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RESEARCH ARTICLE

EFFECT OF CELERY FLAVONOID ON LIVER ENZYME GOT AND GPT IN MICE

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ABSTRACT

Celery is good source of flavonoid. They have also other positive effects on human health. The aim of current research was to study and compare an antioxidant activity. The hexanolic extracted and purification flavonoides from celery plant by using Thin layer chromatography (TLC). The study showed the effects of different three concentrations (10,20,30) of celery flavonoid extract on the effectiveness of liver enzymes (GPT,GOT) in blood and liver mice through injection Subcutaneous of laboratory mice during period of tow weeks in same age mice. The results of experiments demonstrate that the indicated there were a significant differences ($P < 0.05$) to concentration of the hexanolic extract on the effectiveness of tow types specific liver enzymes (GPT,GOT). The study showed of their inhibitory effects on certain enzymes and antioxidative activity. In mice treated with carbon tetrachloride CCl_4 , Through experiments show that the percentage of 30% is the best ratio of antioxidant.

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INTRODUCTION

Celery (*Apium graveolens* L.) was introduced from Caucasia into China in about fifteenth century. Ancient medicinal herbs records and modern research have proved that Celery has a series of medicinal properties, such as reducing blood press, sedation, promoting digestion, diuresis and moistening lung. Celery, is hepaxanthic herb grown as abiennial and under certain conditions, as an annual. Celery is a native of Eurasia and is grown mainly in coastal regions. Celery is widely cultivated in the temperate Zones as an important garden crop and the bleached leaf stalks are relished as a popular vegetable. *A. graveolens* is one of ingredients in 8 of the 33 indian polyherbal formations with reputed life-protecting activity. Celery is also used as an effective remedy for various ailments such as bronchitis, liver and spleen disease, arthritic pain and this natural holistic approach to health is becoming mor and mor popular now adays (kolarovic *et al.*, 2010). (Jung 2011). Flavonoids (or bioflavonoids), collectively known as Vitamin p and citrin, are a class of plant secondary metabolites which are ubiquitous in photosynthesizing cell and are commonly found in fruits, vegetables, nuts, seeds, stems, flowers, tea, Wine, propolis and honey. For centuries, preparations containing these compounds as the principal physiologically active constituents have been used to treat human disease. (Mink *et al.*, 2007). The function of flavonoids in flowers is to provide colors attractive to plant pollinators. In leaves, these compounds are increasingly believed to promote physiological

survival of the plant, protecting it from, for example, fungal pathogens and UV-radiation, flavonoids are in photosensitization, energy transfer, the actions of plant growth hormones and growth regulators, control of respiration and photosynthesis, morphogenesis and determination. (Das *et al.*, 2009). The basic structural feature of flavonoid compounds is the 2-phenyl-benzopyrane or flavane nucleus, which consists of tow benzene rings (A and B) linked through a heterocyclic ring (C) Increasingly, this class of natural products is becoming the subjects of anti infective, and many groups have isolated and identified the structures of possessing antifungal, antiviral and antibacterial activity. (Shohaib *et al.*, 2011).

The therapeutic effect of many traditional drugs are attributed to this group of compounds because of their inhibitory effects on certain enzymes and antioxidative activity. They have been shown to posses antibacterial, antifungal, antiviral and anti-inflammatory activities. Their antiallergic, antioxidative and antimutagen activities have been proven. Reduced risk of breast, prostate and colon cancers is related to isoflavonoid activity. Flavonoids have been studied in the prevention of menopausal symptoms and osteoporosis. It was shown that their biological activity depended on the location of the free hydroxyl groups on ring A more so than that on ring B. (Dragan *et al.*, 2007). Celery flavonoids will play very important role in theoretical and practical significance for Celery. The distribution of flavonoids from different Celery resources in Hunan was studied, and characteristics fingerprints of Apigenin, Apiin from Celery with chromatographic technique. Biological activity and pharmacological functions of Celery flavonoids compounds on resistance liver cancer, leukemia, lowering lipid, anti-inflammatory were studied.

