

## The effect of different levels of *Matricaria chamomilla* on functional status of liver in rats injected with carbon tetrachloride (Ccl4).

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**Abstract: Background:** The word chamomile actually refers to a range of different daisy-like plants, which are a member of the Asteraceae family. There are many different species of chamomile, the two most commonly being German chamomile (*Matricaria chamomilla*) and Roman chamomile (*Chamaemelum nobile*). They have been used since Ancient times for their calming and anti-inflammatory properties, and each offer their own additional health benefits. The plant's healing properties come from its daisy-like flowers, which contain volatile oils (including bisabolol, bisabolol oxides A and B, and matricin) as well as flavonoids (particularly a compound called apigenin) and other therapeutic substances. **OBJECTIVE:** This investigation aims to study the possible therapy of different levels of *Matricaria chamomilla* on functional status of liver in rats injected with carbon tetrachloride (Ccl4). **DESIGN:** Thirty-six male albino rats were treated subcutaneous injection of carbon tetrachloride (Ccl4) in paraffin oil 50% V/V (2ml / kg b. wt.) twice a week for two weeks to induce chronic damage of the liver. After the injection of Ccl4, blood samples were obtained by retro orbital method to ensure occurrence of liver injury and to estimate liver function. The obtained data were statistically analyzed using computerized SPSS **RESULTS:** Rats given Ccl4 and fed on 5% *Matricaria chamomilla* showed the highest significant increase in the mentioned relative organ weight as compared to all levels of treatment. Rats given Ccl4 then fed on a combination of all levels of treatment showed the highest decrease of AST, ALT and ALP enzyme levels in the serum. The decrease in serum AST, ALT and ALP enzyme levels. rats given Ccl4 prior to feeding on all levels of treatments revealed the highest decrease in serum lipoprotein HDL, LDL, VLDL fraction levels.

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**Keywords:** *Matricaria chamomilla*, Liver diseases, Therapeutic Uses of *Matricaria chamomilla*

### - Introduction

Chamomile (*Matricaria chamomilla* L.) is a well-known medicinal plant species from the Asteraceae family often referred to as the “star among medicinal species.” Nowadays it is a highly favored and much used medicinal plant in folk and traditional medicine. Its multitherapeutic, cosmetic, and nutritional values have been established through years of traditional and scientific use and research. Chamomile has an established domestic (Indian) and international market, which is increasing day by day. The plant available in the market many a times is adulterated and substituted by close relatives of chamomile. (raedon, *et al.*, 2011). Chamomile has calming and soothing properties. It is used for nervousness, headaches, anxiety, and hysteria. It is also beneficial for colds and flu. Its antispasmodic properties. benefit cramps and spasms, probably due to the easily assimilable form of calcium found in it. ( **Reader's Digest Association.,2012**). Chamomile is renowned for its medical and household uses. The apparently endless list of conditions it can help all fall into areas that the relaxing, carminative and anti-inflammatory actions can aid. It is an excellent, gentle sedative, useful and safe for use with children. (Wang, *et al.*, 2014).

The liver has a complex role in the function of the body,” said Jordan Knowlton, an advanced registered nurse practitioner at the University of Florida Health Shands Hospital. “Detoxification, metabolism (including regulation of glycogen storage), hormone regulation, protein synthesis, digestion, and decomposition of red blood cells, to name a few.” It produces bile, a chemical substance that breaks down fats and makes them more easily digestible. liver also produces and synthesizes multiple important elements of plasma, (Williams, *et al.*,2014) Your liver helps your body digest food, store energy, and remove poisons. Liver function tests are blood tests that check to see how well your liver is working. They check for liver damage, and can help diagnose liver diseases. You may have liver function tests as part of a regular checkup. Or you may have them if you have symptoms of liver disease. Doctors also use the tests to monitor some liver diseases, treatments, and possible side effects of medicines. ( Jump up, *et al.*,2016).

### *Aim of study*

This investigation aims to study the possible therapy of different levels of *Matricaria chamomilla* on functional status of liver in rats injected with carbon tetrachloride (Ccl4).

## - Materials And Methods

### 1- Materials

#### 1.1. Plants

The tested plant in this investigation was *Matricaria chamomilla*. These plant were selected to study their effects against liver disorders. These plants were purchased as dried material from the local market, while others were obtained as raw plants from greengroceries.

#### 1.2. Diets

##### 1.2.1. Basal Diet

**Table (1): The composition of basal diet**

Compounds	Amount
Protein	20%
Corn oil	4.7 %
Salt mixture	3.5 %
Vitamin mixture	1 %
Cellulose	5 %
Choline chloride	2 %
Sucrose	10%
Corn starch	Up to 100%

Source: **Reeves et al., (1993)**.

