



ANTIBACTERIAL ACTIVITY OF FRUITS AGAINST *Escherichia coli*

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ABSTRACT

Numerous fruits are unquestionably utilized to prevent food borne illness diseases. Fruits were analyzed for their antibacterial activity. The antibacterial activity was determined by disc diffusion method. Nearly eight fruits with their various concentrations (25%, 50%, 75%, 100%,) were prepared in order to check their antibacterial activity opposing *E. coli*. Mango (*Mangifera indica* L), Apricot (*Prunus armeniaca*), Grapes (*Vitisvinifera*), Apple (*Malus domestica*), Peach (*Prunus Persica*), Lemon (*Citrus limonum*), Melon (*Cucumismelo*) and watermelon (*Citrullus lanatus*) were the selected fruits. The highest inhibition zone was observed in the juice extract of Apricot with concentration of 100%. The mean value of inhibition zone was (8.2 ± 1.1121) . The minimum inhibition was surely noticed in the juice extract of mango with also concentration of 100%. The mean value of inhibition zone was (5 ± 0.9574) . Other fruits showed different inhibition zones along with different concentrations and observed that the effect of fruits against *E. coli* was concentration dependent. Response against increase and decrease in concentration was varied among all the fruits.

Keywords: fruits, antibacterial activity, *Escherichia coli*.

INTRODUCTION

E. coli is an enteric gram - negative, multi-talented *Bacillus* and optimum grasped as noninvasive symbiotic, *E. coli* assuredly breeds in human and also including in animal gut lumen in mass culture. Conceivably, more baleful bacteria are evolved from propagation. Multitudinous *E. coli* is opportunistic microbes of animals and humans. On the other hand, most of them are symbiotic. What delighted one extremely is that the growth of a critical disease, cancer, might be hindered by fruits and vegetables, since these carry the attendance of conceivably ant carcinogenic substances as vitamin E, Isothiocyanates, dithiothioes and carotenoids (Rashidkhani *et al.*, 2005). *Ziziphhus mauritiana* lam, a tropical fruit tree species, referring to the family Rhamnaceae, manipulated anode and tonic to bring about one of the ingredients utilized in chest disorder, joshanda (Mahesh *et al.*, 2008). *Terminalia chebula* Retz has traditionally been used in every nook and corner of the world in the treatment of the asthma, vomiting, hiccup, and heart, bleeding piles, gout, sore throat and bladder diseases (Aneja *et al.*, 2009).

MATERIALS AND METHODS

Fruits of various plants were collected from one of the well-known cities of Pakistan Quetta on 5th of June 2012. Most importantly, Juice from the fruits was taken out with the help of electrical equipment, Juicer. The juices were undoubtedly utilized for the test of antimicrobial effect. Different concentration of solution was prepared (25%, 50%, 75%, and 100%). Different concentration of juices dissolved in distilled water then dipped the disc with the help of forceps into solution are kept into each petri- dish. The compound was indubitably permitted to spread out for 5 minutes and then the plates were held for incubation at 37°C. Inhibition zone was arranged around the disc were certainly calculated in millimeter.

The experiment was undoubtedly performed in order to determine the antibacterial effect of the extracts of fruit on *E. coli*. Fruits (juice) were collected from the well-known city Quetta and were utilized. During the Laboratory work, the following steps were performed.

Preparation of extracts

100% solution

In 100% solution, juice was used with in pure concentration.

75% solution

75 ml juice was mixed with 25 ml of distilled water and was further used for antibacterial activity.

50% solution

50 ml of juice was mixed with 50 ml of distilled water and was further used for antibacterial activity.

25% solution

25 ml of juice was mixed with 75ml of distilled water, which was further used for antibacterial activity.

Preparation of solid medium of agar

28 grams of nutrient agar were mixed in a 1000 ml (1L) with distilled water. After this, the solution was put on hot plate for short periods. Then it was kept in an autoclave for nearly 45 minutes. Media was finally poured into the petri plates.

Preparation of filter paper discs

Watmann's filter paper discs were certainly used. Kirby Bauer antibiotic sensitivity test procedure was followed for Inoculation of *E. coli* into the petri plates.



THE KIRBY BAUER ANTIBIOTIC SENSITIVITY TEST PROCEDURE

Purpose

The main motive of this test procedure is undoubtedly to become familiar with the Kirby- Bauer producer for the assessment of the antimicrobial activity of chemotherapeutic agents.

