

**Possible antioxidant and anticancer effects of plant extracts from *Anastatica hierochuntica*, *Lepidium sativum* and *Carcia papaya* against Ehrlich ascites carcinoma cells**

Rasha Aly Ahmed El Sayed<sup>1</sup>, Zeinab Eid Madboly Hanafy<sup>1</sup>, Hend Fouad Abd El Fattah<sup>2</sup> and Asmaa Kutb Mohamed Amer<sup>1</sup>

<sup>1</sup>Zoology and Entomology Department, Faculty of Science, Al-Azhar University (Girl's), Cairo, Egypt

<sup>2</sup>Tissue Culture and Cytogenetics Unit, Pathology department, National Cancer Institut, Cairo University, Cairo, Egypt<sup>2</sup>.

[Asmaakutb86@gmail.com](mailto:Asmaakutb86@gmail.com)

**Abstract:** This study is undertaken to investigate the antioxidant and antimutagenic effects of three natural extracts of *Anastatica hierochuntica* (AH), *Lepidium sativum* (LS) and *Carcia papaya* (CP) against *in vivo* Ehrlich ascites carcinoma (EAC) in Swiss albino mice. EAC was induced by intraperitoneal injection of EAC-cells in the female mice. EAC-bearing mice were orally treated with 500 mg/kg body weight of AH, LS and CP extracts for 7 days after EAC intraperitoneal transplantation. Ninety female mice were divided into nine groups (10 mice/group), control group, oil, AH, LS and CP groups, EAC group (mice were inoculated with  $2.5 \times 10^6$  intraperitoneally (i.p), EAC+AH, EAC+LS and EAC + CP. The antitumor activity of AH and LS was pronounced in the results of this study as indicated by the increase of EAC tumor-bearing mice lifespan. Liver enzymes were greatly improved by treatment with AH and LS. However, the increase in glutathione peroxidase (GPx) activity that was accompanied by the marked decrease of MDA indicate the antioxidant activity of these plants. Furthermore, the reduction in MPO level in serum of EAC bearing mice revealed an obvious anti-inflammatory activity of these plants specially CP. Also, the three plants decreased chromosomal aberration and DNA fragmentation induced by EAC in mice.

[Rasha Aly Ahmed El Sayed, Zeinab Eid Madboly Hanafy, Hend Fouad Abd El Fattah and Asmaa Kutb Mohamed Amer. **Possible antioxidant and anticancer effects of plant extracts from *Anastatica hierochuntica*, *Lepidium sativum* and *Carcia papaya* against Ehrlich ascites carcinoma cells.** *Cancer Biology* 2020;10(1):1-16]. ISSN: 2150-1041 (print); ISSN: 2150-105X (online). <http://www.cancerbio.net>. 1. doi:[10.7537/marscbj100120.01](https://doi.org/10.7537/marscbj100120.01).

**Key words:** *Anastatica hierochuntica*, *Lepidium sativum*, *Carcia papaya*, Ehrlich ascites carcinoma, ALT, AST, MDA, GPx, MPO, chromosomal aberrations, DNA fragmentation

## 1. Introduction

Recently, the use of synthetic or natural agents (alone or in combination) to treat the development of cancer is a promising strategy. Various class of anticancer drugs or chemotherapeutic agents are available widely which either undergoes alkylation of DNA or microtubules arrest or altering the cells at various mitotic phases (Estrela *et al.*, 1992 and Samanta, *et al.*, 2016). With regard to the bad effect of chemotherapeutic drugs, major attention has been drawn recently to natural products with antioxidant and anti-inflammatory potential that may be treat various kinds of diseases (Huang *et al.*, 2013).

Brassicaceae family trigger a great attention as the phytochemicals present in the Brassica vegetables showed potential in chronic disease, oxidative stress, the risk of cancer, carcinogenic mutations and proliferation of cancer cells prevention (Chauhan *et al.*, 2016 and Šamec *et al.*, 2018). In addition, vegetables in the Brassicaceae family contains vitamins, catalase, superoxide dismutase and peroxidase so these vegetables are a prominent source for antioxidants activity (Ching *et al.*, 2007). With regard to the above mention, three plant of this family,

*Anastatica hierochuntica*, *Lepidium sativum* and *Carica papaya* were chosen in this study. AH is a desert plant. It is a species of *Anastatica* genus that commonly called as Kaff Maryam (Mary's hand), Rose of Jericho and Genggam Fatimah (Friedman and Stein, 1980). This herb has been well regarded for its aid in the management of various ailments (Al Gamdi *et al.*, 2011) as well as important properties like antimelanogenesis (Nakashima *et al.*, 2010), nitric oxide inhibitor (Yoshikawa *et al.*, 2003a), hepatoprotective (Yoshikawa *et al.*, 2003b and Sobhy *et al.*, 2011), gastro protective (Shah *et al.*, 2014), anti-inflammatory (Rizk *et al.*, 1985 and Abou-Ellella *et al.*, 2016) and immunostimulatory action (Abdulfattah, 2013). However, LS (garden cress) that is commonly known as "Hab el Rashaad" or "Thufa", (Gilani *et al.*, 2013) showed a great antioxidant (Zia-Ul-Haq *et al.*, 2010 and Agarwal and Verma, 2011) and anti-inflammatory (Raval *et al.*, 2013) properties as a result for the presence of many components such as tannins, benzyl isothiocyanate and flavonoids (Adamu and Boonkaewwan, 2014; Bahrami *et al.*, 2016 and Raish *et al.*, 2016). CP is a plant called as pawpaw and has been cultivated in

most of the tropical countries (Roshan *et al.*, 2014). Many researchers studied the anticancer effect of Papaya leaves (Bergonio and Perez, 2016 & Kavimandan and Saraf, 2016). Papaya leaves have shown the presence of anti-cancer, hepatoprotection, anti-inflammatory and antioxidant properties *in-vitro* and *in-vivo* studies (Nugroho *et al.*, 2017).

## 2. Materials and methods

### Plants Extracts

The dried aerial parts of AH, seeds of LS and CP leaves were purchased from Egyptian local market and grounded to fine powders. Each powder was soaked in dichloromethane for 24 hours to remove the chlorophyll, resinous and waxy materials, and then extracted with ethyl acetate several times. The extracts were filtered and evaporated under reduced pressure until the complete removal of ethyl acetate and kept in a sealed vessel for further investigation (Liu *et al.*, 1989).

### Animals

Female Swiss albino mice were used in the study aged 8-10 weeks' old with weight of 20-25 g were obtained from the animal house of National Cancer Institute, El-Giza, Egypt. Mice were kept for accommodation one week before onset of the experiment. Animals provided with standard feed pellets and water *ad libitum* under 12h of light/dark cycle. Animal use followed guidelines stated by Institutional Animal Care and Use Committee (IACUC) guidebook 8<sup>th</sup> edition (2011) found at NIH website <http://www.grants.nih.gov/grants/olaw/Guide-for-the-Care-and-use-of-laboratory-animals>.

### Experimental design

Ninety mice were divided into 9 groups (10/group), Group I (control); Group II: (oil); Group III (AH); Group IV (LS); Group V (CP); Group VI (EAC); Group VII (EAC+AH); Group VIII (EAC+LS) and Group IV (EAC+CP). The three plant extracts were applied orally by gastric intubation at dose of 500 mg/kg body weight /day. Five animals of each group were anesthetized with diethyl ether and sacrificed after seven days of tumor transplantation, whole blood was withdrawn and left to coagulate at 37°C for 15 min. Serum was then separated and stored at -20°C until further analysis (Medhat *et al.*, 2017) days, bone marrow was flushed from for mice femur for chromosome preparation and 5 mice/group were left for calculation of life span.

The animals were weighed at the beginning and the end of the treatment. The animal organs; liver, spleen, and kidneys were excised, washed with normal

saline then weighed. Also, the organ-to-body weight ratios were evaluated according to formula adopted (Ashafa *et al.*, 2012).

### Life span:

The effect of each extract on the percentage increase in life span was calculated on the basis of mortality of the experimental mice (Radha, *et al.*, 2011) was evaluated. ILS (%) = [(Mean survival time of treated group/Mean survival time of control group) - 1] × 100. Mean survival time (days) = (first death + last death)/2 (Samanta *et al.*, 2016).

### Biochemical Parameters

Aspartate transaminase (AST) and alanine transaminase (ALT) were determined in the serum of different groups by the method of Reitman and Frankel (1957), glutathione peroxidase (GPx) activity (Paglia and Valentine, 1967), MDA content (Uchiyama and Mihara, 1978) and Myeloperoxidase (MPO) activity (Manktelow and Meyer, 1986) were also analyzed.

### Chromosome aberration (CA) assay

The CA assay was carried out as described by Evans (1987). Animals were injected with 2mg/kg b.w. of colchicine 1.5 h prior to sacrifice. Bone marrow cells were collected by flushing with phosphate buffer from femur bone, treated with 0.56% KCl (pre-warmed at 37°C) and incubated for 20 min at 37°C, centrifuged, fixed in freshly prepared acetomethanol (1:3, v/v), refrigeration for 30 min., spread on slides and staining was done in 5% buffered Giemsa stain (pH 7.0). Metaphase plates were studied per animal (5 animals/group).

### DNA fragmentation

Animal tissues were used to determine the quantitative profile of the DNA fragmentation (Gibb *et al.*, 1997) and DNA fragmentation was qualitatively analyzed according to Lu *et al.* (2002).

