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ROLE OF *CAPPARIS SPINOSA* LEAVES IN AMELIORATING TRICHLOROACETIC ACID INDUCED HEMATOTOXICITY IN SWISS ALBINO MICE

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Abstract:-

The present study was amied to evaluate the effect of leaves powder of Capparis spinosa which using in Libya as folk medicine for many ailments as cancer treatment against morphological alterations in blood and bone marrow cells in the blood and bone marrow smears of male mice intoxicated with trichloroacetic acid (TCA). A hundred male mice were divided in to 5 groups. Group I was considered as a control and received only distilled water, group II treated orally with honey (40 mg/kg bw) for 3 weeks, group III treated orally with a mixture of C. spinosa leaves powder and honey(40 mg/kg bw) for 3 weeks, group IV treated orally with TCA in drinking water (500mg/kg bw for 3 and 6 weeks, then left for 3 weeks for self-recovery), group V was given TCA in drinking water (500mg/kg bw) for 6 weeks, then treated with a mixture of C. spinosa and honey for 3 weeks. Blood and bone marrow samples were collected for the smears preparations and evaluated morphological alterations in blood and bone marrow cells. Administration of TCA only in drinking water showed many abnormalities in blood and bone marrow smears which were more pronounced in TCA for 6 weeks and TCA recovery groups. Administration of mixture of leaves powder of C. spinosa and honey to TCA intoxicated mice lessened most abnormalities (anisocytosis and poikilocytosis) in blood and bone marrow smears. Result of the present work suggested that the dose of leaves powder of C. spinosa used in this investigation have no hematotoxicity and have some beneficial effect to inhibition TCA induce hematotoxicity in mice.

Key words*: Capparis spinosa, trichloroacetic acid, blood bone marrow smear, mice.*

INTRODUCTION

Capparis spinosa L. family Capparidaceae is well known with its common name Kabbar or Capper in different countries and it is one of the most common aromatic plants growing in wild dry regions around the west or central Asia and the Mediterranean basin [1, 2]. It has been known for centuries in traditional phytomedicine which exploited its properties for several purposes as anti-inflammatory [3, 4] and anticarcinogenic activity [5]. Caper plant has been used in folk medicine from ancient times due to their strong antioxidant properties, and high phenolics and flavonoid contents [6, 7]. Capers has been used traditionally for the curing of various human ailments including gastro-intestinal problems, inflammation, anemia, liver dysfunction, rheumatism, heart disease, kidney disorders, and diuretic [6, 8]. Also, Capper is used in phytomedicine as anti-oxidative and was found to induce an increase in both enzymatic and non-enzymatic antioxidant levels observed [9]. Hematological profile converted to more or less normal levels in Dalton's ascites lymphoma (DAL) tumor bearing mice after treated with methanol extract of *Capparis sepiaria* (MECS) at the doses of 200 and 400 mg/kg body weight per day for 14 days, also after treatment with MECS antioxidant levels increased significantly. The results indicate that MECS exhibited significant antitumor activity in DAL-bearing mice [10]. In the chronic toxicity study, the haematological parameters (hemoglobin concentration, clotting time, neutrophils, easinophils, lymphocytes, monocytes, red and white blood cells) in the rats treated orally with ethanolic extract of *Capparis aphylla Roth* (EECA) at doses of 100, 200 and 400 mg/kg once in a week for 6 weeks did not differ significantly from that of the control group and all the values remained within normal limits throughout the experimental period, indicating that the EECA was not toxic to the circulating red cells, nor interfered with their production [11]. On the other hand hematological changes indicated the development of hypochromic erythrocytopaenia in goats were given daily oral doses ranging from 0.05 to 5 g per kg per day of the dried leaves of *C. tomentosa* and died or were killed at various times after dosing [12]. Trichloroacetic acid (TCA) is a colorless to white crystalline solid with a sharp, pungent odor [13] formed from organic material during water chlorination [14] and has been detected in groundwater, surface water distribution systems, swimming pool water and drinking water [15]. TCA was detected in vegetables, fruits, and grains [16] and can be taken up into foodstuffs from the cooking water [17]. Therefore, human exposure to TCA can also occur via food tap water consumption. TCA administered at dose level 2,000 ppm (300 mg/kg-day) in drinking water for 50 days significantly increased serum AST, ALT that probably resulted from damage to liver cells [18]. Further, **Celik** [18] found that TCA treatments caused different effects on antioxidant defense system and lipid peroxidation in various tissues of rats at the end of the TCA treatment. In the literature, it is reported that TCA exposure caused different effects on gross and microscopic examinations, serum chemistry, hematology and biochemical analysis [19, 20]. It was observed that TCA caused histological alterations in the liver such as centrilobular necrosis, vacuolation in hepatocytes and loss of hepatic architecture [21]. Moreover, degeneration of renal tubules in rats treated with TCA dose level 3.8 mg/kg-day for 10 weeks was evident [21]. It was found that oral administered of TCA (2000 ppm) for 52 days consecutively caused a significant disturbances in hematological parameters and led to hematotoxic in rat [19]. **Mather *et al.*** [22] reported that TCA and DCA (dichloroacetic acid) produced substantial systemic organ toxicity to the liver and kidney during a 90-day sub-chronic exposure. An increase in incidence of benign and malignant liver tumors was observed in mice orally administered trichloroethylene. The IARC has concluded that there is sufficient evidence in experimental animals for the carcinogenicity of trichloroethylene [23].

MATERIAL AND METHODS

Experimental animals:

Healthy adult male Swiss albino mice (*Mus-musculus*) 8 to 10 weeks old and weighing 22±4 gm were obtained from the animal breeding house of Faculty of Veterinary Medicine, Omar Al-Mukhtar University, Al-Bayda-Libya. They were housed in the laboratory animal room in clean plastic cages (10 mice/ cage) under controlled conditions of temperature (20 ± 2)°C and photoperiod (14 h light : 10 h dark) cycle. The animals were maintained on standard commercial pellet diet and clear drinking water and allowed a free access to food and water. The mice were acclimatized for 1 week prior to the start of experiments.

