RBC) Count ($10^6/\text{mm}^3$)

At day 14, group 1 rats recorded an insignificant ($p>0.05$) decrease in mean RBC count compared with the control. Group 2 and 3 rats recorded a higher mean RBC count, though these increases were not significant ($P>0.05$) compared with the control. The mean RBCs ($10^6/\text{mm}^3$) \pm SD ranged from a minimum of 4.52 ± 0.51 , for group 1 rats given 1ml (0.8%) of IZOL, to a maximum of 5.78 ± 0.74 , for group 3 rats given 3ml (2.5%) of IZOL per 123.12g of feed (Table 7). At day 28, group 1, and 2 rats recorded a decrease in mean RBC count. While that of group 1 was significant ($P<0.05$), that of group 2 was not significant ($P>0.05$) compared with control. Group 3 recorded a significantly ($P<0.05$) higher mean RBC count compared with the control. The mean RBCs \pm SD ranged from a minimum of 5.65 ± 0.28 , for group 1 rats given 1ml (0.8%) of IZOL, to a maximum of 7.03 ± 0.06 , ($10^6/\text{mm}^3$) for group 3 rats given 3ml (2.5%) of IZOL per 123.12g of feed (Table 7).

Mean Platelet Count (mm³)

At day 14, group 2 rats recorded an insignificant ($P>0.05$) decrease in platelet count compared with control. Conversely, group 1, and 3 recorded insignificant ($P>0.05$) increases when compared with control. The mean platelet (mm^3) \pm SD ranged from a minimum of 55000.00 ± 7000.00 , for group 2 rats given 2ml (1.7%) of IZOL, to a maximum of $191000.00, \pm 121500.34$, for group 1 rats given 1ml (0.8%) of IZOL per 123. 2g of feed (Table 8). At day 28, group 2 and 3 rats recorded insignificant ($P>0.05$) decreases compared with control. Group 1 rats recorded an insignificant ($P>0.005$)

increase in mean platelet count compared with control. The mean platelet (mm^3) \pm SD ranged from a minimum of 520000.00 ± 90000.00 , for group 2 rats given 2ml (1.7%) of Izol, to a maximum of 745000.00 ± 95000.00 , for group 1 rats given 1ml (0.8%) of Izol per 123.12g of feed (Table 8).

Mean red cell indices

Mean Cell Haemoglobin concentration (MCHC)

At day 14, group 1 and 3 rats recorded insignificant ($P > 0.05$) decreases in MCHC compared with the control. Conversely, group 2 rats recorded a significant ($P < 0.05$) increase in MCHC compared with the control. The MCHCs (%) \pm SD ranged from a minimum of 24.80 ± 4.21 , for group 1 rats given 1ml (0.8%) of IZOL, to a maximum of 35.76 ± 0.05 , for group 2 rats given 2ml (1.7%) of IZOL per 123.12g of feed (Table 9). At day 28, all the groups recorded significant ($P < 0.05$) decreases in MCHC compared with the control. The MCHCs (%) \pm SD ranged from a minimum of 25.21 ± 0.32 , for groups 3 rats given 3ml (2.5%) of IZOL per 123.12g of feed, to a maximum of 33.04 ± 1.98 , for the control group which was not exposed to IZOL (Table 9).

Mean Cell Haemoglobin (MCH)

At day 14, all the groups recorded insignificant ($P > 0.05$) decreases in MCH compared with the control. The MCHs (pg) \pm SD ranged from a minimum of 20.80 ± 0.53 , for group 2 rats given 2ml (1.7%) of IZOL per 123.12g of feed, to a maximum of 21.84 ± 4.78 , for the control group which was not given IZOL (Table 9). At day 28, all the groups recorded significant decreases ($P < 0.05$) in MCH compared with the control. The MCHs (pg) \pm SD ranged from a minimum of 17.21 ± 0.00 ,

for group 3 rats given 3ml (2.5%) of IZOL per 123.12g of feed, to a maximum of 19.90 ± 0.61 , for the control group which was not given IZOL (Table 9).

