Open Access

Journal of the Faculty of Medicine Baghdad
Print ISSN: 0041-9419, Online-ISSN: 2410-8057
DOI: https://doi.org/10.32007/jfacmedbagdad.6512008.

Research Article

Antibacterial Effect of Ethyl Acetate Fraction of *Medicago Sativa* on *Escherichia Coli* in Urinary Tract Infections

Alaa G. Ahmed[1], Mohammed Q. Yahya[1], Wifaq M.Ali Alwattar [2]
[1]*Department of Pharmacology, College of Medicine, University of Baghdad, Baghdad, Iraq.*
[2]*Department of Microbiology, College of Medicine, University of Baghdad, Baghdad, Iraq.*

Received: Nov. 2022
Revised: Jan.2024
Accepted: April 2023
Published: April 2023

Abstract:

Background: One of the most frequent bacterial infections is urinary tract infections, both in public and in medical settings. It is vital to employ new techniques to overcome drug resistant bacteria, which has emerged as one of the major health issues facing the world. Medicinal plants (Medicago sativa) are recognized as one of the most abundant sources of antibiotics in this regard.

Objective: To evaluate the antibacterial effect of the fraction of ethyl acetate of *Medicago Sativa* extract against Escherichia coli isolated from urine samples of patients with urinary tract infections.

Methods: In the current study, 85 urine samples were taken from patients attending one of the teaching hospitals in the Medical City complex in Baghdad. The study was conducted between December 2021 and May 2022. The urine samples were cultured using semi-quantitative culture techniques, and conventional microbiological methods were employed to identify the bacteria. The method of Kirby-Bauer Disk diffusion was used to determine the susceptibility to antibiotics. The antibacterial effect of Medicago Sativa was tested by preparing Medicago sativa crude extract in 85% ethanol in the Soxhlet apparatus. The extract is then fractionated into ethyl acetate fraction, then the effectiveness against uropathogenic *E. coli* was tested at different concentrations (25mg/ml, 50mg/ml and 75mg/ml).

Result: Of the 85 UTI patients from whom the urine samples were taken, 60% were females and 40% were males. The majority of the cases were under 40 years old (56.7%). All isolates showed complete meropenem sensitivity. The extracts from Medicago Sativa showed significant antibacterial activity against *E. coli.*

Conclusion: *Medicago sativa* extracts have an excellent opportunity as an antibacterial agent to uropathogenic *E. coli*, according to the findings of the current investigation.

Keywords*: Escherichia coli*; Ethyl acetate fraction; *Medicago sativa* extract; UTIs.

Introduction:

Urinary tract infections (UTIs) are one of the most frequent infections and a substantial reason for mortality and morbidity (1). A UTI occurs in about 40% of women at some point in their life. (2). UTIs affect all age groups, females get UTIs more commonly and frequently than males (3).“The most common cause of urinary tract infections is uropathogenic E. coli (UPEC)”. The infection can affect both the upper and lower urinary tract. Upper urinary tract infections are referred to as pyelonephritis while lower urinary tract infections are called cystitis. (4). UPEC strains harbour many virulence characteristics that enable the establishment of an infection, including adherence, toxins, and host defense avoidance mechanisms (5). “Antibiotics such as trimethoprim, sulphamethoxazole, and ciprofloxacin are among the most commonly recommended therapeutics for UTIs. However, the rise of antibiotic resistance and high recurrence rates of such common infections greatly impact society” (6). Today, finding new antimicrobial agents to overcome infection is still the greatest priority (7). In traditional medicine, herbs have been employed for a long time and documented phytochemical and pharmacological investigations confirm this use. In addition, they are effective for clinical research and the creation of commercial medications (8). Alfalfa, scientifically known as *Medicago sativa*, is a perennial herb plant with flowers that belongs to the Fabaceae family of peas. One of the most well-known medical plants, alfalfa may reach a height of three feet and has vibrant green leaves and bluish-violet flowers (9). the *Medicago sativa* extract exhibited significant antimicrobial activity against microbial pathogens and can be initiated as a choice to chemical antimicrobial drugs. Since the problem of antimicrobial drug resistance is expanding, our research aimed to evaluate the antibacterial influence of *Medicago sativa* on uropathogenic *E. coli* cultured from urine samples of patients visiting the Medical City hospitals in Baghdad.

**Corresponding Author: alla.Ghaith1206b@comed.uobaghdad.edu.iq*

Materials and methods

Plant materials: The whole Medicago sativa plant, which belongs to the Fabaceae family, was gathered in Kirkuk, and its identity was verified by the pharmacognosy department at Baghdad College of Pharmacy. The plant was harvested in November (2021), during the flowering season, cleaned, dried at room temperature in the shade, mechanically ground, and weighed.

