EFFECT OF CRUDE METHANOLIC EXTRACTS OF SOME MEDICINAL PLANT FLOWERS ON SOLID EHRLICH TUMOR IN MICE.

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ABSTRACT

Crude methanolic extracts of *Cynara Cardunculus, Achillea millefolium, Calendula officinalis,* and *Matricaria chamomilla* flowers belong to family Compositeae were used in this work to study their efficiency to relief solid ehrlich tumor in mice. The more effective extract was with that of *C. officinalis* which decreased tumor size from 513.6 to 124.5mm at a dose of 200mg/kg body weight comparing with doxorubicin which reduced tumor size to 98.2mm in concentration of 15mg/kg after 21 days, followed by *M. chamomilla* which showed the mean value of 157.1mm. While, the lowest values were with *C. cardunculus and A. millefolium* flowers extract which were 184.3 and 210.9 mm, respectively. Also, the more effective treatment were doxorubicin in concentration of 15mg/kg and *C. officinalis*, followed by *M. chamomilla*, *C. cardunculus and A. millefolium* flowers methanolic extract for blood tests in tumor mice, e.g. antioxidant markers (MDA, TAC, SOD), liver functions (ALT, AST, Total proteins, Albumin and Globulins), kidney functions (Creatinine and Urea) lipid profile (triglycerides, total cholesterol, HDL, LDL and vLDL) haematological parameters (HB, RBCs, Plt and WBCs),comparing with control tumor mice at a dose of 200mg/kg body weight for 21 days.

Keywords: medicinal plants and solid ehrlich ascitic tumor.

INTRODUCTION

The solid ehrlich ascitic tumor derived from a spontaneous murine mammary adenocarcinoma, is maintained in the ascetic form by passages in Swiss mice, by weekly transplantation of Ehrlich Ascitic Carcinoma (EAC) cells (0.2 ml of 1×10^6) i.p. cells/mice, (Dagli *et al.,* 1992).

Doxorubicin is one of the most frequently used anticancer drugs. It is used for the treatment of different haematopoietic and solid cancers, such as breast cancer, osteosarcomas, aggressive lymphomas and leukemias. It has the ability to intercalate between the DNA strands and hydrolyzes them in a nuclease like manner (Fornari *et al.,* 1994 and Lentacker *et al.,* 2010).

Nascimento *et al.,* (2006), found that medicinal plants containing the flavonoids and terpenoids products which have diverse pharmacological properties including antioxidant and cancer chemo preventive effects.

Globe Artichoke (*Cynara Cardunculus*) is widely used as food and medicine in folk medicine. Artichoke flowers and leaves extract has shown antioxidant, antibacterial, anti-HIV, bile-expelling, hepatoprotective, urinative and choleretic activities, as well as the ability to inhibit cholesterol biosynthesis and LDL oxidation (Christaki *et al.,* 2012). Artichoke leaves, stalks and inflorescences extracts as antioxidant, anti-proliferative, anti-migratory and anti-angiogenic activities (Zélia *et al.,* 2012). Cynara ethanolic extract reduced total cholesterol and triglyceride levels in rats from 66.5 to

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58.3 and 100.8 to 81.2 mg/kg, respectively. these effects were attributed for the active ingredients e.g. cynarin, flavonoids (Dalen *et al.,* 2012).

Achillea (*Achillea millefolium*) species are used in folk medicine, the infusion of dried flowers are considered suitable for treatment of antioxidant, antifungal, anti-inflammatory, anti-edematous, antiviral, antimicrobial, hemorrhoids, dyspepsia, dysmenorrhea, gastritis and immunomodulatory properties (Vitalini *et al.,* 2011). Also, (Florea *et al.,* (2013) found antitumor activity of wild and commercial *A. millefolium* in human tumor cell for breast carcinoma (MCF-7), non-small cell lung cancer (NCI-H460), colon carcinoma (HCT-15), cervical carcinoma (HELA), hepatocellular carcinoma (HEPG2) and hepatotoxicity.

Pot marigold (*Calendula officinalis*) species are used for cutaneous wound healing, collagen efficiency, jaundice, blood purification, cytotoxic, antitumor and antispasmodic effects (Gharineh *et al.,* 2013). Likewise, (Preethi *et al.,* 2006) studied the effect of *C. officinalis* administration on antioxidant system in blood pointed to catalase, superoxide dismutase, glutathione peroxidase and glutathione. The pharmacological activities of *C. officinalis* such as anti-HIV, anti-cancer, anti-inflammatory, haepatoprotective, spasmolytic and spasmogenic activities were reported by (Muley *et al.,* 2009).

Chamomile (*Matricaria chamomilla*) has been used traditionally as a medicinal and pharmaceutical preparation due to its anti-inflammatory and antispasmodic properties (Harbourne *et al.,* 2009). Similarly, (Guimarães *et al.,* 2013) tested the antitumour potential of wild german chamomile methanolic extract on MCF-7 (breast carcinoma), NCI-h460 (non-small cell lung cancer), HCT-15 (colon carcinoma), HELA (cervical carcinoma), HEPG2 (hepatocellular carcinoma) and PLP2 (hepatotoxicity).

Aim of this study is to examine the efficiency of methanolic extracts of *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* flowers at doses of 200 mg/kg body weight on solid tumor mice after 21 days through the determination of tumor size, survival rate and survival time, antioxidant markers, liver functions, kidney functions, lipid profile and haematological analysis comparing with doxorubicin in concentration of 15mg/kg.

MATERIALS AND METHODS

Sampling:

The present investigation was carried out using *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* flowers belong to family Compositeae. Samples were kindly obtained from Agricultural Research Center, Giza, Egypt. Flower samples were air dried in the shade and ground into a fine powder.

Powdered was air dried flowers (2 Kg) of each plant were extracted by soaking at room temperature for six times with methanol (30 L), then the methanolic extracts were concentrated to nearly dryness under reduced pressure using the rotary evaporator at 45°C to achieve the crude methanolic extracts which kept for further investigation (El-Khateeb *et al.,* 2014).

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Experimental animals:

A number of 49 female albino mice (20-25g) were obtained from the animal house of Faculty of Pharmacy, Mansoura University, Egypt. All mice were housed in microlon boxes in a controlled environment (temperature 25±20ºC and 12 h dark/light cycle) with standard laboratory diet and water *ad libitum*. Saluja *et al.,* (2010).

Solid Ehrlich Carcinoma (SEC) Tumor Model:

A model of SEC where 0.2mg (1×10^6) of the Ehrlich carcinoma cells obtained from Experimental Oncology Unit of National Cancer Institute, Cairo University, Egypt were implanted subcutaneously into the right thigh of the lower limb of mice. A palpable solid tumor mass (about 100 mm3) was developed within 12 days (Osman *et al.,* 1993).

Experimental protocol:

Female albino mice were divided into 7 groups contain 7 animals of each, as follows:

Group1(normal):represents normal mice by means non tumors.
Group2:(control): SEC-bearing mice.

