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In Vivo toxicity Studies of *Citrullus colocynthis* Schard

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ABSTRACT
Citrullus colocynthis is an important medicinal cucurbit belonging to the family Cucurbitaceae. The present work was carried out to investigate pharmacognostical and pharmacological activity of Citrullus colocynthis schard. The in vivo toxicity study of Citrullus colocynthis schard was carried out on healthy wistar pale skinned rodents (rats) somewhere in the range of 150-200 gram were randomly allocated to different groups containing three rats in each group. The toxicity studies of leaf, stem and fruit of C. colocynthis schard. The leaf extract shows loss of locomotion action, ataxia and at the most elevated doses administered, in appetence, laziness, hypothermia, and cyanosis and in the end demise. During the study of stem extract, animals that endure had a few side effects, including diarrhea. The death rate in male rodents was most extreme in groups treated with 300 mg/kg.
Keywords*: Citrullus colocynthis schard, In Vivo, Toxicity Studies.*

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INTRODUCTION

Citrullus colocynthis contained sugar, protein, isolated amino corrosive, tannins, saponins, phenolics, flavanoids, flavone glucosides, terpenoids, alkaloids, anthranol, steroids, cucurbitacins, saponarin, cardicglycoloids, follow components and numerous other substance groups. It had cell reinforcement, Antidiabetic, antimicrobial, anticancer, hostile to inflammatory, pain relieving, gastrointestinal, regenerative and defensive and numerous other pharmacological impacts. [1,8]

It was local to dry territories of North Africa and it has been known in the Mediterranean area since Biblical occasions. Yearly or perpetual (in wild) herbaceous vine; stems rakish and unpleasant; leaves harsh, 3-to 7-lobed, 5-10 cm long, centre projection in some cases praise, sinuses open; blossoms monoecious, single, peduncled, axillary, corollas 5-lobed; ovary villous; natural product a pepo, almost globular, 4-10 cm in measurement with to some degree circular crevices, about size of little orange, green and yellow variegated getting yellow when ready, with hard skin, mash light in weight, springy, handily broken, light yellowish-orange to light yellow; strongly severe; seeds various, ovoid, compacted, smooth, dim earthy colored to light yellowish-orange, borne on parietal placenta [9].

The root was utilized in aggravation of the bosoms, joints torment; remotely it was utilized in ophthalmia and in uterine agonies. The foods grown from the ground were scoured with water and applied to bubbles and pimples. A glue of the root is applied to the developed stomach area of children. The organic product was likewise utilized in ascites, biliousness, jaundice, cerebral clog, colic, clogging dropsy, fever, worms and sciatica. Root was likewise given in instances of stomach extension, hack, asthma, irritation of the bosom, ulcers, urinary ailments and ailment. [3,10-14].

The present work was carried out to investigate pharmacognostical and pharmacological activity of *Citrullus colocynthis* schard.

MATERIAL AND METHODS

Toxicity study[15]

The toxicity study for each crude extract was performed according to the organization for economic co-operation and development (OECD) revised draft guidelines 423 B ("Up and Down" method). Healthy wistar pale skinned rodents (rats) somewhere in the range of 150-200 gram were randomly allocated to different groups containing three rats in each group. Single dose individual concentrate was ingested in separate group to evaluate the wellbeing and viability. In view of LD50 cut-off estimation of the individual

plant concentrate, the compelling dose was determined as 1/10 portion for the each plant concentrate. The protocol according to the annexure was pursued.

Toxicity study of leaf of *Citrullus colocynthis* [15]

Animal selection

Male Albino-Wistar rodents weighing between 180 to 210 g were utilized for acute poisonousness study. Rodents were housed in hanging transparent plastic pens (55 × 33× 19 cm) in the animal room and accustomed for 3 weeks before explore. The litter was reestablished like clockwork. They were taken care of with a standard pellet and faucet water not indispensable. All rodents were kept in standard ecological conditions. The animal care and test conventions were according to control and supervision of experiments on animals (CPCSEA) and institutional animal ethical committee (IAEC). IAEC approval number for the animal study is IAEC/RCPIPER/2019-20/15.

Determination of LD50 [15]

The leaf concentrate of *Citrullus colocynthis* to be tested is soluble in a couple of drops of ethanol and diluted in saline, by gavage at a dosage for each group. Male rodents were weighed 186.25 ±2.80 g, distinguished by marking with a fluid solution of picric acid and isolated into groups of 10 rodents each and fasting for brief period before oral organization of single portions of all the leaf concentrate of Citrullus colocynthis. Five gatherings of rodents are treated with basic application and progressively with the accompanying dosages: 500, 1000, 1800, 2000 and 3000 mg/kg. The control group got physiological saline with a couple of drops of liquor. After organization of the concentrate, rodents were watched exclusively consistently during the main day and consistently for 14 days. Conduct and clinical indications of rodents are noted all through the span of the trial. LD50 and its range are determined by the realistic technique for Litchfield and Wilcoxon [15]

Acute toxicity study [16]

The male rodents were assigned haphazardly to six groups of 10 creatures and were given orally a solitary portion of 440 mg/kg (1/3 DL50) body weight of the leaf of *C. colocynthis*, yet not deadly portion to attempt to explore the objective organs. The control group (10 rodents) got saline water. Rodents were watched and recorded deliberately 1, 2, 3, 4, 5 and 6 h and day by day after test substance administration. The visual perceptions remembered changes for skin and hide (hair), eyes and mucous films, and furthermore respiratory, circulatory, autonomic and focal sensory system. The treatment bunches were relinquished after 24 h, 3, 5, 7, 10 and 14 days of the treatment. In people, after intense harming by leaf concentrate of C. colocynthis, hospitalization and recuperation takes between 3 to 6 days. [16]