**Table (2): The composition of salt mixture (g/100 g):**

Compounds	Amount
CaCO <sub>3</sub>	600 mg
K <sub>2</sub> HPO <sub>4</sub>	645 mg
Ca HPO <sub>4</sub> 2H <sub>2</sub> O	150 mg
MgSO <sub>4</sub> 2H <sub>2</sub> O	204 mg
Nacl	334 mg
Fe (C <sub>6</sub> H <sub>5</sub> O <sub>7</sub> ) <sub>2</sub> 6H <sub>2</sub> O	55 mg
KI	1.6 mg
MnSO <sub>4</sub> 4H <sub>2</sub> O	10 mg
Zncl <sub>2</sub>	0.5 mg
Cu SO <sub>4</sub> 5H <sub>2</sub> O	0.06 mg

Source: **(Hegsted et al., 1941)**.

**Table (3): The composition of vitamin mixture**

Vitamin	Amount
Vitamin E	10 Iu
Vitamin K	0.50 Iu
Vitamin A	200 Iu
Thiamin	0.50 mg
Pyridoxine	1.00 mg
Niacin	4.00 mg
Calcium panthothenic acid	0.40 mg
Vitamin D	100 Iu
Choline chloride	200 mg
Folic acid	0.02 mg
Inositol	24 mg
Para-amino – benzoic acid	0.02 mg
Vitamin B12	2.00 µg
Biotin	0.02 mg

Source: **(Campbell, 1963)**.

The basal diet was prepared according to **Reeves et al., (1993)**. It was consisted of 20% protein (casein), 10% sucrose, 4.7% corn oil, 2% choline chloride, 1% vitamin mixture, 3.5% salt mixture and 5% fiber (cellulose). The remainder was corn starch as it was recorded in table (1).

##### 1.2.2. Experimental diet

Experimental diet prepared from basal diet plus the powdered plants added at a percentage of 10% and is shown in table (4).

##### 1.3. Carbon tetra chloride (Ccl<sub>4</sub>)

Carbon tetrachloride (Ccl<sub>4</sub>) was obtained from El-Gomhoryia Company for Chemical Industries, Cairo, Egypt as 10% liquid solution. It was dispensed in white plastic bottles each containing one liter as a toxic chemical material for liver poisoning according to **Passmore and Eastwood (1986)**. In the same time, it is mixed with paraffin oil which obtained from the pharmacy for dilution during the induction.

##### 1.4. Rats

Mature male albino rats of Sprague - Dawley strain weighing 150-160 g. B.Wt. at age of 14-16 weeks were obtained from Laboratory of Animal Colony, Helwan, Egypt. The animals were allocated in plastic cages with metallic strainless covers and kept under strict hygienic measures. Rats were fed the basal diet for 7 days before the beginning of the experiment for adaptation. Diets were presented to rats in a special non-scattering feeding cups to avoid loss of food and contamination. Water was provided *ad libitum* via a narrow mouth bottle with a metallic tube tightly fixed at its mouth by a piece of rubber tube. Animals were subjected to a 12 hours light and 12 hours dark schedule and kept for 7 days before the start of the experiment for acclimatization as noted before.

## 2- Methods

### 2.1. Preparation of plant

The plant materials were grinded in a mixer to give a powder and were kept in dusky stoppered glass bottles in a cool and dry location till use, according to **Russo (2001)**, who reported that all herbs and plants are best kept in a cool, dry and dark location to reduce oxidation of their contents.

### 2.2. Grouping and feeding of rats

The experiment was performed in Animal House. All rats were fed for one week on basal diet before starting the experiment, then divided into two main groups, the first group (n= 6 rats) was fed on the basal diet only as a control negative (C –ve ) normal rats for 28 days. The rats of second main group (n= 36 rats) were injected s/c by Ccl<sub>4</sub> to induce liver damage according to **Jayasekhar et al., (1997)**.

**Table (4): The composition of basal and Experimental diet:-**

Component (g)	Basal diet	5% <i>Matricaria chamomilla</i>	10% <i>Matricaria chamomilla</i>	15% <i>Matricaria chamomilla</i>	20% <i>Matricaria chamomilla</i>	25% <i>Matricaria chamomilla</i>
Test ingredients	---	10	10	10	10	10
Casein	20	20	20	20	20	20
Corn oil	4.7	4.7	4.7	4.7	4.7	4.7
Mineral mix	3.5	3.5	3.5	3.5	3.5	3.5
Vitamin mix	1	1	1	1	1	1
Cellulose	5	5	5	5	5	5
Cholin chloride	2	2	2	2	2	2
Sucrose	10	10	10	10	10	10
Corn starch	Up to 100	Up to 100	Up to 100	Up to 100	Up to 100	Up to 100

Rats with liver intoxication were dispersed into six groups (n= 6 rats) as follow:-

**Group (2):** was kept without any treatment as a control positive (C +ve group) and fed on basal diet for 28 days.

**Group (3):** was fed on basal diet plus 5% *Matricaria chamomilla*.

**Group (4):** was fed on basal diet plus 10% *Matricaria chamomilla*.

**Group (5):** was fed on basal diet plus 15% *Matricaria chamomilla*.

**Group (6):** was fed on basal diet plus 20% *Matricaria chamomilla*.

**Group (7):** was fed on basal diet plus 25% *Matricaria chamomilla*.

### 2.3. Induction of liver intoxication in rats

Thirty-six male albino rats were treated subcutaneous injection of carbon tetrachloride (Ccl<sub>4</sub>) in paraffin oil 50% V/V (2ml / kg b. wt.) twice a week for two weeks to induce chronic damage of the liver according to the method described by **Jayasekhar et al., (1997)**.