Group3: SEC-bearing mice treated with doxorubicin (DOX) (molecular weight: 543.5262 and chemical formula: C_{27}-H_{29}-NO_{11}) in concentration of 15 mg/kg. Doxorubicin was administered intraperitoneally in six equal injections (each containing 2.5 mg/kg body weight) to animals over a period of 3 weeks for accumulative dose of 15 mg/kg body weight according to Timao and Singal, (2000).

Group 4: SEC-bearing mice treated intraperitoneally with methanolic extract of *C. cardunculus* flowers (200 mg/kg) (Sethuramani *et al.,* 2014).

Group 5: SEC-bearing mice treated with methanolic extract of *A. millefolium* flowers (200 mg/kg).

Group 6: SEC-bearing mice treated with methanolic extract of *C. officinalis* flowers (200 mg/kg).

Group 7: SEC-bearing mice treated with methanolic extract of *M. chamomilla* flowers (200 mg/kg), single dose daily for 3 weeks.

Blood samples were collected from the eye canthus by heparinized tubes after 21 days from the beginning of the experiment. Then, each blood samples were divided into two portions. First portion was centrifugation to obtain the blood serum. Serum samples were kept at refrigerator under freezing conditions for the determination the parameters included antioxidant markers (MDA, TAC and SOD), liver functions (ALT, AST, proteins, albumin and Globulins), kidney functions (creatinine and urea) and lipid profile (Triglycerides, total cholesterol, HDL-c, LDL-c and VLDL-c). Second portion was treated with 10% of ethylene diamine tetracetic acid (EDTA) with a good shaking to determine complete blood count (CBC) as a haematological analysis.

At the end of the experimental period, mice were anaesthetized with diethyl ether. Then, selected organs (liver and kidney) were excised, rinsed in isotonic saline solution and stored in 10% formalin for preparing to histopathological examination.

Tumor Volume, survival rate and survival time:

Antitumor effects of different treatments were evaluated by tumor growth inhibition. Tumors were measured individually using a caliper. Tumor

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volume was determined by the following equation: (Tumor Volume = length × width^2 × 0.52) according to method of (Jia *et al.,* 2005).

Chemical analysis of blood:

Determination of malondialdehyde (MDA), superoxide dismutase (SOD) activity was assayed by the method of Habig *et al.,* (1974) and Nishikimi *et al.,* (1972).

Liver functions (ALT+AST) were determined as described by Randox (United Kingdom) according to the method of Reitman and Frankel, (1957). Also, George (1939) method was used to determine of protein and albumin.

Kidney functions (creatinine and blood urea) were determined by a colorimetric method according to Patton and Crouch (1977) as described in a commercial kits by Human (Germany).

Lipid profile (triglycerides, total cholesterol, HDL, LDL and vLDL) were determined by enzymatic colorimetric method of Richmond, (1973) described in a commercial kits by Human (Germany).

Haematological analysis (Hb, RBC, PCV, MCV, MCHC, Plt, MPV, PCT, PDW, WBC, LYM, MON, GRA) were through using apparatus namely ABX Micros 60 which a fully automated Haematological analyzer from Sysmex Corporation International Company according to Nakul *et al.,* (2003).

Statistical analysis of obtained data were done using the statistical software package CoStat (2005). All comparisons were first subjected to one way ANOVA and significant differences between treatment means were determined using Duncan's multiple rang test at $p<0.05$ as the level of the significance (Duncan, 1955).

RESULTS AND DISCUSSION

Effect of crude methanolic extract of investigated flowers as antitumor:

The yield of investigated flower methanolic extracts were 17.9%, 16.5%, 19.8%, 18.3%, for *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla*, respectively.

Data in table (1) revealed that the tumor size in zero time for all mice was ranged from 55.8 and 85.7 mm. From the same table, it was clear that the tumor size for control tumor mice (Group 2) was raised from 85.7 to 513.6mm after 21 days. While, the treatment of doxorubicin in concentration of 15mg/kg inhibit tumor size to 98.2mm after 21 days. Also, the methanolic extracts of *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* flowers at doses of 200 mg/kg reduced the size to 184.3, 210.9, 124.5 and 157.1mm, respectively, at the end of the experimental period.

Data in table (1), the survival rate of normal mice which was 100%. While, it decreased in control tumor mice to 28.5%. Whereas, the highest effective of doxorubicin in concentration of 15mg/kg and *C. officinalis* at doses 200mg/kg on survival rate, were, 85.7% and 85.7%, respectively. Likewise, the lowest effective of methanolic extracts for *C. cardunculus, A. millefolium* and *M. chamomilla* flowers at doses 200 mg/kg on survival rate, were 57.1, 42.8 and 57.1 %, respectively.

The survival time of control tumor mice was 29 days. Although, the highest effective of doxorubicin in concentration of 15mg/kg and *C. officinalis,* at doses 200mg/kg on survival time, raised to 70 and 69 days, Likewise, the

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lowest effective of methanolic extracts for *C. cardunculus, A. millefolium* and *M. chamomilla* flowers at doses 200mg/kg on survival time, were 43, 36 and 55 days, respectively.

The present data showed the highest impact on tumor size, survival rate and survival time, of doxorubicin in concentration of 15mg/kg and methanolic extracts for *C. officinalis* at doses 200 mg/kg, followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers at dose 200 mg/kg, respectively.

Table.1.Effect of investigated extracts on solid ehrlich carcinoma and administration tumor size, survival rate and survival time in mice.

	Tumor size (mm)		No. of mice	No. of survived mice	Survival rate %	Survival time (day)
	Zero time	21 days				
Group 1	0^{k}	0^{k}	7	7	%100	0
Group 2	$85.7^{g}\pm7.5$	$513.6^{a}\pm14.6$	7	2	%28.5	29
Group 3	$78.5^{h}\pm5.6$	$98.2^{j}\pm3.7$	7	6	%85.7	70
Group 4	$55.8^{l}\pm3.9$	$184.3^{c}\pm6.5$	7	4	%57.1	43
Group 5	$72.5^{h}\pm6.7$	$210.9^{b}\pm7.4$	7	3	%42.8	36
Group 6	$59.1^{l}\pm4.2$	$124.5^{e}\pm4.2$	7	6	%85.7	69
Group 7	$64.4^{l}\pm5.8$	$157.1^{d}\pm4.1$	7	4	%57.1	55
LSD, 0.05	**3.39**					

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: mice treated with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

These findings were in agreement with Saluja *et al.,* (2010) who studied the effect of acetone and ethanol extracts from *Vitex negundo* leaves (carbohydrate, glycoside, alkaloid, amino acids, flavonoids, fixed oil, tannins, gum and mucilage, phytosterols etc.) and 5-fluorouracil (5-FU, 20 mg/kg/day) on survival time which were 37.5, 34.16 and 44.33 days compared to tumor control after 21 days, respectively.