Principle

The available chemotherapeutic agent probably differs in their scope or potential of antimicrobial activity. Some chemotherapeutic agent has a limited spectrum of microbial activity, being effective agent's only one group of microorganism. Other exhibits or has broad and wide spectrum activity agents a range of microorganism. The drug susceptibilities many disease causing microorganisms are known, but it sometimes becomes surely necessary to test a multitude of agents in order to determine the drug of choice. The producer of standardized filter paper disc agar diffusion is known as Kirby -Bauer. Most importantly, this method undoubtedly permits the rapid and quick determination of the efficiency of a drug by measuring diameter of the zones of inhibition that result from diffusion of the agent into soaked in concentration of various substances that can prevent the growth of bacteria,

antibiotic, and then kept on the surface of the agar plates that have been seeded with organism to be tested.

Muller- Hutton agar is the medium of choice with the pH of 7.2 - 7.4 is poured up to 5mm to plates and refrigerator in solidification. Before utilizing them, the plates are transferred to a box like machine incubator at 37c for approximately 10-20 minutes to dry of that developed mixture on the agar surface. The plates with the help of a cotton swab are then inoculated with inoculums in order to ensure the full growth and development of the organism. As well-spaced intervals, the discs are transferred to the surface of the agar plates, after incubation with the assistance of the incubator, the plates are carefully examined with the growth inhibition, which is indicated or represented by the clear and a vivid zone around each disc.

The size of the zone determines the susceptibility of an organism to the drug.

MCFARLAND BARIUM SULFATE STANDARD

For the procedure of API staph- indent 1% aqueous barium chloride must be prepared and prepare 1% aqueous sulfuric acid solution, utilizing the following amount of barium chloride and sulfuric acid. Then test tubes were labeled from 1 through 10.

Preparation McFarland standards

Tubes	Barium chloride 1 % (ml)	Sulfuric acid 1% (ml)	Density of bacteria million/m
1	0.1	9.9	300
2	0.2	9.8	600
3	0.3	9.7	900
4	0.4	9.6	1, 200
5	0.5	9.5	1, 500
6	0.6	9.4	1, 800
7	0.7	9.3	2, 400
8	0.8	9.2	2, 400
9	0.9	9.1	1, 700
10	1.0	9.0	3, 000

One of the common test tubes screw capped test tube, was utilized in which the solution was taken and with the help of cotton swab, for the inoculation, a small or little amount of bacteria from the culture was taken. Then after this, the comparison between the turbidity of this test tube against the turbidity of 10 test tube of McFarland standard took place. After the comparison it was noticed that test tube of McFarland series which matched with the prepared or inoculated test tube is the same one (Parekh *et al.*, 2006).

RESULTS AND DISCUSSION

Antibacterial activity of different fruits

This study was conducted to analyze the antibacterial activity of different eight available seasonal fruits (Mango, Apricot, Grapes, Peach, Apple, Lemon, Melon and water melon) against *Escherichia coli*. Fruits were collected for their bacterial activity in the summer.



Mango juice with different concentration (25%, 50%, 75%, 100%)

It was observed that 25% extract of mango had (6.6mm) inhibition of zone, 50% extract had (7.2mm), 75% had (5.8mm) and 100% extract had (5mm) inhibition zone. Among all the concentration only 50% extract was observed with the maximum inhibition zone as (7.2mm). It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

Apricot juice with different concentration (25%, 50%, 75%, 100%)

It was noticed that 25% extract of apricot's mean inhibition zone was (8.2mm), 50% extract concentration had (8mm) inhibition zone, 75% extract concentration had (6mm) inhibition zone, 100% extract concentration had (8.4mm) inhibition zone. The maximum inhibition was observed in 100% concentration with (8.4mm) inhibition zone. It was compared with control (*Ox tetracycline* and Offoxyimo Sodium)

Grapes juice with different concentration (25%, 50%, 75%, and 100%)

It was observed that 25% extract concentration of grape's inhibition zone with mean value was (6.8mm), 50% extract concentration had (6.6mm) inhibition zone, 75% extract concentration had (6.6mm) inhibition zone, 100% extract concentration had (7.6mm) inhibition zone. The maximum inhibition was observed in 100% concentration with (7.6mm) inhibition zone. It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

Peach juice with different concentration (25%, 50% 75% 100%)

It was observed that 25% extract of peach had (6.6mm)) mean value of inhibition zone, 50% extract concentration had (6.8mm) inhibition zone, 75% extract concentration had (6.2mm) inhibition zone, 100% extract concentration had (6mm) inhibition zone. The maximum inhibition was observed in 50% concentration with (6.8mm) inhibition zone. It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

