Antibacterial Activity of Pomegranate Peel (*Punica Granatum*) and Apple Peel *(Malus Domestica*) Exract Against Urinary Tract Infection

KIRAN , SONIA SHARMA

M.Sc. Microbiology, Associate Professor

Department of Biotechnology and Microbiology

MIET, Meerut, INDIA

How to Cite this Article:

KIRAN, (2026). Antibacterial Activity of Pomegranate Peel (Punica Granatum) and Apple Peel (Malus Domestica) Exract Against Urinary Tract Infection. International Journal of Creative and Open Research in Engineering and Management, 02(05). https://doi.org/10.55041/ijcope.v2i5.794

https://doi.org/10.55041/ijcope.v2i5.794

ABSTRACT

Pomegranate (*Punica granatum)* and Apple (*Malus Domestica*) cultivation has increased significantly due to its well-recognized medicinal properties and health promoting benefits. In addition to the edible fruit and juice, pomegranate peel and apple peel a major byproducts of the food processing industry is rich in bioactive compounds including phenolic, tannins, and flavonoids compounds. These phytochemical possess strong antioxidant and broad spectrum antibacterial activities. In this study, antibacterial activity was investigated with pomegranate peel and apple peel extracts against pathogens causing urinary tract infections. The Antibacterial activity was evaluated by means of the agar diffusion method. The antibacterial potential of the peel extract was assessed against specific bacterial strains- *E. coli* and *Streptomyces Albus. Punica granatum* and *Malus domestica* extract were prepared using two different organic solvents- Ethanol and Methanol. Both extracts were effective against all bacteria tested, but the pomegranate methanol extract and pomegranate ethanol extract worked the best as compared to apple methanol extract and apple ethanol extract by inhibiting bacterial growth the most. It also exhibits resistance to certain antibiotics, such as Ciprofloxacin. The present study demonstrates that *Punica granatum* fruit peel extract exhibits significant antibacterial activity against certain pathogenic bacteria.

KEYWORDS: Antibacterial activity, Ciprofloxacin, Phytochemicals, Tannins and Flavonoids.

INTRODUCTION:

A urinary tract infection (UTI) is a common bacterial infection that affects parts of the urinary system, most often the bladder and urethra, and is typically caused by bacteria like *E. coli* entering the tract. Urinary tract infection (UTIs) account for most of the common infectious diseases found within the community or healthcare setting. In empirical antibacterial treatment in both primary and secondary care setting, UTIs are the commonest clinical indication, and urine samples usually constituent the single largest category of specimens examined in most medical microbiology laboratories (Haider G et al., 2010).Uncomplicated UTIs typically occur in healthy adult non-pregnant women, while complicated UTIs (cUTIs) may occur in all gender and age groups and are frequently associated with either structural or functional urinary tract abnormalities. UTIs are the most common problem during pregnancy as result of physiologic changes which are related to pregnancy that makes healthy women more susceptible to acquired various injury. UTIs are caused by different type of gram positive and negative bacteria like *Escherichia coli, Streptomyces albus, Pseudomonas aeruginosa, Enterococcus, Staphylococcus,* and *Streptococcus* (Ali MM et al., 2011). The pathogenic bacteria can adhere, grow and resist against host defenses which will result in colonization and infection of the urinary tract. It has been reported in several studies that the Gram-negative bacteria of *E. coli* cause 70-90% of upper and lower UTIs (Stamm WE et al., 2001). Medicinal plants have always been a good source to find new remedies for human health problems. Recently, wide range of these plants have been screened for antibacterial property. Pomegranate *(Punica granatum)* has a well- knows medicinal history. It is rich in bioactive molecules, phenolic compounds, and flavonoids and has many medicinal properties. The level of phenolic compounds changes according to cultivars and fruit parts demonstrated that peel extract contains higher total phenolics, flavonoids posseses stronger biological activities (Daham SS et al.,2010). Various extracts prepared from pomegranate fruit peels were evaluated for their antibacterial activity against some pathogens using several methods. While Apple (*Malus Domestica*) fruits consumed worldwide in different forms *i.e.* fresh, in juices and cider. Their beneficial properties to human health related to the high content of phenol compounds. Apple contain many types of phenolic derivates and flavonoids. And, apple peel is highly nutritious, containing significant amounts of dietary Fiber, Vitamins A and C, potassium, and calcium. Apples have been found to very strong antioxidant activity and antibacterial properties (Alberto et al.,2006). Extracts from the organically grown apple exhibited a clear antibacterial activity against *E. coli* and *Streptomyces albus.* The objective of this study was to explore the efficacy of using aqueous pomegranate peel extract to reduce pathogenicity of E. coli responsible for UTI and attempt to find a safety method to solve the problem of multi-drug resistance pathogen.