Mean Cell Volume (MCV)

At day 14, the group 2 rats recorded an insignificant ($P > 0.05$) decrease in MCV compared with the control. Group 1 and 3 rats recorded insignificant ($P > 0.05$) increases in MCV compared with the control. The MCVs (fl) \pm SD ranged from a minimum of 5.82 ± 0.14 , for group 2 rats given 2ml (1.7%) of IZOL, to a maximum of 8.39 ± 0.61 , for group 1 rats given 1ml (0.8%) of IZOL per 123.12g of feed (Table 9). At day 28, all the groups recorded significant ($P < 0.05$) increases in MCV compared with the control. The MCV (pg) \pm SD ranged from a minimum of 6.03 ± 0.18 , for the control, to a maximum of 6.85 ± 0.25 , for group 1 rats given 1ml (0.8%) of IZOL per 123.12g of feed (Table 9).

The result of the daily oral administration of different concentrations of Izol to various groups of albino rats showed a regular increase in mean body weight (g) compared with the control. The effect seems not to be dose-dependent as the least concentration or dose produced the highest percentage appreciation in weight. This finding does not agree with that of Kurliandskii *et al.* (1976) on the subchronic toxicity study of Izol on Sprague-Dawley rats. The difference in observation could be due to the difference in age, doses/concentration, mode of exposure, or duration of exposure. The haematological results showed a dose-dependent significant ($P < 0.05$) increases in percentage packed cell volume (%PCV), red blood cell (RBC) count, and mean cell volume (MCV) compared with those of the control.

Table.1 Mean Body Weight (g) of adolescent male albino rats after acclimatization and during treatment with Izol for 28 days

Particulars	Duration of Exposure (days)	Groups	Dose per 123.12 g of feed	Mean body weight (g)
After acclimatization	7	Group 1	1 ml (0.8%)	90.00 ± 18.17
		Group 2	2 ml (1.7%)	116.67 ± 19.41
		Group 3	3 ml (2.5%)	105.00 ± 22.14
		Control	0 ml (0%)	97.50 ± 13.32
During treatments/Exposure	7	Group 1	1 ml (0.8%)	91.67 ± 18.62
		Group 2	2 ml (1.7%)	121.67 ± 19.92
		Group 3	3 ml (2.5%)	110.83 ± 22.45
		Control	0 ml (0%)	100.00 ± 13.78
During treatments/Exposure	14	Group 1	1 ml (0.8%)	101.67 ± 19.15
		Group 2	2 ml (1.7%)	124.17 ± 19.85
		Group 3	3 ml (2.5%)	119.17 ± 22.23
		Control	0 ml (0%)	93.33 ± 51.34
During treatments/Exposure	21	Group 1	1 ml (0.8%)	95.83 ± 20.10
		Group 2	2 ml (1.7%)	124.17 ± 22.23
		Group 3	3 ml (2.5%)	107.50 ± 27.34
		Control	0 ml (0%)	108.33 ± 22.73
	28	Group 1	1 ml (0.8%)	119.17 ± 32.77
		Group 2	2 ml (1.7%)	154.17 ± 15.30
		Group 3	3 ml (2.5%)	134.17 ± 21.31
		Control	0 ml (0%)	124.17 ± 19.34

Mean within the same column and in each treatment days are not significantly different ($P > 0.05$) compared with control.

Table.2 Percentage (%) increase in Mean Body Weight of adolescent male albino rats exposed to Izol for 28 days

GROUPS	Doses per 123.12 g of feed	Final mean body weight (g)	Initial mean body weight (g)	% increase
Group 1	1ml (0.8%)	119.17 ± 32.77	90.00 ± 18.17	32.41
Group 2	2ml (1.7%)	154.17 ± 15.30	116.67 ± 19.41	32.14
Group 3	3ml (2.5%)	134.17 ± 21.31	105.00 ± 21.14	27.78
Control	0ml (0%)	124.17 ± 19.34	97.50 ± 13.32	27.35

Table.3 Mean packed cell volume (%) of adolescent male albino rates exposed to Izol

Groups	Does per 123.12 g of feed	Duration of exposure	
		Day 14	Day 28
Group 1	1ml (0.8%)	37.67± 1.53	38.67 ± 0.58
Group 2	2ml (1.7%)	33.00 ± 2.00	42.67 ±1.53*
Group 3	3ml (2.5%)	44.00± 1.00*	48.00± 1.00*
Control	0ml (0%)	37.67 ±5.51	39.00 ±1.00

Mean with (*) within the same column are significantly different (p<0.05) compared with control.