Apple juice with different concentration (25%, 50%, 75%, 100%)

It was observed that 25% extract of apple had (5.6mm) mean value of inhibition zone, 50% extract

concentration had (6.4mm) inhibition zone, 75% extract concentration had (5.2mm) inhibition zone, 100% extract concentration had (7.2mm) inhibition zone. The maximum inhibition was observed in 100% concentration with (7.2mm) inhibition zone. It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

Lemon juice with different concentration (25%, 50%, 75%, 100%)

It was observed that 25% extract of lemon had (5.8mm) mean value of inhibition zone, 50% extract concentration had (5.8mm) inhibition zone, 75% extract concentration had (6.6mm) inhibition zone, 100% extract concentration had (5.4mm) inhibition zone. The maximum inhibition was observed in 75% concentration with (6.6mm) inhibition zone. It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

Melon juice with different concentration (25%, 50%, 75%, 100%)

It was observed that 25% extract of melon had (6.4mm) mean value of inhibition zone, 50% extract concentration had (6.2mm) inhibition zone, 75% extract concentration had (6.6mm) inhibition zone, 100% extract concentration had (5.4mm) inhibition zone. The maximum inhibition was observed in 75% concentration with (6.6mm) inhibition zone. It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

Watermelon with different concentration (25%, 50%, 75%, 100%)

It was observed that 25% extract of watermelon had (6.2mm) mean value of inhibition zone, 50% extract concentration had (6.4mm) inhibition zone, 75% extract concentration had (5.6mm) inhibition zone, 100% extract concentration had (7.8mm) inhibition zone. The maximum inhibition was observed in 100% concentration with (7.8mm) inhibition zone. It was compared with control (*Oxytetracycline* and Offoxyimo Sodium).

So among all the apricot had the maximum inhibition in 100% concentration with 8.4mm inhibition zone this was followed by again apricot in 25% concentration with 8.2mm inhibition zone. While mango had minimum inhibition zone of 5mm in 100% concentration.



Table-1. Showing mean and standard derivation of fruits extracts (juices) with different concentration Of 25%, 50%, 75% and 100% along with control of *Ox tetracycline*.

Control Ox tetracycline	Mean/S.D $\bar{X} \pm \bar{d}$	Fruit	Conc (%)	Mean/S.D $\bar{X} \pm \bar{d}$
5mm	5±0	Mango	25	6.6±0.9574
			50	7.2±0.9574
			75	5.8±0.9574
			100	5±0.9574
		Apricot	25	8.2±1.1121
			50	8±1.1121
			75	6±1.1121
			100	8.4±1.1121
		Grapes	25	6.8±0.4761
			50	6.6±0.4761
			75	6.6±0.4761
			100	7.6±0.47611
		Apple	25	5.6±0.8869
			50	6.4±0.8869
			75	5.2±0.8869
			100	7.2±0.8869
		Peach	25	6.6±0.3651
			50	6.8±0.3651
			75	6.2±0.3651
			100	6±0.3651
		Lemon	25	5.8±0.5033
			50	5.8±0.5033
			75	6.6±0.5033
			100	5.4±0.5033
		Melon	25	6.4±0.5259
			50	6.2±0.5259
			75	6.6±0.5259
			100	5.4±0.5259
		Watermelon	25	6.2±0.9309
			50	6.4±0.9309
			75	5.6±0.9309
			100	7.8±0.9309



Table-2. Showing mean and standard derivation of fruits extracts (juices) with different concentration of 25%, 50%, 75%, and 100% along with control of Offoxyimo sodium).

Control Offoxyimo Sodium	Mean/S.D $\bar{X} \pm \sigma$	Fruit	Conc (%)	Mean/S.D $\bar{X} \pm \sigma$
7mm	7 \pm 0	Mango	25	6.6 \pm 0.9574
			50	7.2 \pm 0.9574
			75	5.8 \pm 0.9574
			100	5 \pm 0.9574
		Apricot	25	8.2 \pm 1.1121
			50	8 \pm 1.1121
			75	6 \pm 1.1121
			100	8.4 \pm 1.1121
		Grapes	25	6.8 \pm 0.4761
			50	6.6 \pm 0.4761
			75	6.6 \pm 0.4761
			100	7.6 \pm 0.47611
		Apple	25	5.6 \pm 0.8869
			50	6.4 \pm 0.8869
			75	5.2 \pm 0.8869
			100	7.2 \pm 0.8869
		Peach	25	6.6 \pm 0.3651
			50	6.8 \pm 0.3651
			75	6.2 \pm 0.3651
			100	6 \pm 0.3651
		Lemon	25	5.8 \pm 0.5033
			50	5.8 \pm 0.5033
			75	6.6 \pm 0.5033
			100	5.4 \pm 0.5033
		Melon	25	6.4 \pm 0.5259
			50	6.2 \pm 0.5259
			75	6.6 \pm 0.5259
			100	5.4 \pm 0.5259
		Watermelon	25	6.2 \pm 0.9309
			50	6.4 \pm 0.9309
			75	5.6 \pm 0.9309
			100	7.8 \pm 0.9309



