

Study of Bioactivity, Anticoagulant and Antioxidant of *Ruta Chalepensis* L. Growing in Syria

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Abstract

In this study (the research laboratories of Chemistry Departments- Aleppo University, Syria, 2018), the active components of the aerial parts of *Ruta chalepensis* L plant was extracted by using different solvents of methanol 70%, ethanol 80, ethyl acetate and hexane. The methanol 70% and ethanol 80% extracts of *Ruta chalepensis* L were produced excellent antimicrobial activities (1.5×10^8 CFU/mL) against three strains of gram-positive bacteria; *Bacillus subtilis*, *Staphylococcus aureus*, and *Streptococcus mutans* and three strains of gram-negative bacteria; *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Escherichia coli*. And three species of fungi (2×10^5) spores/mL; *Aspergillus fumigatus*, *Aspergillus niger* and *Candida albicans*. All extracts showed DPPH radical scavenging activity in a concentration-dependent manner. The extracts were used for anticoagulant assays PT and APTT assays with different concentration of the extracts. The methanol 70% and ethanol 80% extracts of *Ruta chalepensis* L prolonged the time taken for blood clotting in all the tested methods. The activity was increasing as the concentration of extracts increased.

Keywords: *Ruta chalepensis* L, antimicrobial, antifungal, antioxidant, anticoagulant, DPPH.

Introduction:

Numerous infections and disorders caused by bacterial and fungal pathogens including Salmonella, Staphylococcus, Bacillus, Klebsiella, Proteus, Pseudomonas Aspergillus, Candida, Cryptococcus and Trichophyton (Bibi *et al.*, 2011). For several decades, natural remedies and medicinal plants were the main, and in fact the only, resource for the physicians (Rothrock *et al.*, 2019). Until the present, most of the people, especially in developing countries, depend on plants for medicines (Amabye *et al.*, 2015).

The significance of plants to homeopathy and modern medicine is correlated to their chemical constituents such as terpenoids, phenols, alkaloids, flavonoids, amino acids, saponins, glycosides, diterpenes, triterpenes and their compatibility with the human body. plants is expected that more than 30% of the worldwide (Rothrock *et al.*, 2019)

The family of *Rutaceae* contains extremely wide of species aromatic plants, mainly in tropical regions. Among them, the rich one is the genus *Ruta* (Fredj *et al.*, 2007). It is now cultivated in many parts of the world (Bently *et al.* 2004). This plant is considered indigenous in South

Europe and North Africa and it grows on waste stony ground (Anonymous. 2004). The Rutaceae is one of the largest plant families with approximately 150 genera and 1,500 species distributed largely in tropical and subtropical parts of the world (Jones. 1995).. This family is known throughout the world for its citrus fruits such as oranges, lemons and grape fruit and also called as citrus family. A variety of plants of the family *Rutaceae* are used in traditional system of medicine world-wide (Dymock *et al.*, 2005).

Ruta chalepensis (Rue) is an aromatic evergreen shrub which is originally from the Mediterranean region and is at present distributed worldwide (Akkaria *et al.*, 2015). In many countries, it is cultivated for its pharmacological and biological activity and it is widely used for treatment of gastric, diuretic, inflammation, headache and rheumatism disorders (Kacem *et al.*, 2015). Analysis of the chemical constituents of *R. chalepensis* L extracts revealed that the aerial parts contain alkaloids, phenols, flavonoids, amino acids, saponins and furocoumarins (Araya *et al.*, 2018).

The current study was conducted for antimicrobial, anticoagulant, antioxidant, and anticoagulant activities of different solvent extracts *Ruta chalepensis* L. aerial parts.

Materials and Methods:

Chemicals and Equipments:

Chemicals: Methanol, Ethanol, Ethyl acetate, hexane, Mueller–Hinton agar, DPPH (1-diphenyl-2-picrylhydrazyl), PT Reagent (Prothrombin Time), APTT Reagent (Activated Partial Thromboplastin Time), Tri-sodium citrate.

