The Histological Effects of Volatile Oil and Seeds of Bitter Fennel (*Foeniculum Vulgar*-var. vulgare) in Hyperlipidemic Rats

Nada Nasser Faris

Alkunooze University College / Department of Medical Laboratory Techniques/Basrah /Iraq Email: nada.f[at]kunoozu.edu.iq

Abstract: The experiment conducted during 2019/2020 in the laboratory of the college of Agriculture / Basrah University. To study the histological effects of volatile oil and seeds of bitter fennel (Foeniculum vulgare var. vulgare) in laboratory rats. Rats were divided into four major groups, control, hyperlipidemic and hyperlipidemic treated with volatile oil of bitter fennel at doses 40 mg. kg-1 b. w. and hyperlipidemic treated with grind seed at dose 1000 mg. kg-1 b. w. duration three weeks. Hypolipidemia was developed by diet contained 1% cholesterol for 21 days. The results showed no changes in control (normal rats) and the tissue remain within normal limits. Whereas, the rat treated with cholesterol only, illustrate clear morbid changes. While liver and kidney in rats treated with cholesterol then with essential oil and grind seed of bitter fennel duration three weeks, showed amelioration in histological picture compared with group treated with cholesterol only (hyperlipidemic group).

Keywords: Cholesterol; Bitter fennel; Hyperlipidemia

1. Introduction

Hypocholesterolaemia is one of the major factors for cardiovascular diseases and major causes for mortality and disability of many people today [1]. There are several ways for hypocholesterolaemia treatment by drug therapy and medical plants. There is an increasing interest towards aromatic and medicinal plants and their active in the industry and scientific research as natural antioxidants and antimicrobial properties these because is the major sources of many natural photochemicals including flavonoids, terpenoids, carotenoids, coumarins, curcumines *etc.* [2, 3, 4].

Fennel Foeniculum vulgare Mill. belongs to the family Apiaceae, is the Mediterranean region and southern Europe. is a small, erect and aromatic herb. A number of chemical constituents and various therapeutic effects of this herb have been reported by different workers. Extensive investigations have been carried out on different parts of herb and as a consequence, varied classes of compounds fatty acids, hydrocarbons and sterols, Furocoumarins; (imperatorin, psoralen, bergapten, xanthotoxin and isopimpinellin), Flavonoids; (isorhamnetin 3-O- α -rhamnoside, quercetin and kaempferol) and quercetin; (3-O-rutinoside, kaempferol 3-O-rutinoside and quercetin 3-O-\beta-glucoside) have been isolated. [5]Volatile oils are composed of several monoterpenes and phenylpropanoids, where trans-anethole, estragole (methylchavicol), fenchone and limonene. Both extracts and volatile oils obtained from the fruits and the aerial parts of the plant, are used in traditional medicine as diuretic, analgesic, antipyretic, anti-diabetic, antioxidant and anti-inflammation activity [6]. The aim of this experiment to study effect of the volatile oil of bitter fennel on the lipid of the induced hyperlipidemic animals.

Bitter Fennel Oil (%)					
Component	%				
Trans-anethol	0.75-0.55				
Fenchone	0.12-0.25				
Estragole	Max 0.6				
a-Pinene	0.10 - 0.1				
Limonene	0.9 –0.5				
Alpha-pinene/ Limonene	< 1.0				
Cis-anethole	Max 0.5				
Anisaldehyde	Max 2.0				
Beta-myrcene	1.4				

Table 1: Compounds identified in essential oils obtained by steam distillation from ripe fruits of bitter fennels

2. Materials and Methods

2.1 Preparation of Seeds and Volatile Oils

Collected seeds from experiment field. Cleaned and dried in the shade in the laboratory under room temperature for three weeks, after completing seeds preserved in dark glass containers and sealed and then placed in a refrigerator temperature of 4°C degree until use. Volatile oil extracted from the dried seed powder using Clevenger [7].20g of plant sample (seeds) added with 500 ml of distilled water, in Clevenger during 2 hours. Then anhydrous sodium sulphate was added to remove the remaining moisture with the volatile oil and then stored at 4°C until use.

2.2 Animals and Treatment Experiments

Were performed on male albino rats aged 4-8 weeks and weighing $55\pm5g$. They were housed in a controlled room with a 12 h light-dark cycle, at room temperature $20 \pm 3 \text{ C}^{\circ}$, and kept on standard diet for two weeks until the experiment start.

2.3 Prescribing Doses of Seeds and Volatile Oils

The dose of volatile oil was chosen according to its LD50 (the medium 50 lethal doses after acute toxicity).