Material used:

Fresh plants of *C. spinosa* were collected from Blgray region Algabal Alakhder in Al-Bayda-Libya between March and April 2012. The plant was authenticated by Department of Botany, Faculty of Science, Omar Al-Mukhtar University, Al-Bayda-Libya.

All unwanted materials like stems, flowers, roots or stones were removed from the leaves. The plants were cleaned, air-dried and then powdered mechanically. Natural bees honey (vehicle) used in this study was purchased from the local honey market in Al-Bayda-Libya. The honey was collected form beehives built on Algabal Alakhder-Libya. This honey is also locally known as Seder honey. It was filtered to remove solid particles.

Preparation of the mixture of *Capparis spinosa* and honey:

Leaves powder of *C. spinosa* (400 mg) were well mixed with 40 gm of seder honey and used at dose level 40 mg/kg bw (0.1 ml/mouse) (equivalent to dose used by a human weighing 70 kg in traditional medicine) the mixture of *C. spinosa* leaves powder and honey was prepared according to the prescriptions given by traditional healers in Libya. A dose was determined according to **Paget and Barnes** [24].

Trichloroacetic acid (TCA)
Was purchased from (Sigma Co, Germany). Mice were given TCA trichloroacetic acid in drinking as 500 mg/kg for 3 and 6 weeks. TCA was chosen because it has been reported to induce toxicity in mice [25,26].

Fig. (1) : Capparis spinosa

Experimental Design:
A hundred male mice were divided into 5 groups of 20 mice each and subjected to the following treatments:
Group I (control group):
Healthy adult male mice received distilled water at dose level 4 ml/kg by oral gavage for 3 and 6 successive weeks and served as negative control (untreated control group).

Group II (vehicle group): Healthy adult male mice were given orally by oral gavage natural bees honey at dose level 4 ml/kg for 3 successive weeks.

Group III (*C. spinosa* and honey group):
Healthy adult male mice were given orally by oral gavage mixture of *C. spinosa* leaves powder and honey at dose level 40 mg/kg bw suspended in 0.1 ml honey once per day for 3 successive weeks.

Group IV (TCA group): Healthy adult male mice were received TCA at dose level 500 mg/kg body weight in drinking water for 3 and 6 successive weeks (Doses were estimated based on default drinking water intake values for mice). After the end of the experimental period the animals in this group left for recovered and known as **recovery group.**

Group V (*C. spinosa* and honey with TCA group) (Treatment group): Healthy adult male mice were received TCA at dose level 500 mg/kg bw in drinking water for 6 successive weeks then treated orally by oral gavage with mixture *C. spinosa* and honey at dose level 40 mg/kg bw once per day for 3 successive weeks.

Preparations and staining blood and bone marrow smears
Twenty four hours after the end of experimental period, un-anesthetized mice from both control and experimental groups were sacrificed by cervical dislocation. Peripheral blood samples from the neck blood vessels and bone marrow from both femurs were collected. Peripheral blood and bone marrow smears were prepared and stained with Giemsa stain according to **Lewis *et al*.** [27]. Stained blood and bone marrow smears were examined under light microscope and the alterations were recognized and photographed using Nikon Eclipse E400, Japan with camera head. In bone marrow preparations at least 500 cells per slide were counted to investigate morphological alterations in erythroid, myeloid, megakaryocytic series and myeloid/erythroid ratio and selected sections were photographed.

RESULTS AND DISCUSSION
Examination of blood smears of control mice (Figs.2-4) revealed that erythrocytes (red blood corpuscles RBCs) are the most common type of blood cells. They are a nucleated, disc shape with smooth contour and showed pale center and peripheral condensation hemoglobin. Negligible numbers of pikilocytosis cells were noticed in blood smears of control mice. Leucocytes (White blood cells WBCs) with normal nuclear appearance and distinct cytoplasm were seen. Platelets (Thrombocytes) are cell fragments that derive from large bone marrow cells called megakaryocytes. Platelets appeared as minute disk shape exhibited peripheral light blue stain cytoplasm hyalomere and central purple granules granulomere. Light microscopic examination of bone marrow smears of control mice (Figs.5 and 6) showed unorganized mixture of hematopoietic cells in different stage of maturation include small size erythroid elements with condensed nuclear chromatine, myeloid elements with relatively large size and pleomorphic nuclei and large megakaryocytes with large irregular nuclei and homogeneous cytoplasm.

The bone marrow is the major hematopoietic organ, and a primary lymphoid tissue, responsible for the production of erythrocytes, granulocytes, monocytes, lymphocytes and platelets [28]. Blood and bone marrow is one of the largest organs in the body and is an important potential target organ of chemical exposure [29]. Assessments of the blood and bone marrow have become routine procedures in the investigation of hematologic disorders in toxicology and safety assessment studies [30]. The hematopoietic tissue consists of a variety of cell types including, the blood cells and their precursors, adventitial/barrier cells, adipocytes, and macrophages. The hematopoietic tissue cells are not randomly arranged but demonstrate a particular organization within the tissue [31]. Hematopoietic tissue is also sensitive to external

influences and can become suppressed in response to dietary restriction, malnutrition, chronic inflammation, toxicity, and proliferative or neoplastic disorders [32, 33, and 34].