Equipments: Ultrasonic Bath (Hwashin Power Sonic 405), Rotary evaporator (Heidolph Laborata 4000, Germany), UV Detecting Chamber 254 nm, and 365 nm lamps.

All the test are applied in Vitro.

Plant Material:

Ruta chalepensis L. aerial parts (stems, leaves, flowers) was collected, beginning of March to end of, April 2018, from the public gardens in Aleppo city, The previous samples were dried in the laboratory atmosphere by placing them in the shade at a temperature of 25 °C until the weight was stable (for one week), then the dried parts were ground by a blade mill to obtain the vegetable powder. The dry plant powder was kept in opaque and airtight glass containers and in a cool place for conducting extraction processes and necessary experiments.

Extracts preparation:

Plant extractions (5g) The aerial parts of plant extracted were performed with (50ml) methanol 70%, ethanol 80%, ethyl acetate 99.99% and hexane 99.98% in glass bottles for one hour in a sonicator bath using 10 ml solvents per gram ground plant material. The extracts were filtered using What man No. 1 filter paper. The residual aerial parts powder were re-extracted twice applying the same procedure (Andersen *et al.*, 2006). Finally, the combined extracts were evaporated under reduced pressure in a rotary evaporator at room temperature. The final concentration extract (15mg/ml). All extracts were freeze-dried and stored at 4°C in the dark until testing (Zhao *et al.*, 2013).

bioactivity of *Ruta Chalepensis* L

Antimicrobial and Antifungal activity by the Agar Well Diffusion method:

The antibacterial and Antifungal activity of extracts was assessed by the Agar Well diffusion assay, against four human pathogenic bacteria, three strains of gram-positive bacteria; *Bacillus subtilis*, *Staphylococcus aureus*, and *Streptococcus mutans* and three strains of gram-negative

bacteria; *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Escherichia coli*. And three species of fungi; *Aspergillus fumigatus*, *Aspergillus niger* and *Candida albicans*. All microorganisms were obtained (Khan et al., 2011).

The assessments were performed using 100.0 µl of tested micro-organisms suspension, which contains culture medium was used Muller Hinton Agar with a concentration of 38g/l in distilled water. Table (1) shows the composition of the medium.

Table 1. Mueller Hinton Agar Composition.

Ingredients	g/l
HM infusion B from	300
Acicase	17.5
Starch	1.5
Agar	17

The medium is prepared by heating, stirring, adjusting the pH, and sterilizing the nutrient medium at a temperature of 121°C for 15 minutes, in an autoclave, then cooling to 45°C, and pouring into Petri dishes. Then the plates containing the medium were inoculated with Muller Hinton Agar with a bacterial suspension with a concentration of 0.5 Mc/Ferland, which is equivalent to (1.5×10^8 CFU/mL) by planning method, covering the entire surface of the nutrient, then making holes in the nutrient medium with a diameter of 6 mm and filling Drilling with plant extracts at a rate of 50µl, then placing the plates in the fridge for 1-2 hours so that the extract spreads in the agar, then the plates containing the bacteria are incubated in the incubator for 24 hours at a temperature of 32.5-37.5°C, and bacteria were inoculated overnight at 37°C in 10.0 µL of the extract (15mg/ml) was added individually to a 6 mm Whatman filter paper then the diameters of the areas of no bacterial growth surrounding the holes are measured Usually measured in (mm) (Nccl., 2001). while plates containing fungal (2×10^5) spores/mL of the tested fungal strains, Sabouraud Dextrose Agar (SDA) was used at a concentration of 30g/l in distilled water, were incubated at 30°C for 24 and 48 h, respectively (Erturk. 2006).

1.4. 2. Antioxidant activity (DPPH (1-diphenyl-2-picrylhydrazyl) radical-scavenging assay)

The antioxidant activity of extracts was estimated according Yen and Duh (1994) with slight modification using the stable DPPH radical, which has an absorption maximum at 515 nm. A solution of the radical is prepared by dissolving 0.4 mg DPPH in 100 ml methanol. A test solution (40 ml) was added to 3 ml of methanolic DPPH. The mixture was shaken vigorously and kept at room temperature for 30 min in the dark. Absorbance of the reaction mixture was measured at 515 nm spectrophotometrically. Absorbance of the DPPH radical without antioxidant, i.e. blank was also measured. All the determinations were performed in threeplicate.