Figure-1 shows mean extracts of fruits with different concentration ((25%, 50%, 75%, 100%) along with control (Ofloxacin Sodium and *Oxytetracycline*).

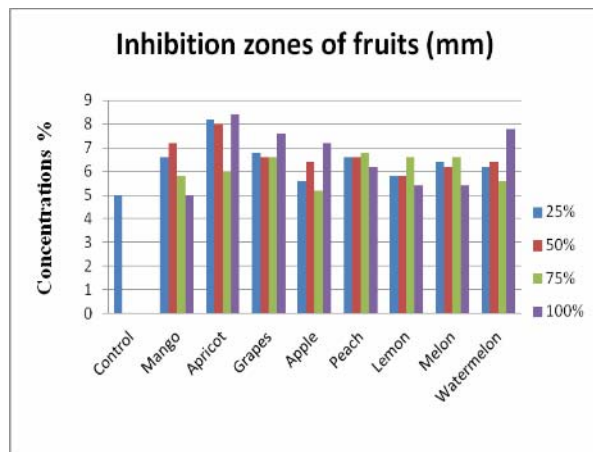


Figure-1 Comparison of inhibition zones of Ofloxacin Sodium and fruits (Juices) with different concentrations.

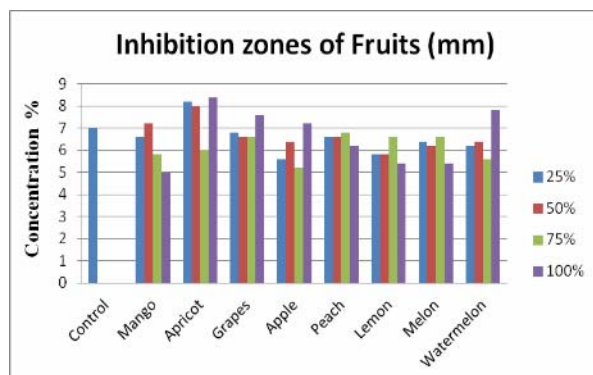


Figure-2 Comparison of inhibition zones of *Oxytetracycline* and fruits (Juices) with different concentrations.

The antibacterial potential of *Terminalia chebula* fruit extract (acetone, ethanol, methanol, cold and hot aqueous) the maximum activity was unquestionably displayed by the acetonic extracts including mean inhibition zone being (25.32mm) (Aneja and Joshi, 2009).

Flower and fruits of *Cordia allamanda* were unsurprisingly utilized to prepare an extract, which were most productive. It displayed highest antibacterial activity opposing *Staphylococcus aureus* (2.53 mm) at 15mg/ml. (Walter *et al.*, 2011).

The entire plant of *Ephedra gerardiana* was unquestionably utilized for study and analysis. *Ephedra gerardiana* displayed highest antibacterial activity opposing *E. coli* (2.57 mm) at 15mg/ml (Walter *et al.*, 2011).

CONCLUSIONS

Several fruits were undoubtedly analyzed for its antibacterial activity opposing *E. coli*, the fruits were collected from a well-known city Quetta. The fruits were Apricot, Mango, Lemon, Melon, Watermelon, Peach, and Apple and Grapes. Various juice extracts of the fruits were unquestionably put to action for the determination of antibacterial activity with undoubtedly various concentrations (25%, 50%, 75%, and 100%).

The maximum inhibition was certainly found in the juice extracts of Apricot essentially with concentration of 100% and the value of mean inhibition zone was (8.4 ± 1.1121). The minimum inhibition was surely noticed in the juice extract of mango with also concentration 100% of inhibition zone of 5 ± 0.9574 . Apricot undoubtedly plays a tremendous role in manufacturing the various products essentially jam and nectar.

RECOMMENDATIONS

- For the determination of antibacterial activity, juice of the various fruits can be unquestionably put to action.
- Roots, leaves, and flower of plants or the complete plant can unsurprisingly be utilized for the determination of antibacterial activity.
- More organisms can undoubtedly be analyzed for this antibacterial activity.

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