After the injection of Ccl<sub>4</sub>, blood samples were obtained by retro orbital method to ensure occurrence of liver injury and to estimate liver function.

### 2.4. Blood sampling

At the end of the experiment period (28 days) rats were sacrificed by ether an anesthesia. Blood samples were obtained by retro-orbital method in a clean dry centrifuge tube. They were left to clot by standing at room temperature for 20 minutes, and then centrifuged at 1500 r.p.m for 15 minutes. Serum samples were collected by a dry clean syringe, poured in Wisserman tubes and then kept frozen in a refrigerator at -10°C till biochemical analysis. Rats were thereafter opened, liver, spleen, heart, lungs and kidneys removed and washed in saline solution, then dried and weighted.

Relative weights of mentioned organs were calculated using the following formula

$$\text{Relative organ weight} = \frac{\text{Organ weight}}{\text{body weight}} \times 100$$

For fixation prior to histopathological investigation, organs were kept in formalin solution (10% V/V) according to methods described by **Drury and Wallington (1967)**.

### 2.5. Biological Evaluation

During the period of the experiment, all rats were weighed once a week and the consumed diets were recorded everyday (daily food intake). At the end of the experiment, biological evaluation of the experimental diets was carried out by determination of body weight gain% (BWG%) and food efficiency ratio (FER). According to **Chapman et al., (1959)**, using the following formulas:-

$$\text{BWG \%} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100$$

$$\text{FER} = \frac{\text{Body weight gain (g)}}{\text{Food Intake (g)}}$$

Food intake was also calculated daily.

### 2.6. Biochemical Analysis

#### 2.6.1. Determination of the activity of liver enzymes

##### A- Estimation of the activity of aspartate aminotransferase (AST)

Determination of AST enzyme was carried out by spectrophotometer using specific kits (BioMerieux) according to **Reitman and Frankel (1957)**.

##### B- Determination of the activity of serum alanine aminotransferase (ALT)

##### C- Determination of the activity of serum alkaline

**phosphatase**

Alkaline phosphatase (ALP) determination procedure based on colorimetric determination of ALP was performed according to the method of Roy (1970).

**2.6.2. Determination of serum total bilirubin**

Serum total bilirubin was determined colorimetrically as described by Doumas *et al.*, (1973) using spectrophotometer adjusted at 578 nm.

**Principle:****2.6.3. Determination of total cholesterol in serum:**

Total cholesterol was determined according to Ratliff and Hall (1973).

**2.6.4. Determination of triglycerides**

Enzymatic colorimetric determination of triglycerides was carried out according to Jacobs and Van Denmark (1960).

**2.6.5. Determination of HDL**

Determination of HDL was carried out according to the method of Fnedewaid (1972) and Gordon and Amer (1977).

**2.6.6. Determination of VLDL and LDL**

The determination of VLDL and LDL was carried out according to the method of Lee and

Nieman (1996).

**2.7. Statistical analysis**

The obtained data were statistically analyzed using computerized SPSS (Statistic Program Sigmastat, statistical soft-ware, SAS Institute, Cary, NC). Effects of different treatments were analyzed by one way ANOVA (Analysis of variance) test using Duncan's multiple range test and  $p < 0.05$  was used to indicate significance between different groups, the following formulas were used (Snedecor and Cochran, 1967).

**Results And Discussion**

In the current study the effect of the different levels of (5%,10%, 15%, 20%,25%) *Matricaria chamomilla*, on functions of the liver status was investigated in liver of Ccl4-intoxicated rats.

**1. Effect on food intake (FI), body weight gain% (BWG%) and feed efficiency ratio (FER)**

Data present in table (5) show the effect of the different levels of (5%,10%, 15%, 20%,25%) *Matricaria chamomilla*, on food intake (FI), body weight gain % (BWG%) and feed efficiency ratio (FER) in Ccl4 - intoxicated rats.

**Table (5): Effect of the different levels of (5%,10%, 15%, 20%,25%) *Matricaria chamomilla* on FI (Food intake), BWG%(Body weight gain) and FER (Feed efficiency ratio) of Ccl4-intoxicated rats. (n=6 rats)**

parameters	FI (g)	BWG (%)	FER
<b>Groups</b>			
<b>Control – ve</b>	17.19±0.041 <b>d</b>	35.78±0.973 <b>a</b>	0.13±0.007 <b>a</b>
<b>Control + ve</b>	15.05±0.038 <b>g</b>	10.24±0.624 <b>d</b>	0.06±0.003 <b>e</b>
<b>5%</b> <i>Matricaria chamomilla</i>	17.61±0.025 <b>c</b>	34.04±0.579 <b>a</b>	0.11±0.004 <b>b</b>
<b>10%</b> <i>Matricaria chamomilla</i>	19.09±0.015 <b>a</b>	33.58±0.455 <b>a</b>	0.11±0.006 <b>b</b>
<b>15%</b> <i>Matricaria chamomilla</i>	18.54±0.032 <b>b</b>	24.88±0.646 <b>b</b>	0.09±0.003 <b>c</b>
<b>20%</b> <i>Matricaria chamomilla</i>	16.46±0.024 <b>e</b>	14.37±0.417 <b>c</b>	0.08±0.004 <b>c</b>
<b>25%</b> <i>Matricaria chamomilla</i>	16.19±0.018 <b>f</b>	13.73±0.531 <b>c</b>	0.07±0.004 <b>d</b>

Values denote arithmetic means ± Standard error of the mean.