The capability to scavenge the DPPH radical was calculated using the following equation [Yen and Duh,1994]. DPPH Scavenged (%) = $((AB-AA)/AB) \times 100 \dots (1)$, where, AB is absorbance of blank at t= 0 min; AA is absorbance of the antioxidant at t= 16 min. A calibration curve was plotted with % DPPH scavenged versus concentration of standard antioxidant (Trolox).

Anti-coagulant assay

Preparation of Pool of Plasma and Red Blood Cell (RBC) Suspension:

Blood samples from healthy volunteers free from medication for at least two weeks and fasted for at least 8 h was taken by venipuncture, and collected into 3.8% tri-sodium citrate (9:1 v/v, blood: anticoagulant) and K3EDTA (1.5 mg EDTA: 1 mL blood) tubes. The human plasma pool was prepared from the supernatants obtained after centrifugation at 4000 g for 15 min at room temperature, and stored at 4 °C until its use (Silva *et al.*, 2014). The red blood cell (RBC) suspension preparation, blood collected with EDTA was centrifuged at 560 g for 10 min at room temperature and the red blood cell pellet was subsequently rinsed three times with PBS. A 20% (v/v) RBC suspension was obtained by dilution with PBS. The RBC was used immediately after preparation.

Prothrombin Time PT Test:

The action in extrinsic pathway was evaluated by PT test, as described by Mahajan *et al.*, 2012 with a few modifications. The test was carried out using commercial reagent kits (Diagnostica Stago, France). Plasma (90 µL) was mixed with 10 µL of samples (10, 20, 30, 40 µg/mL) in saline containing a DMSO concentration of 2% (v/v) and incubated at 37 °C for 5 min at 37 °C. Then, 200 µL of PT assay reagent (rabbit brain extract and calcium chloride) pre-warmed at 37 °C for 10 min. The tube was shaken to mix the contents and it was tilted gently back and forth and the stopwatch was stopped as soon as the clot formation began. Plasma alone was used as control (absence of anticoagulant activity). Heparin 10 µg/mL (Chandra Bhaga, Pharma pvt. Ltd, Mumbai, India) was used as positive control, and DMSO 2% was used in place of the extracts for the negative control.

Activated Partial Thromboplastin Time APTT Test:

The action in intrinsic and common pathways was evaluated by APTT test, as described by Rosselli *et al.*, 2007, with a few modifications. The test was carried out, using commercial reagent kits (Diagnostica Stago, France). Plasma (90 µL) was mixed with 10 µL of samples (10, 20, 30, 40 µg/mL) in saline containing a DMSO concentration of 2% (v/v) and incubated at 37 °C for 5 min at 37 °C, before the addition of pre-warmed APTT reagent (rabbit brain extract and ellagic acid) and incubation at 37 °C for 2 min, Pre-warmed 37 °C, 25 mM calcium chloride was then added.

The tube was shaken to mix the contents and it was tilted gently back and forth and the timer at the moment of adding 100 µL calcium chloride solution 25 mM was inserted into the tube and stopped when the clot formation began. Plasma alone was used as control (absence of anticoagulant activity). Heparin 10 µg/mL (Chandra Bhaga, Pharma pvt. Ltd, Mumbai, India) was used as positive control, and Normal saline water 0.9% was used in place of the extracts for the negative control.

statistical analysis:

The statistical program (spss, 1998) was used to analyze the data and used the least significant difference test (L.S.D) at the level of significance 0.01 to compare between the mean readings.

Results and Discussion:

Plant extraction:

The percentage yield of extraction was determined using methanol 70%, ethanol 80%, ethyl Acetate and hexane for the aerial parts extracts of *Ruta Chalepennsis L* plant of, and the results shown in the following table were obtained.