Extraction and fractionation: Three hundred and fifty grams of coarsely powdered plant materials that had been shade-dried for 24 hours were defatted with hexane and then left to dry at room temperature. In a Soxhlet system, the defatted plant materials were extracted using 1.5 liters of 85% ethanol until they were completely depleted.

A dark greenish-yellow residue known as the crude fraction was produced after the alcoholic extract was vaporized to dryness at a lower pressure and a temperature below 40 °C. The crude fraction was divided using solvents of varying polarities, such as the petroleum ether F1 (boiling point from 40 - 60 ºC), the Chloroform F2, the Ethyl acetate F3, and the Ethanol F4 (2x100 ml) for each fraction. Anhydrous sodium sulfate was used to dry the fractions (10, 11).

A preliminary qualitative examination of phytochemicals

Plant crude and fractional extracts were subjected to chemical assays to determine their active components (steroids, terpenoids, flavonoids, and alkaloids) among others using accepted practices (10).

Chemical tests

Alkaloid test: **The test of Dragendorff's:** 1 - 2 drops of Dragendorff's reagent were applied to 2 ml of alcoholic extract. The presence of precipitation in orange colour suggests that alkaloids were successfully detected.

Mayer's test: 1 - 2 droplets of Mayer's reagent were applied to two ml of alcoholic extract. The formation of a creamy white precipitate suggests that alkaloids were successfully detected.

Flavonoid test: The test of lead acetate t: One ml of 10% lead acetate solution was applied to 2 ml of an alcoholic extract to check for the presence of flavonoids. The appearance of precipitation that is yellow-white was interpreted positively.

Steroids test: Liebermann-Burchard:2 ml of the extract were treated with 1-2 ml of acetic anhydride, 1 ml of chloroform, and two drops of concentrated H2SO4 (concentrated H2SO4 were slowly added along the test tube's side). A dark green colour indicates a positive outcome.

Terpenoids test: Salkowski test: 2 ml of the extract was blended with 2 ml of chloroform followed by a careful addition of 2 ml of concentrated H2SO4. The existence of terpenoids was confirmed by the formation of a red-brown layer at the surface

Condition of high-performance liquid chromatography

- The main component of fast liquid chromatography (FLC) was isolated.

The column under the optimum condition column: Nuclear C18-DB, 3 nm particle size (50 x 4.6 mm I.D) column.

A linear gradient of the mobile phase and the solvent A (0.05%) trifluoro acetic acid (TFA acid) in deionized water: Solvent B (0.05%) TFA in methanol, pH 2.5.

Program with a gradient from (0%) B to (100%) B for 15 minutes.

1.1 ml/min flow rate

Detecting UV at 355 nm

Urine sample collection: Patients visiting the Medical City hospitals with clinical features of UTI received sterile disposable containers and requested a midstream specimen of urine. The container was labelled properly with the name of the patient and the date of collection and the specimens were instantly examined.

Culturing the urine samples: The semiquantitative culture techniques were employed for cultivating urine samples to detect significant bacteriuria using a traditional method. Before inoculating the agar plates, the bacteria must be brought into a homogeneous suspension, the urine sample was mixed rather than centrifuged. After touching the loopful of samples to the growth medium's (MacConkey agar, blood agar) surface, the inoculum diffused throughout the entire plate. After 24 hours of aerobic incubation at 37 ºC, the inoculation plates were checked for the isolates using morphological, microscopic, and vitek2 techniques. *E. coli* isolates were the only isolates included in the analysis.

Testing for antibiotic susceptibility: The modified Kirby-Bauer discs diffusion method suggested by CLSI 2014 (12) was used to evaluate the antibiotic susceptibility of the bacterial isolates to six antibiotic discs: Meropenem (MEM, 10mcg), ceftriaxone (CRO, 10mcg), Amikacin (AK, 10mcg), ciprofloxacin (CIP, 10mcg), nitrofurantoin (F, 100mcg), Trimethoprim (TMP,10mcg). It was determined that ***E. coli*** isolates that displayed resistance to more than two antibiotics was a multidrug-resistant strain.

Testing the antibacterial effectiveness: At first formation of Inoculum and Test Solutions: The cultures of test organisms grown on sterile MacConkey agar surfaces were separated. By employing the streak plate method, the MacConkey agar plate was kept for 24 hours at 37 °C. The same morphological type of well-isolated overnight cultured colonies was then chosen from the cultured media. A flamed wire loop was used to touch each colony, and the bacterial colony was placed into test tube that had been sterilized and contained five ml of sterile 0.9%NaCl solution. bacterial suspension was thoroughly and uniformly mixed in the test tubes using a vortex mixer. The suspension of bacteria was then modified using McFarland turbidity (0.5) criteria. By using a (0.5) McFarland turbidity calibre, background in white colour in the presence of sufficient lighting, it was possible to adjust and compare the turbidity of inoculum tubes (13).