The examination of blood smears of mice treated with honey revealed no abnormalities in RBCs and WBCs. Also, platelets with normal appearance were seen (Fig. 7). However, normal equilibrium state between myeloid and erythroid elements and megakaryocytes with normal appearance were observed in bone marrow smears of mice treated orally with honey for 3 weeks (Fig. 8). Blood smears of mixture of *C. spinosa* leaves and honey treated group showed RBCs with slight anisocytosis and poikilocytosis including target cells (Fig. 9). However, examination of bone marrow smears of mixture of *C. spinosa* leaves powder of and honey revealed nearly normal cellularity (erythriod/myeloid ratio) and normal appearance of megakaryocytes (Fig. 10). Similarly, hematopoiesis and leucopoiesis were also not affected in rats treated orally with ethanol extract of *Capparis aphylla Roth* at doses of 100, 200 and 400 mg/kg once in a week for 6 weeks [11], even though the hematopoietic system is one of the most sensitive targets for toxic compounds [35] and an important index of physiological and pathological status in man and animals [36]. Administration of TCA only in drinking water showed many abnormalities in blood and bone marrow smears which were more pronounced in TCA for 6 weeks and TCA recovery groups. The examination of blood smears demonstrated that administration of TCA induced many poikilocytosis in RBCs appeared in form target cells (codocytes), crenation, elliptocytes and rouleaux appearance of RBCs with prominent hypochromasia and anisocytosis. Also, aggregation of blood platelets and increase precursor of white blood cells were noticed. An activation of hemopoiesis and increase of megakaryoblastes and megakaryocytes were also observed in bone marrow smears of male mice treated with TCA only for 3 or 6 weeks (Figs. 11-14, 15 a-15d and 16 a, b). Blood smears of TCA recovery group showed also hypochromic of RBCs and many poikilocytosis include target cells and rouleaux appearance of red blood corpuscles was remain observed. Many WBCs showed abnormal nuclear feature as less condensed chromatin. However, decrease in aggregation of blood platelets and precursor of WBCs were noticed (Fig. 17). Bone marrow smears of TCA recovery group showed nearly normal equilibrium state between myeloid and erythroid elements, however an activation of hemopoiesis and destructed of some myeloid elements were seen in bone marrow smears of some animals (Fig. 18). From the results of above study it can be concluded that TCA exposure can severely damage heme synthesis and alter the RBCs and WBCs. The alterations in hematological changes serve as the earliest indicator of toxic effects on tissue [37]. **Ferzand *et al*. (2008)** reported that toxic substance can bind to animal and human hemoglobin and other blood cell proteins. This binding might cause distortion of blood cell shape/morphology [38]. Anemia may result when the cell membranes of RBCs become more fragile as the result of damage to their membrane [39]. It was found that administration of mixture of *C. spinosa* and honey to TCA intoxicated mice lessened most abnormalities in blood and bone marrow smears. Blood smears of male mice treated with TCA and mixture of *C. spinosa* leaves and honey (Treatment group) showed ansocytosis hypochromic of RBCs with obvious diminution of poikilocytosis (Fig.19) compared to TCA only treated groups. Bone marrow smears of treatment group showed normal appearance of hemopoietic cells with slight decrease in erythroid elements. Megakaryocytes with normal appearance was also seen (Fig. 20). This study clearly demonstrates that exposure to TCA is extremely hazardous in causing alterations in the peripheral RBCs. **Nikinmaa** [40] suggested that toxic materials directly or indirectly damage the membrane structure, ion permeability and cell metabolism of erythrocytes thus may cause morphologically damaged erythrocyte formation. Structural defects and changes in surface shapes of erythrocytes have been reported by [41]. A change in the composition of hemoglobin inside red blood cells membrane was observed which affects the physiological function of and capacity to transport oxygen, as observed during decrease in hemoglobin concentration [42]. Erythrocytes are the most abundant cells in the human body possess desirable physiological and morphological characteristics [43]. Oxidants produce alterations in erythrocyte membranes as manifested by decreased cytoskeletal protein content [44]. Since TCA was found to be lessened antioxidant defense system and induced oxidative stress in various tissues of rats [18]. TCA treatment also resulted in dose-dependent increases in lipid peroxidation production at doses ranging between 77-410 mg/kg/day in the 4-13 weeks treatment periods in liver tissue of mice [45]. The ability of different phenolic substances to scavenge various types of oxidation-initiating radicals has been reported in the polar phase [46]. It was demonstrate that binding of the flavonoids to the RBC membranes significantly inhibits lipid peroxidation, and at the same time enhances their integrity against lyses [47]. This may be explain less RBCs abnormalities in blood smears of mice intoxicated with TCA and treated with mixture of *C. spinosa* and honey (Treatment group). Since Caper is very good sources of glucosinolates, flavonoids (rutin, kaempferol), and quercetin phenolic acids, alkaloids [48][49][50]. These compounds are powerful antioxidants. Furthermore, rutin strengthen capillaries and inhibits platelet clump formation in the blood vessels [49]. These findings may be explain obvious diminution of abnormalities in the blood (specially aggregation of blood platelets) and bone marrow smears of mice intoxicated with TCA and treated with mixture of leaves powder of *C. spinosa* and honey (treatment group). In the present work administration of TCA induced disturbance in WBCs. Leukocytes are the first line of cellular defence that respond to infectious agents, tissue injury, or inflammatory process [51] and immune processes [52] lead us to suspect that TCA may affect these functions. Exorbitance compounds (such as quercetin) are flavonoid antioxidant which existing in the *C. spinosa* may reduce the negative effects of oxidants and control cell/ tissue damages [53]. These flavonoids not only play a direct role as ROS scavengers but also inhibit the enzymes involved in the production of these oxidative substances protecting the DNA against the toxic effects of these compounds [54]. Since lipid peroxidation, a complex radical chain reaction leading to oxidation of cell membrane lipids, is considered a critical mechanism of injury that occurs in cells during oxidative stress [55]. Flavonoids are phytophenolic compounds with strong antioxidant effects that function against oxidative stress [56].

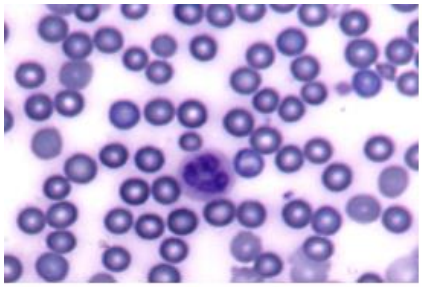

Fig. (2): Blood smear of mouse from control group showing red blood corpuscles with normal peripheral condensed hemoglobin and pale central pallor, Neutrophil with lobulated nucleus (Giemsa stain, X1000).