Muresan *et al.,* (2006), tested the effect of doxorubicin, grape seed, doxorubicin + Grape seed and vehicle (carboxymethylcelullose 0.5%) on tumor volume which were 2.16, 1.64, 1.55 and 2.83ml, respectively, compared with tumor control (2.67ml). Furthermore, Hussein *et al.,* (2014), reported the effect of purslane seed oil (200g/kg), 5-Flourourasil (20mg/kg) and purslane seed oil+5-fluorouracil on tumor volume which were 4.5, 2.3 and 2.7ml compared with tumor control (8.3 ml).

Also, the obtained data were agreed with those described by Mbarek *et al.,* (2007), who found the effects of (*Nigella sativa* L.) extracts belong to family Asteraceae on tumor volume that injected with 30µL and 50µL were 0.22 and 0.16cm^3, respectively compared with 2.5 cm^3 for tumor control.

Effect of crude methanolic extracts on antioxidant markers (MDA, TAC and SOD):

Data in table (2) cleared that the malondialdehyde (MDA) of normal and control tumor mice were 3.7 and 7.2µmol/ml. While, the most effective treatments were doxorubicin in concentration of 15mg/kg and *C. officinalis* at dose of 200mg/kg which decreased MDA levels to 3.9 and 4.1µmol/ml, respectively. Also, the effect of methanolic extracts for *C. cardunculus, A.*

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millefolium and *M. chamomilla* flowers at a dose of 200mg/kg reduced MDA to 4.3, 5.4 and 4.5µmol/ml, respectively.

The same table exhibited the total antioxidant capacity (TAC) that reduced from 0.529mmol/L of normal mice to reach 0.404mmol/L after bearing tumor. The most effective treatments were doxorubicin (15mg/kg) and *C. officinalis* (200 mg/kg) which raised TAC to 0.518 and 0.506mmol/L, respectively. Furthermore, the effect of methanolic extracts of *C. cardunculus, A. millefolium* and *M. chamomilla* flowers at a dose of 200mg/kg increased TAC values to 0.482, 0.463 and 0.491mmol/L, respectively.

From table (2), it was noticed that the total superoxide dismutase (SOD) of normal mice was 1.67U/g which decreased to 1.23U/g in control tumor mice. Though, the effect of doxorubicin in concentration of 15mg/kg and *C. officinalis* at a dose of 200mg/kg raised SOD values to 1.59 and 1.56U/g, respectively. Moreover, the effect of *C. cardunculus, A. millefolium* and *M. chamomilla* were 1.42, 1.39 and 1.49U/g, respectively.

Table2.Effect of investigated extracts on antioxidants markers (MDA, TAC and SOD) in mice.

Groups	MDA (µmol/ml)	TAC (mmol/L)	SOD (U/g)
Group 1	3.7^{d}±1.40	0.529^{a}±0.041	1.67^{a}±0.05
Group 2	7.2^{a}±2.03	0.404^{b}±0.039	1.23^{a}±0.08
Group 3	3.9^{d}±0.95	0.518^{a}±0.009	1.59^{a}±0.54
Group 4	4.3^{c}±1.56	0.482^{b}±0.082	1.42^{a}±0.41
Group 5	5.4^{b}±1.87	0.463^{b}±0.063	1.39^{a}±0.26
Group 6	4.1^{c}±1.33	0.506^{a}±0.056	1.56^{a}±0.36
Group 7	4.5^{c}±1.72	0.491^{b}±0.007	1.49^{a}±0.29
LSD, 0.05	3.78	1.27	3.02

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

The data, showed that doxorubicin in concentration 15mg/kg and methanolic extract of *C. officinalis* at doses 200mg/kg, the highest impact antioxidant activity, e.g. MDA, TAC and SOD, followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers 200mg/kg, respectively.

This finding was in the same line of, Metwally *et al.,* (2014), who found effect of neem, sec and sec+neem administration on MDA content in liver, were 49.23, 104.11 and 71.01µmol/g, respectively. While, effect of neem, sec and sec+neem administration on SOD activity in liver, were 2.62, 1.87and 3.43 U/g, respectively. compared with normal control for MDA and SOD, were 39µmol/g and 2.17U/g, respectively.

Obtained data were agreed with those by Amal and Eman (2010), who found the effects of *zizyphus* leaves extract and EAC on serum total antioxidant, were 1.06 and 0.80mM/L, compared with normal control, 1.07mM/L, respectively.

Abd el-aziz *et al.,* (2014), found that the effects of rosemary extract on SOD, CAT, MDA and TAC of ehrlich ascites carcinoma (EAC)- bearing mice, were 26.06u/g, 35.02u/g, 0.1036mmol/g, 8.24mmol/g and 1.946 mM/L, respectively. While, the effects of ehrlich ascites carcinoma (EAC)- bearing

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mice, on SOD, CAT, MDA and TAC of normal mice, were 18.00u/g, 22.08u/g, 0.0636mmol/g, 11.1mmol/g and 1.568mM/L, respectively. However, the effect on normal mice, were 38.90u/g, 42.56u/g, 0.1366mmol/g, 4.24mmol/g and 2.406mM/L, respectively.

Effect of crude methanolic extract of flowers on liver functions:

Alanine amino transferase (ALT) and aspartate amino transferase (AST) activities are known as cytosolic marker enzymes reflecting hepatocellular necrosis as they are released into the blood after damaging of the cell membrane. Therefore both enzymes are used as indicators for hepatic damage (Andallu and Vardacharyulu, 2001).

From tables (3), it was clear that ALT, AST, Total proteins, Albumin and Globulins increased significantly from 17.86, 35.15, 6.28, 2.97 and 3.31U/L, respectively In normal mice to 37.38, 78.12, 4.01, 1.99 and 2.02U/L, respectively in tumor mice. Such significant increase of ALT and AST activities and suggested possible necrotic injury of the liver or cholestasis with hepatocellular necrosis reported by (Van Hoof and De Broe, 1994).

Data in tables, (3) showed that the alanine amino transferase was raised from 17.86 for normal mice to reach 37.38U/l, after bearing tumor. While, the most effective treatments was doxorubicin and *C. officinalis* on ALT, were 21.34 and 22.64U/l, respectively. Likewise, the effect of methanolic extracts for *C. cardunculus, A. millefolium,* and *M. chamomilla*, were 24.30, 27.30 and 29.64U/l, respectively.

The table showed that the aspartate amino transferase AST of normal mice was 35.15U/g which raised to 78.12U/l in control tumor mice. Though, the effect of doxorubicin in concentration of 15mg/kg and *C. officinalis* at a dose of 200mg/kg were 44.91 and 41.82U/l, respectively. Furthermore, the effect of *C. cardunculus, A. millefolium* and *M. chamomilla* were 45.24, 51.34 and 53.82U/l, respectively.

Sethuramani et al., (2014), who found the effects for aqueous extract of *Oldenlandia umbellata* on AST and ALT, were 71.25 and 50.05U/l, respectively. While, the effect of ethanolic extract were 68.22 and 48.50U/l, respectively. Also, they found that effects of fluorouracil at 20mg/kg on AST and ALT, were 11.5 and 17.32U/l, respectively. compared to tumor control on the same serum enzymes were 7.15 and 30.62U/l, respectively.