## 3. Results:

### Effect of plants on body and organ weight:

LS and CP increased animal body weight, although organs weight did not show any significant changes compared to the control group. In EAC infected female mice, EAC increased the body and liver weight significantly comparing to control group, AH and CP did not affect significantly on body weight comparing to the EAC group, but in EAC+LS mice, body weight was significantly reduced as compared to EAC only mice group. Spleen weight was decreased for AH treated mice, while in LS and CP groups it did not show significant change as represented in table (1)

Table (1) shows the mean body and organs weights for mice treated with the three plants extracts with and without EAC:

Groups	Body weight	Liver weight	Liver ratio	Spleen weight	Spleen ratio	Kidney weight	Kidney ratio
control	21.66 ± 1.18	1.17 ± 0.07	0.054	0.2 ± 0.02	0.009	0.13 ± 0.01	0.006
EAC	34.5 ± 2.5 <sup>a</sup>	1.54 ± 0.23 <sup>a</sup>	0.044	0.16 ± 0.02	0.004	0.13 ± 0.01	0.003
oil	27.25 ± 0.85 <sup>b</sup>	1.38 ± 0.07	0.05	0.2 ± 0.03	0.007	0.13 ± 0.006	0.004
AH	23 ± 0.4 <sup>b</sup>	1.15 ± 0.03 <sup>b</sup>	0.05	0.11 ± 0.01 <sup>a</sup>	0.004	0.12 ± 0.009	0.005
LS	37 ± 2.3 <sup>a</sup>	1.42 ± 0.07	0.038	0.17 ± 0.01	0.004	0.13 ± 0.006	0.003
CP	33.75 ± 4.5 <sup>a</sup>	1.46 ± 0.09	0.043	0.18 ± 0.02	0.005	0.14 ± 0.002	0.004
EAC+AH	35 ± 1.22 <sup>a</sup>	1.42 ± 0.15	0.04	0.23 ± 0.04	0.006	0.13 ± 0.01	0.003
EAC+LS	27.75 ± 2.17 <sup>b</sup>	1.31 ± 0.15	0.047	0.15 ± 0.04	0.005	0.12 ± 0.005	0.004
EAC+CP	31.75 ± 1.93 <sup>a</sup>	1.39 ± 0.07	0.044	0.14 ± 0.01	0.004	0.105 ± 0.006 <sup>ab</sup>	0.003

AH: *Anastatica hierochuntica*; LS: *Lepidium sativum*; CP: *Carica papaya*; EAC: Ehrlich ascites carcinoma.

#### Determination of life span:

The effect of AH, LS and CP extracts on life span of EAC-bearing mice were investigated (table 2). The administration of LS was found to increase the percentage of life span by 37.14% followed by AH

that increased life span of EAC-bearing mice by 28.7%. However, the administration of CP did not show an increase in life span as compared to the EAC group.

**Table (2):** Mean survival time and percentage of increased life span for mice treated with the three plants extracts with and without EAC:

Groups	Mean survival time (days)	%ILS
EAC	17.5 ± 0.23	0
EAC+AH	22.5 ± 0.11 <sup>b</sup>	28.57142857
EAC+LS	24 ± 0.11 <sup>b</sup>	37.14285714
EAC+CP	17.5 ± 0.23	0

#### Biochemical parameters:

An elevation of liver damaging marker enzymes (ALT and AST) were reported by injection of EAC cells in mice when compared to the normal group as represented in table (3). The treatment with *Anastatica hierochuntica* significantly increased AST and ALT activity, however, in AH+EAC group, the enzyme level of both AST and ALT enzymes significantly decreased comparing to the EAC group. By comparing, the serum levels of ALT and AST in EAC-bearing mice with the EAC+LS, a significant reduction in the ALT and AST enzyme levels were recorded. No significant change in the AST level were reported at the LS group as compared to the control group. CP leaf extract significantly increased the serum activity of AST and reduced ALT compared to control group and restored the AST and the ALT level in EAC mice, recording the best effect of all.

With regard to the glutathione peroxidase (Gpx), a significant reduction in the serum level of GPx in the mice of EAC group was recorded. Moreover, the extract caused a significant elevation to Gpx activity in EAC-bearing mice as compared to the EAC group. Meanwhile, the serum MDA level of the EAC mice showed marked increase compared with mice in the control group. The results indicated that mice treated with AH+EAC, LS+EAC and CP+EAC showed a

significant reduction in the mean values of serum MDA level compared to the EAC group.

Anti-inflammatory activity of the three plant extracts was investigated by MPO activity. CP+EAC achieved the highest degree of reduction in MPO level even than that of control mice, followed by LS then AH which recorded a significant reduction compared to EAC group.

#### Chromosome aberration:

Low percentages of structural chromosomal aberration were scored (0.67±0.33) in control group, while the oil group recorded (0.83±0.31) (Table 4). On the other hand, after 7 days of EAC inoculation, major chromosomal aberrations in bone marrow were recorded as represented in table (4). This aberration includes: cells with more than one aberration, structural (chromatid gaps, chromatid breaks, deletions, fragments and centromeric attenuation). Meanwhile, EAC inoculation showed a significant numerical aberration (polyploidy and aneuploidy) that reached 8.17±1.58 as compared to control.

No significant changes were recorded in groups treated with AH, LS and CP compared to the control as represented in table (4), while the three extracts revealed a significant decrease in aberration level in AH+EAC, LS+EAC and CP+EAC mice compared to the EAC group. CP extract effect was the best of all in

chromosomal aberration reduction.

**Table 3:** Effect of *Anastatica hierochuntica* L. (AH), *Lepidium sativum* (LS) and *Carica papaya* (CP) in biochemical parameters in experimental mice with Ehrlich ascites carcinoma (EAC) at seven days treatment:

Group	AST	ALT	GPx	MDA	MPO
C	193.3± 3.3	42.3± 0.88	15.4± 0.31	2.71± 0.19	7.54 ± 0.12
EAC	339.6± 0.33 <sup>a</sup>	74.6± 0.33 <sup>a</sup>	2.78 ± 0.3 <sup>a</sup>	12.2 ± 0.12 <sup>a</sup>	11.1 ± 0.1 <sup>a</sup>
Oil	189.6 ± 5.7 <sup>b</sup>	34.3± 0.88 <sup>b</sup>	12.57 ± 0.34 <sup>a</sup>	7.59± 0.25 <sup>a</sup>	8.83 ± 0.22 <sup>a</sup>
AH	234.3 ± 2.3 <sup>ab</sup>	67 ± 3 <sup>a</sup>	12.22 ± 0.21 <sup>ab</sup>	6.75 ± 0.17 <sup>ab</sup>	8.07 ± 0.37 <sup>b</sup>
LS	203.3 ± 3.3 <sup>b</sup>	52.3 ± 3.7 <sup>ab</sup>	13.66 ± 0.2 <sup>ab</sup>	7.49 ± 0.23 <sup>a</sup>	7.46 ± 0.08
CP	229.6 ± 2.9 <sup>ab</sup>	27 ± 0.57 <sup>ab</sup>	12.87 ± 0.05 <sup>ab</sup>	8.5 ± 0.04 <sup>a</sup>	8.76 ± 0.36 <sup>a</sup>
EAC+AH	160 ± 5.7 <sup>ab</sup>	40.3 ± 4.9 <sup>b</sup>	8.55 ± 0.39 <sup>ab</sup>	9.47 ± 0.2 <sup>ab</sup>	10.11 ± 0.16 <sup>ab</sup>
EAC+LS	195 ± 5 <sup>ab</sup>	25.6 ± 1.2 <sup>ab</sup>	11.5 ± 1.18 <sup>ab</sup>	8.03 ± 2.63 <sup>ab</sup>	8.13 ± 0.9 <sup>b</sup>
EAC+CP	176.6 ± 6.6 <sup>b</sup>	30 ± 5 <sup>ab</sup>	13.26 ± 0.03 <sup>ab</sup>	3.36 ± 0.2 <sup>b</sup>	5.53 ± 0.1 <sup>ab</sup>

### Chromosome aberration:

Low percentages of structural chromosomal aberration were scored (0.67±0.33) in control group, while the oil group recorded (0.83±0.31) (Table 4). On the other hand, after 7 days of EAC inoculation, major chromosomal aberrations in bone marrow were recorded as represented in table (4). This aberration includes: cells with more than one aberration, structural (chromatid gaps, chromatid breaks, deletions, fragments and centromeric attenuation).

Meanwhile, EAC inoculation showed a significant numerical aberration (polyploidy and aneuploidy) that reached 8.17±1.58 as compared to control.

No significant changes were recorded in groups treated with AH, LS and CP compared to the control as represented in table (4), while the three extracts revealed a significant decrease in aberration level in AH+EAC, LS+EAC and CP+EAC mice compared to the EAC group. CP extract effect was the best of all in chromosomal aberration reduction.

**Table (4):** Mean ±SE. of Chromosomal aberration frequency of in bone marrow cells of mice treated with the three plant extracts (*Anastatica hierochuntica*: AH, *lepidium sativum*: LS and *Carica papaya*: CP) for seven days with/without Ehrlich ascites carcinoma (EAC)

Groups	Abnormal cells	Cells with more than one aberration	Chromatid type aberrations		Chromosome type aberrations			Total structural aberrations	Numerical aberrations		Total num. aberr.	Total aberration Excluding g.
			Chd.g.	Chd.br	D	F	CA		Poly-ploidy	Aneu-ploidy		
Control	.67±.33	0.0±0.00	0.00±0.00	.33±.21	0.00±0.00	.17±.17	.17±.17	.67±.33	0.00±0.00	0.00±0.00	0.00±0.00	.67±.33
EAC	14.33a±.92	1.5 a ± 0.43	0.50 a ± 0.22	.50±.22	0.67 a ± 0.21	.67 a ± 0.21	1.33 ±.67	4.67 a ± 1.17	1.00 a ± 0.00	7.17a ± 1.58	8.17 a ± 1.58	13.83 a ±.75
Oil	1.00b ±.45	0.2 b ± 0.17	0.17 b ± 0.17	.00±.00	0.17 ± 0.17	.33 ±.21	.17 ±.17	.83 b ±.31	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	.83 b ±.31
AH	.67b ±.33	0.0±0.00	0.00±0.00	.00±.00	0.33 ± 0.21	.17 b ±.17	.17 ±.17	.67 b ±.33	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	.67 b ±.33
LS	1.00b ±.52	0.0±0.00	0.00±0.00	.00±.00	0.33 ± 0.21	.17 b ±.17	.50 ±.34	1.00 b ±.52	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	1.00 b ±.52
CP	.33b ±.21	0.0±0.00	0.00±0.00	.00±.00	0.00 ± 0.00	.17 b ±.17	.17 ±.17	.33 b ±.21	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	.33 b ±.21
AH + EAC	5.33ab ± 2.43	0.8 a ± 0.48	0.00±0.00	.00±.00	0.50 ± 0.34	.17 b ±.17	1.50 a ± 1.02	2.50 ± 1.63	0.00 ± 0.00	2.00 ab ± 0.68	2.00 ab ± 0.68	5.17 ab ± 2.27
LS + EAC	6.33ab ± 1.69	0.2 b ± 0.17	0.33 ± 0.21	.33 ±.21	0.00 ± 0.00	.33 ±.21	.67 ±.33	2.17 b ±.79	0.17 ab ± 0.17	3.83 ab ± 0.95	4.00 ab ± 0.93	6.00 ab ± 1.51
CP + EAC	2.33b ±.76	0.2 b ± 0.17	0.17 ± 0.17	.00±.00	0.00 ± 0.00	.00 ±.00	0.5 ± 0.22	1.17 b ±.60	0.00 ± 0.00	1.00 ± 0.52	1.00 b ± 0.52	2.17 b ±.70

The number of scored cells is 250/group

G: group, Chd. g.: chromatid gap, Chd. br: chromatid break, D.: deletion F.: fragment, C. A.: centromeric attenuation.