Toxicity study of stem extract of *Citrullus colocynthis* [17]

Determination of LD50 [17]

The stem concentrate of *Citrullus colocynthis* to be tried is soluble in a couple of drops of methanol and weakened in saline and regulated at various portions, by gavage at a portion for each group. Male rodents were weighed 186.25 ±2.80 g, distinguished by naming with a fluid solution of picric acid and isolated into gatherings of 10 animals each and fasting for brief period before oral organization of single portions of the stem-concentrate of *Citrullus colocynthis*. Five gatherings of rodents are treated with application and progressively with the accompanying dosages: 500, 1000, 1800, 2000 and 3000 mg/kg. The control group got physiological saline with a couple of drops of liquor. After administration of the concentrate of stems, animals were watched separately consistently during the main day and consistently for 14 days. Conduct and clinical manifestations of animals are noted all through the term of the test. LD50 and its range are determined by the realistic technique for Litchfield and Wilcoxon (1949).

Acute toxicity in male rats [17]

The rodents were partitioned in arbitrary into 5 groups, every one of 10 rodents. Four groups got per os 131 mg/kg (≈ 1/10 DL50) of the ethanolic concentrate of stem of *Citrullus colocynthis* however were killed following 24 hours, 5 days, 10 days and 14 days of the treatment. One group was kept up as typical control and got ordinary saline. Toward the finish of every single exploratory period, creatures were anesthetized with urethane at the portion 760 mg/kg. Two sorts of blood were acquired from the retro-orbital vein, an example for hematology containing ethylene diaminetetraacetic corrosive for estimation (Erythrocyte (RBC) and leukocyte (WBC) checks, hemoglobin fixation (Hb), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin focus (MCHC), mean platelet volume (MPV) and platelets hematocrit (PCT) with mechanical assembly MEDONIC (Beckman Coulter – USA) and test for serum and utilized for estimation of exercises Glutamic-oxaloacetic transaminase (GOT), Glutamic-Pyruvic Transaminase (GPT) (utilizing business Kits – SGM Rome-Italy) , antacid phosphatase (ALP) (utilizing business Kits – CYPRESS DIAGNOSTIC Langdrop – Belgium), groupings of complete protein, urea, glucose, creatinine, sodium, potassium, calcium and phosphorus with device TECHNICON RA-1000-USA. After blood assortment, the creatures were relinquished by cervical separation. After dissection, all tissues were inspected terribly and significant organs (liver, cerebrum, heart, kidneys,

spleen, testicles and lung) were weighted. The relative organ (weight of organ as an extent of the all-out weight of each rodent) was determined and contrasted and the estimation of the control.

Toxicity study of fruit extract of *Citrullus colcoynthis*[18]

Determination of oral LD50

So as to decide the zero and 100% mortality, genuine preliminaries were completed on 6 groups of animals every one of four rodents for every region. Rodents dosed orally by means of intra-gastric intubation utilizing rodent stomach tube with 24, 20, 16, 12, 8 and 4 g./kg.b.wt. for every area. In every preliminary a group of 6 control rodents controlled refined water just was incorporated. The genuine assurance of LD50 was done. All rodents were held under close perception for clinical signs. Dead rodents were exposed to after death assessment. [18]

Acute toxicity in male rats [18]

Forty eight evidently sound albino rodents were utilized to contemplate the subchronic toxicological impacts of fruit pulp of C. colocynthis remove. Rodents were similarly arranged into four groups. The initial three groups were given orally (¼ of the LD50) of the concentrate of C. colocynthis fruit, while the fourth group was kept as control and got just refined water. The concentrate was given each week till 10 weeks, at that point rodents were yielded and EDTA-blood was gathered for hematological assessment. Extra blood tests were gathered without anticoagulant for biochemical assessment of serum.

RESULTS AND DISCUSSION

Toxicity study

Toxicity study of leaf extract of *Citrullus colocynthis*

Determination of LD50

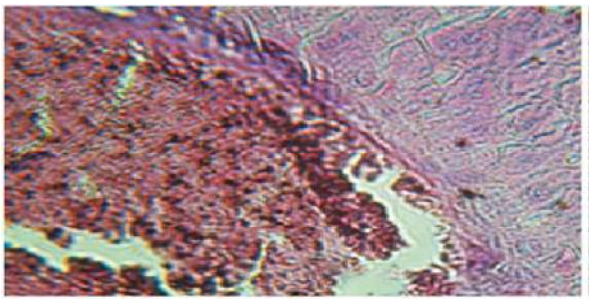

Fig.1 Liver parenchyma traversed by congestive blood vessels in male rats treated with leaf extract. H&E (× 400)

The animals were held under close perception for 72 hours in the wake of dosing to check for side effects, conduct changes and demise. The dosages tried created, right on time inside the first hour of treatment the accompanying signs, which exacerbates with the death of the time: loss of locomotion action, ataxia and at the most elevated doses administered, in appetence, flaw of the hind limbs, laziness, hypothermia, cyanosis and in the end demise. Extreme diarrhea was the most genuine side effects; subsequent to developing it, the exploratory animals died. The animals that endure had a few side effects, including mild diarrhea, however had the option to recuperate. The intensity of the harmful impacts was dose subordinate. Rodent's mortality after various dosages of ethanol concentrate of leaf of *Citrullus colocynthis* was plotted against probability values. The death rate in male rodents was greatest (100%) in bunches treated with 300 mg/kg. The acute median lethal doses (LD50) of the concentrate were seen as 1311.45 mg/kg at 95% certainty limit: [1037.80 to 1657.27 mg/kg] for rodents' males. The posthumous discoveries were as clog in the liver just as in renal parenchyma (Fig. 1, 2).