Determination of inhibition zone (Zone of inhibition test): Meropenem disc 10 mcg was used as a standard antibiotic for the inhibition zone determination to compare the results. The Medicago sativa ethyl acetate fraction was investigated for its antibacterial properties against Gram-negative bacteria ***E. coli*** by using the agar well diffusion method, this method was described by (13, 14). The plates were swabbed with small amounts of the bacterial solutions. Each plate was equally seeded and

streaked with sterile cotton swabs, to achieve a uniform dispersion of inoculums. This was done four times while streaking, spinning the plate by around 60 degrees each time. Finally, the agar's rim was swabbed, then wells were created on an agar surface by applying a 6 mm diameter (sterilized cork borer). Three various concentrations (25mg/ml, 50mg/ml, 75mg/ml) of the extract were prepared by dissolving the extract in DMSO. The plant extract solutions in 3 different concentrations were carefully applied to the corresponding plate media's wells using a micropipette antibiotic disc (meropenem 10 g/disc) was applied to the inoculated agar plate surface with a supplying tool (sterilized forceps) and firmly pressed to make full touch with the surface of the agar. Before incubation, the tested extract and antibiotic discs were given for around 20 minutes to disperse. The plates were then incubated for around 24 hours at 37 °C. After an overnight incubation, Caliber was used to measure the widths of the inhibitory zones in millimeters, and the results were recorded separately, Meropenem (10μg/ml) served as positive control and dimethyl sulfoxide served as a negative control.

Calculation of the minimal inhibitory concentration (MIC): Using a 96-well microplate and the described procedure (13), the minimum inhibitory concentration was assessed. For each well on every plate, 100μl of (MHB) was added, and (100 μl) of plant extract, containing (50mg of plant extract per ml), was then put into the first column of the microplates. Thus, total volume of (200μl) was produced for each well in the first column. Serial dilution was carried out with double folding from the first column up to the sixth column. The last volume (100 μl) of the broth and plant extract were collected from the sixth column and discarded. Column 7 contains broth only. A suspension of bacteria (50 μl), which was made up by mixing on milliliter of MHB with (100 μl) of fresh inoculums, was aseptically poured into each well up to the seventh column. then kept the plates in an incubator for 24 hours at 37 °C, and loss of turbidity was used to calculate the MIC.

Analysing the Data: The data was analyzed by employing Statistical Package for Social Sciences (SPSS) version 26. The data are presented as concentration than in concentration (25 mg/ml), table (5). (mean±standard deviation, ranges, frequencies and percentages). Analysis of Variance (ANOVA) (two-tailed) and post hoc test (LSD) least significant difference compared the continuous variables based on this method. P-values lower than 0.05 were regarded as significant.

Results

Only 30 of the 85 urine samples taken from patients tested positive for *E. coli*, the remaining samples were *E. coli* negative and were excluded from the study.

Among those, the highest proportion was from the age group of 20-39 years (56.7%), figure (1). The male:female ratio was 1:1.5, with 60% females and 40% males.

Table (1) displays shows that sensitivity to meropenem was found in 100% of the urine samples, while it was 80% for nitrofurantoin and 83.3% for amikacin.

Iraqi *Medicago sativa* pre-eliminatory analysis of phytochemicals demonstrated the existence of flavonoids, alkaloids, terpenoids, and steroids in the crude extract, but only flavonoids in the ethyl acetate fraction, table (2). HPLC analysis indicates the presence of gallic acid, salicylic acid, caffeic acid, pyrogallol, quercetin, myricetin, and naringin, figure (2) and table (3).by preparing different concentrations of ethyl acetate fraction, the researchers were able to assess the antibacterial activity of *Medicago sativa* extract against *E. coli*,as showen in figure (3). Table (4) and figure (4) compare the mean inhibition zone at various concentrations of ethyl acetate fraction with meropenem. Meropenem clearly had a larger mean inhibition zone than other concentration (32.96 mm compared to 17.32 mm for concentrations of 75 mg/ml, 15.39 mm for 50 mg/ml, and 12.92 mm for 25 mg/ml, P=0.001).

For the verification of the mean differences of the inhibition zone among groups, post hoc tests (LSD) were conducted. The results showed that the mean of the inhibition zone was significantly higher in meropenem than in ethyl acetate fraction concentrations of (75mg/ml, 50mg/ml and 25 mg/ml) (P< 0.05), higher in (75 mg/ml) concentration than in concentrations of (50 mg/ml and 25mg/ml), and higher in (50 mg/ml)