Table.4 Mean Erthrocyte Sedimentation Rate (mm) of Adolescent male albino rats

Group 1	Dose per 123.12 g of feed	Duration of exposure	
		Day 14	Day 28
Group 1	1ml (0.8%)	1.57 ± 0.40	1.40±0.10
Group 2	2ml (1.7%)	2.00 ±0.50	1.23 ±0.15
Group 3	3ml (2.5%)	1.33 ±0.58	1.00 ± 0.00
Control	0ml (0%)	1.80 ± 0.52	1.23±0.15

Table.5 Mean Haemoglobin (g/dl) of Adolescent male albino Rats exposed to Izol for 28 days

Groups	Doses per 123.12 g of feed	Duration of Exposure	
		Day 14	Day28
Groups1	1ml (0.8%)	9.30± 0.20	9.77 ±0.00*
Groups2	2ml (1.7%)	11.80 ± 0.70	11.40±0.10
Groups3	3ml (2.5%)	12.10 ±1.10	12.10± 1.10
Control	0ml (2.5%)	11.00 ±1.50	12.9±1.10

Mean with (*) within the same column are significantly different (p<0.05) compared with control.

Table.6 Mean total White Blood Cell Count ($10^3/mm^3$) of adolescent male albino rats exposed to Izol for 28 days

Groups	Does per 123.12 g of feed	Duration of Exposure	
		Day 14	Day 28
Group 1	1ml (0.8%)	12300.00±1350.31*	7700.00 ±1900.00
Group 2	2ml (1.7%)	5600.00±400.00	10200.00±1100.00
Group 3	3ml (2.5%)	5933 ±850.49	7933.00±1950.21
Control	0ml (0%)	7100±900.00	9933.00±195021

Table.7 Mean Red Blood Cell (RBC) Count ($10^6/\text{mm}^3$) of adolescent male albino rats exposed to Izol for 28 days

Groups	Does per 123.12 g of feed	Duration of Exposure	
		Day 14	Day 28
Group 1	1ml (0.8%)	4.52 ±0.51	5.65 ±0.28*
Group 2	2ml (2.5%)	5.68 ± 0.48	6.39 ±0.06*
Group 3	3ml (2.5%)	5.78 ± 0.74	7.03 ± 0.06*
Control	0ml (0%)	5.18 ±1.24	6.47 ± 0.36

Mean with (*) within the same column are significantly different ($p < 0.0$) compared with control.

Table.8 Mean Platelet count (mm^3) of adolescent male albino rats exposed to Izol for days

Groups	Does per 123.12 g of feed	Duration of Exposure	
		Day 14	Day 28
Group1	1ml (0.8%)	191000.00 ± 121500.34	745000.00 ±95000.00
Group2	2ml (1.7%)	55000.00 ±7000.00	520000.00 ±90000.00
Group 3	3ml (2.5%)	168000.00 ±116500.00	590000.00 ± 90000.00
Control	0ml (0%)	91000.00±2000.00	615000.00±215000.00

Table.9 Mean Red Cell Indices of Adolescent Male Albino Rats

Red cell indices	Groups	Dose per 123.12 g of	Days 14	Days 28
MCHC (%)	Group 1	1ml (0.8%)	24.80 ± 4.21	25.24 ±2.14*
	Group 2	2ml (1.7%)	35.76±0.05*	26.74±0.32*
	Group 3	3ml (2.5%)	27.53±2.83	25.21±0.32*
	Control	0ml (0%)	29.24±0.40	33.04±1.98
MCH (Pg)	Group 1	1ml (0.8%)	20.97±5.03	17.263 ± 0.83*
	Group 2	2ml (1.7%)	20.80 ±0.53	17.85 ±0.32
	Group 3	3ml (2.5%)	20.87±3.68	17.21±0.00*
	Control	0ml (0.8%)	21.84 ±4.78	19.90 ± 0.61
MCV(fl)	Group 1	1ml (0.8%)	8.39 ±0.61	6.85 ±0.25*
	Group 2	2ml (1.7%)	5.82 ±0.14	6.68 ± 0.12*
	Group 3	3ml (2.5%)	7.55±0.77	6.83 ± 0.09*
	Control	0ml (0%)	7.48 ± 1.71	6.03 ±0.18