Obtained data were agreed with those by Gupta *et al.,* (2004), who found that, effects of methanol extract for *Bauhinia racemosa* (100 mg/kg) on ALT and AST U/l, were 71.1 and 43.3 U/l, compared with Vehicle control on ALT and AST U/l, were 65.3 and 39.50 U/l, respectively.

Effect on total proteins, albumins and globulins activity:

Also, data cleared that the total proteins in normal mice was 6.28g/dl, while it showed that doxorubicin in concentration of 15mg/kg and methanolic extract of *C. officinalis,* at adose of 200mg/kg the values for total proteins in Ehrlich solid tumor mice, were 5.98 and 6.01g/dl, respectively. Followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers at doses 200mg/kg, were 5.54, 5.21 and 4.94g/dl, respectively. as compared to normal and control tumor mice, was 4.01g/dl.

The same table reveled that the total albumins in normal mice was 2.97g/dl. The effect of doxorubicin in concentration of 15mg/kg and

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methanolic extract of *C. officinalis* at a dose of 200mg/kg showed values for total albumins in solid Ehrlich tumor mice, were 2.66 and 2.76g/dl, respectively. This followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers at a dose of 200 mg/kg, that were 2.45, 2.08 and 2.36g/dl, for methanolic extract, respectively. This was compared with control tumor mice, which was 1.99g/dl.

From table (3), it was notified that the total Globulins was reduced from 3.31 in normal mice to reach 2.02g/dl after bearing tumor. While, the most effective treatments was doxorubicin and *C. officinalis* on globulin, were 3.1 and 3.75g/dl, respectively. Likewise, the effect of methanolic extracts for *C. cardunculus, A. millefolium,* and *M. chamomilla*, were 2.55, 2.66, and 2.25g/dl, respectively.

Table.3.Effect of extracts on liver functions for solid ehrlich tumor mice.

Groups	ALT (U/L)	AST (U/L)	Total proteins (g/dl)	Albumin (g/dl)	Globulins (g/dl)
Group 1	17.86^{j} ± 0.97	35.15^{e}± 0.12	6.28^{a} ± 0.24	2.97^{a} ± 0.08	3.31^{a}± 0.16
Group 2	37.38^{a} ± 0.22	78.12^{a}± 0.16	4.01^{c} ± 0.16	1.99^{b} ± 0.14	2.02^{b} ± 0.02
Group 3	21.34^{f} ± 0.33	40.92^{d}±0.34	5.98^{b} ± 0.79	2.88^{a} ± 0.09	3.1^{a} ± 0.7
Group 4	27.30^{c} ± 0.02	51.34^{b}±0.11	5.21^{b}± 0.55	2.56^{a}± 0.43	2.55^{b} ± 0.41
Group 5	29.64^{b} ± 0.80	53.82^{b}±0.23	4.94^{c}± 0.49	$2.\ 28^{a}$± 0.17	2.66^{b} ± 0.32
Group 6	22.64^{e} ± 0.80	41.82^{d}±0.03	6.01^{a} ± 0.33	2.76^{a} ± 0.10	3.75^{a} ± 0.23
Group 7	24.30^{d} ± 0.02	45.24^{c}± 0.01	5.54^{b} ± 0.17	2.78^{a} ± 0.03	2.25^{b} ± 0.12
LSD, 0.05	**1.03**	**3.26**	**1.47**	**1.07**	**0.4**

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

The doxorubicin of 15mg/kg and methanolic extract for *C. officinalis* at a dose of 200mg/kg, showed the highest impact Total proteins, Albumin and Globulins, followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers at a dose of 200 mg/kg, respectively.

The finding was in the same line with Saluja *et al.,* (2010), who found that effect of ethanolic, acetone (500 mg/kg) from the leaves of *Vitex negundo* and 5- fluorouracil (5-FU, 20 mg/kg/day) on Total proteins, were 9.1, 9.6 and 8.65g/dl, respectively. Was compared tumor control, 13.95g/dl.

Obtained data were agreed with those by Abd el-aziz, et al., (2014), who found that effects of rosemary extract on Albumin of ehrlich ascites carcinoma (EAC)- bearing mice, was 2.570g/dl, was compared normal and control mice were 3.196 and 2.090g/dl, respectively.

Abd el-aziz, et al., (2014), who stated that, the antioxidant's activity of polar extracts of rosemary is related with the content of phenolic compounds (carnosol, carnosic acid). Constituents in rosemary have shown a variety of pharmacological activities of cancer chemoprevention and therapy in vitro and, in vivo models.

Effect of crude methanolic extract of investigated flowers on kidney functions:

Determination of serum creatinine and urea were used as indicators for kidney functions. The effect of methanolic and aqueous extracts of all

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samples under investigation on serum creatinine and urea levels in Ehrlich solid tumor during the experimental periods are tabulated in tables (4).

It could be noticed that the injection with tumor induced a significantly increase in serum creatinine and urea levels from 1.46 and 49.91 to 0.58 and 78.45 mg/dl compared with non- tumor mice. This could be attributed to the presence of renal damage as a result of cancer cell invasions, Griffin *et al.,* (1995).

From tables (4), cleared that the serum creatinine in normal mice was 0.58mg/dl, although the methanolic extracts of *C. cardunculus*, *A. millefolium, C. officinalis,* and *M. chamomilla* flowers, at a dose of 200mg/kg, reduced serum creatinine to 1.09, 1.12, 0.83 and 0.92mg/dl, respectively. While, the doxorubicin in concentration of 15mg/kg reduced serum creatinine to 0.79 mg/dl, after 21 days, as compared to tumor control mice, which was 1.46mg/dl.

It also revealed that, the level urea of normal mice, was 78.45g/dl, while, the effect of doxorubicin and *C. officinalis* on ehrlich tumor, were 75.65 and 74.92g/dl. Also, the effects of methanolic extracts of *C. cardunculus, A. millefolium* and *M. chamomilla*, were 59.81, 56.64, and 67.44g/dl, respectively. as compared to tumor control mice, that was 49.91g/dl.

Table.4.Effect of investigated extracts on kidney functions in Ehrlich solid tumor mice.

Groups	Creatinine (mg/dl)	Urea (mg/dl)
Group 1	$0.58^{b}\pm 0.14$	$78.45^{a}\pm 0.34$
Group 2	$1.46^{a}\pm 0.28$	$49.91^{d}\pm 5.36$
Group 3	$0.79^{b}\pm 0.18$	$75.65^{a}\pm 2.13$
Group 4	$1.09^{a}\pm 0.16$	$59.81^{c}\pm 0.26$
Group 5	$1.12^{a}\pm 0.12$	$56.64^{c}\pm 2.66$
Group 6	$0.83^{b}\pm 0.15$	$74.92^{a}\pm 0.20$
Group 7	$0.92^{b}\pm 0.13$	$67.44^{b}\pm 0.26$
LSD, 0.05	2.07	4.98

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

Such data revealed that doxorubicin and methanolic flowers extract of *C. officinalis* were the most effective treatment on serum creatinine and urea levels. This followed by *M. chamomilla, cardunculus* and *A. millefolium* flowers, after 21 days at a dose of 200mg/kg body weight for plants and 15mg/kg for (DOX) respectively.