Table 2. The percentage yield of extracting the aerial parts of *Ruta Chalepennsis L* plant

The aerial parts of <i>Ruta Chalepennsis L</i> extracts			
Methanol 70%	Ethanol 80%	Ethyl acetate	Hexane
38.73	45.66	4.96	4.42

The Polar solvents such as methanol and ethanol were compared with non-polar solvents such as ethyl acetate and hexane. The Ethanol 80% was used and Methanol 70% because according to previous studies it was observed that the mixing of water with Methanol and Ethanol increases their effectiveness (Yen *et al.*, 1994).

It was noted from the previous table that ethanol 80% extract achieved the highest extraction yield, The high effectiveness of the ethanol 80% and methanol 70% may be due to the plant's richness in phenolic compounds and flavonoids dissolved in polar solvents (Yen *et al.*, 1994). followed by methanol 70% extract, then ethyl acetate, and finally hexane.

In previous studies (Muhamad *et al.*, 2019) was undertaken to prepared crude extracts of camphor leaves with different polarity organic solvents using a hot extraction (Soxhlet) and cold extraction (maceration) method, The extraction yield is a measure of the solvent efficiency to extract chemical constituents from the samples. A result of different extraction method by using different polarity organic solvent such as hexane, chloroform and ethanol on the camphor leaves were obtained as shown in Table 3. The yield obtained was differ for each solvent and both methods used. The yield for hot and cold extraction using ethanol solvent has the highest yield with 8.52% and 49.56%, respectively. However, cold extraction gave high percentage than hot extraction because of the high impurities includes in the extract. It shows that both extractions were efficient and gave more influence on the percentage yield using ethanol solvent compare to hexane and chloroform extract with only small amount of yield obtained. In cold and hot extraction technique, the extract yield percentage of all crude extracts was found to be in order: ethanol > chloroform > hexane.

Table 3. Yield value of ethanol, chloroform and hexane extracts of Camphor leaf

	Solvent	Extract
Hot Extraction	Hexane	1.35
	Chloroform	1.86
	Ethanol 96%	4.26
Cold Extraction	Hexane	0.42
	Chloroform	1.19
	Ethanol 96%	24.78

Antimicrobial activity:

The extracts of methanol 70%, ethanol 80%, ethyl Acetate and hexane of *Ruta Chalepennsis L* plant-controlled conditions antimicrobial activity assessment, the parameters examined were the zone of inhibition, zone diameter (mm).

Table 4. Zones of growth inhibition (mm), showing antibacterial activity of *Ruta Chalepennsis L* plant extracts (15mg/ml).

Test organism	The aerial parts of <i>Ruta Chalepennsis L</i> extracts				
	Zone inhibition (mm)a ± standard deviation				
	Methanol 70%	Ethanol 80%	Ethyl acetate	Hexane	RA
Gram-positive					
<i>Bacillus subtilis</i>	24±0.20	26±0.08	18±0.05	16±0.03	32
<i>Staphylococcus aureus</i>	22±0.03	25±0.01	16±0.08	14±0.09	30
<i>Streptococcus mutans</i>	25±0.02	28±0.04	20±0.09	18±0.01	34
Gram- negative					
<i>Pseudomonas aeruginosa</i>	23±0.05	25±0.07	21±0.01	19±0.03	30
<i>Klebsiella pneumoniae</i>	14±0.07	17±0.05	12±0.08	11±0.07	22
<i>Escherichia coli</i>	16±0.03	20±0.09	14±0.05	13±0.02	26
RA = 30 µg/dick of antibiotics gentamicin and mikacin was used 30 for gram-positie and negative bacteria bacteria as references respectively.					

A significant variation was observed in the antibacterial properties activities of *Ruta Chalepennsis L* extracts. Based on the zone of inhibition diameter, Depending on the diameter of the inhibition zone, ethanol 80% extract was observed. It achieved the highest antibacterial activity for grampositive, and gramnegative bacteria, It was noted that ethyl acetate and hexane extracts had lower inhibitory activity in gram-positive and gram-negative bacteria compared with ethanol and methanol extracts. This indicates that the polar extracts are more effective as anti-bacterials, while the weak polar extracts show resistance against bacteria.