### DNA fragmentation

The results revealed that the control group observed very low DNA fragmented bands (Figure 4). The rate of DNA fragmentation in control group and oil group was significantly lower than EAC (Figure 3). Moreover, DNA fragmentation in the group treated

with CP was relatively similar to that in control and oil treated groups.

The treatment of mice with EAC induced highest rate of DNA fragmentation with high significant differences than control and all other treated groups as well as it showed high increase in the fragmented DNA bands (Figure 4). On the other hand, treatment of EAC-exposed animals with the three plant extracts AH, LS and CP decreased significantly the rate of DNA fragmentation as well as decreased the damage in the DNA bands induced by EAC cells.

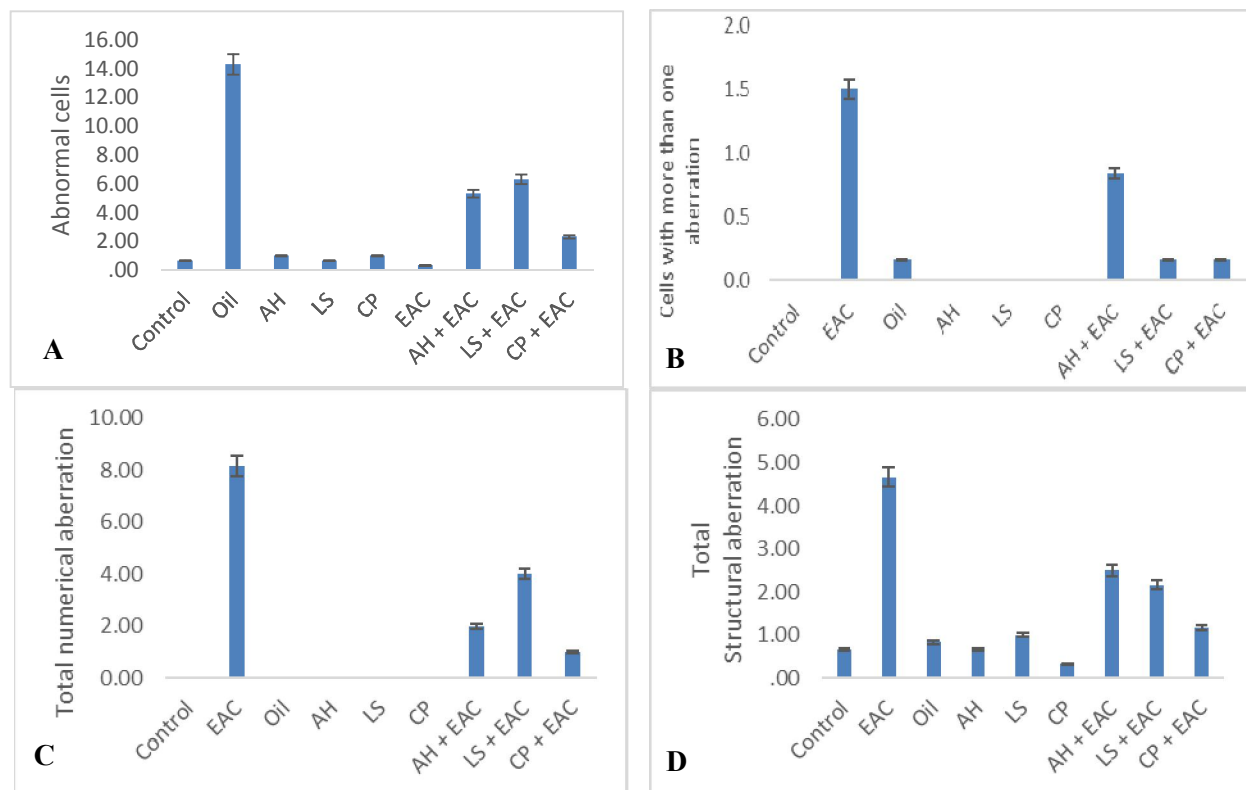


Fig. (1): Effect of *Anastatica hierochuntica* L. (AH), *Lepidium sativum* (LS) and *Carica papaya* (CP), extracts (7 days treatment) on bone marrow chromosomes of mice with/without Ehrlich ascites carcinoma (EAC) where A: abnormal cells; B: cell with more than one aberration, C: total numerical aberration and D: total structural aberration.

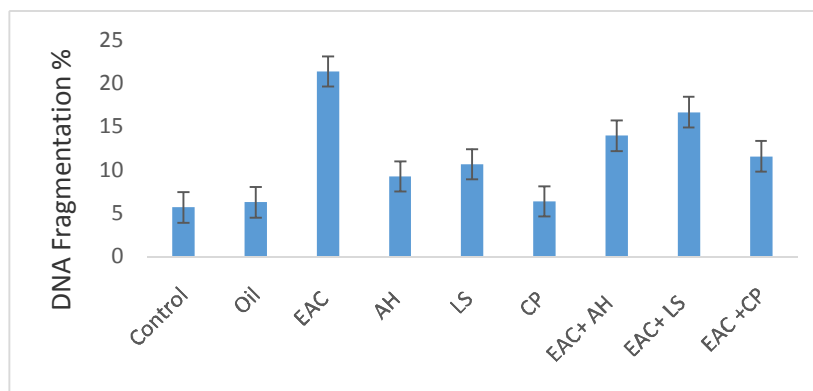
Furthermore, treatment of EAC-bearing mice with CP exhibited highest protection, where it decreased significantly the rate of DNA fragmentation

as well as decreased the damage in the DNA bands induced by EAC more than the other two extracts AH and LS as shown in table (5).

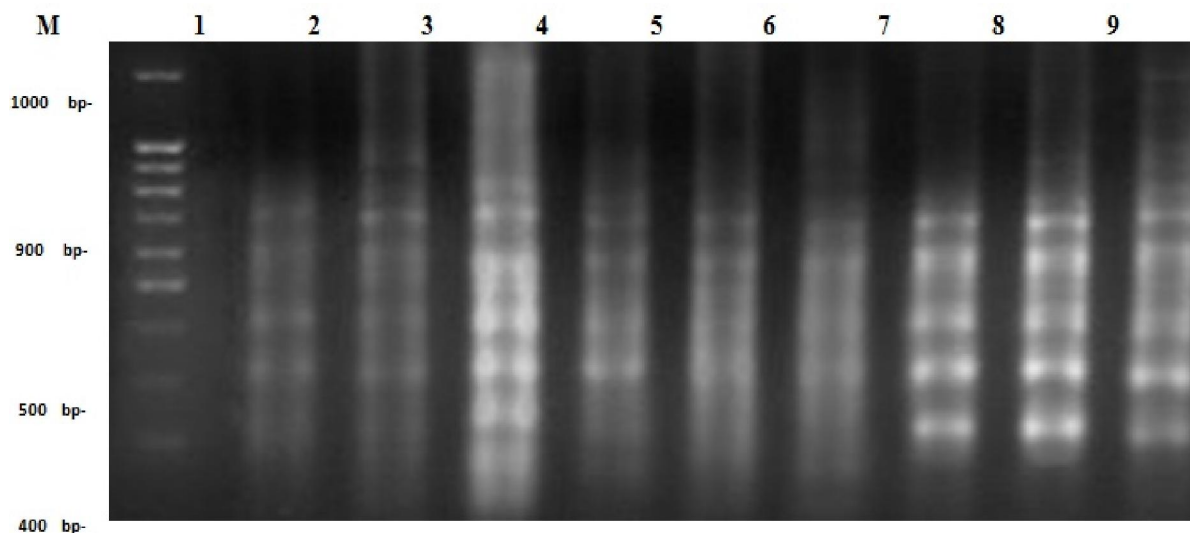
**Table 5:** DNA fragmentation detected in liver tissues of mice with/without EAC treated/untreated with *Anastatica hierochuntica* L. (AH), *Lepidium sativum* (LS) and *Carica papaya* (CP) extracts for seven days:

Treatment	DNA Fragmentation % (M ± SEM)	Change	Inhibition %
Control	5.7±0.29 <sup>c</sup>	0.0	0.0
Oil	6.3±0.30 <sup>c</sup>	0.6	0.0
EAC	21.4±0.32 <sup>a</sup>	15.6	173.68
AH	9.3±0.31 <sup>c</sup>	3.6	0.0
LS	10.7±0.28 <sup>bc</sup>	5.0	0.0
CP	6.4±0.67 <sup>c</sup>	0.6	0.0
EAC+ AH	14.0±0.58 <sup>b</sup>	8.3	45.61
EAC+ LS	16.7±0.33 <sup>b</sup>	11.0	92.98
EAC+CP	11.6±0.65 <sup>bc</sup>	6.0	5.26

Means with different superscripts (a, b, and c) between groups in the same column are significantly different at P<0.05.



**Figure 3:** DNA fragmentation in liver tissues exposed to EAC and treatments with different plants extracts (AH, LS and CP). Results are expressed as mean±SEM of data from analyzed samples. <sup>a,b,c</sup> Mean values within tissue with unlike superscript letters were significantly different ( $P < 0.05$ ).



**Figure 4:** DNA fragmentation detected with agarose gel of DNA extracted from liver tissues by DNA gel electrophoresis laddering assay. Lane 1 represents control group. Lane 2 represents oil treatment. Lane 3 represents group treated with EAC. Lanes 4-6 represent groups treated with AH, LS and CP extracts, respectively. Lanes 7-9 represent groups treated with AH+EAC, LS+EAC and CP+EAC respectively.

#### 4. Discussion

Ehrlich ascites carcinoma (EAC) is one of the commonly used experimental breast tumor that derived from spontaneous mouse adenocarcinoma that characterized by accumulation of ascitic fluid in the peritoneal cavity (Ulakoglu and Altun, 2004). The *in vivo* antitumor and antiproliferative activity was evaluated against EAC using mean survival time (MST) and increased life span percentages (%ILS) as reported by Abu Osman *et al.* (2011). Treatment of EAC-bearing mice with AH and LS extracts separately by oral gastric tube at 500 mg/kg. body weight in

EAC-infected mice, revealed an increase in the mean survival time (MST) and life span (%ILS) with a reduction in body weight that may be attributed to up-regulation of humoral factors and diminished rate of cell division by the effect of the extracts (Bhattacharya and Halder, 2012). It is suggested that the extract might be absorbed by the tumor cells which involved in lysis of these cells by cytotoxic mechanisms (Samanta *et al.*, 2016). Meanwhile, this effect may be attributed to the induction of apoptosis or the inhibition of neovascularization and tumor progression or an indirect local effect, which may

involve macrophage activation and vascular permeability inhibition as reported by many studies (**Dolai et al., 2012** and **Samanta et al., 2016**). This effect probably caused by the quercetin content in these plants that was reported to induced cytotoxicity in tumor cells without significantly affecting the normal cells (**Elsisy et al., 2017**).

