Antioxidant Effect of A. graveolens (Dill) on Adult Male Rats induced to Paracetamol

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Abstract. An apparent side effect of paracetamol overdose is liver damageIn this study, we examine how paracetamol effects on oxidative enzyme malaldehyde (MDA) in liver and antioxidant enzyme glutation (GSH) and glucose of male rats that drenched *A. graveolens* (Dill) , 40 laboratory male rats were randomly assigned into four groups in the animal house at Karbala University's. (G) G1 control given water and food , G2 injection with paracetamol 500 mg/kg , G3 injection with dill oil 150 mg/kg , G4 injection (paracetamol 500 mg/kg and dill oil 150 mg/kg) , Dill was discovered to be able to protect liver enzymes from paracetamol's toxic effects by decreasing MDA and glucose concentration and increasing GSH enzyme, according to the study's findings. Antioxidant properties of dill volatile oil are being studied.

Keywords. Dill, Paracetamol ,Volatile oils ,MDA, GSH and glucose.

1. Introduction

The use of medicinal plants (herbs) for dietary therapy has a long history and currently occupies a significant portion of pharmacy and medical sciences, which is known as alternative medicine [1].

[2] Aromatic oils or volatile oils For its distinctive aromatic smell for ease and for their convenience ,essential oils, and the neutralization of the products of the meiotic metabolism that are produced by some naturally or either hydrolyzed. The well-known plants, especially the aromatic plants, and their great importance As raw materials in many scientific, industries, medical and economical aids [3]. *A. graveolens* (Dill) is a family of umbelliferae annual herbs that comes from Mediterranean branching stems., tripinnate leaves and linear leaflets [4]. The total height of this herb is 150 cm and the seeds are brown and flowers are yellow [5,6], leaves are used in salads and tea [7]. Dill leaves, flowers, and seeds are among the components of the plant from which volatile oils (also known as aromatic oils) can be extracted. The yield of volatile oil extraction varies depending on the section of the plant [8]. Dill seed powder is also in reduce High blood pressure and added to food, calm the nerves, remove chronic headaches ,push the person to a quiet sleep and also reduce .It reduces blood sugar by balancing insulin hormone production, strengthening the spleen and heart, and treating asthma, The secretion of milk in women and the highlighting of urine, and the ash resulting from burning the seeds increases the speed of wound healing festering, as well as for the treatment of hemorrhoids and the disintegration of kidney stones and in cases of purulent conjunctivitis, as it contains a high percentage of the anti-cancer Caroline, as well as its use against snake bite [9].

Living in an oxidizing stress environment causes cells to produce modest amounts of free radicals, which can damage cellular macromolecules like carbohydrate, lipid, protein, and Deoxyribonucleic acid (DNA)

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[10]. Free radicals can also cause DNA damage.

By preventing oxidative stress damage, antioxidant enzyme defense mechanisms shield cells from cellular ROS and lower the buildup of oxidatively damaged molecules in cells. Antioxidant enzymes play a crucial role in keeping ROS, which are extremely toxic and harmful to cells, from causing cell damage. [11].

Antioxidants are divided into Enzymatic and Non-Enzymatic categories. Enzyme oxidation and other enzymes that induce catalase (CAT) and super oxide dismutase (SOD) are examples of antioxidants, while ROS plays an essential role in the conversion of two non-interacting molecules. In addition to enzymatic antioxidants, non-enzymatic antioxidants such as vitamins E, C, and A are also important. Other antigens and non-enzymatic chmotathione and albumin-based antioxidants Paracetamol, which has a vital role in treating oxidative damage in the body, is used to treat moderate fever and pain. Aspirin and ibuprofen's gastrointestinal adverse effects are absent when taken in large dosages, making it a suitable overdose medication [14]. However, acute overdoses of paracetamol, whether accidental or planned, are rather common, and can be exceedingly dangerous. Adults may suffer severe hepatocellular necrosis if they consume 10–15 grams of paracetamol, and 20–25 grams are lethal [15].