Means with different letters (a, b, c, d) in the same column differ significantly at  $p \leq 0.05$  using one way ANOVA test, while those with similar letters are non-significant.

It could be observed for rats intoxicated with Ccl4 (C +ve) group that food intake (FI) was 15.05±0.038 g/day compared to 17.19±0.041 g/day in (C -ve) normal rats. These results denote that there were significant decrease in FI of rats intoxicated with Ccl4 (C +ve) group. All rats poisoned by Ccl4 and fed on all tested (5%,10%, 15%, 20%,25%) *Matricaria chamomilla* had a significant increase in FI. Rats given Ccl4 and fed on (10%) *Matricaria chamomilla* showed the highest increase in FI compared to all levels of *Matricaria chamomilla*, which reached to 19.09±0.015

g/day.

Concerning body weight gain (BWG%), it is clear in the same table that in (C -ve) group (BWG%) was 35.78±0.973% but in (C +ve) group was 10.24±0.624%. The obtained results showed that there were a significant increase in BWG% in Ccl4-intoxicated rats and fed on all levels of *Matricaria chamomilla* compared to (C +ve) group. There were non-significant changes between rats fed on 5% and 10% *Matricaria chamomilla* which were 34.04±0.579 and 33.58±0.455% respectively. In the same time,

these mentioned level showed the highest significant increase compared to other levels. Also, there were non-significant changes between rats fed on levels 20% and 25% for BWG% which were  $14.37 \pm 0.417$  and  $13.73 \pm 0.531\%$  respectively.

Regarding feed efficiency ratio (FER), it was found from data of same table that in rats injected with Ccl4 without treatment (C +ve) group, FER was  $0.06 \pm 0.003$  while in normal rats (C -ve) it was  $0.13 \pm 0.007$ . These results denote that there was a significant decrease in FER of rats poisoned by Ccl4 as compared to normal rats. Rats intoxicated by Ccl4 and fed on formulas 5% and 10% showed non-significant increase in FER which were  $0.11 \pm 0.004$  and  $0.11 \pm 0.006$  respectively. Similarly FER of groups fed with 15% fo and 20% of *Matricaria chamomilla*

showed values of  $0.09 \pm 0.006$  and  $0.008 \pm 0.004$  respectively, differences were also non-significant. Meanwhile, rats poisoned by Ccl4 and fed on 25% *Matricaria chamomilla* showed significant increase compared to (C +ve) rats which were  $0.07 \pm 0.004$  and  $0.06 \pm 0.003$  respectively.

Similar results were obtained by **Dickerson and Lee (1988)** reported that many patients with acute or chronic liver disease are ill, and commonly lose weight. Moreover, **Clevely and Richmond (1998)** concluded that *Matricaria chamomilla* medicinally valuable and for treating disorders of the liver.

## 2. Effect on relative organs weight

Data listed in table (6) show the effect of feeding by plant concentration on relative organs weight of Ccl4-intoxicated rats.

**Table (6): Effect of feeding with different levels of *Matricaria chamomilla* on relative organs weight of Ccl4-intoxicated rats. (n=6 rats)**

parameters Groups	Relative organs weight (g/100 g. B.Wt.)				
	Liver	Spleen	Lungs	Heart	Kidneys
Control – ve	$3.79 \pm 0.083$ b	$0.62 \pm 0.059$ a	$0.87 \pm 0.041$ a	$0.91 \pm 0.041$ a	$0.94 \pm 0.124$ a
Control + ve	$3.49 \pm 0.077$ e	$0.49 \pm 0.053$ d	$0.67 \pm 0.009$ d	$0.49 \pm 0.021$ d	$0.79 \pm 0.073$ d
5% <i>Matricaria chamomilla</i>	$4.09 \pm 0.092$ a	$0.62 \pm 0.018$ a	$0.83 \pm 0.014$ a	$0.87 \pm 0.017$ a	$0.93 \pm 0.064$ a
10% <i>Matricaria chamomilla</i>	$3.64 \pm 0.045$ c	$0.58 \pm 0.019$ b	$0.75 \pm 0.064$ b	$0.83 \pm 0.018$ a	$0.84 \pm 0.082$ b
15% <i>Matricaria chamomilla</i>	$3.72 \pm 0.059$ b	$0.56 \pm 0.031$ b	$0.74 \pm 0.022$ b	$0.61 \pm 0.018$ c	$0.83 \pm 0.023$ b
20% <i>Matricaria chamomilla</i>	$3.62 \pm 0.046$ c	$0.52 \pm 0.019$ c	$0.72 \pm 0.008$ b	$0.57 \pm 0.021$ c	$0.73 \pm 0.007$ c
25% <i>Matricaria chamomilla</i>	$3.59 \pm 0.021$ d	$0.51 \pm 0.011$ c	$0.69 \pm 0.012$ c	$0.77 \pm 0.018$ b	$0.71 \pm 0.009$ c

Values denote arithmetic means  $\pm$  Standard error of the mean. Means with different letters (**a, b, c, d**) in the same column differ significantly at  $p \leq 0.05$  using one way ANOVA test, while those with similar letters are non-significant.