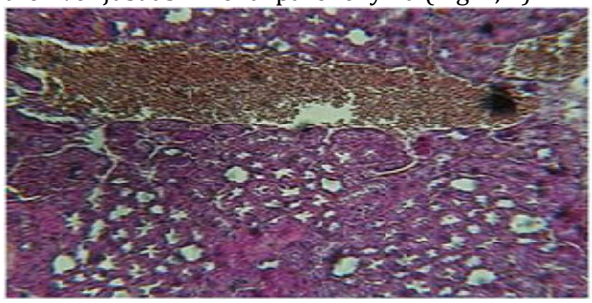

Fig.2 Renal parenchyma traversed by blood vessels with distended lumen engorged with blood in male rats treated with leaf extract. H&E (× 100)

B) Acute toxicity study

The principle indications of toxicity saw after oral administration of single portion tried (440 mg/kg ≈ 1/3 DL50) were: diarrhea, unsettled hair, speeding up of pulse breathing trouble, soft defecation and clustering together toward one side of the cages. None of the rodents in totally treated groups passed on over the span of the examination.

Table.1 Effect of acute administration of CCT fruit methanolic extract (440 mg/kg) on body weight of male rats. Values are mean ± SEM

Group	1st day	3rd day	5th day	7th day	10th day	14th day
Control	244.8216.72					258.241 9.96
1st day	238114.30					
3rd day	244.40+12.04	237.86+9.10				
5th day	255.40+5.65		247.34+6.05			
7th day	237.60±8.32			231.20±9.30		
10th day	242.54±10.18				237.40±9.34	
14th day	242.88±10.15					240.46±9.39

There were no measurably huge contrasts in normal body weight of the control group and *Citrullus colocynthis* leaf-extract treated groups during the acute toxicity. By and by, a slight misfortune in body weight was noted in CCT leaf extract regarded rodents when contrasted with control group (Table. 41). Macroscopic assessment of different organs in situ didn't show any morphological changes in organs of treated animals contrasted and those of control rodents. The impacts of CCT leaf extract on relative organ weights (liver, kidney, lung, heart, mind, testis and spleen) are introduced in table 1. Huge measurable decreases were noted on the general weights of liver, kidney, lungs, spleen and testicles among the treated groups. Following fourteen days of the treatment, every single treated group has recouped the typical relative organ weight.

Heamatological changes

The blood image of leaf concentrate of C. colocynthis treated rodents after acute administration of leaf alcoholic concentrate is appeared. The total RBC count, WBC, HCT and HGB content were altogether lower ($P < 0.05$) in the treated groups than the control one.

Table 43.Effect of oral administration of CCT leaf ethanolic extract (200 mg/kg) on some haematological parameters in male rats. Values are mean ± SEM.

Changes in serum constituents

The impacts of CCT leaf concentrate on liver capacity biomarkers in serum of albino rodents after oral administration of (1/3 DL50): AST, ALT and ALP are given in figure 3. It was seen that the estimations of AST (GOT) and ALT (GPT) were higher however not altogether unique in the fundamental treatment groups contrasted and those of control group. Nonetheless, the increase of the degrees of GOT (AST) and GPT (ALT) were huge ($P < 0.05$) following 3 days of the treatment. Moreover, the soluble phosphatase values were essentially higher ($P < 0.05$) nearly in most of the tested groups (first, 3rd and fifth day), and begin to come back to the typical values following 10 to 14 days of the treatment. Be that as it may, complete proteins levels were no altogether unique in contrast with control group (Fig. 4). At long last, practically all treatment groups (aside from the gathering relinquished following 14 days) have registered a critical decrease in blood glucose level when contrasted and control (Fig. 5). The consequences of the impacts of CCT leaf extract on the principle kidney work parameters are appeared in figure 6. From this figure, creatinine level was fundamentally higher ($P < 0.05$) in treatment groups contrasted and the control. The urea level has registered a noteworthy increase ($P < 0.05$) in the three first groups and demonstrated typical values for the other treatment groups. Be that as it may, the uric acid has shown a no noteworthy variety for all treatment groups.

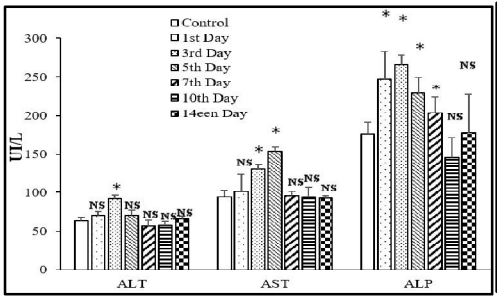

Fig.3 Effect of leaf ethanolic extract of CCT (200mg/kg) in male rats.