This finding was in the same line of Fakhry *et al.,* (2011), who found that effects of levamisole and chlorambucil on creatinine level were 1.91 and 1.65 mg/dl, compared to normal and tumor control mice, were 0.63 and 1.36 mg/dl, respectively.

Jayanthi *et al.,* (2014), who found that the effect of extract BG (250mg/kg) on Creatinine and urea in EAC inoculated mice, were 8.050 and 140.7mg, respectively. While, they found that effects of Cisplatin (3.5mg/kg) on serum creatinine and urea in EAC inoculated mice, were 7.250g/dl and

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129.7g/dl, respectively, as compared to tumor control were 9.250g/dl and 189.3g/dl, respectively.

Effect of crude methanolic extracts on lipid profile:

Data recorded in table (5), revealed that serum triglycerides, total cholesterol, LDL and vLDL values increased from 69.43, 98.5, 13.35 and 19.7mg/dl in non-tumor mice to 98.01, 156.6, 45.35 and 31.32mg/dl in tumor mice, respectively, by injection with Ehrlich solid tumor mice $1X10^6$ at 12^{th} days.

On the other hand, tables (5), declare that there is a highly significant decrease in serum HDL level and reached 21.35mg/dl for tumor mice comparing to 36.38 mg/dl for normal mice at the beginning of experiment.

Abu-Sinna *et al.,* (2003), who reported that the consumption of free amino acid for building the proteins of rapidly dividing tumor cells might result in the disturbance of the enzyme activity in the liver.

Effect on total cholesterol and triglycerides:

From tables (5), it could be noticed that t-cholesterol decreased with increasing the concentration of methanolic extract of investigated flowers and the experimental period for all samples under investigation. Accordingly, the treatment of tumor mice of doxorubicin in concentration of 15mg/kg, methanolic flowers extract of *C. officinalis* at a adose of 200mg/kg have the most effective, while the reduction of t-cholesterol levels achieve about 70.65 and 76.42mg/dl, respectively. Also, for *C. cardunculus* and *A. millefolium* with the same reducing value for t-cholesterol levels which were 80.14 and 82.44mg/dl, respectively. While, *M. chamomilla* flowers have a moderate value for reducing t-cholesterol levels, was 76.42mg/dl, for methanolic flowers extracts, respectively.

The data, for doxorubicin in concentration of 15mg/kg and methanolic extract of *C. officinalis* at a adose of 200mg/kg, showed the highest impact t-cholesterol, followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium*, respectively. as compared to normal and tumor control, which were 69.43 and 98.01mg/dl, respectively.

From tables (5), it could be noticed that triglycerides decreased by increasing the concentration of methanolic extract of investigated flowers and the experimental period for all samples under investigation. Accordingly, the treatment of tumor mice of doxorubicin (15mg/kg), methanolic flowers extract of *C. officinalis* (200mg/kg) have the most effective, where the reduction of triglycerides levels achieve about 109.7 and 113.2mg/dl, respectively. Also, for *C. cardunculus, A. millefolium* and flowers 200 mg/kg with the same reducing value for triglycerides levels which were 128.2 and 133.6mg/dl, respectively. While, *M. chamomilla* flowers have a moderate value for reducing triglycerides levels, was 121.6mg/dl, for methanolic flowers extract, respectively.

The data, the doxorubicin (15mg/kg) and methanolic extract for *C. officinalis* (200 mg/kg), showed the highest impact triglycerides, followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers 200mg/kg, respectively. as compared to normal and tumor control, were 98.5 and 156.6mg/dl, respectively.

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Obtained data were agreed with those by Hussein *et al.,* (2014), who found that effects of Ehrlich ascites carcinoma (EAC), on plasma total cholesterol and triglycerides of normal mice, were 73.76 and 67.24mg/dl, respectively. While, the effects of Purslane seed oil (200g/kg), on control mice, were 95.27mg/dl, 85.24mg/dl, 29.11mg/dl and 49.112mg/dl, respectively. Also, the effects of 5-Flourourasil (20mg/kg), on plasma total cholesterol and triglycerides of control mice, were 81.46mg/dl and 59.57mg/dl, respectively. as compared to normal mice, which were 124.43mg/dl and 109.16mg/dl, for plasma total cholesterol and triglycerides, respectively.

Sethuramani *et al.,* (2014), they said that effects of aqueous extract of *Oldenlandia umbellata* 200mg/kg on t-cholesterol and triglycerides, were 122.48mg/dl and 168.15mg/dl, while, the effects of ethanolic extract of *Oldenlandia umbellata* 200 mg/kg, were 124.88 and 162.68mg/dl, Also, who found the effects of fluorouracil at 20mg/kg on lipid proteins, were 11.28and 4.25mg/dl, respectively. as compared to tumor control on the same lipid proteins, were 14.62 and 2.32mg/dl, respectively.

Effect on HDL, LDL and vLDL-cholesterol:

Data in table (5), showed that oral administration of methanolic extract of all samples led to a gradual increase of serum HDL. Raising both of concentration of extracts and period of the experiment caused an increase in serum HDL, which reached 30.66, 28.41, and 31.09mg/dl for methanolic extract of *C. cardunculus, A. millefolium,* and *M. chamomilla* flowers (200mg/kg), respectively. While, treatment with doxorubicin (15mg/kg) and *C. officinalis* (200mg/kg) on serum HDL, were 35.41 and 34.71mg/dl, as compared to normal and tumor control, were 36.38 and 21.35mg/dl respectively.

The data in table (5), showed similar which effect on serum LDL. Raising both of concentration of extracts and period of the experiment caused an increase in serum LDL, were 22.84, 23.31, and 20.72mg/dl of methanolic extract for *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* flowers (200 mg/kg), respectively. While, treatment with doxorubicin (15mg/kg) and *C. officinalis* (200mg/kg)on serum LDL, was 13.9 and 14.16mg/dl, respectively. as compared to normal and tumor control, which were 13.35 and 45.35mg/dl, respectively.

Data for vLDL values as a result of treatment with methanolic extract for *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* flowers (200 mg/kg) in tumor mice, were 25.64, 26.72, 22.64 and 24.32mg/dl respectively. While, the effect of doxorubicin 15mg/kg on serum vLDL, was 21.94 mg/dl, as compared to normal and tumor control which were 19.7 and 31.32mg/dl respectively.

The data, of doxorubicin 15mg/kg and methanolic extract for *C. officinalis* 200mg/kg, showed the highest impact HDL, LDL and vLDL, followed by *M. chamomilla*, *C. cardunculus* and *A. millefolium* flowers 200 mg/kg, respectively. as compared to normal and tumor control.

