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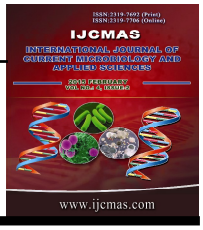

Original Research Article

Screening for Fecal Coliforms from Different Drinking Water Sources in Madurai City and Testing the Efficacy of Plant Extracts against the Isolates

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A B S T R A C T

Water is indispensable for life. Water can support the growth of beneficial and harmful microorganisms. The presence of disease causing microbes in water is unhealthy and even life-threatening. Bacteria that live in the intestinal tracts of human and other warm-blooded animals such as *Escherichia coli* can contaminate water, if feces enter the water. The present study was undertaken for screening and identifying of the fecal coliforms in five samples of drinking water from different areas in Madurai city. All the water samples show the microbial contamination but two samples show the presence of fecal coliforms. *E. coli* which is substantial indicators of fecal contamination was found in Sample I and II. Chlorination of water can bring down the fecal coliform counts. But the side effects are irritation in oesophagus, burning sensation in the mouth and throat. To overcome these harmful effects, plants could be used as alternatives for decontamination of water. Plant extracts like Lemon (*Citrus aurantifolia*), Neem (*Azadirachta indica*), Guava (*Psidium guajava*), Papaya (*Carica papaya*), Sponge Gourd *(Luffa cylindrica)*, Drumstick (*Moringa oleifera*) and Bitter gourd (*Momordica charantia*) were tested for their antibacterial activity against fecal coliforms. Among these, ethanol peel extract of *Citrus aurantifolia* shows antibacterial activity against fecal coliforms. Hence it can be used as better alternative for chemical disinfectants.

Introduction

The World Health Organization (WHO) has estimated that up to 80% of all disease and sickness in the world is caused due to inadequate sanitation, polluted water or unavailability of water. Approximately three out of every five persons in the developing countries do not have access to drinking water and only one in four has any kind of sanitary facility. About 1.6 billion people are forced to use contaminated water, this is so because most of the water sources are polluted with either industrial or domestic waste, thus unfit for use (Cheesbrough, 1984, UNICEF, 2009). Water can support the growth of many types of microorganisms. The presence of disease

causing microbes in water is unhealthy and even life threatening. (Chapelle, 2000). *E.coli* is a specific coliform species that is almost always present in fecal material and its presence likely indicates potential contamination of fecal origin. If *E.coli is* detected in a drinking water sample, this is considered as an evidence of a critical public health concern. Fecal contamination and waterborne pathogens can cause a variety of illnesses, including acute gastrointestinal illness (AGI) with diarrhoea, abdominal discomfort, nausea, vomiting, and other symptoms. Most AGI cases are of short duration and result in mild illness. Other more severe illnesses caused by waterborne pathogens include hemolytic uremic syndrome (HUS), kidney failure, hepatitis, and bloody diarrhoea (EPA, 2013). Fecal coliform bacteria, members of the family *Enterobacteriacae,* which include *Escherichia coli*, *Citrobacter*, *Enterobacter* and *Klebsiella* species, are often used as indicators which are gram negative bacteria found in the digestive tracts of all warm-blooded animals. (Geldreich *et al.,* 2008). Reduction of fecal coliform in drinking water may require the use of chlorine and other disinfectant chemicals. Such materials may kill the fecal coliform and disease causing bacteria found in the drinking water. (EPA, 2007). But consumption of chlorinated drinking water generally results in irritation of the oesophagus, a burning sensation in the mouth and throat, and spontaneous vomiting. (Muegge, 1986). There is an increased risk of bladder cancer to be associated with the consumption of chlorinated drinking water. (Cantor *et al* .,1987). Instead of using chemical disinfectants, plants could be used as an alternative for the removal of fecal coliforms. The need to exploit the potential of plants may offer cheap, and environment friendly methods of tackling water contamination and help to overcome the hazards of using chlorine (Chang, 1993). This research was undrtaken for screening and identification of fecal coliforms in drinking water in Madurai city and examining the antibacterial activity of natural plant extracts against fecal coliforms and to develop inexpensive disinfectants to purify water.

Materials and Methods

In the present study identification of fecal coliforms in drinking water was undertaken in five different areas of Madurai city which are most human populated areas. The process of collecting sample was carried out with meticulous care to avoid microbial contaminants from outside sources (Othman E.A.R and Hamid.M.E 1999). Presence of fecal coliform in different drinking water sample was analysed by presumptive, confirmatory and complete tests. Biochemical characteristics of the isolates were determined by IMViC tests and Gram staining technique .Antibacterial activity of plant extracts were tested against isolated fecal coliforms (Sarwar *et al* 2004).

Results and Discussion

In this study Table – 1 Plate – I and Plate – II showed the presence of fecal coliforms in Sample- 1 and Sample -2. Other water samples showed negative for the presence of fecal coliforms. But they showed other bacterial contamination.