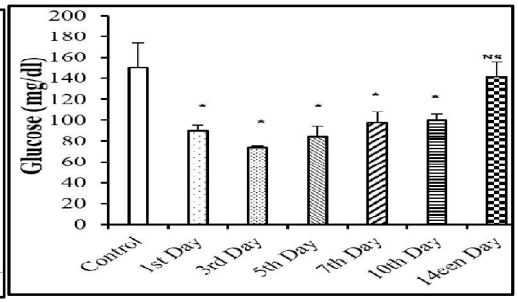

Fig.4 Effect of leaf CCT ethanolic extract (200mg/kg) on glucose level in male rats

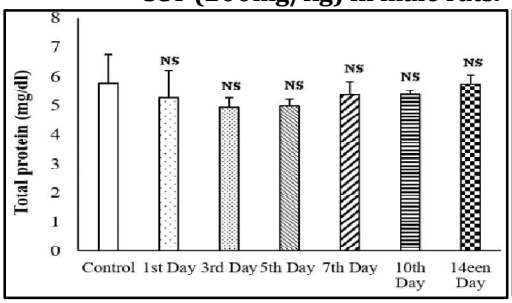

Fig.5 Effect of leaf CCT ethanolic extract (200mg/kg) on total protein level in male rats.

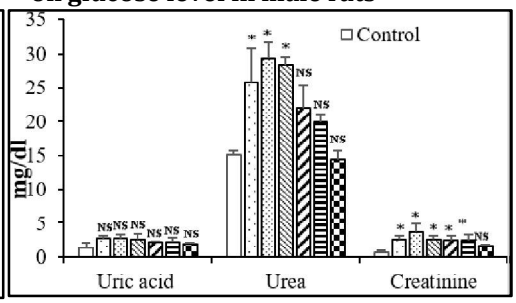

Fig.6 Effect of leaf CCT ethanolic extract on some renal function parameter in male rats

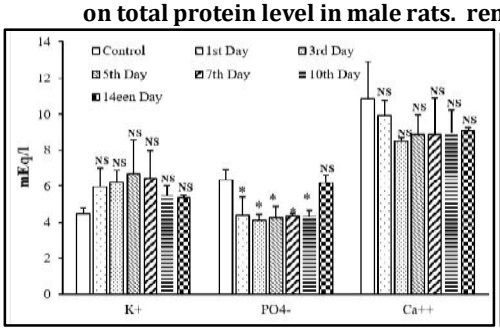

Fig.7 Effect of leaf CCT ethanolic extract (200mg/kg) on sodium level in male rats

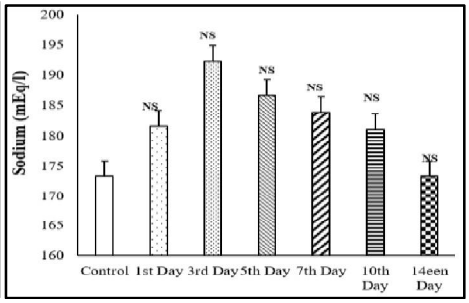

Fig.8 Effect of leaf CCT ethanolic extract (200mg/kg) on some electrolyte's level in male rats

It shows up from the figure 7 and 8 that CCT ethanolic leaf extract didn't influence altogether the electrolyte concentrations, yet it actuated a non-huge increase in sodium and potassium levels and a decrease in calcium level. Phosphorus was the single electrolyte which has introduced a noteworthy decrease ($P < 0.05$) in its level relatively to control values.

II) Pathological changes

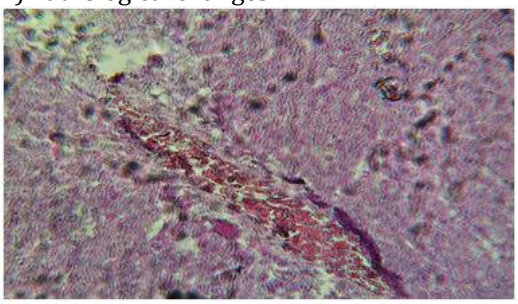

Fig.9 Liver parenchyma traversed by congestive blood vessels

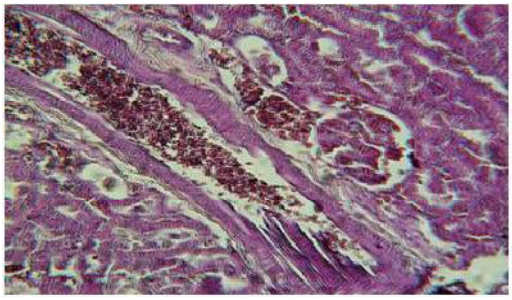

Fig.10 Congestive renal parenchyma after one day of acute treatment with CCT leaf extract (200 mg/kg)

The histological assessment of liver from male rodents was acted in both control and treated groups. Hepatic parenchyma crossed by congestive blood vessels and a congestive renal parenchyma were seen in the group relinquished following 24 hours (Fig.9, 10). The renal parenchyma turns out to be attentively congestive in the group relinquished after 5 days (Fig.11). Clog vanishes totally following 14 days of treatment in both the liver and kidneys (Fig.12, 13). Control group gave a liver parenchyma protected

architecture made of Remack's radial spans joining towards a central vein and renal parenchyma with a saved architecture (Fig.14, 15). None of the rodents had heart or splenic injuries.