The serum level of liver enzymes (ALT and AST) that are considered as reliable indices of hepatotoxicity (**Singh et al., 2011** and **Dolai et al., 2012**), showed a significant increase in EAC – bearing mice as compared with that of the normal mice that may indicates the loss of functional integrity of the liver as reported by **Halaby et al. (2015)**. However, **Salem et al., (2011)** and **Dolai et al., (2012)** attributed this raised activities of liver enzymes to the hepatocellular damages. Meanwhile, **Abu-Sinna et al. (2003)** have suggested that the consumption of free amino acids in building the proteins of rapidly dividing tumor cells might result in the disturbances of the enzyme activity in the liver. Treatment of EAC-infected with AH, LS and CP extracts separately for seven days in the present study resulted in reduction in AST and ALT activities. Reduced level of these hepatic enzymes in serum is one of the indications of the antitumour potential (**Sundaram et al., 2012**). The hepatoprotective effect of AH may be revealed to its content of Anastatins A and B that effect on d-galactosamine induced cytotoxicity in hepatocytes as reported by **Yoshikawa et al. (2003b)** or via antioxidant property of its polyphenolic compounds (**Shah et al., 2014**). LS extract significantly reduced the mean values of serum AST and ALT in EAC-bearing mice revealing hepatoprotection, chemopreventive and chemotherapeutic activity as confirmed by **Behrouzian et al. (2014)**, **Halaby et al. (2015)** and **Abd El-Kaream (2019)**. This effect may be due to the presence of phenolic compound in the extract (**Al-Asmaria et al., 2015**) or prevention the oxidative stress related liver damage (**Lee et al., 2013**). In agreement with these results, **Sakran et al. (2014)** and **Raish et al. (2016)** proved that LS seed extract (LS) has the ability to reduce hepatotoxicity by limiting damage to the liver cells, with a significant improvement and normalizing of the liver enzyme levels that effect may attributed to the presence of isoflavonoids and glycosilated phenols (**Aranda et al., 2007**; **Sakran et al., 2014**) such phenols was found to markedly restoring these enzymes to their normal levels (**Al-Otaibi et al., 2019**).

EAC-bearing mice in the present study recorded a significant decreased level of glutathione peroxidase. This result agrees with that of **Das et al. (2014)** who stated that, the cellular proliferation of tumors is involved in the decrease of GPx activity. Meanwhile, **Samudrala et al. (2015)** found that EAC-mice had a

decrease in the activity of SOD, CAT and GSH, that may be due to elevated MDA level that is claimed to be an inhibitor to protective enzymes, hence, it could have both mutagenic and carcinogenic effects (**Ziech et al., 2010**).

AH, LS and CP extracts increased the level of Gpx in EAC mice with the effect increasing respectively. AH effect may be due to its content of flavonoids, chlorogenic acids, and phenolic compounds with antioxidant (**Jaiswal et al., 2011**) and free radical scavenging potential (**Nakashima et al., 2010** and **Jaiswal et al., 2011**). Luteolin was found to increase the levels of SOD, CAT, GPx as mentioned by **Sharma et al. (2007)**.

On the other hand, there is a significant increase in the antioxidant enzymes in the EAC-mice administered LS compared with EAC-mice only. This increase is due to the ability of LS extract to enhance the endogenous antioxidant activity owing to the presence of alkaloids, amino acids, flavonoids, glycosides, phytosterols, saponins and tannins (**Thoppil and Bishayee, 2011** and **Abd El-Kaream, 2019**) beyond its free radical scavenging property and the reduction of lipoperoxide formation (**Lee et al. 2013** and **Ebtesam et al., 2016**).

Also, these results were in agreement with **Ojo et al. (2017)** who found that administration of *C. papaya* extract considerably increased antioxidant enzymes, signifying the capability of *C. papaya* leaf extract to reduce oxidative stress in Ehrlich-exposed animals by its content of antioxidant compounds that impact on the gene expression associated with oxidative stress (**Manu and Kuttan, 2008** and **Shan et al., 2009**).

MDA is a main product of lipid peroxidation that has the potential not only to evaluate the extent of oxidative injury, but also to predict the potential efficiency of therapeutic strategies aimed at restricting the oxidative stress (**Yazdanparast et al., 2007** and **Halaby, et al., 2013**). The present study recorded a significant increase in the amount of MDA in the EAC-bearing mice. This come in line with **Mansour and Anis (2010)** and **Aldubayan et al. (2019)** who demonstrated a rise in MDA level associated with impaired hepatic function, indicating general toxicity in EAC mice. Meanwhile, **Al-Rasheed et al. (2018)** attributed this elevation to the peroxidation of cell membranes and plasma lipoproteins that released into the blood stream or defect in the antioxidant system as stated by **Kumaraguruparan et al., (2002)**. However, treatment with AH, LS and CP extracts, significantly reduced MDA levels in EAC mice. The significant reduction of MDA level by AH extra that may be attributed to their content of flavonoids: Anastatin A and anastatin B, glucosinolates and their derived isothiocyanate that were confirmed to possess antioxidant (free radical scavenging) properties

(Adewusi and Afolayan, 2010 and Mota *et al.*, 2011) they act by inhibition of lipoxygenase and cyclooxygenase pathways, and they are useful to explicate fatty acid peroxidation process (Szabo, 1989 and Mohammed *et al.*, 2010). LS extract results was in line with Halaby *et al.*, (2015) that recorded a significant reduction in the serum MDA level. It may be suggested that the activities of the LS plant are due to its free radical scavenging activities and the rich content in antioxidants as vitamin C, E and carotenoids, also phytochemical constituents such as (polyphenols and flavonoids) that have been reported to protect the body system against reactive oxygen species, as mentioned by Donno *et al.*, (2013). Administration of *C. papaya* extract reduced MDA levels, signifying the capability of *C. papaya* leaf extract to reduce oxidative stress in EAC-mice as confirmed by Elsisy *et al.* (2017). This effect may be due to its content of quercetin that reported to attenuate the oxidative stress and it caused a significant decrease of MDA (Abd-Elbaset *et al.* 2015).

EAC was found to increase the amount on MPO in mice serum. This may be due to the fact that several human tumors present high levels of ROS (Reuter *et al.* 2010). In this context, the immune system is activated through the inoculation of mice with Ehrlich tumor (Schneider and Oliveira 2004). Previous studies that have shown that most of the plant extracts reduce EAC induced myelotoxicity due to their immune boosting, antioxidant and free radical scavenging activity. (Dolai *et al.*, 2012). In the present study, AH, LS and CP recorded a significant reduction in the amount of MPO. LS may act through suppression of the neutrophil infiltration and inflammatory cytokine release to the injured myocardium (Mohamed and Safwat, 2016) or by increasing the weight of the spleen (Mahassni and Khudauardi, 2017) which may result from the increased production and storage of immune system cells increase in the mean WBC count and attenuates the myeloperoxidase (MPO) level (Raish *et al.*, 2016). LS extract effect may be due to the presence of lipid compounds including alpha linoleic acid which has anti-inflammatory activity (Ferrucci *et al.*, 2006 and Zhao *et al.*, 2007). The role of alpha linoleic acid is to down regulate the expression of inflammatory cytokines such as IL-6, IL-1 $\beta$  and TNF- $\alpha$  (Zhao *et al.*, 2007). CP exhibited a significant inhibition in MPO value in EAC-bearing mice. A study on the *C. papaya* leaves extract showed that it increased the levels of white blood cells, thrombocytes, lymphocytes and neutrophils of rats. (Nwiloh *et al.*, 2009). *Papaya* is known for its antimicrobial, anti-inflammatory, immunomodulatory (Otsuki *et al.*, 2010), and antioxidant properties (Owoyele *et al.*, 2008 and

Oboh *et al.*, 2013). Anti-inflammatory and antioxidant properties of papaya are attributed to primary and secondary bioactive components such as papain, chymopapain, papaya lipase, and carotenoids (Nafiu and Rahman, 2015).

The flavonols and flavonoids have stimulant effect on blood cell production. (Sundarmurthy *et al.*, 2017). Researchers have found that *C. papaya* leaf extract can mediate Th1-type shift in human immune system, which could potentially help regulate the immune system, treat or prevent cancer, and also serve as an immune adjuvant for vaccine therapy (Otsukia *et al.*, 2010). Furthermore, it is shown to influence platelet production and aggregation by activating arachidonate 12-lipoxygenase (ALOX 12) and platelet-activating factor receptor (PTAFR) genes (Sundarmurthy *et al.*, 2017). However, saponins (present in AH and CP) demonstrated to have immunomodulatory potential via cytokine interplay (Sun *et al.*, 2009), cytostatic and cytotoxic effects on malignant tumor cells (Bachran *et al.*, 2008). Quercetin (present in AH, LS and CP) inhibits pro inflammatory cytokine gene expression through the modulation of NF- $\kappa$ B system (Nair *et al.*, 2006).

In the present study, the EAC inoculated intraperitoneal in female mice caused high incidence of chromosomal aberration in mice bone marrow cells. These findings imply that Ehrlich's solid tumor (EST) caused loss in chromatin as results of a damage to either chromosome structure (fragmentation) or dysfunction of the spindle apparatus or centromere kinetochore complexes (Fenech, 2000). Many studies suggested that EAC induced oxidative stress in mice (Gupta *et al.*, 2004 a, b) that can cause hypoxia, reoxygenation and damage to cellular macromolecules and lead to increased genetic instability (Vafa *et al.*, 2002; Moeller *et al.*, 2004 and Corn and El-Deiry, 2007).

Kondoh *et al.* (2013) reported that hypoxia induced chromosomal abnormalities (aneuploidy) in endothelial cells through the induction of reactive oxygen species and excess signaling of vascular endothelial growth factor in the tumor microenvironment. This concept was in agreement with Salah *et al.* (2011) who found that, oxidative stress produced great chromosomal aberrations, this damage DNA and block the action of RNA polymerase and so prevent replication process. Also, Pang, 1995 stated that, cells exposed to reactive oxygen species will incur a range of genetic aberrations, which may either be structural (e.g. deletion, DNA strand breaks) or numerical (e.g. chromosomal loss/ gain).