Obtained data were agreed with those by **Hussein *et al.,* (2014),** who found that effects of Ehrlich ascites carcinoma (EAC), on HDL and LDL of normal mice, were 20.64mg/dl and 39.67mg/dl, respectively. While, the

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effects of Purslane seed oil (200g/kg), on the same compound of control mice, were 29.11mg/dl and 49.112mg/dl, respectively. Also, the effects of 5-Flourourasil (20mg/kg), on HDL and LDL of control mice, were 25.68mg/dl and 43.87mg/dl, respectively. as compared to normal mice, which were 34.29mg/dl and 68.31mg/dl, for HDL and LDL respectively.

Abdel-Maksoud *et al.,* (2015), they found that effect of cranberry, extract (75mg/kg) on HDL-C and LDL-C of tumor mice, were 32.5 and 133.5mg/dl, while, the effect of 5-fluorouracil (20mg/kg), were 35.8 and 90.91mg/dl, also, the effect of cranberry, extract (75mg/kg) + 5-fluorouracil (20mg/kg), on HDL-C and LDL-C of tumor mice, were 37.6 and 90.10mg/dl, respectively. where compared with normal mice, were 35.16 and 91.85mg/dl, and tumor control mice, were 30.38 and 166.77mg/dl, respectively.

Table.5.Effect of investigated extracts on lipid profile in solid ehrlich tumor mice.

Groups	Total Cholesterol (mg/dl)	Triglycerides (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	vLDL (mg/dl)
Group 1	$69.43^{d} \pm 0.22$	$98.5^{f} \pm 01.55$	$36.38^{a} \pm 3.53$	$13.35^{c} \pm 0.12$	$19.7^{c} \pm 1.18$
Group 2	$98.01^{a} \pm 0.06$	$156.6^{a} \pm 9.18$	$21.35^{b} \pm 4.49$	$45.35^{a} \pm 1.06$	$31.32^{a} \pm 0.48$
Group 3	$70.65^{c} \pm 2.33$	$109.7^{e} \pm 0.22$	$35.41^{a} \pm 3.35$	$13.9^{c} \pm 0.01$	$21.94^{b} \pm 0.21$
Group 4	$80.14^{b} \pm 9.07$	$128.2^{c} \pm 5.04$	$30.66^{a} \pm 8.17$	$22.84^{b} \pm 0.92$	$25.64^{b} \pm 1.32$
Group 5	$82.44^{b} \pm 3.16$	$133.6^{b} \pm 5.07$	$28.41^{b} \pm 3.56$	$23.31^{b} \pm 4.05$	$26.72^{b} \pm 2.09$
Group 6	$71.01^{c} \pm 0.03$	$113.2^{d} \pm 0.03$	$34.71^{a} \pm 6.44$	$14.16^{c} \pm 9.07$	$22.64^{b} \pm 0.36$
Group 7	$76.42^{c} \pm 0.43$	$121.6^{c} \pm 4.19$	$31.38^{a} \pm 3.53$	$20.72^{b} \pm 5.46$	$24.32^{b} \pm 1.08$
LSD, 0.05	3.82	7.35	2.87	2.36	1.98

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

Effect of crude methanolic extract of investigated flowers on haematological parameters:

The complete blood count (CBC) was used as a broad screening test to check such disorders as anemia, infection and many other diseases. It is actually a panel of tests that examines different parts of the blood, which play an important role in metabolism and important indicators of health in both human or animals (Bain *et al.,* 2006).

The complete blood count (CBC) includes the following tests:

Effect of HB, RBCs, PCV, MCV and MCHC:

Data in table (6), conveyed that the haemoglobin level (Hb), in normal mice, was 12.89 g/dl and decreased to 8.30 g/dl, in tumor control mice. While, doxorubicin in concentration of 15mg/kg and *C. officinalis* at a dose of 200 mg/kg were the most effective treatment on ehrlich tumor increase haemoglobin levels to 12.75 and 12.75g/dl, respectively. Likewise, the effect of methanolic extracts for *C. cardunculus, A. millefolium* and *M. chamomilla* were 11.97, 9.97 and 10.98g/dl, respectively.

From the same table, it was clear that the total red blood cells (RBCs), were reduced from 6.19×10^{6}/µl for normal mice to reach 3.11×10^{6}/µl after bearing tumor. Whereas, the effect of doxorubicin and *C. officinalis* were 4.79

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and 4.69×10^6/µl on ehrlich tumor mice, respectively. Also, the effect of methanolic extracts for *C. cardunculus, A. millefolium,* and *M. chamomilla*, were 3.95, 3.39 and 4.20×10^6/µl, respectively.

The same table showed that the total packed cell volume (PCV) of normal mice was 41.17 which decreased to 29.00% for control tumor mice. While, the effect of doxorubicin in concentration of 15mg/kg and *C. officinalis* at a dose of 200mg/kg raised PCV to 40.33 and 39.89%. Also, the effect of methanolic extracts for *C. cardunculus, A. millefolium,* and *M. chamomilla*, were 35.00, 34.17, and 39.89%, respectively.

Table.6.Effect of investigated extracts on haemoglobin and red blood cells in solid ehrlich tumor mice.

Groups	Hb (g/dl)	RBCs (10^6/µl)	PCV (%)	MCV(µm^3)	MCHC (g/dl)
Group 1	12.89^{a} ±0.23	6.19^{a} ± 0.29	41.17^{a} ±1.49	59.94^{a} ± 0.11	34.11^{a} ± 0.59
Group 2	8.30^{e} ± 0.29	3.11^{c} ± 0.20	29.00^{c} ±0.84	33.95^{c} ± 0.08	19.29^{c} ± 0.17
Group 3	12.75^{a} ±0.43	4.79^{b} ± 0.77	40.33^{a} ±0.49	58.84^{a} ± 0.47	33.93^{a}± 0.57
Group 4	10.98^{c} ±0.33	3.95^{c} ± 0.82	35.00^{b} ±0.57	45.33^{b} ± 0.28	29.43^{b} ± 0.52
Group 5	9.97^{d} ± 0.52	3.39^{c} ± 0.96	34.17^{b} ±1.97	43.48^{b} ± 1.11	27.96^{b} ± 0.62
Group 6	12.75^{a}± 0.91	4.69^{b} ± 0.67	39.89^{b} ±0.63	58.79^{a} ± 0.41	33.85^{a} ± 0.25
Group 7	11.97^{b} ±0.47	4.20^{b} ± 0.86	37.67^{b} ±1.16	55.11^{a} ± 0.45	31.87^{a} ± 0.36
LSD, 0.05	2.68	3.19	1.23	1.73	6.39

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

Previous data revealed that the total mean corpuscular volume (MCV) of normal and control tumor mice were 59.94 and 33.95µm^3 respectively. Although, the effect of doxorubicin and C. officinalis on MCV values were 58.84 and 58.79µm^3, respectively. Moreover, the effect of *C. cardunculus, A. millefolium* and *M. chamomilla* were 45.33, 43.48 and 55.11µm^3, respectively.