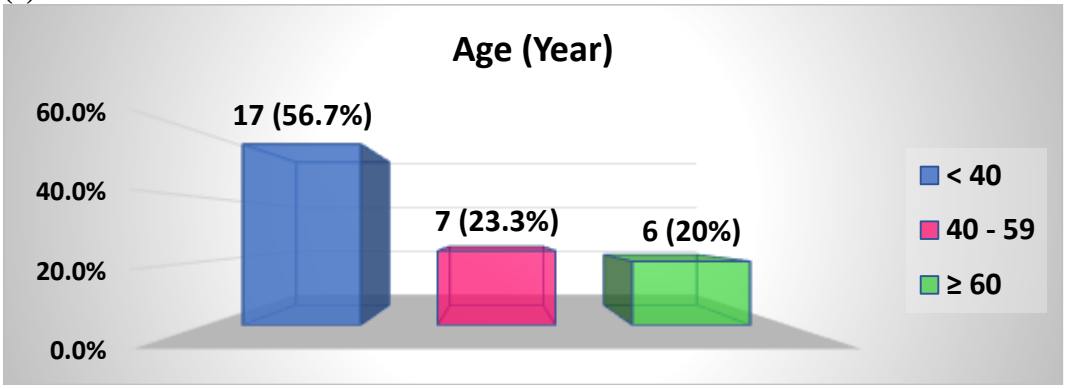

Figure (1): Age distribution of the cases with a positive E. coli urine sample

Table (1): Susceptibility to antibiotics in the E. coli positive urine samples

Drug	Symbol	No. of bacterial isolates and their percentages		
		Sensitive (%) n= 30	Intermediate (%) n= 30	Resistant (%) n= 30
Meropenem	MEM	30 (100.0)	0 (0)	0 (0)
Ceftriaxone	CRO	10 (33.3)	1 (3.3)	19 (63.4)
ciprofloxacin	CIP	14 (43.3)	0 (0)	16 (56.7)
Trimethoprim	TMP	16 (56.7)	0 (0)	14 (43.4)
Nitrofurantoin	F	24 (80.0)	3 (10.0)	3 (10)
Amikacin	AK	25 (83.3)	3 (10.0)	2 (6.7)

Table (2): Phytochemical screening of crude extract and different fraction

Crude and fractions	Alkaloids	flavonoids	Steroids	Terpenoids
Crude Extract	+	+	+	+
F1	+	-	+	+
F2	+	-	+	+
F3	-	+	-	-
F4	-	+	-	-

+ and - represent the presence and absence of phytoconstituents respectively.

Table (3): HPLC of ethyl acetate fraction

Sequence	Subject	Retention time	Area under UV
1	Gallic acid	2.007	33379
2	Pyrogallol	4.09	46455
3	Caffeic acid	5.163	25141
4	Salicylic acid	6.252	23256
5	Naringin	6.992	16266
6	Myricetin	7.902	43078
7	Quercetin	8.828	25338

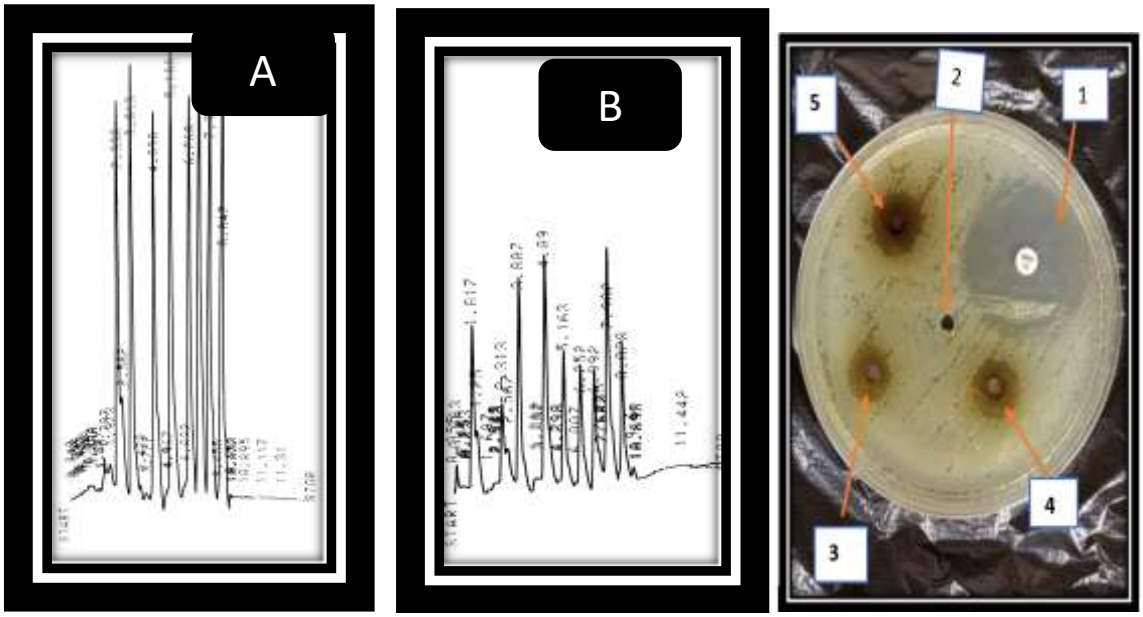

Figure (2): HPLC analysis of extract A. Standard, B. Ethyl acetate fraction

Figure (3): Sensitivity of *E. coli* to different concentrations of ethyl acetate fraction of *Medicago Sativa* 1- meropenem. 2-dimethyl sulfoxide. 3-25mg/ml, 4-50 mg/ml, 5-75mg/ml