It could be noticed that for (C -ve) normal rats, the relative weight of liver, spleen, lungs, heart and kidneys were  $3.79 \pm 0.083$ ,  $0.62 \pm 0.059$ ,  $0.87 \pm 0.041$ ,  $0.91 \pm 0.041$  and  $0.94 \pm 0.124$  g/100g B.Wt., respectively. In (C +ve) rats, the relative weight of the previously mentioned organs were  $3.49 \pm 0.077$ ,  $0.49 \pm 0.053$ ,  $0.67 \pm 0.009$ ,  $0.49 \pm 0.021$  and  $0.79 \pm 0.073$  g/100g B.Wt., respectively. These results denote that there were a significant decrease in relative liver, spleen, lungs, heart and kidneys weight of rats poisoned by Ccl4 as compared to the control -ve normal rats.

For relative weight of liver, there were non significant differences between rats poisoned by Ccl4 then fed on 10%,20% of *Matricaria chamomilla* which were  $3.64 \pm 0.045$  and  $3.62 \pm 0.046$  g/100g B.Wt., respectively. Rats given Ccl4 and fed on 5%

*Matricaria chamomilla* showed the highest significant increase in the mentioned relative organ weight as compared to all levels of treatment, while rats given Ccl4 and fed on level 25% of *Matricaria chamomilla* showed the lowest significant increase as compared to all levels.

Regarding relative spleen weight, there were non significant changes between rats injected with Ccl4 and fed on 10%,15% *Matricaria chamomilla* which were  $0.58 \pm 0.019$  and  $0.56 \pm 0.031$  g/100g B.Wt., respectively. Meanwhile, there were non significant changes between rats injected with Ccl4 then fed on 20 % and 25% *Matricaria chamomilla* which were  $0.52 \pm 0.019$  and  $0.51 \pm 0.011$  g/100g B.Wt., respectively. Rats poisoned by Ccl4 and fed 5% *Matricaria chamomilla* showed the highest significant increase in the mentioned relative organ weight which was  $0.62 \pm 0.018$  g/100g B.Wt., showing non -significant changes as compared to control -ve normal rats.

Concerning relative lungs weight, there were non significant changes between rats injured by Ccl4 then



fed on levels 10%, 15% and 20% *Matricaria chamomilla* which were  $0.75 \pm 0.64$ ,  $0.74 \pm 0.022$  and  $0.73 \pm 0.008$  g/100g B.Wt., respectively. Rats fed on 5% *Matricaria chamomilla* showed the highest significant increase in mentioned relative organ weight which was  $0.83 \pm 0.014$  g/100g B.Wt. Mean while, it didn't reflect any significant change as compared to control -ve normal rats which was  $0.87 \pm 0.41$  g/100g B.Wt.

According to data present in the same table (6), it is clear in relative heart weight showed non-significant changes between rats given Ccl4 then fed on levels 5%, and 10% *Matricaria chamomilla* which were  $0.87 \pm 0.17$  and  $0.083 \pm 0.018$  g/100g B.Wt., respectively. At the same time, there were also non-significant differences between rats given Ccl4 then fed on levels 15% and 20% which were  $0.61 \pm 0.018$  and  $0.57 \pm 0.021$  g/100g B.Wt., respectively. The highest significant value in for mentioned organ noticed in rats given Ccl4 then fed on 5% and 10% *Matricaria chamomilla* as compared to control +ve group.

In the same table (6), we can see that the values of kidneys relative weight showed non-significant differences between rats given Ccl4 then fed on 10% and 15% *Matricaria chamomilla* which were

$0.84 \pm 0.082$  and  $0.83 \pm 0.023$  g/100g B.Wt., respectively. Also, there were non-significant changes between rats injured by Ccl4 then fed on 20% and 25% *Matricaria chamomilla* which were  $0.73 \pm 0.007$  and  $0.71 \pm 0.009$  g/100g B.Wt., respectively. Rats given Ccl4 then fed on 5% *Matricaria chamomilla* showed the highest significant increase among the mentioned relative weights which was  $0.93 \pm 0.064$  g/100g B.Wt., and showed non-significant change as compared to that of (C-ve) normal rats. In this concern, it could be observed that carbon tetrachloride Ccl4 decreased liver weight and induced the atrophy.

### 3. Biochemical analysis

Tables from (7) to (10) show the effect of different levels of *Matricaria chamomilla* on liver enzymes, total protein & total bilirubin, total cholesterol & triglycerides and lipoprotein fractions in Ccl4-intoxicated rats.