Fig.11 Liver parenchyma with discreetly congestive acute architecture

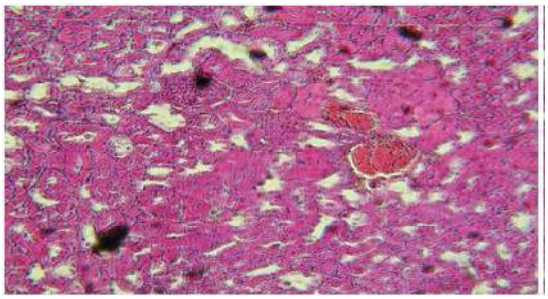

Fig.12 Renal parenchyma after 5 days of treatment with CCT fruit extract(200 mg/kg)

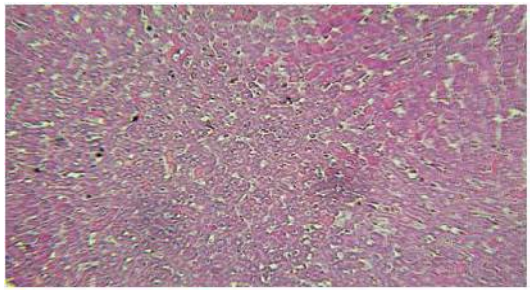

Fig.13 Hepatic parenchyma without any particularity

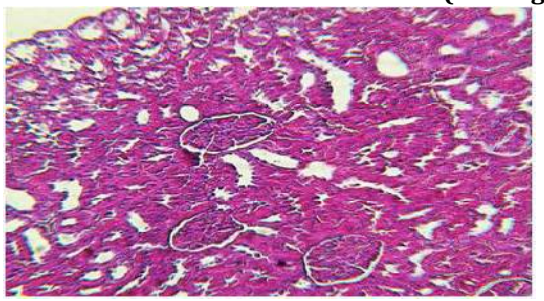

Fig.14 Normal or discreetly congestive kidney parenchyma after 14 days of the of CCT fruit extract(200 mg/kg)

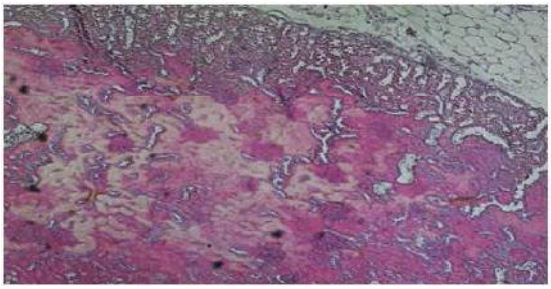

Fig.15 Preserved morphology of control rat liver

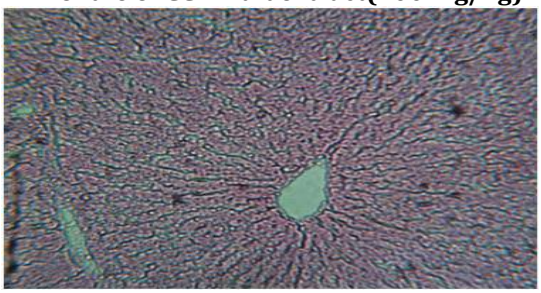

Fig.16 Normal kidneys without any congestion in control rats without any congestion

Toxicity study of stem extract of *Citrullus colocynthis*

Determination of LD50

The animals were held under perception for 72 hours subsequent to dosing to check for indications, behavioral changes and death. The dosages tested created, right on time inside the initial not many long periods of treatment the accompanying signs which intensifies with the passing of the time: loss of locomotion action, ataxia, and at the most noteworthy doses administered, laziness, hypothermia and demise. Extreme diarrhea was the most genuine indications; in the wake of creating it, all the exploratory animals sacrificed.

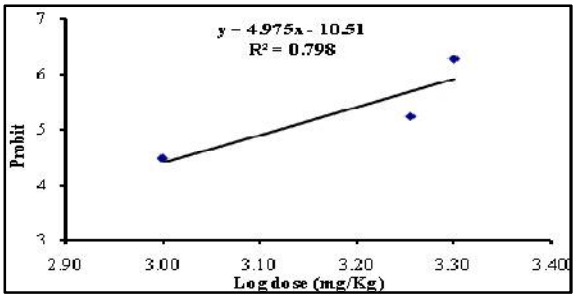

Fig 17. Median lethal dose (LD50) of the stem ethanolic extract of *C. colocynthis*in male Wistar rats

The animals that endure had a few side effects, including gentle diarrhea, however had the option to recuperate. The power of the harmful impacts was dose subordinate. Rodent's mortality after various portions of CCT stem ethanol extract was plotted against probability values. The death rate in male rodents was most extreme (100%) in groups treated with 300 mg/kg. The acute median lethal dose (LD50) of the concentrate was seen as separately 200 mg/kg (at 95% confidence cutoff of (1037, 80 to 1657, 27 mg/kg) for male rodents.(Fig. 16)