On the other hand, table (6) declare that the total mean corpuscular hemoglobin concentration (MCHC) was reduced from 34.11g/dl for normal mice to reach 19.29g/dl after bearing tumor. Whereas, the effect of doxorubicin and *C. officinalis* on MCHC were 33.93 and 33.85g/dl, respectively. Also, the effect of *C. cardunculus, A. millefolium* and *M. chamomilla* were 29.43, 27.96 and 31.87g/dl, respectively.

The data revealed that doxorubicin in concentration of 15mg/kg and methanolic extract for *C. officinalis* at a dose of 200mg/kg, showed the highest values for Hb, RBCs, PCV, MCV and MCHC followed by *M. chamomilla* which displayed the medium values tailed by the lowest levels for *C. cardunculus* and *A. millefolium* flowers (200mg/kg), respectively, compared with normal and tumor control mice.

This finding was in the same line with Saluja, *et al.,* (2010), who described that ethanolic extract (500 mg/kg) of *Vitex negundo* leaves on Hb, RBCs, WBC, PCV, were 12.91g/dl, 3.88mm^3, 9.09mm^3 and 21.3mm, respectively. Otherwise, Effect of 5-fluorouracil (20 mg/kg/day) on the same haematological parameters were 14.0, 4.11, 8.23, and 19.5, respectively, compared to tumor control mice (8.35, 2.67, 27.19 and 31.5, respectively).

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Obtained data were agreed with those reported by Venu *et al.,* (2012), who found the effect of methanol extract of *cleome viscosa* (400 mg/kg) on haematological parameters of EAC-bearing mice e.g. Haemoglobin, RBCs, WBC were 14.4g%, 5.9U/l and 6.4U/l, respectively. compared to tumor mice (9.7g%, 3.5U/l and 15.6U/l, respectively).

Sevastre *et al.,* (2010), found that effect of *V. album* extract on MCV, MCH and MCHC were 48.14, 16.40 and 38.89g/dl, respectively. While, the effect of doxorubicin on the same haematological parameters were 51.00, 16.72 and 32.70g/dl, respectively. Though, bearing tumor mice were 44.20, 14.56 and 33.00g/dl, respectively.

Effect of Plt, MPV, PCT and PDW:

Data in table (7), clear that the platelet blood (Plt) level in normal mice was 1279×10^3/µl and decreased to 173×10^3/µl in tumor control mice. While, the effect of doxorubicin in concentration of 15mg/kg on Plt was 1232×10^3/µl. Similarly, the effect of methanolic extracts for *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* flowers at a dose of 200mg/kg on ehrlich tumor were 1019, 981, 1201 and 1127×10^3/µl, respectively.

The table showed that the mean platelet volume (MPV) was reduced from 9.9µm^3 for normal mice to reach 5.2µm^3 after bearing tumor. Though, the effect of doxorubicin (15mg/kg) and *C. officinalis* (200mg/kg) have the equal value of 8.0µm^3. Also, the effect of methanolic extracts for *C. cardunculus, A. millefolium* and *M. chamomilla* were 7.2, 6.5, and 7.7µm^3, respectively.

The same table showed that the platelets hematocrit value (PCT) in normal mice was 8.4 which reduced to 6.4% after bearing tumor. While, the effect of doxorubicin 15mg/kg was 8.01%. Correspondingly, the effect of methanolic extracts for *C. cardunculus, A. millefolium, C. officinalis* and *M. chamomilla* (200mg/kg) were 6.52, 6.16, 7.97 and 6.68%, respectively.

On the other hand, table (7) declare that the total platelet distribution width (PDW) of normal and control tumor mice were 12.9 and 6.2%, respectively. Although, the treatment with doxorubicin slightly similar normal mice value (12.0%). As well, the effect of methanolic extracts for *C. cardunculus, A. millefolium, C. officinalis,* and *M. chamomilla* (200mg/kg), were 9.5, 9.0, 11.8 and 10.7%, respectively.

Table.7.Effect of investigated extracts on platelets in solid ehrlich tumor mice.

Groups	Plt (103/ µl)	MPV (µm3)	PCT (%)	PDW (%)
Group 1	$1279^{a} \pm 93.31$	$9.9^{a} \pm 0.23$	$8.4^{a} \pm 0.08$	$12.9^{a} \pm 0.44$
Group 2	$173^{e} \pm 17.55$	$5.2^{e} \pm 0.36$	$4.6^{e} \pm 0.02$	$6.2^{f} \pm 0.13$
Group 3	$1232^{a} \pm 16.29$	$8.0^{b} \pm 0.24$	$8.01^{a} \pm 0.04$	$12.0^{a} \pm 0.06$
Group 4	$1019^{c} \pm 19.43$	$7.2^{c} \pm 0.39$	$6.52^{c} \pm 0.04$	$9.5^{d} \pm 0.37$
Group 5	$9081^{d} \pm 12.17$	$6.5^{d} \pm 0.31$	$6.16^{c} \pm 0.05$	$9.0^{d} \pm 0.18$
Group 6	$1201^{a} \pm 90.88$	$8.0^{b} \pm 0.97$	$7.97^{b} \pm 0.06$	$11.8^{b} \pm 0.89$
Group 7	$1127^{b} \pm 77.09$	$7.7^{c} \pm 0.33$	$6.68^{c} \pm 0.06$	$10.7^{c} \pm 0.65$
LSD, 0.05	1.87	5.13	4.82	1.61

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

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From the data, the doxorubicin in concentration of 15mg/kg and methanolic extract for *C. officinalis* 200 mg/kg showed the highest values for Plt, MPV, PCT and PDW followed by *M. chamomilla* presented the medium values. Finally, the lowest levels achieved by treatment with *C. cardunculus* and *A. millefolium* flowers at a dose of 200mg/kg, respectively compared with normal and tumor control.

Such findings were in the same line with Sevastre *et al.,* (2010), who found that effects of *V. album* extract on PLT, PCT, MPV and PDW were 723.60×10^9/l, 0.45%, 6.20% and 6.06%, respectively. While, the effect of doxorubicin on the same haematological were 498.50×10^9/l, 0.32%, 6.38% and 6.40%, respectively compared to tumor control mice 389.33×10^9/l, 0.23%, 5.97% and 5.84%, respectively.

Effect of WBCs, Lym, Mono and GRA:

Data in table (8) cleared that white blood cells (WBCs) level in normal mice was 11.23 which raised to 24.23×10^3/µl in control tumor mice. While, the effect of doxorubicin in concentration of 15mg/kg and *C. officinalis* at a dose of 200mg/kg were 12.46 and 12.16×10^3/µl, respectively. Also, the effect of methanolic extracts for *C. cardunculus, A. millefolium,* and *M. chamomilla*, were 14.83, 17.89 and 13.39×10^3/µl, respectively.

From table (8), it was informed that the level of Lymphocytes (Lym) increased from $5.35\times\mu m^3$ for normal mice to reach 7.11×10^3/µl after bearing tumor. Even though, the effect of doxorubicin (15mg/kg) and *C. officinalis* (200mg/kg) were 5.51 and 5.64×10^3/µl, respectively. Finally, the effect of *C. cardunculus, A. millefolium* and *M. chamomilla* were 6.10, 6.51 and5.98×10^3/µl, respectively.