Oral administration of LS, AH and CP extracts at 500 mg/kg b.wt. was found to reduce chromosome aberration induced in mice bone marrow cells after



EAC intraperitoneal inoculation with their effect increasing respectively indicating that the best effect was for CP extract.

In agreement with **Salah et al. (2011)**, AH extract and revealed an inhibition in the frequency of chromosomal aberrations significantly in both somatic and germ cells of diabetic rats. The therapeutic effect of AH may have attributed to its content of polyphenols including flavonoids which have known protective activity (**Zayachkivska et al., 2005**). The positive effect of LS on mice bone marrow chromosomes may be due to its antitumor activities. LS extract activity was studied by **Ait-Yahia et al. (2015)** who showed its antitumor activities towards Hep2.

The present results on chromosomes was in agreement with the study of **Ojo et al., (2017)** who found that the treatment with *C. papaya* extract significantly decreased chromosomal aberrations and, micro nuclei induction in arsenic induced genotoxicity in Wistar rats.

The leaves of *Carica papaya* L. contain the alkaloid carpaine (**Ramasawamy and Sirsi, 1960**), that has been found to possess antitumor activity *in-vitro* against mouse lymphoid leukemia L1210, lymphocytic leukemia P388 and Ehrlich ascites tumor cells. The leaves of papaya also have been shown to contain many active components that can increase the total antioxidant power in blood and reduce lipid peroxidation level, such as papain, chymopapain, cystatin,  $\alpha$ -tocopherol, ascorbic acid, flavonoids, cyanogenic glucosides and glucosinolates which elucidate its highest effect on chromosome improvement (**Seigler et al., 2002** and **Sundarmurthy et al., 2017**).

The three plant extracts were reported to contain quercetin and luteolin that were known with their chemo preventive activity against chromosomal alterations. Luteolin suppressed genetic damage in the micronucleus assay and chromosome aberrations induced by fish extract. Mutton extract-induced micronuclei and chromosome aberrations were reduced by luteolin and quercetin (**Taj et al., 1996**). Another studies have shown that quercetin administration to mice led to a decrease in the amount of chromosomal aberrations induced by methotrexate in bone marrow cells and diminished the number of cells with methotrexate-induced aberrations (**Sekeroğlu & Sekeroğlu, 2012**).

In this study, we observed high level of DNA fragmentation induced in mice liver after 7 days of EAC intraperitoneal injection. These results come in a close resemblance to the study done by **Hanafy (2009)** who found an increase in DNA fragmentation in mice liver of EAC-mice and the finding obtained by **Khalil et al., (2014)** who found high degree of DNA damage

in bone marrow cells of Ehrlich's solid tumor (EST). Free radicals may lead to a decline in the level of glutathione and increased oxidative damage to normal proliferating cells and may lead to oxidative damage to macromolecules, including DNA damage, which in turn contributes to the formation of DNA strand breaks, (**Sallmyr et al., 2008** and **Papież, 2014**).

More evidence suggests that the potentially cancer-inducing oxidative damage might be prevented or limited by antioxidant. Antioxidant may mediate their effect by direct reaction with ROS, quenching them or chelating the catalytic metal ions (**Sun et al., 2002**). It has been shown that antioxidant rich diets can reduce oxidative damage to DNA, thus preventing a critical step in the onset of carcinogenesis (**Meyskens et al., 2005**). Many cancer chemopreventive agents possess antioxidant potential due to their bioactive phytochemicals that may play a vital role in protecting the cell from oxidative stress (**Ozben, 2007**).

The present study found a positive effect of the three extracts on DNA fragmentation, they significantly reduced the incidence of DNA fragmentation. Previous studies have suggested that the DNA damage was decreased by antioxidant supplementation (**Bingöl et al., 2014; Abraham et al., 2016** and **Koyuncu, 2018**). AH and CP recorded better effect than LS effect, which may be due to their content of Saponins that interfere with the replication of cellular DNA and they prevent the proliferation of cancer cells. (**Yıldırım and Kutlu, 2015**). The higher protective effect against DNA damage was exerted by CP leaf extract that may be owing to its content of tannins which have the properties of the binding metallic ions and also the display of anti-oxidant activities (**Yıldırım and Kutlu, 2015**). *Carica papaya* L or papaya is one of the plants that are known for anticancer therapy by increasing the apoptosis and inhibiting the proliferation (**Puspitasari and Peristiowati, 2016**).

Quercetin that present in the three plant extracts AH, LS and CP. **Srivastava et al. (2016)** reported a change in DNA bands in quercetin antitumor efficacy in animal models and cancer cell lines. **Lin et al. (2002)** reported the role of quercetin in protection against cooking oil fumes-induced DNA damage in human lung adenocarcinoma. Quercetin also significantly reduced the oxidative DNA damage caused by etoposide both *in vitro* and *in vivo* (**Papież, 2014**). **Sashi et al. (2016)** studied the potential roles of dietary agents (including Quercetin) exerting antioxidant properties that may impede cancer progression, and reported an anti-oxidative effect against oxidative stress status in cancer. The efficacy of Quercetin as antitumor agent where it caused an elevation in Quinone Reductase 1 (QR1). QR is an

important phase II cytoprotective enzyme that converts quinones to hydroquinones, reducing oxidative cycling. It exhibits cancer protective activity mainly by inhibiting the formation of intracellular semiquinones radicals, and by generating  $\alpha$ -tocopherol hydroquinone, which acts as a free radical scavenger. Induction of QR1 often coincides with induction of other phase II enzymes, and is therefore useful in the study of chemopreventive agents (Cuendet *et al.*, 2006).

#### Conclusion

The natural extracts of *Anastatica hierochuntica* (AH), *Lepidium sativum* (LS) and *Carica papaya* (CP) revealed great antioxidant, anti-inflammatory, antimutagenic and antitumor effects against *in vivo* Ehrlich ascites carcinoma (EAC) in Swiss albino mice. These effects were indicated by an increase of EAC tumor-bearing mice lifespan, increase in glutathione peroxidase (GPx) and marked reduction of MDA and MPO levels in the sera of EAC bearing mice. Also, the three plants specially *Carica papaya* decreased both of chromosomal aberration and DNA fragmentation in EAC tumor-bearing mice.

#### References

1. Abd El-Kaream, S.A. (2019): Biochemical and biophysical study of chemopreventive and chemotherapeutic anti-tumor potential of some Egyptian plant extracts. *Biochemistry and Biophysics Reports*, 18:100637.
2. Abd-Elbaset, M.; Arafa, E-SA.; El Sherbiny, G.A.; Abdel-Bakky, M.S. and Elgendy, A.N.A.M. (2015): Quercetin modulates iNOS, eNOS and NOSTRIN expressions and attenuates oxidative stress in warm hepatic ischemia reperfusion injury in rats. *J. basics. appl. Sci.*, 4: 246–255.
3. Abu-Sinna, G.; Esmat, A.M.; Al-Zahaby, S.; Soliman, N.A. and Ibrahim, T.M. (2003): Fractionation and characterization of *Cerastes cerastes* snake venom and the antitumor action of its lethal and non-lethal fractions. *Toxicon*, 42: 207-215.
4. Abdulfattah, S.Y. (2013): Study of immunological effect of *Anastatica hierochuntica* (Kaff Maryam) plant methanolic extract on albino male mice. *Journal of Biotechnology Research Center*, 7(2):3-10.
5. Abou-Elella, F.; Hanafy, E.A. and Gavamukulya, Y. (2016): Determination of antioxidant and anti-inflammatory activities, as well as *in vitro* cytotoxic activities of extracts of *Anastatica hierochuntica* (Kaff Maryam) against HeLa cell lines. *Journal of Medicinal Plants Research*, 10(7):77-87.
6. Abraham, S.K.; Khandelwal, N.; Hintzsche, H. and Stopper, H. (2016): Antigenotoxic effects of resveratrol: assessment of *in vitro* and *in vivo* response. *Mutagenesis*, 31: 27-33.
7. Abu Osman, M.; Rashid, M.M.; Abdul Aziz, M.; Habib, M.R. and karim, M.R. (2011): Inhibition of Ehrlich ascites carcinoma by Manilkara zapota L. stem bark in Swiss albino mice. *Asian Pacific Journal of Tropical Biomedicine*, 1(6): 448 – 451.
8. Adamu, M. and Boonkaewwan, C. (2014): Effect of *Lepidium sativum* L. (Garden Cress) seed and its extract on experimental *Eimeria tenella* infection in broiler chickens. *Kasetsart J. Nat. Sci.*, 48:28–37.
9. Adewusi, E.A. and Afolayan, A.J. (2010): A review of natural products with hepatoprotective activity. *J. Med. Plants Res.* 4: 1318-1334.
10. Agarwal, J. and Verma, D.L. (2011): Antioxidant activity-guided fractionation of aqueous extracts from *Lepidium sativum* and identification of active flavonolglycosides. *Acad. Arena* 3:14–18.
11. Ait-Yahia, O.; Bouzroua, S. A.; Belkebir, A.; Kaci, S. and Aouichat, A. B. (2015): Cytotoxic activity of flavonoid extracts from *Lepidium sativum* (Brassicaceae) seeds and leaves. *International Journal of Pharmacognosy and Phytochemical Research*, 7(6):1231-1235.
12. Al-Asmari, A. K.; Athar, T.; Al-Shahrani, H. M.; Al-Dakheel, S.I. and Al-Ghamdi, M. A. (2015): Efficacy of *Lepidium sativum* against carbon tetra chloride induced hepatotoxicity and determination of its bioactive compounds by GC–MS. *Toxicology Reports, Toxicology Reports*, 2: 1319–1326.
13. Al Gamdi, N.; Mullen, W. and Crozier, A. (2011): Tea prepared from *Anastatica hierochuntica* seeds contains a diversity of antioxidant flavonoids, chlorogenic acids and phenolic compounds. *Phytochemistry*, 72(2-3):248-254.
14. Al Gamdi, N.; Mullen, W. and Crozier, A. (2011): Tea prepared from *Anastatica hierochuntica* seeds contains a diversity of antioxidant flavonoids, chlorogenic acids and phenolic compounds. *Phytochemistry*. 72(2-3):248-254.
15. Aldubayan, M.A.; Elgharabawy, R.M.; Ahmed, A.S. and Tousson, E. (2019): Antineoplastic Activity and Curative Role of Avenanthramides against the Growth of Ehrlich Solid Tumors in Mice. *Oxidative Medicine and Cellular Longevity*. Volume 2019, Article ID 5162687, 12 pages.
16. Al-Otaibi, M.S.A.; Al-Quraishy, S.; Al-Malki, E.S. and Abdel-Baki, A.S. (2019): Therapeutic