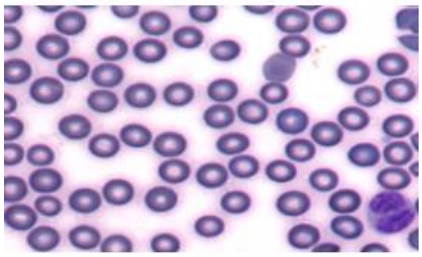

Fig. (3): Blood smear of mouse from control group showing normochromatic anucleated RBCs with central pallor and smooth contour, Eosinophil with bilobed nuclus (Giemsa stain, X1000) .

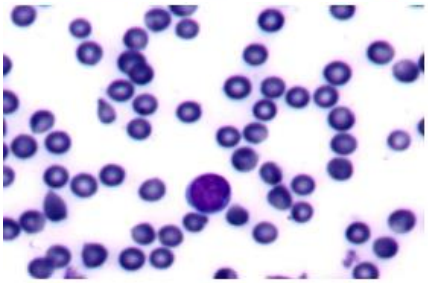

Fig. (4): Blood smear of mouse from control group showing normochromatic red blood corpuscles RBCs with pale central pallor and smooth contour , Platelets with peripheral light blue stain hyalomere and central purple granules granulomere , Lymphocyte(Giemsa stain, X1000).

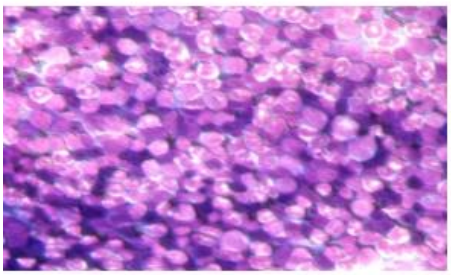

Fig. (5): Bone marrow smear of male mouse from control group showing normal equilibrium state between normal small dark erythroid elements and myeloid elements in different stage of maturation (Giemsa stain, X400).

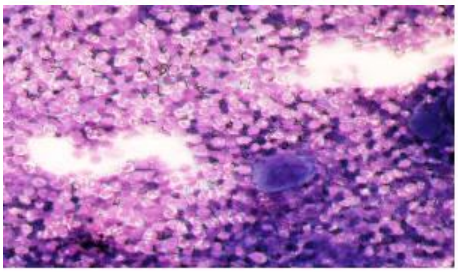

Fig. (6): Bone marrow smear of female mouse from control group showing hematopoietic cells in different stage of maturation and large megakaryocytes with normal appearance. (Giemsa stain, X400).

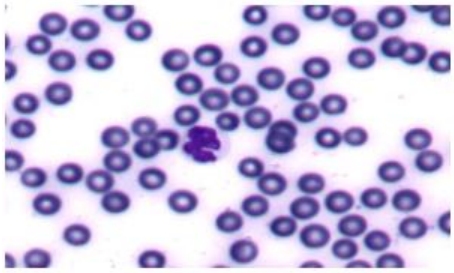

Fig. (7): Blood smear of male mouse treated with honey showing normal red blood corpuscles RBCs, Neutrophil with lobulated nucleus (Giemsa stain, X1000).

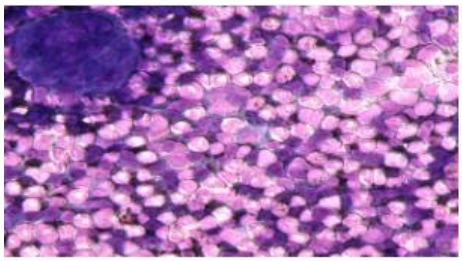

Fig. (8): Bone marrow smear of male mouse treated with honey for 3 weeks exhibiting normal erythroid elements, myeloid elements and Megakaryocytes. (Giemsa stain, X400).

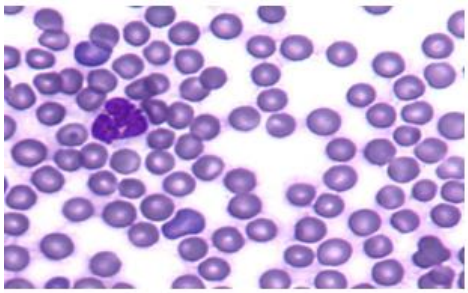

Fig. (9): Blood smear of male mouse treated with mixture of leaves powder of *Capparis spinosa* and honey showing normal red blood corpuscles RBCs with slight anisocytosis Neutrophil with lobulated nucleus (Giemsa stain,X1000).

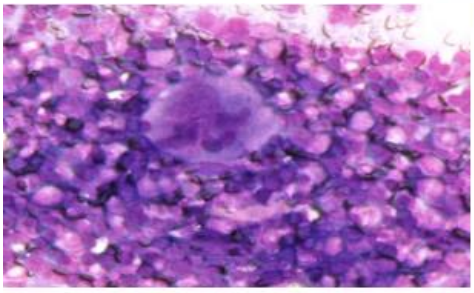

Fig. (10): Bone marrow smear of male mouse treated with mixture of leaves powder of *Capparis spinosa* and honey for 3 weeks showing normal erythroid and myeloid elements; Megakaryocytes (Giemsa stain, X400).

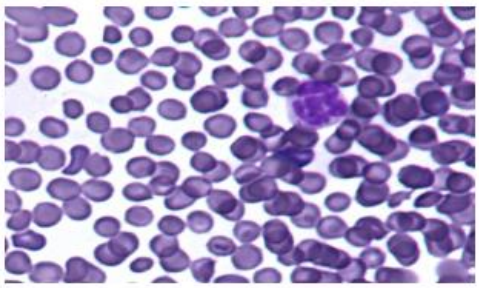

Fig. (11): Blood smear of male mouse treated with aqueous extract of leaves of *Capparis spinosa* showing anisocytosis red blood corpuscles RBCs and few poikilocytosis RBCs, Neutrophil with lobulated nucleus (Giemsa stain, X1000).