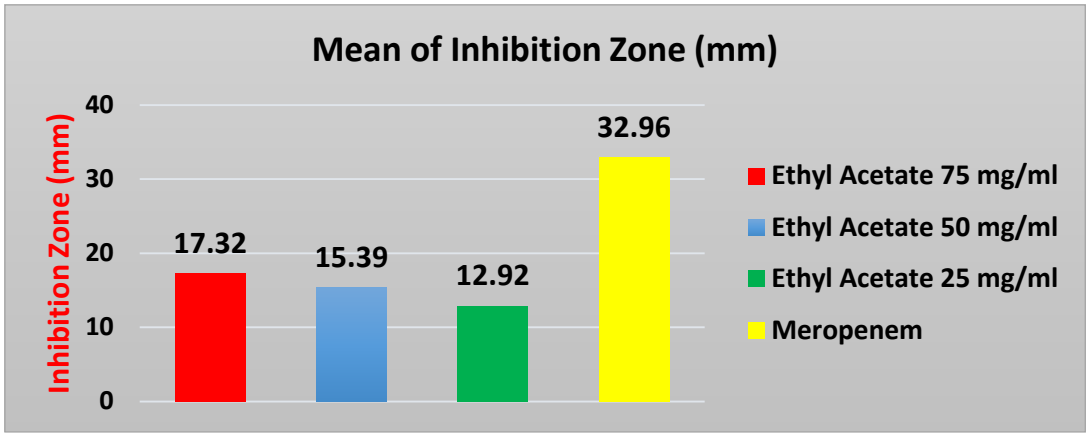

Figure (4): Inhibition zone between different concentrations of ethyl acetate fraction and meropenem

Table (4): Comparison in a mean of inhibition zone between different concentrations of ethyl acetate fraction and meropenem

Ethyl acetate Fraction Concentration (mg/m	Mean of inhibition zone (mm) Mean ± SD	F - test	P - Value
75	17.32 ± 1.5	670.87	0.001
50	15.39 ± 1.3		
25	12.92 ± 1.6		
Meropenem	32.96 ± 2.6		

Table (5): Post hoc tests (LSD) to confirm the differences that occurred between groups in the mean of the inhibition zone

The mean inhibition zone (mn	Ethyl Acetate Fraction Concentration (Mean ± SD)				P-Value
	75 mg/ml	50 mg/ml	25 mg/ml	Meropenem	
	17.32 ± 1.5	15.39 ± 1.3	-	-	0.001
	17.32 ± 1.5	-	12.92 ± 1.6	-	0.001
	17.32 ± 1.5	-	-	32.96 ± 2.6	0.001
	-	15.39 ± 1.3	12.92 ± 1.6	-	0.001
	-	15.39 ± 1.3	-	32.96 ± 2.6	0.001
	-	-	12.92 ± 1.6	32.96 ± 2.6	0.001

Table (6): Bacterial growth in different ethyl acetate fraction concentrations

The concentration of ethar fraction(mg/ml)	Bacterial growthn= 30
60	-
30	-
15	+
7.5	+
3.75	+
1.875	+

Growth = +, no Growth = -

Discussion:

One of the most frequent bacterial infection around the world is urinary tract infection. As a result of the improper or excessive use of antibiotics for treating infectious disorders, several bacterial pathogens are currently developing resistance to the available antibiotics. Scientists are advancing their studies of potential bacterial targets to combat the mutated bacterium and turning their focus on widely used plant extracts and bioactive plant components (13). Medicago sativa (lucerne), which has been used for centuries as a diuretic, antibacterial, anti-inflammatory, antifungal, and anti-asthmatic medicine, has long been regarded as a medicinal plant (15-17). In numerous regions of the world, including China, India, Mexico, Iraq, Turkey, and America, this herb has been used in traditional medicine for a very long time. The most common use is in the treatment of gastrointestinal, vascular, reproductive, and urinary tract complaints (18).