2. MATERIALS AND METHODS:

2.1 Collection of fruit waste and preparation of powder:

Fresh pomegranates and Apples were collected from Binoli markets Baghpat, Uttar Pardesh. The fruits were washed with water and dried using a clean cloth. The peels were manually separated, dried for a few days in open air shade and then powdered in a blender.

A- Pomegranate | **B- Pomegranate peel** | **C- Dried powder**

Figure2. 1. **Preparation of dried powder**

2.2 Preparation of extract:

Fruit peel extract was prepared by immersing 10gm powder in 100ml with two different organic solvents i.e. Methanol and Ethanol for 24 hours, after filtration the extract were evaporated by the help of water bath, ethanol for 2 hours at 60ºC and methanol for 3 hours at 60ºC. Then, the remaining concentrated extract was stored at 20ºc in refrigerator.

A- pomegranate extraction of methanol | **B- filtration of methanol extract.** | **C- liquid suspension of methanol extract.**

Figure 2.2 Preparation of pomegranate extract

2.3 Phytochemical screening:

This extract will be tested for the presence of bioactive compounds by using following reagent test.

Test for steroids:

Salkowski's Test:

Dissolve 1ml of the extract in 2ml chloroform in a dry test tube. And, dissolve 2ml concentrated H2SO4 down the side of the test tube. After that, the appearance of a reddish-brown ring at the interface indicates the presence of steroids.

Test for Alkaloids:

Dragendroff's Reagent Test

Mix 1ml of the filtrate extract with 2ml of 1% HCL and heat gently. Then, 1ml dragendroff's reagent were then added to the mixture. The appearance of an orange or reddish-brown colour indicates the presence of alkaloids.

Test for carbohydrates:

Fehling's Test:

Mixing a 1ml Fehling's A and Fehling's B. And, adding a 2ml extract. Then, heat a boiling water bath for 3-5 minutes. After that observe a brick-red precipitate, indicates the presence of reducing sugars.

Test for protein and amino acids:

Ninhydrin Test:

Take 2ml extract in a test tube. Then, add 2ml ninhydrin solution. Heat the mixture in a boiling water bath for 5 minutes under gentle conditions. Then, cool to observe purple colour for free amino acids present.

Test for Tannins:

Ferric chloride Test:

Take 1ml of extract in a test tube. And, add 1-2 drop of 5% fecl3. Mix gently and observe green-black colour condensed Tannins present.

Test for saponin:

Foam test:

1ml of the aqueous extract was taken with the help of pipette into a clean test tube. Then, add 4ml distilled water. Shake vigorously for 15-30 seconds. Then, allow to stand for 5 minutes. A foam layer is observed, and its height (1cm) indicates the presence of saponins.

Test for flavonoids:

Transfer 1ml of the extract into a 20ml test tube. Then, add 1-2 drops of 5% fecl3 solution mix gently. Observe any colour change immediately.

2.4 Determination of antibacterial activity using agar diffusion method

The two bacterial strains *E. coli* and *Streptomyces albus* were taken from MIET lab and inoculated into 100 ml of nutrient agar broth (NAM) then *E. coli* and *Streptomyces albus* incubated for 18-24 at 37ºC. An aliquot of 0.1 ml of the bacterial cultures was spread on the surface of NAM plates and the surface of the plates were dried under a Laminar air flow (LAF). Three wells of size 6mm were cut with the help of sterilized Cork borer then two wells filled with apple and pomegranate methanol and ethanol extract. One well was taken as control and filled with methanol and ethanol. Then, the plates were incubated for 24 hours at 37ºC. After incubation the plates were observed for inhibition zone. Pomegranate shows high inhibition zone as compared to apple.