- potential of the methanolic extract of *Lepidium sativum* seeds on mice infected with *Trypanosoma evansi*. *Saudi Journal of Biological Sciences*, 26: 7, 1473-1477.
17. Al-Rasheed, N. M.; El-Masry, T. A.; Tousson, E.; Hassan, H. and Al-Ghadeer, A. (2018): Hepatic protective effect of grape seed proanthocyanidin extract against Gleevec-induced Apoptosis, Liver Injury and Ki67 alterations in rats. *Brazilian Journal of Pharmaceutical Sciences*, 54(2):17391.
  18. Aranda, E.; García-Romera, I.; Ocampo, J.A.; Carbone, V.; Mari, A.; Malorni, A.; Sannino, F.; De Martino, A. and Capasso, R. (2007): Chemical characterization and effects on *Lepidium sativum* of the native and bioremediated components of dry olive mill residue. *Chemosphere*, 69:229-239.
  19. Ashafa, A.O.T.; Orekoya, L.O. and Yakubu, M.T. (2012): Toxicity profile of ethanolic extract of *Azadirachta indica* stem bark in male Wistar rats. *Asian Pac. J. Trop. Biomed.*, 2:811-7.
  20. Bachran, C.; Bachran, S.; Sutherland, M.; Bachran, D. and Fuchs, H. (2008): Saponins in tumor therapy. *Mini Rev. Med. Chem.*, 8: 575-584.
  21. Bahrami, S.; Jalali, M.H.R.; Ramezani, Z.; Boroujeni, M.P. and Toeimepour, F. (2016): In vitro scolicidal effect of *Lepidium sativum* essential oil. *J. Ardabil Univ. Med. Sci.* 15:395-403.
  22. Behrouzian, F.; Razavi, S. M. A. and Phillips, G. O. (2014): Cress seed (*Lepidium sativum*) mucilage, an overview. *Bioactive Carbohydrates and Dietary Fibre*, 3(1): 17-28.
  23. Bergonio, K.B. and Perez, M.A. (2016): The potential of male papaya (*Carica papaya* L.) flower as a functional ingredient for herbal tea production. *Indian journal traditional knowledge* 15(1): 41-49.
  24. Bhattacharya, S. and Halder, P.K. (2012): The triterpenoid fraction from *Trichosanthes dioica* root exhibits antiproliferative activity against Ehrlich ascites carcinoma in albino mice: involvement of possible antioxidant role. *J. Expe. Therap. and Enco*, 9: 281 - 290.
  25. Bingöl, G.; Gülkaç, M.D.; Dillioğlugil, M.O.; Polat, F. and Kanli, A.Ö. (2014): Effect of resveratrol on chromosomal aberrations induced by doxorubicin in rat bone marrow cells. *Mutat Res Genet Toxicol Environ Mutagen*, 766: 1-4.
  26. Chauhan, E.S.; Tiwari, T. and Singh, A. (2016): Phytochemical screening of red cabbage (*Brassica oleracea*) powder and juice - A comparative study. *J Med Plants Stud.* 4(5):196-199.
  27. Ching, S.; Herbariae, I.R. and Plantarum, S. (2007): Origins and diversity of brassica and its relatives. In. 2007:1-33.
  28. Corn, P.G. and El-Deiry, W.S. (2007): Microarray analysis of p53-dependent gene expression in response to hypoxia and DNA damage. *Cancer Biol. Ther.* 6: 1858-1866.
  29. Cuendet, M.; Oteham, C.; Moon, R. and Pezzuto, J. (2006): Quinone Reductase induction as a biomarker for cancer chemoprevention. *J. Nat. Prod.*, 69: 460-463.
  30. Das, M.K.; Mukkanti, K.; Rao, G.S.; Sahu, P.K. and Silpavathi, L. (2014): Evaluation of antitumor and antioxidant potential of a polyherbal extract on Ehrlich's ascites carcinoma xenografted mice. *Journal of Pharmacy and Nutrition Sciences*, 4: 20-26.
  31. Dolai, N.; Karmakar, I.; Kumar, R.B.S.; Kar, B.; Bala, A. and Haldar, P.K. (2012): Evaluation of antitumor activity and in vivo antioxidant status of *Anthocephalus cadamba* on Ehrlich ascites carcinoma treated mice. *Journal of Ethnopharmacology*, 142: 865-870.
  32. Donno, D.; G. Beccaro, M.; Mellano, S.; Canterino, A.; Cerutti and Bounous, G. (2013): Improving the nutritional value of kiwifruit with the application of agroindustry waste extracts. *Journal of Applied Botany and Food Quality*, 86: 11 - 15.
  33. Elsisy, M.K.; Ibrahim, W.M.; Salama, A.F. and Kasem, S.M. (2017): The Antitumor Potential of Quercetin on Solid Ehrlich Tumor in Female Mice. *J. Can. Sci. Res.*, 2(1):1-7.
  34. Estrela, A.A.; Pooley, H. M.; de Lencastre, H. and Karamata, D. (1991): Genetic and biochemical characterization of Bacifh subtilis 168 mutants specifically blocked in the synthesis of the teichoic acid poly (3-O-~glucopyranosyl-N-acetylgalactosamine 1-phosphate): gneA, a new locus, is associated with UDP-N-acetylglucosamine 4-epimerase activity. *J Gen Microbiol* 137: 943-950.
  35. Evans, E.P. (1987): Karyotyping and sexing of gametes, embryos, fetuses and *in situ* hybridization to chromosomes. In: *Mammalian Development. A Practical approach*, Non K.M (Ed) pub. IRL, Press Oxford, pp. 93-100.
  36. Fenech, M. (2000): The in Vitro Micronucleus Technique. *Mutation Research*, 455: 81-95.
  37. Ferrucci, L.; Cherubini, A.; Bandinelli, S.; Bartali, B.; Corsi, A.; Lauretani, F.; Martin, A.; Andres-Lacueva, C.; Senin, U. and Guralnik, J.M. (2006): Relationship of plasmalipolysaturated fatty acids to circulating inflammatory markers, *J. Clin. Endocrinol. Metab.*, 91:439-446.

38. Friedman, J. and Stein, Z. (1980): The influence of seed-dispersal mechanisms on the dispersion of *Anastatica hierochuntica* (Cruciferae) in the Negev Desert, Israel. *The Journal of Ecology*, 43-50.
39. Gibb, R.K.; Taylor, D.D.; Wan, T.; Oconnor, D.M.; Doering, D.L. and Gercel-Taylor, C. (1997): Apoptosis as a measure of chemosensitivity to cisplatin and taxol therapy in ovarian cancer cell lines. *Gynecologic Oncology*, 65: 13-22.
40. Gilani, A.H.; Rehman, N.U.; Mehmood, M.H. and Alkharfy, K.M., (2013): Species differences in the antidiarrheal and antispasmodic activities of *Lepidium sativum* and insight into underlying mechanisms. *Phytother. Res.*, 27:1086–1094.
41. Gupta, M.; Mazumder, U.K.; Kumar, R.S. and Kumart, S. (2004b): Antitumor activity and antioxidant role of *Bauhinia racemosa* against Ehrlich ascites carcinoma in Swiss albino mice. *Acta pharmacol. sin*, 25: 1070 – 1076.
42. Gupta, M.; Mazumder, U.K.; Kumar, R.S.; Sivakumar, T. and Vamis, M.L.M. (2004a): Antitumor activity and antioxidant status of *Caesalpinia bunducella* against Ehrlich ascites carcinoma in Swis albino mice. *J. pharmacol. Sci.*, 94: 177 – 184.
43. Halaby, M.S.; Farag, M. H. and Mahmoud, S.A.A. (2015): Protective and Curative Effect of Garden Cress Seeds on Acute Renal Failure in Male Albino Rats. *Middle East Journal of Applied Sciences.*, 5(2): 573-586.
44. Halaby, M.S.; Farag, M.H. and Mohammed, A.Z. (2013): Influence of Kiwifruits on hypercholesterolemia in male albino rats. *Egyptian J. of Nutrition and Health*. Published by Society of Feeding mind, Combating malnutrition. 8 (1): 21 – 36.
45. Hanafy, Z. E. (2009): Ginger extract Antimutagens as Cancer Chemopreventive Agent against Ehrlich Ascites Carcinoma. *Academic J. Cancer Research*; 2(2): 61-67.
46. Huang, S.S.; Chiu, C.S.; Lin, T.H.; Lee, M.M.; Lee, C.Y.; Chang, S.J.; Hou, W.C.; Huang, G.J. and Deng, J.S. (2013): Antioxidant and anti-inflammatory activities of aqueous extract of *Centipeda minima*. *J. Ethnopharmacol*, 147: 395–405.
47. Jaiswal, A.K.; Rajauria, G.; Abu-Ghannam, N. and Gupta, S. (2011): Phenolic composition, antioxidant capacity and antibacterial activity of selected Irish Brassica vegetables. *Nat. Prod Commun.*, 6: 1299-1304.
48. Jaiswal, A.K.; Rajauria, G.; Abu-Ghannam, N. and Gupta, S. (2011): Phenolic composition, antioxidant capacity and antibacterial activity of selected Irish Brassica vegetables. *Nat. Prod Commun.*, 6: 1299-1304.
49. Kavimandan, B. and Saraf, M. (2016): Studies on Biological Efficacy of Various Leaf Extracts of *Carica Papaya L.* *International Conference on Global Trends in Engineering, Technology and Management*, pp: 510-516.
50. Khalil, W.K.B.; Ghaly, I.S.; Diab, K.A.E. and E Lmakawy, A.I. (2014): Antitumor activity of *Moringa Oleifera* leaf extract against Ehrlich solid tumor. *Int. J. Pharm.*, 4(3): 68-82.
51. Kondoh, M.; Ohga, N.; Akiyama, K.; Hida, Y.; Maishi, N.; Towfik, A.M.; Inoue, N.; Shindoh, M. and Hida, K. (2013): Hypoxia-induced reactive oxygen species cause chromosomal abnormalities in endothelial cells in the tumor microenvironment. *PLOS ONE*, 8(11) e80349.
52. Koyuncu, I. (2018): Resveratrol Attenuates Bleomycin-Induced Genotoxicity, Pulmonary Fibrosis and DNA Damage in Balb/C Mice with Ehrlich Ascites Carcinoma. *Bezmialem Science*, 6(4): 262-271.
53. Kumaraguruparan, R.; Subapriya, R.; Kabalimoorthy, J. and Nagini, S. (2002a): Antioxidant profile in the circulation of patients with fibroadenoma and adenocarcinoma of the breast. *Clin. Biochem.*, 35:275–279.
54. Lee, C.C.; Shen, S.R.; Lai, Y.J. and Wu, S.C. (2013): Rutin and quercetin, bioactive compounds from tartary buckwheat, prevent liver inflammatory injury,” *Food & Function*, 4(5):794–802.
55. Lin, S.Y.; Tsai, S.J.; Wang, L.H.; Wu, M.F. and Lee, H. (2002): Protection by quercetin against cooking oil fumes-induced DNA damage in human lung adenocarcinoma CL-3 cells: role of COX-2. *Nutr. Cancer*. 44(1):95-101.
56. Liu, Y.L.; Neuman, P.; Borbara, N.T. and Mabry, J.J. (1989). Technique for flavonoids analysis. *Rev. Latinamer. Quim. Suppl.* 1: 90 – 130.
57. Lu T.; Xu Y.; Mericle M.T. and Mellgren R.L. (2002): Participation of the conventional calpains in apoptosis. *Biochimica et Biophysica Acta*, 1590:16-26.
58. Mahassni, S.H. and Khudauardi, E.R. (2017): A pilot study: The effects of an aqueous extract of *Lepidium sativum* seeds on levels of immune cells and body and organs weights in Mice. *J. Ayurvedic. Herb. Med.*, 3:27–32.
59. Manktelow, A. and Mayer, A.A. (1986): Lack of correlation between decreased chemotaxis and susceptibility to infection in burned rats. *J Trauma* 26:143-148.
60. Mansour, S.Z. and Anis, L.M. (2010): Possible effect of 5, 6 – dimethyle – 4 isothiocyanate thieno [ 2,3 – d] pyrimidine and / or irradiation