Differences in the physiology and anatomy between the two genders have been linked to females having a higher prevalence of UTIs. Women have UTIs more commonly than males do because the female urethra is short and wide towards the anus allowing bacteria to access the bladder more easily (20). The highest prevalence of UTI is seen within reproductive-age females and the highest frequency of symptomatic (UTI) in sexually active young women may account for the age-related findings (21). The patterns of bacterial sensitivity to antibiotics in the current study is in agreement with those reported by other studies (19). Polse et al reported that all of the isolates in their study were meropenem-sensitive (22). the appearance of growth inhibition zones in the agar well diffusion method serves as a visual representation of this activity. The findings of the current study were in accordance with those of Khan et al (23). Other earlier research that stated a strong correlation between antibacterial activities and extracts concentration supported this finding that the antimicrobial activity of the extracts rose as the concentration of extract increased (24), as higher concentrations of the extarcts contain more dissolved active ingredients (25). The ability of Medicago

sativa extract to combat a variety of bacteria, including Bacillus licheniformis, Pseudomonas aeruginosa, Lactococcus lactis, Klebsiella pneumonia, and Bacillus cereus, was investigated by Joy and George (26) in India. The findings of this investigation demonstrate the potency of Medicago sativa extract's anti-bacterial effect on the bacteria mentioned. In the ethyl acetate fraction, flavonoids were found via phytochemical analysis and HPLC analysis. Flavonoids are a class of heterocyclic chemical molecules found in plants. According to early studies, up to three different mechanisms may be responsible for flavonoids' direct antibacterial activity. These included the inhibition of energy metabolism (caused by NADH-cytochrome c reductase inhibition), inhibition of nucleic acid synthesis (induced by topoisomerase inhibition), and damage to the cytoplasmic membrane (caused by perforation and/or a reduction in membrane fluidity) (27). The results obtained from HPLC analysis of the ethyl acetate fraction identified the presence of Myristicin which is a naturally occurring compound found in common herbs and spices. Myristicin is a documented component of nutmeg herb and it is responsible for its antimicrobial properties. It has excellent antibacterial properties by attacking the cell wall of bacteria (28). The MIC of an ethyl acetate fraction of *Medicago sativa* that inhibits *E. coli* growth was 30 mg\ml concentration. In contrast to our findings, it was discovered that the root extract has an MIC of 125 mg/ml against Haemophilus influenza, Moraxella catarrhalis, and Streptococcus pneumonia. The diverse bacterial strain sources or the usage of a different part of the plant could be the cause of the variations in MIC values (29).

Conclusion

The recent study demonstrated the value of using Medicago sativa extract as a replacement for antibacterial medication to fight bacteria that are becoming antibiotic-resistant and beginning to impair human health. According to the findings, *Medicago sativa* extract is pharmacologically useful for treating uropathogenic E. coli-related illnesses.

Authors' declaration:

We hereby confirm that all the Figures and Tables in the manuscript are mine/ ours. Besides, the Figures and images, which are not mine /ours, have been given permission for re-publication attached with the manuscript.-Authors sign on ethical consideration's approval-Ethical Clearance: The project was approved by the local ethical committee in the pharmacology department at Baghdad College of Medicine according to the code number 178.....20/10/2021

Conflicts of Interest: None.

Funding: None.

Author's contributions:

Study conception & design: (Mohammed Q. Alatrakji). Literature search: (Alaa Ghaith Ahmed). Data acquisition: (Alaa Ghaith Ahmed). Data analysis & interpretation: (Alaa Ghaith Ahmed & Mohammed Q. Alatrakji). Manuscript preparation: (Mohammed Q. Alatrakji). Manuscript editing & review: (Wifaq M.Ali Alwattar).

References

1. AlSarray SA, Al Aubydi MA, Tothli KJ. Relation Between Aerobic Bacteria, IFN-γ, TNF-α and Miscarriage in Sample of Iraqi Women. Iraqi Journal of Science. 2019:43-9.

2. Lagha R, Ben Abdallah F, Al-Sarhan BO, Al-Sodany Y. Antibacterial and biofilm inhibitory activity of medicinal plant essential oils against Escherichia coli isolated from UTI patients. Molecules. 2019;24(6):1161. https://doi.org/10.3390/molecules24061161

3. Lüthje P, Brauner A. Novel strategies in the prevention and treatment of urinary tract infections. Pathogens. 2016;5(1):13. https://doi.org/10.3390/pathogens5010013

4. Carroll KC, Butel J, Morse S. Jawetz melnick and adelbergs medical microbiology 27 E: McGraw-Hill Education; 2015.

5. Robinson JL, Le Saux N. Management of urinary tract infections in children in an era of increasing antimicrobial resistance. Expert review of anti-infective therapy. 2016;14(9):809-16. https://doi.org/10.1080/14787210.2016.1206816

6. Flores-Mireles AL, Walker JN, Caparon M, Hultgren SJ. Urinary tract infections: epidemiology, mechanisms of infection and treatment options. Nature reviews microbiology. 2015;13(5):269-84. https://doi.org/10.1038/nrmicro3432

7. Liya SJ, Siddique R. Determination of antimicrobial activity of some commercial fruit (apple, papaya, lemon and strawberry) against bacteria causing urinary tract infection. European Journal of Microbiology and Immunology. 2018;8(3):95-9. https://doi.org/10.1556/1886.2018.00014

8. Ali MS. In vivo antimicrobial inhibition of Punica granatum extracts as mouthwash. Russian Open Medical Journal. 2017;6(4):403. https://doi.org/10.15275/rusomj.2017.0403

9. Ehsan K, Ehsan O, Armin O, Vahid O, Rudi H, Hani N. Insight into the functional and medicinal properties of Medicago sativa (Alfalfa) leaves extract. Journal of Medicinal Plants Research. 2013;7(7):290-7.