#### 3.1. Effect on liver enzymes (AST, ALT and ALP)

Table (7) show the effect of different levels of *Matricaria chamomilla* on serum liver enzymes including aspartate amino trans- aminase (AST), alanine amino transferase (ALT) and alkaline phosphatase (ALP) enzymes in Ccl4-intoxicated rats.

**Table (7): effect of different levels of (5%,10%,15%,20%,25%) *Matricaria chamomilla* on serum levels of aspartate amino transaminase (AST), alanine amino transferase (ALT) and alkaline phosphatase (ALP) enzymes of Ccl4-intoxicated rats. (n = 6 rats)**

Parameters	AST (U/L)*	ALT (U/L)*	ALP (U/L)*
<b>Groups</b>			
<b>Control – ve</b>	65.6±1.8 e	36.5 ± 1.6 e	84.5 ± 1.9 e
<b>Control + ve</b>	130.6 ± 2.1 a	70.5 ± 2.4 a	159.4 ± 2.7 a
<b>5%</b> <i>Matricaria chamomilla</i>	124.6 ± 2.3 b	65.5 ± 2.8 b	154.7 ± 2.5 b
<b>10%</b> <i>Matricaria chamomilla</i>	118.3 ± 2.4 c	46.7 ± 2.2 c	146.3 ± 2.8 c
<b>15%</b> <i>Matricaria chamomilla</i>	114.8 ± 2.1c	40.5 ± 2.6 c	139.5 ± 2.2 c
<b>20%</b> <i>Matricaria chamomilla</i>	113.5 ± 1.6 c	35.5 ± 1.9 c	135.5 ± 1.2 c
<b>25%</b> <i>Matricaria chamomilla</i>	94.7 ± 2.4 d	30.3 ± 2.7 d	114.2 ± 2.9 d

(U/L)\* means unit per liter. Values denote arithmetic means ± Standard error of the mean., Means with different letters (**a, b, c, d**) in the same column differ significantly at  $p \leq 0.05$ . using one way ANOVA test, while those with similar letters are non-significant.

It is clear from table (7) that in rats intoxicated with Ccl4 without treatment, the serum levels of AST, ALT and ALP enzymes were  $130.6 \pm 2.1$ ,  $70.5 \pm 2.4$  and  $159.4 \pm 2.7$  U/L, respectively. In (C -ve) normal rats,

the serum levels of the mentioned previously enzymes were  $65.6 \pm 1.8$ ,  $36.5 \pm 1.6$  and  $84.5 \pm 1.9$  U/L, respectively. These finding denote that there were significant increases of AST, ALT and ALP enzymes in the serum of rats poisoned by Ccl4 as compared to (C -ve) normal rats. Rats given Ccl4 then fed on (5%,10%,15%, and 20% ) *Matricaria chamomilla* had a significant decrease in serum levels of AST, ALT and ALP activities. Rats given Ccl4 then fed on a

combination of all levels of treatment showed the highest decrease of AST, ALT and ALP enzyme levels in the serum. The decrease in serum AST, ALT and ALP enzyme levels reached to  $94.7 \pm 2.4$ ,  $30.3 \pm 2.7$  and  $114.2 \pm 2.9$  U/L, respectively.

These results are confirmed by the findings of **Laçine Aksoy., (2012)** investigated the effects of *Matricaria chamomilla* L. extract (MCE) on lipid peroxidation, antioxidant enzyme systems, and several liver enzymes in carbon tetrachloride (CCl<sub>4</sub>)-treated rats. Rats were divided into five groups. The first group (control group) was fed on standard feed. The rats in the other groups (CCl<sub>4</sub>, MCE50, MCE100, and MCE200) were injected intraperitoneally with 0.8 mL kg<sup>-1</sup> CCl<sub>4</sub>. Moreover, rats in the MCE50, MCE100, and MCE200 groups were gavaged with 50 mg kg<sup>-1</sup>, 100 mg kg<sup>-1</sup>, and 200 mg kg<sup>-1</sup> MCE, respectively. Serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) levels, whole

blood malondialdehyde (MDA) and glutathione (GSH) levels, and erythrocyte superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase (CAT) activity levels were measured after 14 days of exposure. ALT and AST in the CCl<sub>4</sub> group increased significantly in comparison to the control group ( $p < 0.001$ ). MCE decreased MDA levels and increased GSH, SOD, and GPx activities. Decreases were observed in CAT activity levels in the CCl<sub>4</sub>, MCE50, MCE100, and MCE200 groups at different significance levels. In conclusion, the findings suggest that, depending on the dose administered, MCE decreases CCl<sub>4</sub>-induced damage and consequent oxidative stress in rats; it affects the antioxidant system positively.

### 3.2. Effect on total protein and total bilirubin

Table (8) show the effect of different levels of (*Matricaria chamomilla*) on serum levels of total protein and total bilirubin of CCl<sub>4</sub>-intoxicated rats.