- on Ehrlich carcinoma in mice. *J. Rad. Res. Appl. Sci.*, 3 (2B): 599 – 618.
61. Manu, K.A. and Kuttan, G. (2008): Ursolic acid induces apoptosis by activating p53 and caspase-3 gene expressions and suppressing NF- $\kappa$ B mediated activation of bcl-2 in B16F-10 melanoma cells. *Int. Immunopharmacol.*, 8: 974-981.
  62. Medhat, A.M.; Azab, K.S.; Said, M.M.; El Fatih, N.M. and El Bakary, N.M. (2017): Antitumor and radiosensitizing synergistic effects of apigenin and cryptotanshinone against solid Ehrlich carcinoma in female mice. *Tumor Biology*, 1–13.
  63. Meyskens, F.L. and Szabo, E. (2005): Diet and cancer: the disconnect between epidemiology and randomized clinical trials. *Cancer Epidemiology, Biomarkers and prevention*, 14:1366-1369.
  64. Moeller, B.J.; Cao, Y.; Li, C.Y. and Dewhirst, M.W. (2004): Radiation activates HIF-1 to regulate vascular radiosensitivity in tumors: role of reoxygenation, free radicals, and stress granules. *Cancer Cell*, 5: 429-441.
  65. Mohamed, E.T. and Safwat, G.M. (2016): Evaluation of cardioprotective activity of *Lepidium sativum* seed powder in albino rats treated with 5-fluorouracil. *Beni-suef university journal of basic and applied sciences*, 5: 208 – 215.
  66. Mohammed, A.A.; Khalil, A.A. El-Beltagi HES (2010) Antioxidant and antimicrobial properties of kaff maryam (*Anastatica hierochuntica*) and doum palm (*Hyphaene thebaica*). *Grasasy Aceites*, 6: 67-75.
  67. Moralli, D.; Yusuf, M.; Mandegar, M.A.; Khoja, S.; Monaco, Z.L. and Volpi, E.V. (2011): An Improved Technique for Chromosomal Analysis of Human ES and iPS Cells. *Stem Cell Rev. and Rep.*, 7:471–477.
  68. Mota, C.S.; Freitas, R.B.; Athayde, M.L.; Boligon, A.A.; Augusti, P.R.; Somacal, S.; Rocha, M.P. and Bauermann, L.F. (2011): Effect of *Vernonia cognata* on oxidative damage induced by ethanol in rats. *Hum. Exp. Toxicol.*, 30: 675-684.
  69. Nafiu, A.B. and Rahman, M.T. (2015): Anti-inflammatory and antioxidant properties of unripe papaya extract in an excision wound model. *Pharm. Biol.*, 53(5): 662–671.
  70. Nair, M.; Mahajan, S.; Reynolds, J.; Aalinkeel, R.; Nair, H.; Schwartz, S. and Kandaswami, C. (2006): The Flavonoid Quercetin Inhibits Proinflammatory Cytokine (Tumor Necrosis Factor Alpha) Gene Expression in Normal Peripheral Blood Mononuclear Cells via Modulation of the NF- $\kappa$ B System." *Clinical and Vaccine Immunology* 13(3): 319-328.
  71. Nakashima, S.; Matsuda, H.; Oda, Y.; Nakamura, S.; Xu, F. and Yoshikawa, M. (2010): Melanogenesis inhibitors from the desert plant *Anastatica hierochuntica* in B16 melanoma cells. *Bioorganic & medicinal chemistry*.18(6):2337-2345.
  72. Nugroho, A.; Heryani, H.; Choi, J.S. and Park, H.J. (2017): Identification and quantification of flavonoids in *Carica papaya* leaf and peroxynitrite scavenging activity. *Asian Pacific Journal Tropical Biomedicine*, 7(3): 208-213.
  73. Nwiloh, B.I.; Nwinuka, N.M. and Monanu, M.O. (2009): The effect of aqueous extract of *Carica papaya* leaves on liver enzymes and blood cell counts of normal albino rats. *Int. J. Biol. Chem. Sci.*, 3(3): 561-566.
  74. Nwiloh, B.I.; Nwinuka, N.M. and Monanu, M.O. (2009): The effect of aqueous extract of *Carica papaya* leaves on liver enzymes and blood cell counts of normal albino rats. *Int. J. Biol. Chem. Sci.*, 3(3): 561-566.
  75. Oboh, G.; Olabiyi, A.A. and Akinyemi, A.J. (2013): Inhibitory effect of aqueous extract of different parts of unripe pawpaw (*Carica papaya*) fruit on Fe<sup>2+</sup>-induced oxidative stress in rat pancreas in vitro. *Pharm. Biol.*, 51: 1165–1174.
  76. Ojo, O.A.; Ojo, A.B.; Awoyinka, O.; Ajiboye, B.O.; Oyinloye, B.E.; Osukoya, O.A.; Olayide, I.I. and Ibitayo, A. (2017): Aqueous extract of *Carica papaya* Linn. roots potentially attenuates arsenic induced biochemical and genotoxic effects in Wistar rats. *Journal of Traditional and Complementary Medicine*, 8(2):324-334.
  77. Otsukia, N.; Dangb, N.H.; Kumagaia, E.; Kondo, A.; Iwataa, S.; Morimoto, C. and Abdul fattah S.Y. (2010): Aqueous extract of *Carica papaya* leaves exhibits anti-tumor activity and immunomodulatory effects *Journal of Ethnopharmacology*, 127:760–767.
  78. Owoyele, B.V.; Adebukola, O.M.; Funmilayo, A.A. and Soladoye, A.O. (2008): Anti-inflammatory activities of ethanolic extract of *Carica papaya* leaves. *Inflammopharmacology*, 16:168–173.
  79. Ozben, T. (2007): Oxidative stress and apoptosis: impact on cancer therapy. *J Pharm Sci.*, 96(9):2181-2196.
  80. Paglia, D.E. and Valentine, W.N. (1967): Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. *J. Lab. Clin. Med.*, 70:158-169.
  81. Pang, W.L., (1995): Induction of numerical chromosomal aberrations by reactive oxygen species.