In previous studies on *Ruta Chalepennsis L* plant (Shorok *et.al.*, 2018) to determine the antimicrobial activity using ethanol 95% mixing with 5% water as a plant solvent , it was noted that the results of . It achieved the highest antibacterial activity for Gram-positive bacteria, respectively

Streptococcus mutans > *Staphylococcus aureus* > *Bacillus subtilis*. The highest antibacterial activity for Gram-negative bacteria respectively *Klebsiella pneumoniae* > *Escherichia col*.

This indicates that the diameter of the inhibition is greater by using ethanol at a concentration of 80% than for ethanol at a concentration of 95%, and thus the antibacterial activity is better.

Antifungal activity:

Methanol 70%, ethanol 80%, ethyl Acetate and hexane extracts of *Ruta Chalepennsis L* plant of aerial parts, were spiked at a rate of 10 µL per filter paper and demonstrated significant antifungal

effects for all tested strains (Table 4). Activity against fungi was compared to that of amphotericin B, a synthetic antifungal drug.

Table 5. In vitro antifungal activity of *Ruta Chalepensis* L plant extracts.

Test Organism	The aerial parts of <i>Ruta Chalepensis</i> L extracts (10 µL/disc)				
	Inhibitory diameters(mm) ± standard deviation				
	Methanol 70%	Ethanol 80%	Ethyl acetate	Hexane	Amphotericin B. (20 µg/disc)
<i>Aspergillus fumigatus</i>	16±0.03	14±0.07	12±0.06	0	22
<i>Aspergillus niger</i>	14±0.03	12±0.03	10±0.09	0	24
<i>Candida albicans</i>	17±0.06	15±0.05	13±0.01	0	26

The methanol 70%, ethanol 80%, ethyl Acetate and hexane extracts of *Ruta Chalepensis* L plant of aerial parts were spiked at a rate of 10 µL per filter paper and demonstrated significant antifungal effects for all tested strains (Table 5). Activity against fungi was compared to that of amphotericin B, a synthetic antifungal drug. The antifungal activities of methanol 70%, ethanol 80%, ethyl Acetate and hexane extracts of *Ruta Chalepensis* L plant of aerial parts showed maximum the inhibition zone in methanol 70% extract respectively *Candida albicans* > *Aspergillus fumigatus* > *Aspergillus niger*.

The result agreed with (Desam *et al.*, 2016) in the study of the antifungal activity of *Ruta Chalepensis* L volatile oils against different types of fungi, where the diameter of the inhibition of *Aspergillus fumigatus* 14.30 mm.

2.3. Antioxidant activity:

The obtained results of the antioxidant activity indicated that methanol 70%, ethanol 80%, ethyl Acetate and hexane extracts of *Ruta Chalepensis* L plant of aerial parts showed DPPH radical scavenging activity in a concentration-dependent manner (Table 6).

Table 6. The scavenging activity of DPPH radicals of *Ruta Chalepensis* L plant extracts.

Concentration (µg/ml)		DPPH Scavenging %				
Extracts	Ascorbic acid	Methanol 70%	Ethanol 80%	Ethyl acetate	Hexane	Ascorbic acid
000	00	00.00	00.00	00.00	00.00	00.00
001	05	40.66±0.03	38.57±0.03	15.73±0.05	09.34±0.05	51.95±0.03
002	10	48.01±0.05	45.97±0.07	21.46±0.01	17.96±0.02	59.03±0.05
004	15	55.37±0.09	52.46±0.08	34.76±0.06	28.61±0.03	60.77±0.03
008	20	67.39±0.02	64.00±0.05	45.68±0.09	35.35±0.08	71.42±0.09
0016	25	75.83±0.05	71.50±0.02	58.11±0.04	52.94±0.04	79.01±0.03
0032	30	80.41±0.01	76.71±0.03	67.80±0.04	63.23±0.02	88.36±0.02

The best antioxidant activity of *R. chalepensis* compared with the ascorbic acid was obtained by methanol 70% extract, and the ethanol 80% extract showed antioxidant activity. All extracts possess very promising antioxidant activities which can be attributed to the presence of phenolic compounds and flavonoids in this plant (Carocho *et al.*, 2013).