**Table (8): effect of different levels of (5%,10%,15%,20%,25%) *Matricaria chamomilla* on serum levels of total protein and total bilirubin of CCl<sub>4</sub>-intoxicated rats. (n = 6 rats)**

Groups	parameters	Total protein (mg/dl)	Total bilirubin (mg/dl)
Control – ve		6.68 ± 1.3 a	0.66 ± 0.011 b
Control + ve		4.65 ± 1.6 b	0.99 ± 0.012 a
5%	<i>Matricaria chamomilla</i>	6.54 ± 1.8 a	0.83 ± 0.013 b
10%	<i>Matricaria chamomilla</i>	6.55 ± 1.2 a	0.80 ± 0.012 b
15%	<i>Matricaria chamomilla</i>	6.53 ± 1.3 a	0.75 ± 0.011 b
20%	<i>Matricaria chamomilla</i>	6.54 ± 1.5 a	0.70 ± 0.014 b
25%	<i>Matricaria chamomilla</i>	6.53 ± 1.1 a	0.69 ± 0.015 b

Values denote arithmetic means ± Standard error of the mean. Means with different letters (**a, b, c, d**) in the same column different significantly at  $p \leq 0.05$ . using one way ANOVA test, while those with similar letters are non-significant

It is clear that the serum levels of total protein and total bilirubin in (C +ve) group were  $4.65 \pm 1.6$  and  $0.99 \pm 0.012$  mg/dl, respectively. While in the (C –ve) normal rats were  $6.68 \pm 1.3$  and  $0.66 \pm 0.011$  mg/dl, respectively. These results revealed that there were significant decrease in total protein but significant increase in total bilirubin in the serum of rats intoxicated by CCl<sub>4</sub> as compared to (C –ve) normal rats. In rats given CCl<sub>4</sub> then fed on all levels of *Matricaria chamomilla*, there were significant increase

in the serum level of total protein, but significant decrease in serum level of total bilirubin. There were non-significant changes between all tested levels. Such results mean that all tested levels produced an improvement in levels of both total protein and total bilirubin in the serum.

### 3.3. Effect on total cholesterol and triglycerides

The effect of different levels of *Matricaria chamomilla* on serum levels of total cholesterol and triglycerides of CCl<sub>4</sub>-intoxicated rats is recorded in table (4).

**Table (9): Effect of different levels of (5%,10%,15%,20%,25%) *Matricaria chamomilla* on serum levels of total cholesterol and triglycerides of Ccl4-intoxicated rats. (n = 6 rats)**

parameters	Total cholesterol (mg/dl)	Triglycerides (mg/dl)
<b>Control – ve</b>	88.98 ± 1.4 <b>d</b>	43.35 ± 1.5 <b>d</b>
<b>Control + ve</b>	105.95 ± 1.6 <b>a</b>	56.60 ± 1.9 <b>a</b>
<b>5%</b> <i>Matricaria chamomilla</i>	101.97 ± 1.8 <b>b</b>	52.60 ± 1.4 <b>b</b>
<b>10%</b> <i>Matricaria chamomilla</i>	98.90 ± 1.2 <b>c</b>	49.50 ± 1.2 <b>c</b>
<b>15%</b> <i>Matricaria chamomilla</i>	102.26 ± 1.3 <b>b</b>	54.30 ± 1.4 <b>b</b>
<b>20%</b> <i>Matricaria chamomilla</i>	95.90 ± 1.5 <b>c</b>	46.50 ± 1.3 <b>c</b>
<b>25%</b> <i>Matricaria chamomilla</i>	90.45 ± 1.1 <b>d</b>	40.50 ± 1.4 <b>d</b>

Values denote arithmetic means ± Standard error of the mean. Means with different letters (**a, b, c, d**) in the same column different significantly at  $p \leq 0.05$ . using one way ANOVA test, while those with similar letters are non-significant.

It is clear from data that in rats injected with Ccl4 without treatment (C +ve) the serum levels of total cholesterol and triglycerides were 105.95±1.6 and 56.60±1.9 mg/dl, compared to 88.98±1.4 and 53.35±1.5 mg/dl in normal rats (C –ve). The obtained results showed that there were significant increase in serum levels of total cholesterol and triglycerides in rats poisoned by Ccl4 as compared to normal rats. In rats given Ccl4 then fed on all levels of treatment, there were significant decrease in serum levels of total cholesterol and triglycerides as compared to (C +ve) group.

There were non-significant differences between 5% and 15% *Matricaria chamomilla* which were 101.97±1.8 and 102.26±1.3 mg/dl for total cholesterol and were 52.60±1.4 and 54.30±1.4 mg/dl respectively for triglycerides. Also, there were non-significant differences between 10% and 20% *Matricaria*

*chamomilla* which showed 98.90±1.2 and 95.90±1.5 mg/dl for total cholesterol and 49.50±1.2 and 46.50±1.3 mg/dl respectively for triglycerides. Meanwhile, 25% *Matricaria chamomilla* showed the highest significant decrease both in total cholesterol and triglycerides as compared to all levels of treatment, and revealed non-significant differences compared with (C –ve) normal rats as regards both parameters. These results obtained in the present work agreed with that of **Tsi, and Tan, (1996)** who administered aqueous celery extract intraperitoneally to genetically hypercholesterolaemic (RICO) and normocholesterolaemic (RAIF) rats via Alzet osmotic pumps over a 13 day period. The serum cholesterol concentration of the celery extract-treated RICO rats was found to be significantly lower ( $P < 0.05$ ) than the control rats. Also.