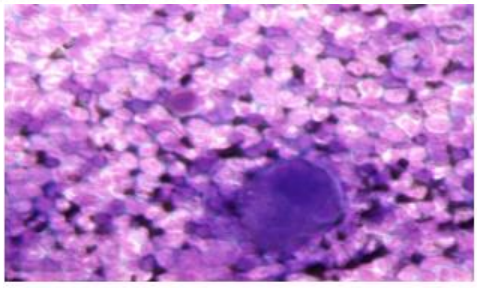

Fig. (12): Bone marrow smear of male mouse treated with aqueous extract of leaves of *Capparis spinosa* for 3 weeks showing few decrease and less stained erythroid elements; Megakaryocytes. (Giemsa stain, X400).

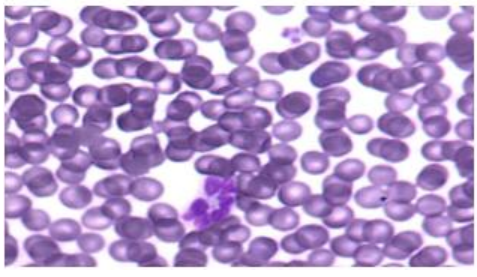

Fig. (13): Blood smear of male mouse treated with TCA at dose level 500mg/kg in drinking water for 3 weeks showing hypochromic red blood corpuscles RBCs, Neutrophil with lobulated nucleus (Giemsa stain,X1000).

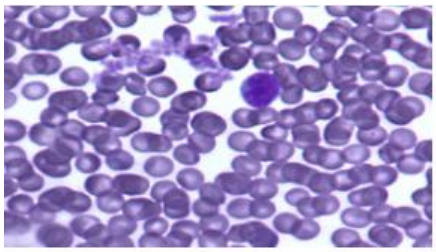

Fig. (14): Blood smear of male mouse treated with TCA for 3 weeks exhibiting aggregation of blood platelets (Giemsa stain,X1000).

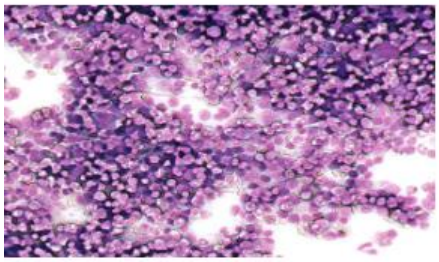

Fig. (15): Bone marrow smear of male mouse treated with TCA for 3 weeks showing an activation of hemopoiesis. (Giemsa stain, X200).

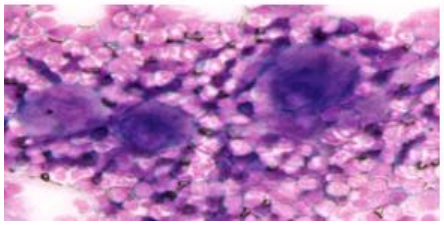

Fig. (16): Bone marrow smear of male mouse treated with TCA for 3 weeks showing increase of megakaryocytes (Giemsa stain, X400).

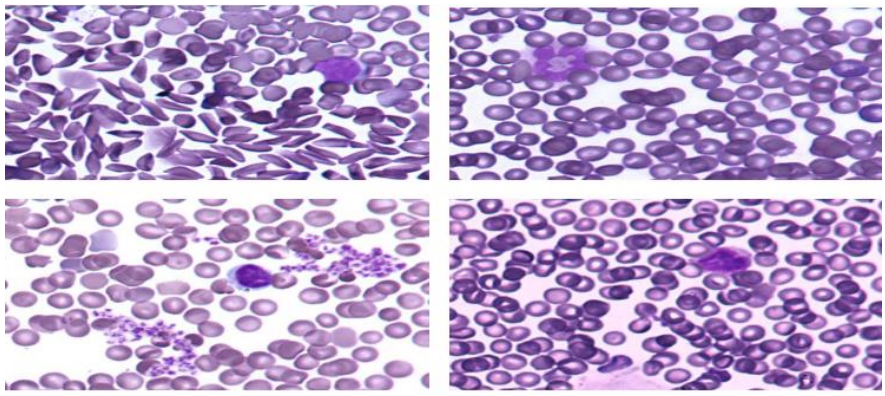

Fig.(17a,b,c,d):Blood smear of male mouse treated with TCA at dose level 500mg/kg in drinking water for 6 weeksshowing hypochromic poikilocytosis of red blood corpuscles include elliptocytes and target cells rouleaux appearance of red blood corpuscles, aggregation of blood platelets; Lymphocytes; precursor of white blood cells. (Giemsa stain, X1000).

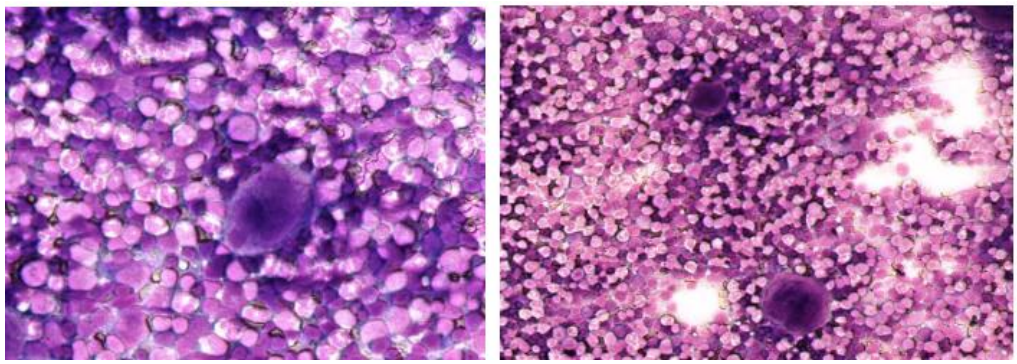

Fig.(18a,b):Bone marrow smear of male mouse treated withTCA at dose level 500mg/kg in drinking water for 6 weeksshowing anactivation of hemopoiesis with the increase of megakaryocytes.(Giemsa stain,X200and 400).