The results of the statistical analysis showed that there were significant differences at the level ($P < 0.01$) between ethanol 80% and hexane in all concentrations, and there weren't significant differences between ethanol 80% and methanol 70%.

2.4. Prothrombin Time PT Test:

Prothrombin time (PT) is known as the external pathway of the coagulation cascade, and it is used to evaluate coagulation factors III, VII, IX, and X. Prothrombin is a protein that is

produced in the liver, and it is the main substance that helps produce the coagulation factors that work on blood clotting. Fibrinogen into fibrin, which is the blood clotting substance (Govindappa *et al.*, 2015)

Methanol 70%, ethanol 80%, ethyl acetate and hexane extracts from *Ruta Chalepennsis L* plant of aerial parts were tested for blood coagulation effects in normal human plasma and found to be significantly prolonged the prothrombin time (PT) of normal human plasma.

Table 7. Effect of *Ruta Chalepennsis L* extracts on Prothrombin Time (PT) of Normal Human Plasma.

Extracts of <i>Ruta Chalepennsis L</i>	Conc (µg/ml)	Prothrombin time (PT) (Second)
		Aerial parts
Methanol 70%	10	38.13
	20	40.35
	30	52.88
	40	55.80
Ethanol 80%	10	40.32
	20	47.76
	30	54.22
	40	67.32
Ethyl acetate	10	21.40
	20	24.74
	30	28.55
	40	31.82
Hexane	10	22.93
	20	25.21
	30	35.83
	40	39.52
Heparin	10	>>100
DMSO 2%	10	15

The extracts of aerial parts and roots of *Ruta Chalepennsis L* were studied for the *in vitro* anticoagulant activity using the prothrombin time assay. Observed from (Table7), the Ethanol 80% showed the highest blood clotting time compared to other solvent extracts, followed by a methanol 70%, Whereas ethyl acetate and hexane extracts exhibited lower activity, when were comparable with polar extracts.

It was also noted from the previous table that with increasing concentrations, the anticoagulant activity increases, as it was noted that the time required for clot formation increases with the increase in concentration, and therefore the blood-diluting activity becomes greater.

The results of the statistical analysis showed that there were significant differences at the level ($P < 0.01$) between ethanol 80% and hexane in all concentrations, and there weren't significant differences between ethanol 80% and methanol 70%, and weren't between ethanol, ethyl acetate.

Activated Partial Thromboplastin Time APTT Test:

APTT is known as the intrinsic pathway of the coagulation cascade, and is used to assess coagulation factors XII, XI, X, IX, and VIII in the presence of calcium ions and platelet membrane phospholipids. The principle of the test: It is an investigative test that helps evaluate a person's ability to adequately form blood clots, by measuring the time required to form a fibrin clot with a sample of plasma to which citrate has been added, and then to measure the thromboplastin time, a suspension of phospholipids is added that activates a series of factors. The above-mentioned coagulation, and an activating agent is also added to the suspension to shorten the time required for blood clotting such as silicon dioxide, silite, kaolin, or ellagic acid, then calcium chloride is added to reverse the effect of anticoagulant citrate with a blood plasma sample, and it is called a partial test because of the lack of The reagent contains tissue factor III (Govindappa *et al.*, 2015).

Methanol 70%, ethanol 80%, methanol, ethyl acetate and hexane extracts from *Ruta Chalepennsis L* plant of aerial parts were tested for blood coagulation effects in normal human plasma and found to be significantly prolonged the activated partial thromboplastin time (APTT) of normal human plasma. The ethanol 80% was used and methanol 70% because according to previous studies it was observed that the mixing of water with Methanol and Ethanol increases their effectiveness as strong anticoagulant agents (Gazard *et al.*, 2005).