82. Papież, M. A., (2014): The effect of quercetin on oxidative DNA damage and myelosuppression induced by etoposide in bone marrow cells of rats. *Acta Biochim. Pol.*, 61(1):7-11.
83. Puspitasari, Y. and Peristiowati, Y. (2016): Effect of Papaya Leaf Extract on Cell Proliferation and Apoptosis Activities in Cervical Cancer Mice Model. *J. Appl. Environ. Biol. Sci.*, 6(9):78-83.
84. Radha, Y.; Manjula, A.; Reddy, B.M. and Rao, B.V. (2011): Synthesis and biological activity of novel Benzimidazoles', *Indian Journal of Chemistry*, 50:1762-1773.
85. Raish, M.; Ahmad, A.; Alkharfy, K.M.; Ahamad, S.R.; Mohsin, K.; Al-Jenoobi, F.I.; Al-Mohizea, A.M. and Ansari, M.A. (2016): Hepatoprotective activity of *Lepidium sativum* seeds against D-galactosamine/lipopolysaccharide induced hepatotoxicity in animal model. *BMC Complem. Altern. Med.*, 16(1):501.
86. Raish, M.; Ahmad, A.; Alkharfy, K.M.; Ahamad, S.R.; Mohsin, K.; Al-Jenoobi, F.I.; Al-Mohizea, A.M. and Ansari, M.A. (2016): Hepatoprotective activity of *Lepidium sativum* seeds against D-galactosamine/lipopolysaccharide induced hepatotoxicity in animal model. *BMC Complem. Altern. Med.*, 16(1):501.
87. Ramasawamy, A.M. and Sirsi, M. (1960): Antituberculosis Activity of Some Chemical Constituents from Higher Plants. *Indian J. Pharm.*, 22: 34-35.
88. Raval, N.D.; Ravishankar, B. and Ashok, B.K. (2013): Anti-inflammatory effect of *Chandrashura* (*Lepidium sativum* Linn.) an experimental study. *AYU*, 34:302-304.
89. Reitman, A. and Frankel, S. (1957): A Colorimetric Method for the Determination of Serum Glutamic Oxalacetic and Glutamic Pyruvic Transaminases. *Amer J. Clin. Path.*, 28(1): 56:63.
90. Reuter, S.; Gupta, S.C.; Chaturvedi, M.M. and Aggarwal, B.B. (2010): Oxidative stress, inflammation, and cancer: how are they linked? *Free Radical Biology & Medicine*, 49: 1603-1616.
91. Riyadh: King Saud University Press, 1.
92. Rizk, A.M.; Williamson, E.M. and Evans, F.J. (1985): Constituents of Plants Growing in Qatar VII an examination of Certain Plants for Anti-Inflammatory Activity. *International Journal of Crude Drug Research*. 23(1):1-4.
93. Roshan, A.; Verma, N.K. and Gupta, A. (2014): A Brief Study on *Carica Papaya*- A Review. *International Journal of Current Trends in Pharmaceutical Research* 2(4):541-550.
94. Sakran, M.; Selim, Y. and Zidan, N. (2014): A new isoflavonoid from seeds of *Lepidium sativum* L. and its protective effect on hepatotoxicity induced by paracetamol in male rats. *Molecules*, 19:15440-15451.
95. Salah, S.H.; Abdou, H.S.; Abd El-Azeem, A.S. and Abdel-Rahim, E.A. (2011): The antioxidative effects of some medicinal plants as hypoglycemic agents on chromosomal aberration and abnormal nucleic acids metabolism produced by diabetes stress in male adult albino rats. *Journal of Diabetes Mellitus*, 1(1):6-14.
96. Salem, F.S.; Badr, M.O. and Neamat-Allah, A.N. (2011): Biochemical and pathological studies on the effects of levamisole and chlorambucil on Ehrlich ascites carcinoma-bearing mice. *Vet. Ital.*, 47: 89-95.
97. Sallmyr, A.; Fan, J. and Rassool, F.V. (2008): Genomic instability in myeloid malignancies: increased reactive oxygen species (ROS), DNA double strand breaks (DSBs) and error-prone repair. *Cancer Lett.* 270(1):1-9.
98. Samanta, A.; Maji, H., S.; De, A. and Kar, S.K. (2016): Evaluation of antitumor and anticancer activity of 4-amino benzoic benzoyl benzimidazole in Ehrlich's ascites carcinoma induced male swiss mice. *JIAPS*, 1(4):1-9.
99. Samanta, A.; Maji, H., S.; De, A. and Kar, S.K. (2016): Evaluation of antitumor and anticancer activity of 4-amino benzoic benzoyl benzimidazole in Ehrlich's ascites carcinoma induced male swiss mice. *JIAPS*, 1(4):1-9.
100. Samanta, A.; Maji, H., S.; De, A. and Kar, S.K. (2016): Evaluation of antitumor and anticancer activity of 4-amino benzoic benzoyl benzimidazole in Ehrlich's ascites carcinoma induced male swiss mice. *JIAPS*, 1(4):1-9.
101. Šamec, D.; Pavlović, I.; Redovniković, I.R. and Salopek-Sondi, B. (2018): Comparative analysis of phytochemicals and activity of endogenous enzymes associated with their stability, bioavailability and food quality in five Brassicaceae sprouts. *Food Chemistry*, 269:96-102.
102. Samudrala, P.K.; Augustine, B.B.; Kasala, E.R.; Bodduluru, L.N.; Barua, C. and Lahkar, M. (2015): Evaluation of antitumor activity and antioxidante status of *Alternanthera brasiliana* against Ehrlich ascites carcinoma in Swiss albino mice. *Pharmacognosy Research*, 7(1): 66-73.
103. Sashi, P.J.; Murali, K.A. and Chendil, D. (2016): Oxidative Stress: A Promising Target for Chemoprevention. *Curr. Pharm. Rep.*, 5: 2-5.
104. Sekeroğlu, Z.A. and Sekeroğlu, V. (2012): Effects of *Viscum album* L. extract and quercetin on methotrexate-induced cyto-genotoxicity in

- mouse bone-marrow cells. *Mutat Res.* 746(1):56-59.
105. Schneider, C.D.E. and Oliveira, A.R. (2004): Radicais livres de oxigênio e exercício: mecanismos de formação e adaptação ao treinamento físico. *Revista Brasileira de Medicina do Esporte*, 10: 87-90.
  106. Seigler, D.S.; Pauli, G.F.; Nahrstedt, A. and Leen, R., (2002): Cyanogenic allosides and glucosides from *Passiflora edulis* and *Carica papaya*. *Phytochemistry*, 60: 873–882.
  107. Shah, A.H.; Bhandari, M.P.; Al-Harbi, N.O. and Al-Ashban, R.M. (2014): Kaff-E-Maryam (*Anastatica hierochuntica* L.): evaluation of gastro-protective activity and toxicity in different experimental models. *Biology and Medicine*. 6(1):1-10.
  108. Shan, J.Z.; Xuan, Y.Y.; Zheng, S.; Dong, Q. and Zhang, S.Z. (2009): Ursolic acid inhibits proliferation and induces apoptosis of HT-29 colon cancer cells by inhibiting the EGFR/MAPK pathway. *J. Zhejiang Univ. Sci. B.*, 10: 668-674.
  109. Sharma, V.; Mishra, M.; Ghosh, S.; Tewari, R.; Basu, A.; Seth, P. and Sen, E. (2007): Modulation of interleukin-1beta mediated inflammatory response in human astrocytes by flavonoids: implications in neuroprotection. *Brain Res. Bull.*, 73: 55-63.
  110. Singh, A.; Bhat, T.K. and Sharma, O.M. (2011): Clinical biochemistry of hepatotoxicity, *J. Clin. Toxicol.*, 4:1–19.
  111. Sobhy, E.A.; Tailang, M.; Benyounes, S. and Gauthaman, K. (2011): Antimalarial and hepatoprotective effects of entire plants of *Anastatica hierochuntica*. *Int. J Res Phytochem Pharmacol.*, 1:24-27.
  112. Spines, G.A. (1999): The dual role of nitric acid in islets  $\beta$ -cells. *News in Physiological Sciences*, 14:49-54.
  113. Srivastava, S.; Somasagara, R.R.; Hegde, M.; Nishana, M.; Tadi, S.K.; Srivastava, M.; Choudhary, B. and Raghavan, S.C. (2016): Quercetin, a Natural Flavonoid Interacts with DNA, Arrests Cell Cycle and Causes Tumor Regression by Activating Mitochondrial Pathway of Apoptosis. *Sci Rep.*, 6: 24049.
  114. Sun, H.X.; Xie Y. and Ye, Y.P. (2009): Advances in saponin-based adjuvants. *Vaccine*, 27(12): 1787-1796.
  115. Sun, J.; Chu, Y. F.; Wu, X. and Liu, R.H. (2002): Antioxidant and antiproliferative activities of common fruits. *Journal of Agriculture and Food Chemistry*, 50: 7449-7454.
  116. Sundaram, M.; Patra, S. and Maniarasu, G. (2012): Antitumor activity of ethanol extract of *Gracilaria edulis* (Gmelin) Silva on Ehrlich ascites carcinoma-bearing mice. *Journal of Chinese Integrative Medicine*, 10 (4):430-435.
  117. Sundarmurthy, D.; Jayanthi, C. R. and Lakshmaiah, K. C. (2017): Effect of *Carica papaya* leaf extract on platelet count in chemotherapy-induced thrombocytopenic patients: A preliminary study. *National Journal of Physiology, Pharmacy and Pharmacology*, 7(7):685-692.
  118. Szabo, S.; Trier, J.S.; Brown, A.; Schnoor, J.; Homan, H.D. and Bradford, J.C. (1985): A quantitative method for assessing the extent of experimental gastric erosions and ulcers. *J. Pharmacol. Methods*, 13: 59-66.
  119. Taj, S. and Nagarajan, B. (1996): Inhibition by quercetin and luteolin of chromosomal alterations induced by salted, deep-fried fish and mutton in rats. *Mutat. Res.*, 369: 97-106.
  120. Thoppil, R.J. and A. Bishayee (2011): Terpenoids as potential chemopreventive and therapeutic agents in liver cancer, *World J. Hepatol.*, 3:228–249.
  121. Uchiyama, M. and Mihara, M. (1978): Determination of malonaldehyde precursor in tissues by thiobarbituric acid test. *Analytical biochemistry*, 86 (1): 271-278.
  122. Ulakoglu, G. and Altun, S. (2004): The effects of epirubicin on proliferation and DNA synthesis of Ehrlich ascites carcinoma cells in vitro and in vivo. *Biologia, Bratislava*, 59 (6): 727—734.
  123. Vafa, O.; Wade, M.; Kern, S.; Beeche, M.; Pandita, T.K.; Hampton, G.M. and Wahl, G.M. (2002): c-Myc can induce DNA damage, increase reactive oxygen species, and mitigate p53 function: a mechanism for oncogene-induced genetic instability. *Mol. Cell*, 9: 1031-1044.
  124. Yazdanparast, R.; Ardestani, A. and Jamshidi, S. (2007): Experimental diabetes treated with *Achillea santolina*: effect on pancreatic oxidative parameters. *J. Ethnopharmacol.*, 112:13–18.
  125. Yildirim, I. and Kutlu, T. (2015): Anticancer Agents: Saponin and Tannin. *International Journal of Biological Chemistry*, 9 (6): 332-340.
  126. Yoshikawa, M.; Morikawa, T. and Xu, F. (2003a): (7R, 8S) and (7S, 8R) 8-5'Linked Neolignans from Egyptian Herbal Medicine *Anastatica hierochuntica* and Inhibitory Activities of Lignans on Nitric Oxide Production. *Heterocycles*, 60(8):1787-1792.
  127. Yoshikawa, M.; Xu, F.; Morikawa, T.; Ninomiya, K. and Matsuda, H. (2003b): Anastatins A and B, new skeletal flavonoids with hepatoprotective activities from the desert plant *Anastatica hierochuntica*. *Bioorganic & medicinal chemistry letters*; 13(6):1045-1049.

128. Zhao, T.D.; Etherton, K.R.; Martin, P.J. and Gillies, S.G. (2007): Dietary linolenic acid inhibits proinflammatory cytokine production by peripheral blood mononuclear cells in hypercholesterolemic subjects, *Am. J. Clin. Nutr.*, 85:385–391.
129. Zia-Ul-Haq, M.; Ahmad, S.; Calani, L.; Mazzeo, T.; Del Rio, D.; Pellegrini, N. and DeFeo, V. (2012): Compositional study and antioxidant potential of *Ipomoea hederacea* Jacq. and *Lepidium sativum* L. seeds. *Molecules*, 17:10306–10321.
130. Ziech, D.; Franco, R.; Georgakilas, A.G.; Georgakila, S.; Malamou-Mitsi, V.; Schoneveld, O.; Pappa, A. and Panayiotidis, M.I. (2010): The role of reactive oxygen species and oxidative stress in environmental carcinogenesis and biomarker development, *Chem. Biol. Interact.* 188:334–339.

12/25/2019