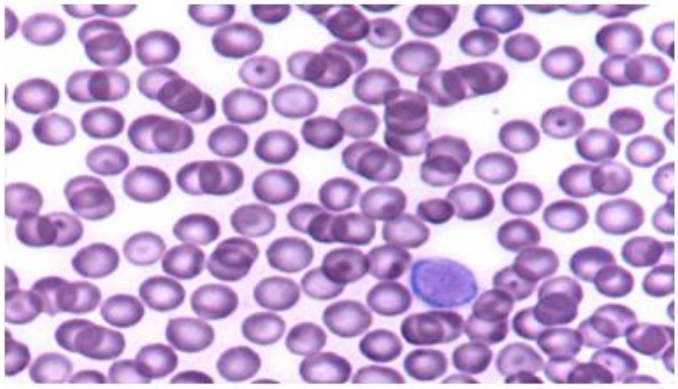

Fig.(19):Blood smear of male mouse treated with TCA at dose level 500mg/kg in drinking water for 6 then left for recovery for 3 weeks TCA (recovery group)showing hypochromic red blood corpuscles RBCs with poikilocytosis, Lymphocyte showing nucleus with less condensed chromatin(Giemsa stain,X1000).

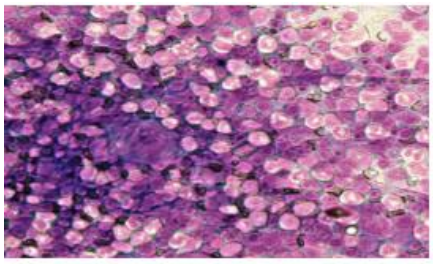

Fig. (20): Bone marrow smear of male mouse of recovery group exhibiting nearly normal equilibrium state between myeloid and erythroid elements. Megakaryocytes (Giemsa stain, X400).

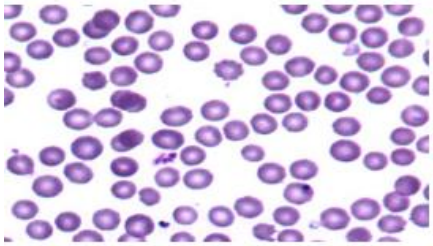

Fig. (21): Blood smear of male mouse intoxicated with TCA at dose level 500mg/kg in drinking water for 6 then treated with mixture of leaves powder of *Capparis spinosa* and honey for 3 weeks(Regeneration group) showing hypochromic ansocytosis red blood corpuscles RBCs with diminution of poikilocytosis, Lymphocytes (Giemsa stain, X1000).

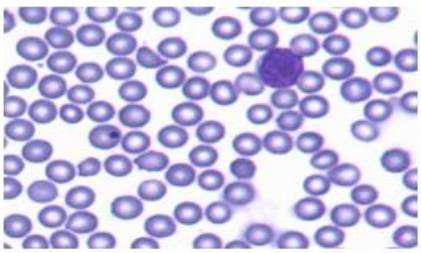

Fig. (22): Blood smear of male mouse intoxicated with TCA at dose level 500mg/kg in drinking water for 6 then treated with mixture of leaves powder of *Capparis spinosa* and honey for 3 weeks(Regeneration group) showing hypochromic ansocytosis red blood corpuscles RBCs with diminution of poikilocytosis, Lymphocytes (Giemsa stain, X1000).

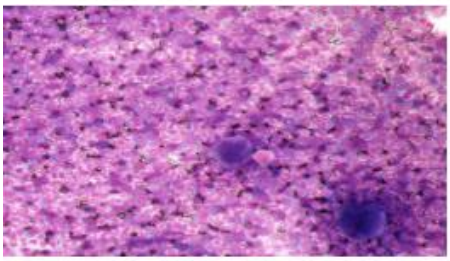

Fig. (23): Bone marrow smear of male mouse intoxicated with TCA at dose level 500mg/kg in drinking water for 6 then treated with mixture of leaves powder of *Capparis spinosa* and honey for 3 weeks(Regeneration group) exhibiting slight decrease in erythroid elements; Megakaryocytes (Giemsa stain, X200).

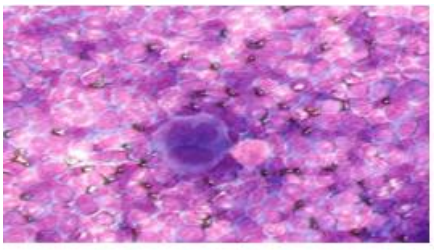

Fig. (24): Bone marrow smear of male mouse intoxicated with TCA at dose level 500mg/kg in drinking water for 6 then treated with mixture of leaves powder of *Capparis spinosa* and honey for 3 weeks(Regeneration group) exhibiting slight decrease in erythroid elements; Megakaryocytes (Giemsa stain, X400).

CONCLUSION

Administration of mixture of leaves powder of *C. spinosa* and honey to TCA intoxicated mice lessened most abnormalities in blood and bone marrow smears. Result of the present work suggested that at the dose used in this work have no hematotoxicity and have some beneficial effect to inhibition TCA induce hematotoxicity. Also, the present investigation

recommended the mixture of leaves powder of *C. spinosa* and honey as traditional medicine for hematopoietic toxicity. However further more investigation must be done.

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REFERENCES

[1].Al-Snafi, A.E. (2015). The chemical constituents and pharmacological effects of Capparis spinosa - An overview. Indian Journal of Pharmaceutical Science and Research, 5(2):93100.

[2].Tlili , N., Elfalleh, W., Saadaoui, E., Khaldi, A., Triki, S. and Nasri, N. (2011). The caper (Capparis L.): Ethnopharmacology, phytochemical and pharmacological properties. Fitoterapia, 82(2):93-101.

[3].Al-Said, M.S., Abdelsattar, E.A., Khalifa, S.I.and El-feraly, F.S. (1988). Isolation and identification of an anti-inflammatory principle from Capparis spinosa. Pharmazie, 43: 640-641.

[4].Boumerfeg, S., Ameni, D., Adjadj, M., Djarmouni, M., Khennouf, S., Arrar, L. and Baghiani, A. (2012). Antihymolyticc and Antioxidant Effects of Medicinal Plant Capparis spinosa L. Journal of Life Sciences, 6: 637-643.