Table-2 shows the results for biochemical analyses of the isolates (fecal coliforms). The isolates from sample-1 and sample-2 show positive for Indole and Methyl Red test and they show negative for the Voges Proskauer and Citrate test.

Table- 3 shows the antibacterial activity of plant extracts against the fecal coliforms.

Among the seven plant extracts, *Citrus aurantifolia* alone shows its antibacterial activity against fecal coliforms.

Table 4 shows antibacterial activity of ethanol peel extract of *Citrus aurantifolia* against fecal coliforms. In 100mg/ml concentration, the zone of inhibition is 24 mm in Sample – I and the zone of inhibition is 22mm Sample –II.

The coliform group is the indicator bacterium to evaluate the quality of drinking water and the presence of coliforms indicates the contact of drinking water with sewage. *E. coli* are members of the coliform bacteria group originating in the intestinal tract of warm blooded animals and are passed into the environment through feces.

The detection of fecal (thermotolerant) coliform organism provides definite evidence of fecal contamination and they are found positive in Sample – I and Sample – II, were further subjected to confirmatory test and metallic sheen was observed in the Sample – I and Sample – II. These samples are subjected to complete test and gas formation was noted. Further Gram staining confirms the presence of fecal coliform in Sample – I and Sample – II.

E. coli is distinguished from other fecal coliforms by Beta-glucuronidase and galactosidase activity. Thus it was able to produce gas formation in lactose broth**.** Cfu/ml was calculated for Sample – I and Sample – II. Potable water should contain 0/ml of fecal coliforms (Bhattarai, 2009). But in Sample – I the fecal coliforms count was 1.6×10^7 /ml. Sample – II the fecal coliform count was 1.3×10^7 / ml. There by the water was contaminated with fecal matter and there are risk of other even more dangerous bacteria, viruses and parasites in the water (Bruno, 1999).

Fecal coliform can be usually killed by treating the drinking water with chemical disinfectants. But due to some disadvantages of using chlorinated water, plant extracts could be used as alternatives for chemical disinfectants

Antibacterial activity of plant extracts like *Citrus aurantifolia, Azadirachta indica, Psidium guajava, Carica papaya, Luffa cylindrica, Momordica chorantia* and *Moringa oleifera* were evaluated against the isolates which are known to cause infections in human along with other pathogens.

In the present study the ethanol peel extract of *Citrus aurantifolia* revealed the antibacterial activity against fecal coliforms. (Table 3 and 4). The ethanol peel extract of *Citrus aurantifolia* exhibited significant inhibitory activity against fecal coliforms (Table 4) because the peel of citrus fruit contains essential oils like limonene, linalool which are toxic to bacteria. Essential oils (limonene) exert their toxic effects at the membrane level where they can increase the permeability of cell membrane. The most well characterized essential oils from citrus fruits are citrullene and limonene which can exert potent antibacterial activity (Barrell *et al.*, 2000).

The greatest threat posed to water resources arises from microbiological contaminations which has been a concern to public health. Water contamination with potentially pathogenic microbes represents an obvious health risk. Water pollution caused by fecal contamination is a serious problem due to the potential for contracting diseases from pathogens. As a result, testing for coliform bacteria can be a reasonable indication of whether other pathogenic bacteria are present or not. Water is a resource of primary necessity now strongly adverse by bacteriological pollutants. To avoid any

enemy or epidemy, it is imperative to disinfect the water before we drink.

The present study demonstrated that *Citrus aurantifolia* represent an economic source of antibacterial compounds that can be a better alternative for chemical disinfectants as they are more eco-friendly.

Table 1 Bacteriological analyses of drinking water for the presence of fecal coliforms

TEST	SAMPLE -1	SAMPLE- 2	SAMPLE-3	SAMPLE-4	SAMPLE-5
PRESUMPTIVE	+	+	_	_	_
CONFIRMATORY	+	+	_	_	_
CONFIRM	+	+	_	_	_

Plate – I

Plate - II

Table.2 Biochemical analyses of the isolates (fecal coliforms)

SAMPLES	INDOLE TEST	METHYL RED TEST	VOGES PROSKAUER TEST	CITRATE TEST
SAMPLE-1	+	+	-	-
SAMPLE-2	+	+	-	-

Table.3 Antibacterial activity of plant extracts against fecal coliforms isolated from drinking water samples

PLANT EXTRACTS	SAMPLE-1	SAMPLE-2
Citrus aurantifolia	+	+
Azadirachta indica	-	-
Psidium guajava	-	-
Carica papaya	-	-
Luffa cylindrica	-	-
Moringa oleifera	-	-
Momordica charantia	-	-

Table.4 Antibacterial activity of ethanol peel extract of *Citrus aurantifolia* against fecal coliforms

Samples concentration (mg/ml)	Zone of Inhibition (mm)	
	SAMPLE-1	SAMPLE-2
20	14	10
40	16	12
60	18	14
80	20	16
100	24	22

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