2.5 Antibacterial activity

The antibacterial activity of the bacterial strains was done by making powder of 250mg ciprofloxacin tablet. 9ml distilled water was taken in a test tube and sterilized by autoclaving at 120ºC for 15-20 minutes. After sterilization, 0.2gm ciprofloxacin powder was added to the test tube.

Then, NAM plates were prepared and three wells in each were cut by Cork borer in 6 plates. In *E. coli* plates two wells were filled with pomegranate methanol and ethanol extract and one well with ciprofloxacin antibiotic. Second plate of *E. coli* was filled with pomegranate methanol extract and 2 wells were filled with antibiotic of different concentration. Third plate was filled with pomegranate ethanol extract and 2 wells were filled with antibiotic of different concentration. Same procedure was repeated in case of *Streptomyces albus.*

3. Result and Observation

Here are the results of methodology used:

3.1 Phytochemicals observations in plant extract

Pomegranate peel and apple peel contains many bioactive phytochemicals such as tannins, flavonoids,

alkaloids, phenols, and saponins. These compounds show antibacterial (antibiotic-like) activity against bacteria

A **B** **C** **D** **E** **F**

Fig: 3.1 Preliminary phytochemical screening showing the presence of (A) amino acids, (B) saponin, (C) flavonoids, (D) tannins, (E) steroids compounds in the extract.

Table: Phytochemicals present or absent in extract:

PHYTOCHEMICALS	USE REAGENTS	PRESENT/ABSENT
1.Alkaloids	Dragendroff's reagent	present
2.Carbohydrates	Fehling's test	present
3.Protein	Ninhydrin test	present
4.Saponin	Foam test	present
5.Flavonoids	Ferric chloride test	present
6.Tannins	Ferric chloride test	present
7.Steroids	Treated with sulfuric ac	present

3.2 Observation of inhibition zone:

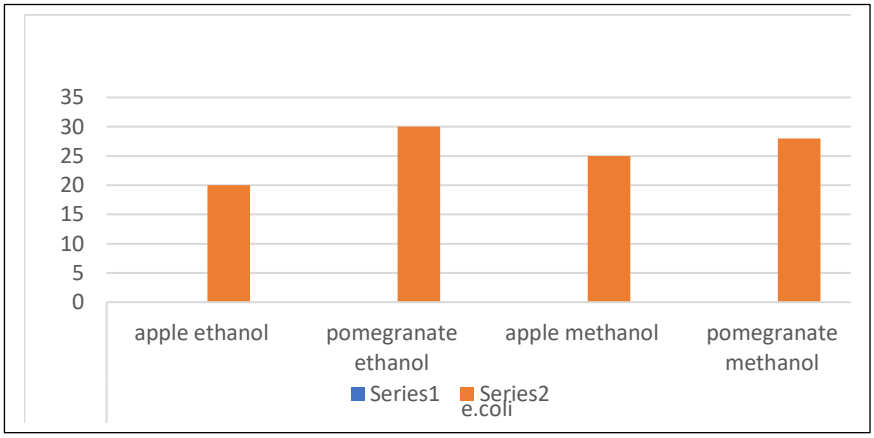
Observe the antibacterial activity of pomegranate methanol extract, pomegranate ethanol extract and concentration of antibiotic ciprofloxacin against antibacterial agent [*Streptomyces albus*]

Observe the antibacterial activity of pomegranate methanol extract, pomegranate ethanol extract and concentration of ciprofloxacin against antibacterial agent [*E. coli*]