[5].Tlili, N., Nasri, N. Saadaoui, E., Khaldi, A., Triki, S. (2009). Carotenoid and tocopherol composition of leaves, buds, and flowers of Capparis spinosa grown wild in Tunisia. J Agric Food Chem .,57(12):5381-5385.

[6].Duman, H., Canatan,D., Alanoglu,G., Sutcu,R. and Nayir.T.(2013). The antioxidant effect of Capparis ovate and deferasirox in patients with Thalassemia major. J.Blood DisordersTransf., 4:1-4.

[7].Zuo, Y. (2014).High-Performance Liquid Chromatography (HPLC): Principles, Procedures and Practices. Nova Science Publishers, Inc. New York.

[8].Sher, H. and Alyemeni, M. (2010). Ethnobotanical and pharmaceutical evaluation of Capparis spinosa L, validity of local folk and Unani system of medicine. J. Med. Plant. Res., 4(17):1751-1756.

[9].Kamalakkannan, N. and Prince, P.S.(2006). Antihyperglycaemic and antioxidant effect of rutin, a polyphenolic flavonoid, in streptozotocin-induced diabetic wistar rats. Basic Clin Pharmacol Toxicol., 98:97-103.

[10]. Sreenivas, S.A., Venu Gopal, Y., Ravindranath, A., Kalpana, G. and Rajkapoor, B. (2012). Antitumor and Antioxidant Activity of Capparis sepiaria Against Dalton's Ascites Lymphoma in Rodents. Academic J. Cancer Res., 5(2):46-52.

[11]. Bhanu Prasad K,. (2011). Acute and chronic toxicity studies of ethanolic extract of Capparis aphylla Roth. International Journal of Advanced Pharmaceutics,1(1):35-39.

[12]. Ahmed, O. M. M., Adam, S. E. I. (1980) .The toxicity of capparis tomentosa in goats. Journal of Comparative Pathology, 90(2):187-195.

[13]. NIOSH. (National Institute for Occupational Safety and Health) (2003). NIOSH pocket guide to chemical hazards. (97-140). Cincinnati, OH. http://www.cdc.gov/niosh/npg/npgdcas.html.

[14]. IPCS. (International Programme on Chemical Safety). (2000). Disinfectants and disinfectant by-products. In Environmental Health Criteria, Geneva, Switzerland: World Health Organization.

[15]. International Agency for the Research on Cancer (IARC) (2004). Some drinking-water disinfectants and contaminants, including arsenic. IARC Monogr Eval Carcinog Risks Hum, 84:1-477.

[16]. Reimann, S.,Grob, K. and Frank, H.(1996).Chloroacetic acids in rainwater. Environ Sci Technol., 30:2340-2344.

[17]. U.S. Environmental Protection Agency (U.S. EPA. 2005). Drinking water addendum to the criteria document for trichloroacetic acid. (EPA 822-R-05-010). Washington, DC: U.S. EPA Office of Water.

[18]. Celik, I. (2007). Determination of toxicity of trichloroacetic acid in rats: 50 days drinking water study. Pestic Biochem Physiol., 89:39-45.

[19]. Celik I. and Temur A. (2009) Determination hematotoxic and hepatotoxic effects of trichloroacetic acid at sublethal dosage in rats. Food and Chemical Toxicology, 47(6):13241326.

[20]. Poon, R., Nadeau, B. and Chu, I. (2000). Biochemical effects of chloral hydrate on male rats following 7-day drinking water exposure. J Appl Toxicol., 20:455-61.

[21]. Acharya, S., Mehta, K., Rodriguez, S., Pereira, J., Krishnan, S. and Rao. C.V. (1997). A histopathological study of liver and kidney in male Wistar rats treated with subtoxic doses of t-butyl alcohol and trichloroacetic acid. Exp Toxicol Pathol. 49(5):369-73.

[22]. Mather, G.G., Exon, J.H. and Koller, L.D. (1990) .Subchronic 90-day toxicity of dichloroacetic and trichloroacetic acid in rats. Toxicology, 64:71-80.

[23]. International Agency for the Research on Cancer (IARC) (1995). Dry cleaning, some chlorinated solvents and other industrial chemicals. Vol 63. IARC. Lyon.

[24]. Paget, G.E. and Barnes, J.M.(1964).Evaluation of drug activities. In: Pharmacometrics Laurence DR, Bacharach AL, editors. New York: Academic Press, pp.161.

[25]. Bull, R.J., Sanchez,I.M., Nelson, M. A. ,Larson, J. L. and Lansing, A. J. (1990) . Liver tumor induction in B6C3F1 mice by dichloroacetate and trichloroacetate. Toxicol., 63(3):341-359.

[26]. Pereira, M.A., Kramer, P.M., Conran, P.B. and Tao, L. (2001). Effect of chloroform on dichloroacetic acid and trichloroacetic acid-induced hypomethylation and expression of the c-myc gene and on their promotion of liver and kidney tumors in mice. Carcinogenesis, 22:1511-1519.