B) Acute oral toxicity study

The fundamental indications of toxicity saw after oral administration of single portion tested (200 mg/kg ≈ 1/10 LD50) were: diarrhea, unsettled hair, quickening of pulse, breathing trouble, soft feces and clustering together. None of the rodents in totally treated groups died the span of the examination. There were no factually noteworthy contrasts in normal body weight of the control group and *Citrullus colocynthis* stem extract treated groups during the intense toxicity. Yet, critical contrasts were recognized in the weight addition of male rodents treated C.C stem extract, when contrasted with control group. Macroscopic assessment of different organs in situ didn't show any morphological changes in organs of treated animals contrasted and those of control rodents. No critical changes were noted on the overall weights of liver, brain, kidney and testicles among the treated groups. Be that as it may, the group relinquished following 10 days of treatment has introduced a noteworthy decrease in the general loads of the kidney, lungs, heart and spleen contrasted with control group. The hematological parameters of rodents treated with *Citrullus colocynthis* stem extract are introduced in. The RBC, HCT (hematocrit), HGB (hemoglobin) and WBC were higher in the treated groups than control group. The major hematological parameters came back to typical after the fourteenth days.The outcomes of the indices of liver function AST (GOT) (glutamic-oxaloacetic transaminase), ALT (GPT) (glutamic-pyruvic) and ALP (alkaline phosphatase) and TP (total protein) are given. It was seen that the estimations of GOT and GPT on Day 1 and 5 were altogether higher contrasted with control. ALP activity was fundamentally higher in treated groups, relinquished following 10 and 14 days of application. TP estimation of the acute toxicity uncovered huge contrasts in the most treated groups contrasted with control.

Impact of CC stem extricate on kidney working parameters shows the mean kidney function estimations of the rodents treated with CC stem extract. From the table urea, creatinine and potassium levels were altogether expanded in groups A, C, and D when contrasted and the control. Calcium was fundamentally higher in groups A, B and C. no critical variety were gotten in the serum sodium and glucose tested when contrasted with control.

Toxicity study of fruit extract of *Citrullus colocynthis*

A) Determination of LD50

The LD50 of the fruit pulp concentrate of C. colocynthis plant was determined as 400 mg/kg.b.wt., separately.

Acute toxicity in male rats

The blood image of C. colocynthis treated albino rodents is appeared. The absolute RBC count was fundamentally higher in group A (7.86 ± 0.5 x 106/mm3) trailed by group B (7.33 ± 0.42 x 106/mm3) and afterward group C (5.26 ± 0.42 x 106/mm3) and those were altogether higher than that of the control one (5.0 ± 0.35 x 106/mm3). The pressed cell volume was higher in group A (52.5 ± 1.75 %) trailed by group B (50.63 ± 0.56 %) and afterward group C (45.86 ± 1.1 %) contrasted and that of the benchmark group (35.92 ± 1.83). The hemoglobin content was higher in group A (8.66 ± 0.19 g./dl) trailed by group B (7.93 ± 0.38 g./dl) at that point group B (7.15 ± 0.41 g./dl) contrasted and that of the control group (5.93 ± 0.32 g./dl).Total and differential leukocytic considers were variable as a part of the treated groups. Total WBC include was higher in group A (10.026 ± 1.22 x 103/mm3) trailed by group B (8.843 ± 0.95 x 103/mm3) and afterward group C (7.237 ± 0.75 x 103/mm3) contrasted with the control group (5.145 ± 0.47 x 103/mm3). As to leucocytic count, monocytes, lymphocytes, neutrophiles, band cells, eosinophiles and basophiles indicated profoundly huge increment in their numbers in treated groups contrasted and those in the control group. The impact of C. colocynthis fruit pulp extract on liver function biomarkers in serum of albino rodents after oral administration of ¼of the LD50 of C. colocynthis fruit pulp extract for 10 weeks was appeared in.SGPT and SGOT values indicated critical increment in completely treated rodents groups than the control group. The estimations of SGPT in Group A, Group B and Group C were 124 ± 30.2, 91.88 ± 10.5 and 74.75 ± 1.27 u/l, individually, while in the control group, it was (46.3±3.15 u/l). Essentially, the SGOT level was additionally expanded fundamentally in completely treated albino rodent groups contrasted with the control one. The mean estimations of SGOT in Group A, Group B and Group C were 275.63 ± 10.19, 225.88 ± 2.35 and 211.25 ± 27.9 u/l, individually, while it was (128.8 ± 3.39 u/l) in the control one. The mean degrees of ALP in the treated albino groups were fundamentally higher (122.25 ± 14.3; 197.0 ± 19.96 and 234.38 ± 18.99 ul/100 ml in Group A, Group B and Group C, separately) than that of the control rodents (32.12 ± 1.26 ul/100 ml). The impact of C. colocynthis extract on blood

glucose level showed that blood glucose level was diminished essentially in treated groups contrasted with the control one. The mean blood glucose levels were 11.63 ± 0.85; 8.24 ± 1.2 and 5.31 ± 0.86 mg/100 ml in Group A, Group B and Group C, individually, while it was 15.2 ± 0.92 mg/100ml in the control group. Furthermore, the all-out serum protein was likewise diminished fundamentally in completely treated groups (6.72 ± 0.36; 5.7 ± 0.32 and 5.68 ± 0.38 g/dl in Group A, Group B and Group C, individually) than that of the control group (7.28 ± 0.24 g/dl). Kidney work parameters in serum of albino rodents after oral administration of ¼ of the LD50 of *C. colocynthis* fruit pulp extract for 10 weeks were appeared in table 51. Serum urea of treated groups expanded altogether contrasted and that of the control one where they were 49.25 ± 1.22; 60.6 ± 1.64 and 78.25 ± 1.13 mg/100 ml in Group A, Group B and Group C, individually, and 37.1 ± 4.1 mg/100 ml in the control one. Serum creatinine was higher in the treated groups (13.85 ± 0.25; 15.04 ± 0.25 and 18.99 ± 0.19 mg/100 ml in Group A, Group B and Group C, individually) than that of the control one (10.12 ± 0.38 mg/100 ml).The ordinary normal level of serum Na+ in control rodents was 141.9 ± 2.1 mEq/L and expanded fundamentally (144.4 ± 0.62, 145.6 ± 1.78 and 146.6 ± 2.86 mEq/L) in Group A, Group B and Group C, individually. The concentration of serum K+ diminished fundamentally in the treated groups of rodents. Its level was 7.41 ± 0.87; 7.05 ± 0.28 and 6.48 ± 0.31 mEq/L in Group A, Group B and Group C, separately, while it was 7.94 ± 0.22 mEq/L in the control rodents.

