



## The Antiseptic Effect of Turmeric (*Curcuma longa*) Extraction on the Bacterial Growth of *Escherichia Coli* (K-12) and *Salmonella Typhi* which Cause Food Poisoning

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### ABSTRACT

Previous research has confirmed that turmeric compounds have antiseptic properties. In this study, the effect of freshly turmeric powder on *Escherichia coli* strain K-12 and *Salmonella typhi* was examined. Fresh turmeric powder were mixed with ethanol and centrifuged to produce a supersaturated turmeric solution. The supersaturated turmeric solution with several concentrations was added to macconky agar, which was used to culture *Escherichia coli* strain K-12, and deoxycholate citrate agar (DC agar) which was used to culture *Salmonella typhi*. The growth of the bacteria was studied by microscopic inspection of bacterial colonies. It was found that turmeric powder is an effective antiseptic agent against *Escherichia coli* K-12 and *Salmonella typhi*. It was determined that even at low concentrations of 50 micro liters of turmeric solution there is a noticeable antiseptic effect. It was also determined that higher concentrations of turmeric had a greater antiseptic property. There were no *Escherichia coli* K-12 and *Salmonella typhi* growth beyond a turmeric concentration of 800 micro liters 400 micro liters respectively.



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## INTRODUCTION

Turmeric *Curcuma longa*, a perennial herb and member of the Zingiberaceae (ginger) family, grows to a height of three to five feet and is cultivated extensively in Asia, India, China, and other countries with a tropical climate. It has oblong, pointed leaves and funnel-shaped yellow flowers<sup>[1]</sup>. The rhizome, the portion of the plant used medicinally, is usually boiled, cleaned, and dried, yielding a yellow powder. Dried *Curcuma longa* is the source of the spice turmeric, the ingredient that gives curry powder its characteristic yellow color. Turmeric is used extensively in foods for its flavor and color, as well as having a long tradition of use in the Chinese and Ayurveda systems of medicine, particularly as an anti-inflammatory and for the treatment of flatulence, jaundice, menstrual difficulties, hematuria, hemorrhage, and colic. Turmeric can also be applied topically in poultices to relieve pain and inflammation<sup>[2]</sup>. Current research has focused on turmeric's antioxidant, hepatoprotective, anti-inflammatory, anticarcinogenic, and antimicrobial properties, in addition to its use in cardiovascular disease and gastrointestinal disorders.

The active constituents of turmeric are the flavonoid curcumin (diferuloylmethane) and various volatile oils, including tumerone, atlantone, and zingiberone. Other constituents include sugars, proteins, and resins. The best researched active constituent is curcumin, which comprises 0.3-5.4 percent of raw turmeric<sup>[2]</sup>.

Curcumin is a potent anti-inflammatory with specific lipoxygenase- and COX-2- inhibiting properties. Animal, in vitro, and in vivo studies demonstrate turmeric's effectiveness at decreasing both acute and chronic inflammation<sup>[3-6]</sup>. Turmeric extract and the essential oil of *Curcuma longa* inhibit the growth of a variety of bacteria, parasites, and pathogenic fungi. A study of chicks infected with the caecal parasite *Eimeria maxima* demonstrated that diets supplemented with 1% turmeric resulted in a reduction in small intestinal lesion scores and improved weight gain<sup>[7]</sup>. Turmeric's protective effects on the cardiovascular system include lowering cholesterol and triglyceride levels, decreasing susceptibility of low density lipoprotein (LDL) to lipid peroxidation<sup>[8]</sup> and inhibiting platelet aggregation<sup>[9]</sup>. These effects have been noted even with low doses of turmeric.

The **goal** of this project is to study the antiseptic effect of different concentrations of turmeric on a particular strain of *Escherichia coli* (K-12) and *Salmonella typhi*. Alcohols, like ethanol was found to be suitable solvent for turmeric as turmeric solution in alcohol is soluble in water. For this project, 200 proof ethanol was used as a solvent for turmeric.

## MATERIAL AND METHOD:

The materials that were used to conduct this experiment are:

- 1- Centrifuge (Beckman Brand, CPKR type).
- 2- Ethanol (Decond Labs Brand, standard 200 proof DSP-MD.43).
- 3- Laboratory test tubes (standard).
- 4- Autoclave (Fisher Scientific Brand).
- 5- Laboratory fume hood (Fisher Scientific Brand, EF-5-8).
- 6- Beaker (500mL capacity, standard).
- 7- Macconky agar, which was used to culture *Escherichia coli* strain K-12, and deoxycholate citrate agar (DC agar) which was used to culture *Salmonella typhi*.
- 8- Nutrient broth tubes.
- 9- Sterile loops (Fisher Scientific Brand).
- 10- Incubator (standard).

### A- *Curcuma* extraction preparation:-

Different amounts of the supersaturated turmeric solution, prepared by centrifuging a mixture of 200 proof ethanol and 100 grams of finely turmeric powder (which collected from local markets) was added to an agar solution to study the effect of turmeric on *Escherichia coli* strain K-12 and *Salmonella typhi*.