[27]. Lewis, S.M., Bain, B.J. and Bates, I.B. (2001). Dacia and Lewis Practical haematology, 9th ed.London Edimburgh New York Philadelphia ST Louis Toronto.
[28]. Travlos,G.S.(2006).Normal structure, function, and histology of the bone marrow. Toxicol Pathol., 34(5):548-565.
[29]. Lund, J. E. (2000). Toxicologic effects on blood and bone marrow, in Schalm's Veterinary Hematology, 5th edition (B. F. Feldman, J. G. Zinkl, and N. C. Jain, eds.), pp.44-50. Lippincott, Williams and Wilkins, Philadelphia.
[30]. Ryan, D. H. (2001). Examination of the blood. In Williams Hematology, 6th edition. (E. Beutler, M. A. Lichtman, B. S. Coller, T. J. Kipps, and U. Seligsohn, eds.), pp. 9-16, McGraw-Hill, New York.
[31]. Weiss, L., and Geduldig, U. (1991). Barrier cells: stromal regulation of hematopoiesis and blood cell release in normal and stressed murine bone marrow. Blood, 78:975-90.
[32]. Wierda, D. (1990). Benzene toxicity to bone marrow stromal cells. Society of Toxicologic Pathologists Great Lakes Region Discussion Group: Bone Marrow Toxicity. Toxicol Pathol., 18:710-711.
[33]. Reagan, W. J. (1993). A Review of myelofibrosis in dogs. Toxicol Pathol., 21:164-9.
[34]. Weiss, D. J. (2000). Aplastic anemia. In Schalm's Veterinary Hematology, 5th edition (B. F. Feldman, J. G. Zinkl, and N. C. Jain, eds.), pp. 212-15. Lippincott, Williams and Wilkins, Philadelphia, PA.
[35]. Harper, H.A. (1973). Review of Physiological Chemistry, 14th ed. Lange Medical Publications, California.
[36]. Adeneye, A.A., Ajagbonna, O.P., Adeleke, T.I. and Bello, S.O. (2006). Preliminary toxicity and phytochemical studies of the stem bark aqueous extract of Musanga cecropioides in rats. Journal of Ethnopharmacology, 105:374-379.
[37]. Paprika, M.V. and Sharma, B.B. (2003). Effect of oral administration of herbicide diclofop on some hematological parameters in mouse. Journal of Cell and Tissue Research, 3:12-17.
[38]. Ferzand, R., Gadahi, J.A., Saleha, S. and Ali, Q. (2008). Histological and haematological disturbance caused by arsenic toxicity in mice model. Pak J Biol Sci., 11(11):1405-1413.
[39]. Yu, M.H. (2005). Soil and water pollution: Environmental metals and metalloids. Environmental Toxicology: Biological and Health Effects of Pollutants. CRC Press, pp.185-226.
[40]. Nikinmaa, M. (1992). How does environmental pollution affect red cell in fish? Aquat Toxicol., 22: 227-238.
[41]. Koc, N.D., Muslu, M.N., Sesal, C. and Kayhan, F.E.(2008). Histopathological effects of Malathion and endosulfan on blood cells of Wistar albino rats (Rattus norvegicus). J Appl Biol Sci .,2:105-108.
[42]. Yao, C., Li, X. and Xiong, Y.(2005). Instant Effects of Radiofrequency Electromagnetic Wave on Hemoglobin in Single Living Intact Red Blood Cell. Chinese Chermical Letters, 16(8):1121- 1124.
[43]. Hamidi, H. and Tajerzadeh, H.(2003). Carrier erythrocytes: An overview. Drug Delivery, 10:9-20.
[44]. Snyder, L.M., Fotier, N.L., Trinor, J., Jacobs, J., Leb, L., Lubin,S. et al., (1985). Effect of hydrogen peroxide exposure on normal human erythrocyte deformability, morphology, surface characteristics and spectrin-hemoglobin crosslinking .Journal of Clinical Investigation, 76:1971-1977.
[45]. Hassoun, EA. Cearfoss,J. and Spildener,J.(2010). Dichloroacetate- and trichloroacetateinduced oxidative stress in the hepatic tissues of mice after long term exposure. J Appl Toxicol., 30(5): 450-456.
[46]. Rice-Evans,C.A., Miller, N.J. and Paganga, G.(1996). Structure-antioxidant activity relationships of flavonoids and phenolic acids.Free Radical Biology and Medicine, 20:933956.
[47]. Kitagawa, H., Sakamoto,H. and Tano,Y. (2004). Inhibitory effects of flavonoids on free radical-induced hemolysis and their oxidative effects on hemoglobin. Chemical and Pharmaceutical Bulletin, 52:999-1001.
[48]. Kulisic-Bilusic, T., Schmoller, K., Schnabele, K., Siracusa, L. and Ruberto, G. (2012). The anticarcinogenic potential of essential oil and aqueous infusion from caper (Capparis spinosa L.). Food Chemistry, 132:261-267.
[49]. Behnaz, M., Davood,E.A. And Anoosh,E. (2012). Caper the Mystique of the recent century. Intl J Agri Crop Sci., 4(10):604-608.
[50]. Yang, T., Wang, C., Liu, H., Chou, G., Cheng, X. and Wang, Z.(2010).A new antioxidant compound from Capparis spinosa. Pharm Biol., 48(5):589-594.
[51]. Ping, K.Y., Darah,I., Chen,Y., Sreeramanan,S. and Sasidharan,S.(2013). Acute and subchronic toxicity study of Euphorbia hirta L. Methanol Eextract in rats BioMed Research International, 2013: 1-14.
[52]. Sharma, R., Panwar, K. and Mogra, S.(2013). Alterations in developing rats after prenatal and postnatal exposure to lead actate and vitamins. IJPSR. 4(8):3214-3224.
[53]. Taghavi, M.M., Nazari M., Rahmani R.,Sayadi A, Hajizadeh MR., Mirzaei,M.R., Ziaaddini,H., Hosseini-Zijoud,S.M. and Mahmoodi, M.(2014). Outcome of Capparis spinosa fruit extracts treatment on liver, kidney, pancreas and stomach tissues in normal and diabetic rats. Med chem., 4:717- 721.
[54]. Panico, A.M., Cardile, V., Garufi, F., Puglia, C., Bonina, F. and Ronsisvalle, G. (2005). Protective effect of Capparis spinosa on chondrocytes. Life Sci., 77(20): 2479-2488.
[55]. Halliwell, B. and Gutteridge, J.M.C. (1989). Free Radicals in Biology and Medicine, 2nd ed., Oxford University Press: London, UK, pp. 126-131.
[56]. Bahar, E., Akter, K.M., Lee, G.H., et al. (2017). B-Cell protection and antidiabetic activities of Crassocephalum crepidioides (Asteraceae) Benth. S. Moore extract against alloxaninduced oxidative stress via regulation of apoptosis and reactive oxygen species (ROS). BMC Complement Altern Med., 17(1):179.