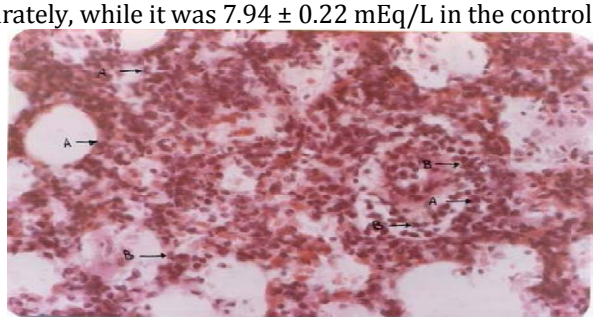

Fig.18 Lungs of albino rats orally administered the extract of *Citrullus colocynthis* fruit pulp extract showing interalveolar leukocytic infiltration mainly lymphocytes (arrow A) and eosinophils (arrow B).

In group B, the inspected lungs were hepatized in surface, red in shading with grayish-white spots on their surface and blood overflowed from the cut-surface. Minutely, diffuse thickening of between alveolar septa because of blockage of the alveolar vessels and leukocytic penetration especially lymphocytes and a couple of eosinophils were identified (Fig.18). Bronchi and bronchioles uncovered hyperplasia of the coating epithelial cells, desquamated cells with eosinophilic material in their lumen notwithstanding peribronchial lymphocytic hyperplasia. Diffuse perivascular edema with leukocytic accumulation for the most part lymphocytes and a couple of eosinophils had been watched. Some interstitial veins uncovered central hyalinization of the tunica media. In group C, compensatory alveolar emphysema had likewise been seen while focal thickening of the between alveolar septa because of lymphocytic penetration notwithstanding diffuse alveolar emphysema were seen group A.

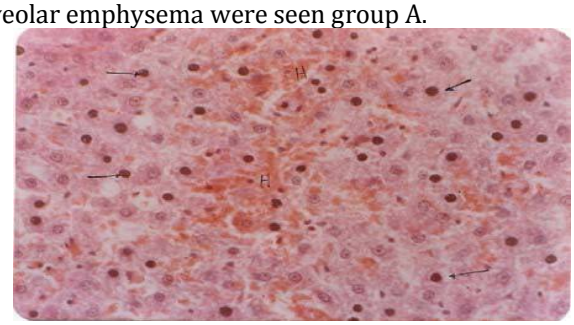

Fig 19. Liver of albino rats orally administered the extract of *Citrullus colocynthis* fruits showing coagulative necrosis in most hepatic cells (arrows) and interstitial haemorrhage (H).

In group B, the inspected liver was friable in consistency and blood overflowed from the cut-surface. The hepatic parenchyma contained grayish-white spots (½ - 1 mm in width) dissipated everywhere throughout the surface. Minutely, some hepatic lobules indicated individualization of the hepatic cells. A portion of these cells experienced necrobiosis and the others indicated coagulative necrosis. Various focal conglomeration of lymphocytes and macrophages supplanted the hepatic parenchyma were exceptional. Coagulative necrosis was confirmed in some hepatic cells by dark little colored nuclei (pyknosis) with cytoplasmosis. Different hepatocytes endured necrobiosis as shady swelling and vacuolar degeneration. Blockage of the central veins and sinusoids and slight conglomeration of lymphocytes close to central veins with central haemorrhage had been taken note. Hypertrophied Kupffer's cells were seen. Different

central interstitial haemorrhage (Fig. 19) and severe clog of central veins and sinusoids had additionally been seen in group C. In group A, the portal tract demonstrated leukocytic penetration essentially lymphocytes. Diffuse coagulative necrosis of every single hepatic cell was confirmed by little dark colored nuclei (pyknotic nuclei) with cytoplasmosis notwithstanding histopathological modifications happened in group A.

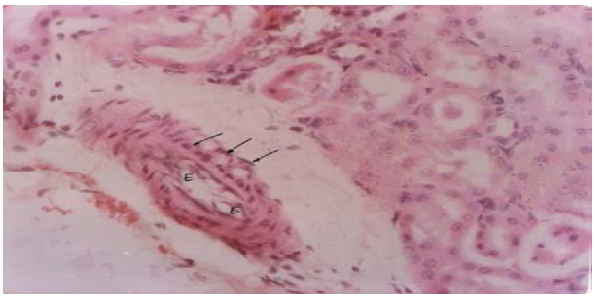

Fig. 20 Kidney of albino rats orally administered the extract of *Citrullus colocynthis* fruit pulp showing endothelosis of the *tunica intema*(E) with vacuoles in the *tunica media* (arrows)

Visibly, the analyzed kidneys were broadened in size, friable in consistency with grayish-white spots on the renal parenchyma in bunch B. Infinitesimally, leukocytic penetration in the middle of renal tubules chiefly lymphocytes and hardly any eosinophiles had been recognized. The renal epithelium of the proximal and distal convulated tubules experienced coagulative necrosis confirm by pyknotic nuclei with cytoplasmosis and shady swelling other than hydropic degeneration. Perivascular edema particularly around the huge between cylindrical veins and albuminous material (eosinophilic material) in the lumen of some renal tubules were overwhelming. In group C, perivascular haemorrhage and hyperplasia of the tunica intema (endothelosis) with vacuoles in the tunica media in some vessels (Fig.20) were likewise watched while in group A, perivascular edema and haemorrhage were predominant.