### 3.4. Effect on lipoprotein fractions (HDLc, LDLc and VLDLc)

Data present in table (10) show the effect of different levels of *Matricaria chamomilla* on the serum levels of lipoprotein fraction (HDLc, LDLc and VLDLc) of Ccl4-intoxicated rats.

**Table (10): Effect of different levels of (5%,10%,15%,20%,25%) *Matricaria chamomilla* on the serum levels of lipoprotein fractions (HDLc, LDLc and VLDLc) of Ccl4-intoxicated rats. (n=6 rats)**

parameters	Lipoprotein fractions (mg/dl)		
	HDLc.	LDLc.	VLDLc.
<b>Control – ve</b>	63.96 ± 1.1 <b>e</b>	16.35 ± 1.2 <b>c</b>	8.67 ± 1.1 <b>d</b>
<b>Control + ve</b>	75.99 ± 1.2 <b>a</b>	18.64 ± 1.4 <b>a</b>	11.32 ± 1.6 <b>a</b>
<b>5%</b> <i>Matricaria chamomilla</i>	74.75 ± 1.3 <b>b</b>	16.70 ± 1.3 <b>c</b>	10.52 ± 1.8 <b>b</b>
<b>10%</b> <i>Matricaria chamomilla</i>	72.10 ± 1.3 <b>c</b>	16.90 ± 1.3 <b>c</b>	9.90 ± 1.1 <b>c</b>
<b>15%</b> <i>Matricaria chamomilla</i>	73.70 ± 1.7 <b>b</b>	17.70 ± 1.3 <b>b</b>	10.86 ± 1.8 <b>b</b>
<b>20%</b> <i>Matricaria chamomilla</i>	70.10 ± 1.2 <b>c</b>	16.50 ± 1.4 <b>c</b>	9.30 ± 1.1 <b>c</b>
<b>25%</b> <i>Matricaria chamomilla</i>	68.85 ± 1.6 <b>d</b>	13.50 ± 1.3 <b>d</b>	8.10 ± 1.2 <b>d</b>



Values denote arithmetic means  $\pm$  Standard error of the mean. Means with different letters (**a, b, c, d**) in the same column different significantly at  $p \leq 0.05$ . using one way ANOVA test, while those with similar letters are non-significant.

It is clear that in rats injected with Ccl4 without treatment (C +ve), the serum levels of HDLc, LDLc and VLDLc were  $75.69 \pm 1.2$ ,  $18.64 \pm 1.4$  and  $11.32 \pm 1.6$  mg/dl, respectively. In normal rats (C -ve) the serum levels of the previously mentioned lipoprotein fractions were  $63.96 \pm 1.1$ ,  $16.35 \pm 1.2$  and  $8.67 \pm 1.1$  mg/dl, respectively. These findings denote that there were significant increase in HDLc, LDLc and VLDLc lipoprotein fractions in the serum of rats poisoned by Ccl4 without treatment as compared to the control -ve normal rats. Rats injected with Ccl4 and fed on 10% and 20% *Matricaria chamomilla* had a significant decrease in high density lipoprotein (HDL-c) while those fed on all levels had a significant decrease in low and very low density lipoprotein (LDLc and VLDLc) as compared to control +ve group. In concern to Lipoprotein fraction HDL, there was non-significant differences between (C +ve) rats given Ccl4 then fed on 5% and 15% *Matricaria chamomilla* as compared to control +ve group which were  $74.75 \pm 1.3$ ,  $73.70 \pm 1.7$  and  $75.69 \pm 1.2$  mg/dl, respectively.

Also, rats injured by Ccl4 and fed on 5%, 10% and 20% *Matricaria chamomilla* showed lipoprotein fraction LDLc of  $16.70 \pm 1.3$ ,  $16.90 \pm 1.3$  and  $16.50 \pm 1.4$  mg/dl, respectively. Regarding lipoprotein fraction VLDc, there were non-significant differences between rats given Ccl4 then fed on 5% and 15% *Matricaria chamomilla* which were  $10.52 \pm 1.8$  and  $10.86 \pm 1.8$  mg/dl, respectively. Also, there were non-significant differences between rats given Ccl4 and fed on 10% and 20% *Matricaria chamomilla* which were  $9.90 \pm 1.1$  and  $9.30 \pm 1.1$  mg/dl, respectively. Finally, rats given Ccl4 prior to feeding on all levels of treatments revealed the highest decrease in serum lipoprotein HDL, LDL, VLDL fraction levels which reached to  $68.85 \pm 1.6$ ,  $13.50 \pm 1.3$  and  $8.10 \pm 1.2$  mg/dl, respectively.

**Williams et al, (2014)**

### Recommendations

1- Nutritional and health educational programs should be organized and directed for the public to protect themselves from liver intoxications and its complications.

2- Plant materials may have certain effective ingredients, which not only correct the impaired liver function.

3- Using the plants especially containing high content of flavonoids and carotenoids reduce serum liver enzymes.

4- Patients with liver disorders are advised to use *Matricaria chamomilla* as medicinal plants for healing

the disease.

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