This is an indication that Izol may stimulate erythropoiesis. Cameron and Watson (2007) had reported a normal PCV range for adult male albino rats as 42.9 - 44.1%. Furthermore, the result of the MCVs which were significantly higher in all the groups compared with the control revealed that the red cells were macrocytic compared with the control. That is, the red cells of all the treatment groups were larger than normal compared with the control. The haemoglobin (Hb), mean cell haemoglobin concentration (MCHC), and mean cell haemoglobin (MCH) results showed significant ($p < 0.05$) decreases compared with those of the control, though the decrease was not dose-dependent. These observations suggest that ingestion of Izol may induce anaemia. Anaemia is a reduction in the concentration of haemoglobin in the peripheral blood below the normal for the age and sex of the patient (Ramnik, 2003). It can also be defined as the deficiency of haemoglobin in the blood due to lack of red blood cells and or their haemoglobin content (Roper, 1998). The fact that the haemoglobin concentration of these rats decreased significantly compared with the control suggests that the anaemia was not haemolytic since red blood cell haemolysis will reflect a rise in the blood haemoglobin concentration (Bolarin, 1997). Thus, there is a clear indication that Izol may act in some ways that may suppress the machineries involved in haemoglobin synthesis. As a result, there is an observed imbalance between the rate of haemoglobin synthesis and the rate of red blood cell formation with the attendant increase in PCV and RBC count and a decrease in Hb concentration. This finding agrees with Bloom and Fawcett (1975), that the concentration of haemoglobin per cell is less than normal when the rate of red cell formation is relatively greater than

the rate of haemoglobin synthesis; under these circumstances, each erythrocyte contain an abnormally small quantity of haemoglobin; the erythrocyte appears paler and is described as hypochromic, compared to normal or normochromic erythrocytes. To lend credence to this, Baker and Silverton (1985) and Monica (2002) had reported low MCHC values in iron deficiency anaemia, and high MCV values in macrocytic anaemia. There was no observed adverse effect on the erythrocyte sedimentation rate (ESR) and platelet count as all the treatment groups recorded values that were not significantly different ($p > 0.05$) compared with those of the control. This observation for platelet count suggests that short-term ingestion of low Izol concentration may not cause cytotoxicity in male rats. Monica (2002) reports that treatment with cytotoxic drugs (e.g quinone), chemicals (e.g benzene), and some herbal remedies cause decreased platelet count in humans. There were significant changes in the results of the leukocyte counts. The total white blood cell count showed significant ($p < 0.05$) increase compared with the control. The observed leukocytosis is an indication that the body defense mechanisms of the rats recognized and responded appropriately to Izol as a foreign chemical agent. OECD (2000) and Uzhdavini *et al.* (1972) had reported elevated leukocyte counts in seminchromic male rats in a study on inhalation toxicity. Similarly, Monica (2002) reported poisoning (e.g chemicals) as one of the major causes of leukocytosis.

The result of this study indicated that short-term ingestion of low Izol concentration causes an increase in weight in adolescent male albino rats. While it produced marked increase in percentage packed cell volume, red blood cell count, and mean cell volume (MCV), it caused a

significant decrease in haemoglobin, mean cell haemoglobin concentration (MCHC), and mean cell haemoglobin (MCH). Therefore, short-term ingestion of low Izol concentrations may stimulate erythropoiesis and at the same time precipitate macrocytic hypochromic anaemia in adolescent male albino rats. Furthermore, it induces leukocytosis and hence lymphocytosis and thereby suggest that in male rats, lymphocytes are probably either the body's first — line of action against disinfectants (Izol), or they are the most relevant type of white blood cells involved in immune responses to disinfectants (Izol). Lastly, short-term ingestion of low Izol concentrations caused no cytotoxicity in adolescent male albino rats.

Recommendation

It is recommended that the practice of applying cresols (Izol, Izaal, or Lysol) to wells after clean up should be discouraged. It is also recommend that the practice of calculating haemoglobin concentration from packed cell volume (PCV) values should be discouraged since this study has shown that it is sometimes misleading and not reliable. Finally, more detailed studies should be carried out on the long-term effects of ingestion of low Izol concentration.

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