2. Material and Method

2.1. Plant Collection

At 45°C, dried Dill from the flowering stage at Karbalaa city in India, Iraq in 6/2/2022 has been mechanically pulverized into a powder [16].

2.2. Dill Extraction

100 gram of Dill was placed soxhlet apparatus in thimble Using a rotary evaporation equipment, the extract was evaporated at 45 Co for 24 hours with 200 ml of (70%) hexane alcohol in a flask. [17]. Using the law indicated in equation No. 1, the percentage is estimated [18-20]:

$$\text{Yield (wt\%)} = \frac{\text{Weight of Oil produced}}{\text{Weight of Seed used}} \times 100$$

3. Preparation of Chemical Reagents

3.1. Alkaloid

3.1.1. Dragendroff's Reagent

This reagent consists of three steps :

- Solution (A) was prepared by adding 0.6 g of bismuth subnitrate and 2 ml of Hydrochloric acid to 10 ml of D .W
- Solution (B) was prepared by adding 6 g of pottasium iodide (KI) to 10 ml of D.W.
- Solution (A) and solution (B) were mixed, and then 7 ml of HCL was added to the mixture, and then the volume was completed with distilled water to 200 ml and preserve in dark container [21].

3.1.2. Phenols

Three to four drops of ferric chloride solution were used to treat the extracts. Phosphors can be seen in the bluish black colour of the solution [22].

3.1.3. Glycoside

One ml of extract of each part was mixed with 5 ml of Benedict reagent. The appearance of red sediment is indicating the presence of reducing sugar [23].

3.1.4. Flavonoids

The presence of flavonoids is indicated by the formation of yellow sediment after treating five milliliters of each extract with one milliliter of potassium hydroxide alcohol. [24].

3.1.5. Saponins

The long-lasting formation of foam after agitating the plant's aqueous solution in a test tube served as evidence that saponins were present [24].

3.1.6. Experiment Design

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These 40 adult male rats, weighing between 220-280 gm and six to eleven weeks of age at the time of the study, were kept alone themselves in a cage with a temperature of 25 C°., where they were fed every two hours. Only G2 received feed and drinking water for paracetamol (500mg/kg) for five weeks, while G1 received normal saline (0.85%).
G3- injection 150mg/kg B.W volatile oil.
G4- injection 150 mg/kg B.W volatile + paracetamol (500mg/kg).

3.1.7. Oxidant Analysis

Malondialdehyde (MDA) , Glutathione peroxidase (GPX) and Lipid Peroxidation (LPO) analysis by using kit from chin Bioassay Technology Laboratory (BT LAB)

3.2. Statistical Analysis

There was a standard deviation in the data. One-way analysis of variances was used to examine the statistical significance of differences between the control and other groups (ANOVA). When using SPSS for Windows version 15.0, significant P values were defined as those below 0.05. (SPSS, Inc., Chicago, Illinois).

4. Results and Discussion

Dill family comprises a wide range of volatile oil components that are natural products and form and accumulate in plants .The results of table (1) showed that the percentage yield of volatile oils supplied the height percentage yield of the volatile oil extract (3.07%).

4.1. Percentage Yield of Volatile Oils Extracted

Table 1. Percentage yield of volatile oils extracted.

extracted	Percentage yield of oils extracted
dill	4/130 *100 = 3. 07%

4.2. Phytochemical Screen

The results of a phytochemical screen study were used to identify the active components in dill volatile oil extract. Table 2 shows the results, which showed that alkaloids, phenols, glycosides, flavonoids, and saponin had favorable results.

Table 2. *M. spicata* phytochemical screening.

Compounds	Result
Alkaloid	+
Flavonoid	+
Phenol	+
Saponin	+
Glycoside	+

Table 3.effect volatile oil on MDA level µmol/ L.

Group	MDA concentration		
	dose times		
	Zero time	Weeks 3	Weeks 5
G1	0.100±1.573	0.164±1.75	0.198±1.412
G2	0.99±1.009	0.125±1.818[b]	0.243±2.988[b]
G3	0.111±1.562	0.95±1.182[a]	0.88±0.632[a]
G4	0.109±1.696	0.88±1.129[a]	0.76±0.841[a]
LSD		2.05	

Different small letter means significant changing .
b means significant increase
a means significant decrease
p≤ 0.05

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Table 4. effect volatile oil on GSH level µmol/ L.