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(Amaowicza *et al.*, 2004., Scott, 2012). The aim of search to studied flavonoid effecting on GPT, GOT blood and liver enzymes.

MATERIALS AND METHODS

The *A. graveolens* leaves were procured from local market in 2010 Baghdad Iraq. The leaves were washed thoroughly in tap water to remove adhering mud particles, rinsed in distilled water, drained, dried in a hot air oven at 50 temperature. The dried leaves were finely powdered. The dried powder (50) gm was extracted with 500 ml hexanol for 10 h in soxhlate. The obtained extracts were filtered through filter paper. The crude extracted concentrated in rotary in temperature 40 C⁰, So green material that give, and suspended in 250 ml Ethanol for 8 h. Solvents were removed in Rotary and extracts were obtained, respectively. The residues were dissolved in 25 ml NaOH 5% in separation funnel, then added 25 ml chloroform for 15 min and show two layers the upper layer (water layer), lower layer (chloroform layer). The upper layer had taken and measured in PH 7 by used HCL, Then added 25 ml Chloroform. Total flavonoid content of celery leaves were determined by using way (Bajis *et al.*, 1999), By mixed 10 ml from extracted with 50 ml Ethyl alcohol 95% in 1:1 vol:vol. TLC sorbents and three mobile phase were used for the analysis of the flavonoid exudates. Toluene : Diethyl ether : acetic acid 10% (50:50:50) V \ V \ V. was used for the development of the exudates on silica gel plates (20×20) cm (0.2) mm layer. Detection with U.V light at (280) nm. Flavonoid reaction products were identified by RF values and chromatography with authentic substances on TLC in different solvent systems.

Lab animal

20 male mice were tested, with two months age and 21 gm weight, for 5 groups every one contain 4 mice in Cages, So these mice that were nutrition on concentrated food and water, then several concentration from flavonoid extracted (10,20,30) mg/Kg (0.1) ml had injected daily subcutaneous s/c for 14 days, these which laboratory animals were autopsied To take the blood and liver to hold the rest of the experiments.

Prepare of blood solution

Subjected mice under anesthesia, then later took heart blood and put in test tubes that contain EDTA. These solution was mixed with phosphate buffer, centrifugation 2000 cycle/min for 10 min. Supernatant was taken and 1 ml distilled water added for it. GPT, GOT in blood extract were analysed with least significant difference in $p < 0.05$.

Liver preparation

Liver mice had taken, then cutting for small pieces and mixed with 3 ml phosphate buffer, then centrifugation, supernatant liver result have determination been the GOT, GPT activity in spectrophotometer.

RESULTS AND DISCUSSION

The accumulation process consider first important steps, Drying plant the second step to lowering moistening of plant, to prevent enzymatic and microbial activity may be happen in

plant tissue (Jung *et al.*, 2011). The isolation and purification of flavonoids from the celery crude extracted was successfully established by using Hexanol to remove volatiles oils and fatty acids were found in celery plant without effecting on flavonoid, so another step using ethyl alcohol as phase solvent, following separation process NaOH sodium hydroxide 5% due to it solve in water layer, and added equal volume from chloroform to remove large amount from chlorophyll material (Kavratskhella 2004), (Das *et al.*, 2009). When show yellowish precipitate after added 10 ml extracted crude with 50 ml ethyl alcohol 95%. These lead to flavonoids found, these results successful with (Al Zubaidy, 2008).

The isolation and purification of flavonoids from the celery crude extract was successfully established by using toluene, diethyl ether, acetic acid 10% (50:50:50) as the two-phase solvent. Celery flavonoid glycosides monomer was tracked in 280 nm wavelength by TLC thin layer chromatography, Celery flavonoids can very good separation. Flavonoid reaction products were identified by RF values (0.68) and cochromatography with authentic substances on TLC in different solvent systems (Figure 1). Researchers are under way to study the effect of flavonoids in GOT and GPT enzymes in blood and liver in mouse. Affecting GPT, GOT enzyme level GPT enzyme found large ratio in liver more than another enzymes so that indicator for liver cell damage, Celery flavonoid play major role in decreased these enzymes ratio when high resonance. (Robert *et al.*, 2001) (Jennifer *et al.*, 2009).