Table.8.Effect of investigated extracts on white blood cells in solid ehrlich tumor mice.

Groups	WBCs (10^3/µl)	Lym (10^3/µl)	Mono (%)	GRA (%)
Group 1	$11.23^{h} \pm 0.25$	$5.35^{c} \pm 0.29$	$0.45^{a} \pm 0.01$	$39.3^{a} \pm 0.25$
Group 2	$24.23^{a} \pm 0.37$	$7.11^{a} \pm 0.01$	$0.76^{a} \pm 0.03$	$25.0^{b} \pm 0.31$
Group 3	$12.46^{j} \pm 0.54$	$5.51^{c} \pm 0.20$	$0.49^{a} \pm 0.01$	$38.8^{a} \pm 0.17$
Group 4	$14.83^{e} \pm 0.75$	$6.10^{b} \pm 0.21$	$0.62^{a} \pm 0.03$	$31.4^{a} \pm 0.21$
Group 5	$17.89^{c} \pm 0.89$	$6.51^{b} \pm 0.46$	$0.65^{a} \pm 0.02$	$30.7^{a} \pm 0.19$
Group 6	$12.16^{j} \pm 0.34$	$5.64^{c} \pm 0.07$	$0.51^{a} \pm 0.00$	$37.8^{a} \pm 0.21$
Group 7	$13.39^{f} \pm 0.93$	$5.98^{c} \pm 0.31$	$0.55^{a} \pm 0.15$	$35.6^{a} \pm 0.49$
LSD, 0.05	1.37	1.26	1.01	1.33

Group 1: normal mice, Group 2: +Ve control mice, Group 3: DOX, Groups 4, 5, 6 and 7: treated mice with methanolic extracts of *C. cardunculus, A. millefolium*, *C. officinalis* and *M. chamomilla*, respectively.

Previous data revealed that the Monocytes (Mono) values of normal mice was 0.45% which raised to 0.76% for control tumor mice. Whereas, the effect of doxorubicin (15mg/kg) and *C. officinalis* (200mg/kg) were 0.49 and 0.51%, respectively. Also, the effect of *C. cardunculus, A. millefolium* and *M. chamomilla*, were 0.62, 0.65 and 0.55%, respectively.

On the other hand table (8) declared that the Granulocytes (GRA) of normal and control tumor mice were 39.3 and 25.0%. While, the effect of doxorubicin and *C. officinalis* were 38.8 and 37.8%, respectively. Similarly,

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the effect of methanolic extracts for *C. cardunculus, A. millefolium* and *M. chamomilla*, were 31.4, 30.7, and 35.6%, respectively.

The data of doxorubicin in concentration of 15mg/kg and methanolic extract for *C. officinalis* at a dose of 200mg/kg showed the highest values of WBCs, Lym, Mono and GRA, followed by *M. chamomilla* which indicated the medium levels, tailed by the lowest values for *C. cardunculus* and *A. millefolium*, respectively.

Sevastre *et al.,* (2010), they found that effects of *V. album* extract on WBC, LYM, MID and GRA, were 7.37, 6.06, 0.14 and 1.17×10^9/l, respectively. While, the effect of doxorubicin on the same haematological parameters were 15.01, 8.93, 0.34 and 5.64×10^9/l, respectively. In spite of tumor control which were 4.87, 4.29, 0.11 and 0.45×10^9/l, respectively.

This finding was in the same line with Venu *et al.,* (2011), who found that effect of methanol extract of *cleome viscosa* (400mg/kg) on WBC, Monocyte, Neutrophil and Lymphocyte were 6.4u/l, 1.9%, 25.1% and 69.4%, respectively when compared with tumor control on the same haematological parameters which were 15.6u/l, 1.2%, 56.4 % and 33.6%, respectively.

The plasma of tumor-bearing humans and other mammalian models exhibits high levels of PGs, especially those of the E-type (mainly PGE2). This is because tumor cells. Siddiqui and Williams (1987) and Kalmar *et al.,* (1984).as well as specialized antigen-presenting cells, stimulated by the manifestation of an initiating tumor, produce PGE2 and PGD2.which present associated with the impairment of immune function, thus leading to immunosuppression and cancer cachexia. Leung and Mihich, (1980) and Scott *et al.,* (1982).

Prostaglandins (PGs) and other eicosanoids are bioactive lipids that impact normal development, tissue homeostasis, inflammation, and cancer progression. Wang and Dubois (2010). Prostaglandins serum levels have been shown to be increased in tumor-bearing mice. Takeuchi *et al.,* (1982). as well as in patients with solid tumors without showing impact on the course or stage of disease. Todo *et al.,* (1987) and Starczewski, *et al.,* (1984).

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تأثير المستخلص الميثانولي الخام لأزهار بعض النباتات الطبية على الأورام السرطانية الصلبة في فئران التجارب

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تم فى هذه الدراسة إستخلاص المركبات الفعالة لأزهار الخرشوف والإيشيليا والأقحوان والبابونج عن طريق النقع فى الميثانول ثم التركيز تحت تفريغ بغرض الحصول على المستخلص الميثانولى الخام للأزهار تحت الدراسة ومقارنتها بالمركب الكيميائى doxorubicin. ثم دراسة تأثير تلك المستخلصات على حجم الأورام السرطانية وكذلك بعض مكونات دم فئران التجارب المصابة بمرض السرطان. وكان مستخلص أزهار الأقحوان أعلى نشاطا في التأثير على حجم الورم السرطانى وتخفيضه الى ١٢٤،٥مم بالحقن بتركيز ٢٠٠مليجرام لكل كيلوجرام مقارنة بالـ doxorubicin بتركيز ١٥ مليجرام/كيلوجرام الذى خفضه الى ٩٨،٢مم بعد ٢١ يوم من المعاملة. يليهم فى التأثير بنسبة متوسطة مستخلص أزهار البابونج والذى كان ١٥٧،١مم. بينما كان التأثير الأقل لمستخلص أزهار الخرشوف والإيشليا بمعدل ١٨٤،٣ و ٢١٠،٩مم على التوالى. كذلك فإن التأثير الأعلى نشاطا يكون للمركب الكيميائى doxorubicin يليه مستخلص أزهار الأقحوان ويكون نشاط مستخلص أزهار البابونج متوسط التأثير. بينما مستخلص أزهار كلا من الخرشوف والإيشليا ذو نشاط أقل تأثيرا فى مدة الحياة وكذلك اختبارات الدم المختلفة فى الفئران المصابة بالسرطان مثل مضادات الأكسدة ، وظائف الكبد ، وظائف الكلى ، صفات الليبيدات ، وصورة الدم من الهيموجلوبين وكرات الدم الحمراء والبيضاء والصفائح الدموية ، مقارنة بالفئران المصابة بالسرطان لمدة ٢١يوم.

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