Table 8. Effect of *Ruta Chalepennsis L* extracts on Activated Partial Thromboplastin Time APTT of Normal Human Plasma

Extracts of <i>Ruta Chalepennsis L</i>	Conc (µg/ml)	Activated Partial Thromboplastin Time APTT (Second)
		Aerial parts
Methanol 70%	10	85.93
	20	89.11
	30	90.22
	40	96.28
Ethanol 80%	10	91.84
	20	95.30
	30	97.22
	40	100.63
Ethyl acetate	10	45.46
	20	49.83
	30	52.03
	40	57.66
Hexane	10	40.10
	20	43.62
	30	48.05
	40	50.19
Heparin	10	>>240
DMSO 2%	10	45

The extracts of *Ruta Chalepennsis L* plant were studied for the *in vitro* anticoagulant activity using the activated partial thromboplastin time (APTT) assay. Observed from (Table 8), the

Ethanol 80% aerial parts extracts of *Ruta Chalepennsis* L showed the highest blood clotting time compared to other solvent extracts, Followed by a methanol 70%, Whereas ethyl acetate and hexane extracts exhibited lower activity, where were comparable with polar extracts.

It was also noted from the previous table that with increasing concentrations, the anticoagulant activity increases, as it was noted that the time required for clot formation increases with the increase in concentration, and therefore the blood-diluting activity becomes greater.

The results of the statistical analysis showed that there were significant differences at the level ($P < 0.01$) between ethanol 80% and hexane in all concentrations, and there weren't significant differences between ethanol 80% and methanol 70%, and weren't between ethanol, ethyl acetate.

Conclusions:

1- The results of the statistical analysis in antibacterial, antifungal, antioxidant and anticoagulant activity, showed that there were significant differences at the level ($P < 0.01$) between ethanol 80% and hexane in all concentrations, but there Weren't significant differences between ethanol 80% and methanol 70%, and achieved Highly effective.

2- It is preferable to use an extract of 80% ethanol and methanol 70% to obtain the best results when performing other studies on this plant.

3- The high effectiveness of the antioxidant compounds may be due to the plant's richness in phenolic compounds and flavonoids(Yen *et al.*,1994).

4- The intrinsic (APTT) and extrinsic anticoagulant (PT) activity may be due to the presence of coumarins. The coumarins are known as strong anticoagulant agents (Gazard *et al.*, 2005).

Recommendations:

It is advised to conduct more research, in-depth biological studies on the *Ruta Chalepennsis* L plant, and to apply the anticoagulant effect in vivo

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دراسة الفعالية الحيوية، المضادة للأكسدة، والمضادة للتخثر لنبات السذاب

Ruta Chalepensis L.

المزروع في سورية

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الملخص

تمت في الدراسة الحالية (جامعة حلب-كلية العلوم، عام 2018) استخلاص المكونات الفعالة من الأجزاء الهوائية لنبات السذاب *Ruta Chalepensis L.* باستخدام محلات مختلفة ميتانول 70%، وإيتانول 80%، وخلات الايتيل، والهكسان، وُحددت فعالية المستخلصات المضادة للبكتيريا إزاء بكتيريا موجبة وسالبة الغرام، وفعاليتها المضادة للفطور، بينت النتائج أن خلاصات الميتانول 70% والايثانول 80% قد حققت أعلى فعالية مضادة للجراثيم والفطور، كما وُحددت الفعالية المضادة للأكسدة باستخدام طريقة كبح الجذر الحر الـ DPPH باستخدام تراكيز مختلفة ولوحظ بأن جميع الخلاصات قد أعطت فعالية مضادة للأكسدة. وُحددت الفعالية المضادة للتخثر باستخدام طريقة تحليل PT، APTT باستخدام تراكيز مختلفة من الخلاصات النباتية وقد بينت النتائج أن خلاصات الميتانول 70% والايثانول 80% قد حققت أعلى فعالية مضادة للتخثر.

الكلمات المفتاحية: *Ruta chalepensis L.*، مضادات الجراثيم، مضادات الفطور، مضادات الأكسدة، مضادات التخثر، DPPH.