2.4 Experimental Protocol

60 rats were divided into four major groups. The first group served as control (CG), this group received standard diet and oral administration of distilled water, the second group HG, was received standard diet contained 1% cholesterol, the third group (HG + VO) the animals received standard diet

contained 1% cholesterol followed by oral administration of volatile oil at doses 40 mg. kg-1 b. w. The fourth group (HG + SE) the animals received standard diet contained 1% cholesterol followed by oral administration of extract of seeds at doses1000 mg. kg-1 bw.

2.5 Histological Study

Fragments of liver and kidney were fixed in Bouin's fluid for 18-24h. Transverse sections (5- μ m-thick) were collected throughout the length of segment, and adjacent slides were stained by hématoxylin and eosin [8].

 Table 2: Plasmatic parameters from CG (control group); HG (hyperlipidemic group); HG + VO and HG+SE (Foeniculum vulgare treated groups) are reported.

Plasmatic parameters	CG	HG	HG+VO (mg.kg ⁻¹ b.w.)			HG + SE
			20	30	40	(1000mg.kg ⁻¹ b.w.)
Glucose(mg/dl)	83.2±5.4	89.8±5.4	55.2±5.4	34.6±5.4	43.5±5.4	44.1±5.5
Total Cholesterol(mg/dl)	140.8±15.0	158.3±15.0	118.7±15.0	60.5±15.0	66.7±15.0	119.0±16.6
Triglyceride (mg/dl)	84.3±10.8	170.8±10.8	91.3±10.8	50.0±10.8	60.7±10.8	122.0±10.6
HDL-Cholesterol (mg/dl)	59.1±10.0	30.3±10.0	27.5±10.0	24.9±10.0	33.8±10.0	47.0±6.6
LDL-Cholesterol (mg/dl)	64.5±12.5	93.8±12.5	72.9±12.5	25.6±12.5	20.7±12.5	47.4±12.5
VLDL-Cholesterol (mg/dl)	16.8±3.5	34.2±3.5	18.2±3.5	10.0±3.5	12.1±3.5	24.4±2.2
Urea(mg/dl)	10.5±1.4	15.0±1.4	8.7±1.4	3.2±1.4	4.5±1.4	13.4±1.2

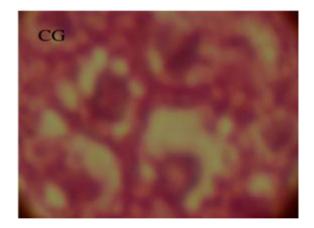
3. Results Discussion

3.1. Histological Effects of Volatile Oil and Seeds Concentration of bitter Fennel (mg. Kg-1 b. w.) in the Cross-Section of Liver

Figure 1, Our result shows a beneficial effect of volatile oil and seeds of F. vulgare in elimination of deposit of the lipids on liver tissue. The reason for the effectiveness of volatile oil in reducing the development of pest disease caused by excess fat to its vital role is important as an anti-oxidant and its role in reducing cholesterol and this is shown by the experiment by the results of testing serum, where it was observed a significant decrease in the level of both total cholesterol and Triglyceride triple protein cholesterol and fat-density cholesterol and the low-lying fat protein density in a very low-lying rats oil pilot and the effect increases with increasing concentration of oil used, compared with a positive comparison (table 2) and the cause to support the activity and effectiveness of the liver to get rid of excess fat by burning and conversion to bile salts and to the important role and the president of volatile oil and its contents of, a-Pinene and Caryophyllene- β in reducing inflammation and inhibiting the work of enzyme 5 Cycloxygenaselipoxygenase and working to convert Arachidonic Acid to Prostaglandin Leukotrienes especially LT-B4 and PG-e which represent the most powerful media inflammatory [9]. The reason for the effectiveness of seeds extract because of its contents Camphor and Carvone and Anethole and Resin, which have highly effective High antioxidant activity and anti-inflammatory which works to prevent and reduce the inflammatory response and changes cholesterol in the liver tissue and this effect is by reducing the concentration of cholesterol fat protein-density low-lying with reduced on centration of cholesterol in blood serum and this is shown by the experiment results, as observed a significant decrease in the level of each of total cholesterol and Triglyceride and LDL and the VLDL (table 2). Or due to the fiber content of

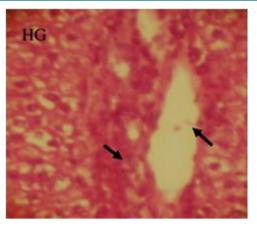
the seeds, which of up to 18.50% of dry weight, which has an effective role in reducing cholesterol in the liver [10].