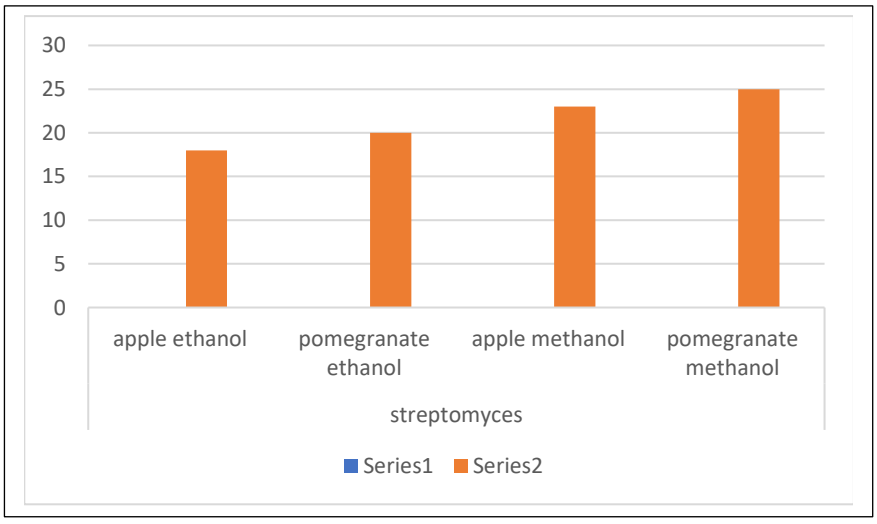
The antibacterial activity of the extract was evaluated by the agar well diffusion method. The extract produced an inhibition zone ranging from 15-50 mm against tested bacterial strains.

Sample	**Microorganism**	**Inhibition zone(mm)**
Pomegranate peel methanol extract	E. coli	22mm
Pomegranate peel methanol extract	Streptomyces albus	27mm
Antibiotic (ciprofloxacin)	E. coli	38mm
Antibiotic (ciprofloxacin)	Streptomyces albus	47mm

3.3 Effect of different extracts on bacterial [*E. coli*] growth



3.3 Effect of different extracts bacterial [*Streptomyces albus*] growth



Conclusion

According to the findings of this study, the pomegranate peel extracts showed antibacterial activities against bacterial (*E. coli, Streptomyces* etc) isolate. However, Pomegranate peel extract had a stronger antibacterial effect than apple peel extract. Therefore, the methanol pomegranate peel extract exhibited bacteriostatic, bactericidal, and anti-virulence activities against urinary tract infection. And, the methanolic extract of pomegranate peel showed higher effect than ethanol extract. The presence of phytochemicals including phenols, tannins and flavonoids may be responsible for these activities. Further studies are required to identify and isolate the active compounds present in the pomegranate's peel which exhibits the antibacterial effect and also to confirm these effects in vivo.

References

1.Haider, G., Zehra, N., Munir, A. A., & Haider, A. (2010). Risk factors of urinary tract infection in pregnancy. *JPMA. The Journal of the Pakistan Medical Association*, *60*(3), 213.

2.Stamm, W. E. (2001). An epidemic of urinary tract infections?. *New England Journal of Medicine*, *345*(14), 1055-1057.

3.Mohamed Ali, M. (2011). Evaluation of antimicrobial susceptibility &rapid urine screening tests in asymptomatic urinary tract infection in pregnant women in Karbala. *karbala journal of pharmaceutical sciences*, *2*(2), 22-34.

4. Cowan, M. M. (1999). Plant products as antimicrobial agents. *Clinical microbiology reviews*, *12*(4), 564-582.

5. Dorman, H. D., & Deans, S. G. (2000). Antimicrobial agents from plants: antibacterial activity of plant volatile oils. *Journal of applied microbiology*, *88*(2), 308-316.

6. Martin, K. W., & Ernst, E. (2003). Herbal medicines for treatment of bacterial infections: a review of controlled clinical trials. *Journal of Antimicrobial Chemotherapy*, *51*(2), 241-246.

7. Dahham, S. S., Ali, M. N., Tabassum, H., & Khan, M. (2010). Studies on antibacterial and antifungal activity of pomegranate (Punica granatum L.). *Am. Eurasian J. Agric. Environ. Sci*, *9*(3), 273-281.

8. Nuamsetti, T., Dechayuenyong, P., & Tantipaibulvut, S. (2012). Antibacterial activity of pomegranate fruit peels and arils. *Science Asia*, *38*(3), 319-22.

9. Álvarez-Ordóñez, A., Carvajal, A., Arguello, H., Martínez-Lobo, F. J., Naharro, G., & Rubio, P. (2013). Antibacterial activity and mode of action of a commercial citrus fruit extract. *Journal of applied microbiology*, *115*(1), 50-60.

10. Basu, A., & Penugonda, K. (2009). Pomegranate juice: a heart-healthy fruit juice. *Nutrition reviews*, *67*(1), 49-56.