10. Abood MA, Kadhim EJ. Phytochemical Investigation of Some Active Components in Iraqi Conyza canadensis (Syn. Erigeron canadensis). 2021.

11. Ghalib SA, Kadhim EJ. The Investigation of Some Phytochemical Compounds Found in Anchusa strigosa L. Grown Naturally in Iraq. Iraqi Journal of Pharmaceutical Sciences (P-ISSN: 1683-3597, E-ISSN: 2521-3512). 2021;30(1):179-88. https://doi.org/10.31351/vol30iss1pp179-188

12. Mubarak KI. Effect of pomegranate peel extract and Thyme leaves extract on growth of Escherichia coli isolated from infant diarrhea. Zanco Journal of Pure and Applied Sciences. 2021;33(s1):103-10. https://doi.org/10.21271/ZJPAS.33.s1.11

13. Debalke D, Birhan M, Kinubeh A, Yayeh M. Assessments of antibacterial effects of aqueous-ethanolic extracts of Sida rhombifolia's aerial part. The Scientific World Journal. 2018;2018. https://doi.org/10.1155/2018/8429809
14. Srinivasan D, Nathan S, Suresh T, Perumalsamy PL. Antimicrobial activity of certain Indian medicinal plants used in folkloric medicine. Journal of ethnopharmacology. 2001;74(3):217-20. https://doi.org/10.1016/S0378-8741(00)00345-7
15. Bora KS, Sharma A. Evaluation of antioxidant and cerebroprotective effect of Medicago sativa Linn. against ischemia and reperfusion insult. Evidence-Based Complementary and Alternative Medicine. 2011;2011. https://doi.org/10.1093/ecam/neq019
16. Bora KS, Sharma A. Phytochemical and pharmacological potential of Medicago sativa: A review. Pharmaceutical biology. 2011;49(2):211-20. https://doi.org/10.3109/13880209.2010.504732
17. Cornara L, Xiao J, Burlando B. Therapeutic potential of temperate forage legumes: a review. Critical reviews in food science and nutrition. 2016;56(sup1):S149-S61. https://doi.org/10.1080/10408398.2015.1038378
18. Rafińska K, Pomastowski P, Wrona O, Górecki R, Buszewski B. Medicago sativa as a source of secondary metabolites for agriculture and pharmaceutical industry. Phytochemistry Letters. 2017;20:520-39. https://doi.org/10.1016/j.phytol.2016.12.006
19. Yadav K, Prakash S. Screening of ESBL producing multidrug resistant E. coli from urinary tract infection suspected cases in southern Terai of Nepal. J Infect Dis Diagn. 2017;2(2):116. https://doi.org/10.4172/2576-389X.1000116
20. Vasudevan R. Urinary tract infection: an overview of the infection and the associated risk factors. J Microbiol Exp. 2014;1(2):00008. https://doi.org/10.15406/jmen.2014.01.00008
21. Dash M, Padhi S, Mohanty I, Panda P, Parida B. Antimicrobial resistance in pathogens causing urinary tract infections in a rural community of Odisha, India. Journal of family & community medicine. 2013;20(1):20. https://doi.org/10.4103/2230-8229.108180
22. Polse R, Yousif S, Assafi M. Prevalence and antimicrobial susceptibility patterns of uropathogenic E. coli among people in Zakho, Iraq. International Journal of Research in Medical Sciences. 2016;4(4):1219-23. https://doi.org/10.18203/2320-6012.ijrms20160813
23. Khan S, Jan G, Bibi H, Sher J, Ullah S, Abidullah S. Phytochemical screening and antimicrobial activity of the Cichorium intybus (Familyasteraceae) and Medicago sativa (Familyfabaceae) Peshawar. Pakistan J Pharmacognosy Phytochem. 2018;7(3):603-16.
24. Bhalodia NR, Shukla V. Antibacterial and antifungal activities from leaf extracts of Cassia fistula l.: An ethnomedicinal plant. Journal of advanced pharmaceutical technology & research. 2011;2(2):104. https://doi.org/10.4103/2231-4040.82956
25. Weli TA, Mohammed AT. Effect of ginger extract on Mutans streptococci in comparison to chlorhexidine gluconate. Journal of Baghdad College of Dentistry. 2013;25(2):179-84. https://doi.org/10.12816/0014953
26. Joy GS, George P. Antimicrobial screening of Alfalfa (Medicago sativa) in various bacterial strains. International Journal of Pharmaceutics and Drug Analysis. 2014:65-9.
27. Cushnie TT, Lamb AJ. Recent advances in understanding the antibacterial properties of flavonoids. International journal of antimicrobial agents. 2011;38(2):99-107. https://doi.org/10.1016/j.ijantimicag.2011.02.014
28. Kareem MWA. The Combination Effect of Nasturtium Officinale (Watercress) Extracts and Calcium Hydroxide against Enterococcus Faecalis and Candida Albicans Isolated from Root Canal (In Vitro Study): University of Baghdad; 2021.
29. Chegini H, Oshaghi M, Boshagh MA, Foroutan P, Jahangiri AH. Antibacterial effect of Medicago sativa extract on the common bacterial in sinusitis infection. International Journal of Biomedicine and Public Health. 2018;1(1):1-5. https://doi.org/10.22631/ijbmph.2018.55334