Figure 1: Histological analysis of liver of rat. cross section of liver colored by hematoxylin and eosin. Control Group (CG), hyperlipidemic Group (HG), hyperlipidemic + Fennel essential oil treated groups (HG+Vo) and hyperlipidemic+ Fennel seeds extract treated groups (HG+SE) [Aggrandizement \times 40].

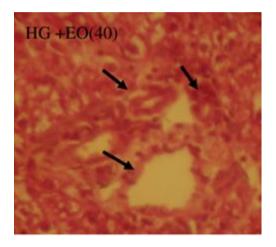


Control group (CG) showed the normal histology of hepatic lobule consisting of central vein with concentrically arranged plate of hepatocytes.

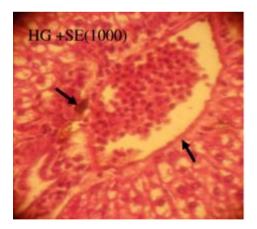
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Hyperlipidemic Group (**HG**) showing vacuolar degeneration of hepatocytes local area of hepatic necrosis associated with mononuclear cells infiltration.



Hyperlipidemic+ volatile oil treated groups (HG+ VO40mg. kg-1 b. w.) showing some degree vacuolar degeneration of hepatocytes



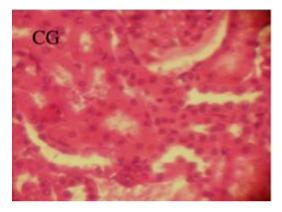
Hyperlipidemic + Seed Extract treated groups (HG+SE1000 mg. kg-1 b. w.) showing vacuolar degeneration of hepatocyte.

3.2. Histological Effects of Volatile oil and Seeds Concentration of bitter Fennel (mg. Kg-1b. w.) in the Cross-Section of Kidney

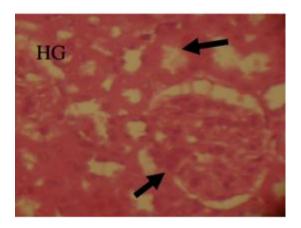
Study shows the survival of total tissue within normal limits in (CG). And (HG) expansion occurred in the urinary

tubules and the accumulation of some inorganic materials. And characterized (HG+VO40 mg. Kg-1 b. w.) to remain kidney tissue within normal limits and did not observed the presence of any pathological changes (figure2). And survival of kidney tissue within normal limits in rat treated with cholesterol 1% and grain seeds of bitter fennel 1000 mg. Kg-1 b. w. compared with HG group. Attributed the cause to the plant known since ancient times its effectiveness vital diuretic and anti-inflammatory, which led to improved renal function and reduce the level of urea in the blood (table2) and get rid of salts and inorganic materials accumulated in kidney [11].

Figure 2: Histological analysis of kidney of rat. section of kidney colored by hematoxylin and eosin. Control Group (CG), hyperlipidemic Group (HG), hyperlipidemic + Fennel essential oil treated groups (HG+VO) and hyperlipidemic + Fennel seeds extract treated groups (HG+SE). [Aggrandizement \times 40].

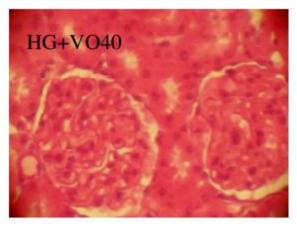


Control Group (normal rat)

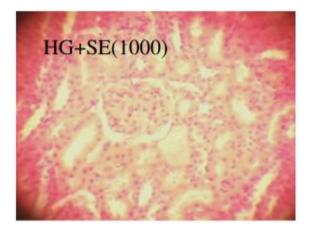


Hyperlipidemic Group (HG), showing expansion occurred in the urinary tubules and the accumulation of some inorganic materials

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Hyperlipidemic + volatile oil treated groups (HG+ VO40mg. kg-1 b. w.) showing remain tissue kidney within normal limits and did not observed the presence of any pathological changes.



Hyperlipidemic + Seed Extract treated groups (HG+SE1000 mg. kg-1 b. w.) showing survival of kidney tissue within normal limits.

4. Conclusion

Based on our findings, we conclude that ingestion of seed and essential oil of Bitter fennel (*Foeniculum vulgare* var. Vulgare) to hyperlipidemic induced rats corrected the hyperlipidemia and pathological abnormalities which could be in part through its antioxidative effect and restoring of redox homeostasis. This makes the possibility of its inclusion in antioxidant drug industry.

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