Group	GSH concentration		
	Treatment times		
	Zero time	Weeks 3	Weeks 5
G1	0.97±3.067	0.222±3.029	0.170±3.654
G2	0.98±3.22	0.87±2.31[a]	0.43 ±2.965[a]
G3	0.102±3.89	0.230±5.34[b]	0.298±7.32[b]
G4	0.110±3.323	0.178 ±4.187[b]	0.202±5.133[b]
LSD		2.4	

Different small letter means significant changing .
b means significant increase
a means significant decrease
p≤ 0.05

Table 6. effect volatile oil on GSH level µmol/ L

Group	Glucose concentration		
	dose times		
	Zero time	Weeks 3	Weeks 5
G1	0.180±4.51	0.233±4.535	0.279±4.327
G2	0.208±4.34	0.265±6.16[b]	0.315±6.21[b]
G3	0.212±4.52	0.153±3.990[a]	0.122±3.37[a]
G4	0.245±4.26	0.179±4.38[a]	0.144±3.908[a]
LSD		1.88	

Different small letter means significant changing .
b means significant increase
a means significant decrease
p≤ 0.05

Compared to G1, G3, and G4, G2 showed a considerable rise in MDA and glucose concentrations (Table 3,5). In comparison to groups G1, G2, and G4, therapy (G3) resulted in a significant drop in the rats' blood serum MDA levels . Table (4) shows that GSH concentration in G2 decreased significantly compared to G1, G3, and G4. Treatment (G3) resulted in significantly higher GSH and glucose levels in the blood serum than G1, G2, and G4.

Antioxidant protective effect of volatile oil on liver enzyme against paracetamol-induced toxicity was the focus of the current study. Toxic and a rise in ROS are the results of treating paracetamol with drinking water. Because they are located in the cytoplasm and are released into circulation after cellular damages indicate the development of inflammatory cells and hepatotoxicity, as well as degeneration in hepatocyte enlargement in necrosis and sinusoids and odema, it has been suggested that an overdose of Paracetamol, one of the most common pain and fever medications, may cause liver damage [27,28].

Both neutrophils and Kupffer cells are activated in the presence of acetaminophen toxicity and other pathophysiological events. Kupffer cells release cytotoxic mediators, such as ROS, as well as proinflammatory mediators, such as chemokines and cytokines, in response to these stimuli. Antioxidant glutathione (GSH) is essential in cells. Acetaminophen's toxicity was dramatically increased after GSH depletion [29]. Overdoses of paracetamol have been associated with liver damage, liver GSH enzyme depletion (caused by the conjugation of GSH enzyme with NAPQI to produce mercapturic acid), and increased lipid peroxidation (LPO). [30].

According to this study, there was a significant increase in the blood serum GSH concentration after treatment with dill, Additionally, the active ingredients serve as a strong source of antioxidants that may be utilized to lower biological oxidative stress free radicals and shield cells from harmful substances [31]. Dill oil may protect liver and brain cells and help reduce paracetamol's damaging effects on liver cells because of the abundance of secondary metabolism compounds such as alkaloids, flavonoids, glycosides, and phenols that have been discovered in the herb [32].

The presence of flavonoids compounds, which have been found to have a variety of biological qualities related to antioxidant mechanisms, gave the plant its activity by inhibiting the leakage of intracellular enzymes brought on by cellular renewal or stable cell membranes [33].

because volatile oils is a good source for phytochemical active compounds and antioxidant compounds, may be due present high activity of volatile oils , act of scavenging triplet oxygen , ROS, free radicals, and peroxides may be rich volatile oils that act as antioxidant capacity , protect hepatocytes from ROS damage by using paracetamol overdose .

Conclusion

– Dill oil contain high antioxidant

– Paracetamol overdose leads to liver toxic

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