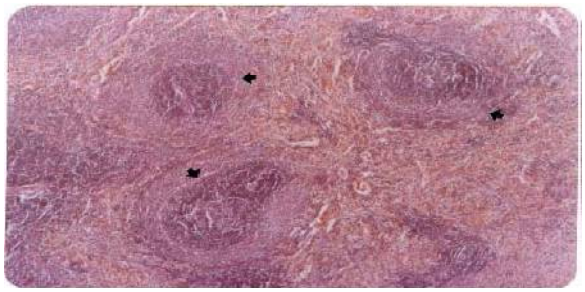

Fig.21 Spleen of albino rats orally administered the extract of *Citrullus colocynthis* fruit pulp showing hyperplasia of the white pulp (arrows).

In totally treated groups, the analyzed spleen was obviously typical while minutely, predominant hyperplasia of the white pulp was observed (Fig. 21).

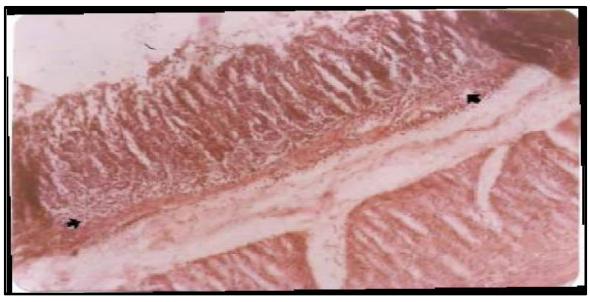

Fig.22 Stomach of albino rats orally administered the extract of *Citrullus colocynthis* fruit pulp showing desquamation of the epithelial cells with exudatein the lumen (thin arrow) and leukocytic infiltration in the *lamina propria*(thick arrow)

Macroscopically, the inspected stomach and digestive tract were obviously ordinary in completely treated groups. Infinitesimally, desquamated epithelial cells with slight exudate in the lumen notwithstanding serious leukocytic penetration in the lamina propria predominantly lymphocytes were watched (Fig.93). The digestive system demonstrated total desquamation of the intestinal villi, lymphocytic penetration in the lamina propria and eosinophilic material in the sub-mucosal layer (Fig.23).

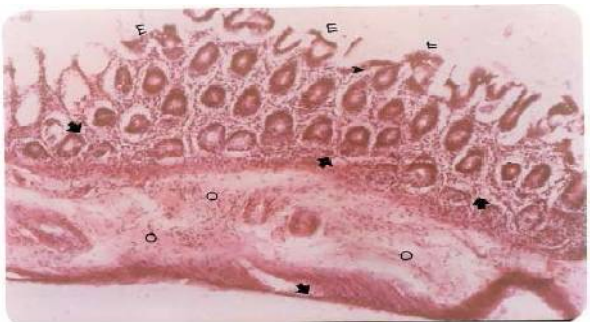

Fig.23 Intestine of albino rats orally administered the extract of *Citrullus colocynthis* fruit pulp showing complete desquamation of intestinal villi (E), lymphocytic infiltration in *lamina propria*(arrows) and edema in submucosal layer (O).

CONCLUSION

Toxicity study of leaf extract of *Citrullus colocynthis*

The animals were held under close perception for 72 hours in the wake of dosing to check for side effects, conduct changes and demise. The dosages tried created, right on time inside the first hour of treatment the accompanying signs, which exacerbates with the death of the time: loss of locomotion action, ataxia and at the most elevated doses administered, in appetence, flaw of the hind limbs, laziness, hypothermia, cyanosis and in the end demise. The principle indications of toxicity saw after oral administration of single portion tried (440 mg/kg ≈ 1/3 DL50) were: diarrhea, unsettled hair, speeding up of pulse breathing trouble, soft defecation and clustering together toward one side of the cages.

Toxicity study of Stem extract of *Citrullus colocynthis*

The animals that endure had a few side effects, including gentle diarrhea, however had the option to recuperate. The power of the harmful impacts was dose subordinate. The death rate in male rodents was most extreme (100%) in groups treated with 300 mg/kg.The fundamental indications of toxicity saw after oral administration of single portion tested (200 mg/kg ≈ 1/10 LD50) were: diarrhea, unsettled hair, quickening of pulse, breathing trouble, soft feces and clustering together. None of the rodents in totally treated groups died the span of the examination.

Toxicity study of fruit extract of *Citrullus colocynthis*

SGPT and SGOT values indicated critical increment in completely treated rodents groups than the control group. As to leucocytic count, monocytes, lymphocytes, neutrophils, band cells, eosinophiles and basophiles indicated profoundly huge increment in their numbers in treated groups contrasted and those in the control group. The impact of C. colocynthis fruit pulp extract on liver function biomarkers in serum of albino rodents after oral administration of ¼of the LD50 of C. colocynthis fruit pulp extract for 10 weeks was appeared.

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CONFLICT OF INTEREST

Authors having no any conflict of interest.

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