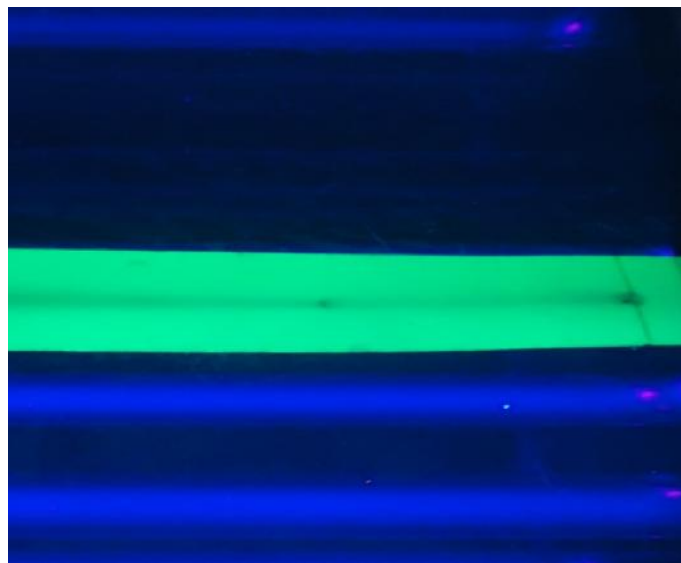


Figure 1. Purification flavonoid on TLC

interaction with other enzyme system. Compared with research on the antioxidant capacities of flavonoid, there has been relatively little research on other beneficial effects of flavonoids. The major effects of flavonoid may be the result of radical scavenging. Another possible mechanism by which flavonoids act is through interaction with various enzyme systems. Another interesting effect of flavonoid on enzymes system is the inhibition of arachidonic acid (Hanneken *et al.*, 2006). Show in Table 1 enzymes GPT, GOT for two weeks report decreasing ($P < 0.05$) after added the material,

comparative with positive controlling during injection Subcutaneous period of different extract concentration. In the experiments periods, lead to that these materials have been effective in liver enzymes these results Conformable with (Jung *et al.*, 2011), compounds because of their inhibitory effects on certain enzymes and antioxidative activity. (Dragant 2007) (Alkubasy *et al.*, 2002).

Table 1. Affected different extracted concentration in GPT, GOT enzymes ratio in blood lab.animal for two weeks (P < 0.05)

Effecting concentration	Quality activation (unit/L) GPT\ Blood	Quality activation (unit/L) GOT\Blood
Positive controlling		
Distal water	79.7±0.24	54.25±5.50
Negative controlling (Ccl4)	128.76±4.57	167.67±1.73
10% flavonoid	104.05±0.12	146.74±2.04
20% flavonoid	97.68±4.25	113.023±0.14
flavonoid 30%	90.35±0.76	79.11±0.40

Table 2. Affected different extracted concentration in GPT, GPT enzymes ratio in liver lab animal for two weeks (P < 0.05)

Effecting concentration	Quality activation (unit/L) GPT\ Blood	Quality activation (unit/L) GOT\Blood
Positive controlling		
Distal water	154.1±0.15	72.13±0.05
Negative controlling (Ccl4)	213.3±5.68	120.1±1.67
10% flavonoid	196.2±3.04	104±1.87
20% flavonoid	184.33±2.30	94.43±0.981
30% flavonoid	178.44±1.31	89.53±0.36

Also when GPT, GOT enzyme measure in lab. Animal liver flavonoid was inhibitory effects on certain enzymes after give lab. Animal Ccl4, So table (2) show 30% the best ratio near from normal, these equal with (Robert *et al.*, 2001), (Pansiri, 2004).

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