How to Cite this Article

Ahmed AG, Al-Atrakji MQ, Al-Wattar WM. Antibacterial Effect of Ethyl Acetate Fraction of Medicago Sativa on Escherichia coli in Urinary Tract Infections. J Fac Med Baghdad [Internet]. 2023 Apr. 27 [cited 2025 Jan. 5];65(1):45-52. Available from: https://iqjmc.uobaghdad.edu.iq/index.php/19JFacMedBaghdad36/article/view/2008

التأثير المضاد للبكتيريا لجزء من أسيتات الإيثيل من ميديكاغو ساتيفا على الإشريكية القولونية في التهابات المسالك البولية

الاء غيث أحمد / قسم علم الأدوية، كلية الطب، جامعة بغداد
د.محمد قاسم يحيى مال الله الأطرقجي/ قسم علم الأدوية، كلية الطب، جامعة بغداد
د.وفاق محمد علي الوتار/ وحدة الأمراض المعدية والسريرية، كلية الطب,جامعة بغداد

الخلاصة

خلفية البحث: من أكثر أنواع العدوى البكتيرية انتشارا عدوى المسالك البولية (UTI)، سواء في المجتمع أو في المستشفيات. نظرا لأن مقاومة الأدوية أصبحت إحدى المشكلات الصحية السائدة في جميع أنحاء العالم، فمن الضروري استخدام طرق جديدة للتغلب على البكتيريا المقاومة للمضادات الحيوية. في هذا الصدد، تعتبر النباتات الطبية (Medicago sativa) من أغنى المصادر لإنتاج المضادات الحيوية.

الاهداف: تقييم تأثير المضاد البكتيري لجزء اسيتات الإيثيل من مستخلص الميديكاغو ساتيفا ضد الإشريكية القولونية المعزولة من عينة بول للمرضى المصابين بعدوى في المسالك البولية.

المواد ومنهجية العمل: خلال هذه الدراسة تم أخذ 85 عينة بول من المرضى الذين يحضرون إلى أحد المستشفيات في مجمع مدينة الطب في بغداد، وأجريت الدراسة بين كانون الأول 2021 ومايو 2022، واستخدمت تقنيات الزراعة شبه الكمية لزراعة عينات البول، وتم استخدام الطرق الميكروبيولوجية التقليدية للتعرف على البكتيريا. تم استخدام طريقة نشر Kirby-Bauer Disk لاختبار قابلية المضادات الحيوية. تم اختبار التأثير المضاد للبكتيريا لـ Medicago Sativa عن طريق تحضير مستخلص خام Medicago sativa في 85% من الإيثانول في جهاز Soxhlet. يتم بعد ذلك تجزئة المستخلص إلى جزء أسيتات الإيثيل، ثم تم اختبار فعاليته ضد الإشريكية القولونية المسببة للأمراض البولية بتركيزات مختلفة (25 مجم/ مل، 50 مجم/ مل، 75 مجم/ مل).

النتائج : كان 60٪ من عينة الدراسة من الإناث مقابل 40٪ من الذكور ومن المرضى المصابين بالتهابات المسالك البولية ولديهم الاشيريكية القولونية. كانت غالبية المشاركين في الدراسة من المجموعة العمرية (20-39 عاما) أي أقل من 40 عاما (56.7٪)، وأظهرت جميع العزلات حساسية كاملة للميروبينيم. أظهرت المستخلصات من ميديكاغو ساتيفا نشاطا مضادا للبكتيريا بشكل كبير ضد الإشريكية القولونية.

الاستنتاجات: تتمتع مستخلصات Medicago sativa بإمكانيات ممتازة كعامل مضاد للجراثيم ضد الإشريكية القولونية المسببة للأمراض البولية، وفقا لنتائج الدراسة الحالية.

الكلمات المفتاحية: الإشريكية القولونية، جزء أسيتات الإيثيل، مستخلص ميديكاغو ساتيفا، عدوى المسالك البولية.