200 proof ethanol was added to the beaker containing the turmeric powder (100 grams) for a total volume of 180 milliliters, and mixed the mixture to create a supersaturated solution the beaker was placed on a lab bench. Then the ethanol and turmeric solution in the beaker was then distributed evenly and poured into four laboratory test tubes. Each test tube had about 45 milliliter of the supersaturated solution and whirled counterclockwise by hand a few times.

The test tubes were then placed into the centrifuge so that both sides of the centrifuge were balanced. The centrifuge was set to the specifications: temperature of twenty-two degrees centigrade, speed of 3870 rotations per minute, timed for seven minutes.

After seven minutes in the centrifuge, the clear turmeric solution was visible at the top and debris settled at the bottom in each of the four test tubes, and the clear solution at the top was carefully poured in a set of new test tubes so that all debris remained in the original test tubes. The clear solution of turmeric in ethanol served as a stock of turmeric for experimentation. This solution was put in a cold room at four degrees centigrade to avoid evaporation of the ethanol.

**B- Culture media preparation and bacterial incubation:-**

Prepare 800 ml of the macconky agar according to the industrial company in beaker to use it to culture the *Escherichia coli strain K-12* after sterile it by autoclave, the agar solution was cooled until the temperature of the agar solution was measured to be 40 degrees. And then 100 ml of macconky agar solution was poured into another beaker and then poured into four Petri dishes, these four Petri dishes were marked as control. The process of pouring 100 ml of macconky agar into a second beaker was repeated; each time a different amount of turmeric stock solution was added to the macconky agar and poured into four already marked Petri dishes. The amount of turmeric solution added were 50 micro liters, 100 micro liters, 200 micro liters, 400 micro liters, 800 micro liters, 1000 micro liters, and 2000 micro liters respectively, yielding 32 dishes including the control, each with a different concentration amount of turmeric extraction.

So prepare 800 ml of the deoxycholate citrate agar (DC agar) according to the industrial company in beaker to use it to culture the *Salmonella typhi* after sterile it by autoclave and use the same method to prepare 32 Petri dishes of deoxycholate citrate agar (DC agar) with a different concentration amount of turmeric extraction.

The stock for the *Escherichia coli K-12* bacteria was prepared by sterilized swab was used to scrape the bacteria from the agar plate from the bacterial bank of sciences collage / Biology department of Al-Qadisiya University and The bacteria was Injection in nutrient broth tubes with nutrients and incubated for forty hours at 37 degrees Celsius, and then make decimal dilution by taking 1 ml of bacterial broth and add to tub which contain 9 ml of distal water and then use 1 ml of second bacterial diluted tub ( $10^{-2}$ ) by Pipette to culture it by spread on each Petri dishes of macconky agar which prepared (the control Petri dish and the dishes with different concentrations of turmeric extraction) and incubate it in incubator at 37degrees Celsius for 24 hour and then notice the bacterial growth on macconky agar with different concentrations of turmeric extraction.

So The stock for the *Salmonella typhi* bacteria was prepared by sterilized swab was used to scrape the bacteria from the agar plate from the bacterial bank of sciences collage / Biology department of Al-Qadisiya University and The bacteria was Injection in nutrient broth tubes with nutrients and incubated for forty hours at 37 degrees Celsius, and then make decimal dilution by taking 1 ml of bacterial broth and add to tub which contain 9 ml of distal water and then use 1 ml of second bacterial diluted tub ( $10^{-2}$ ) by Pipette to culture it by spread on each Petri dishes of Deoxycholate Citrate Agar (DC agar) which prepared (the control Petri dish and the dishes with different concentrations of turmeric extraction) and incubate it in incubator at 37 degrees Celsius for 24 hour and then notice the bacterial growth on Deoxycholate Citrate Agar with different concentrations of turmeric extraction.

**RESULT AND DISCUSSION:**

After incubation the Petri dishes in incubator at 37 degrees Celsius for 24 hour and then notice the bacterial growth of *Salmonella typhi* on Deoxycholate Citrate Agar and the growth of *Escherichia coli strain K-12* and enumerate the total bacterial account on each Petri dish which contain different concentrations of turmeric extraction.

The salmonella appear like yellow or colorless colonies on Deoxycholate Citrate Agar, and the *Escherichia coli strain K-12* appear like deep red colonies on macconky agar.

**Table (1): Explain the total bacterial account of *Escherichia coli strain K-12* and *Salmonella typhi* and its relationship with the concentrations of turmeric extraction.**

concentrations of turmeric extraction	The average of <i>Escherichia coli strain K-12</i> growth on macconky agar	The average of <i>Salmonella typhi</i> growth on DC agar
Turmeric 1 Control	$12 \times 10^{-2}$	$20 \times 10^{-2}$
Turmeric 2 50 micro liters	$10 \times 10^{-2}$	$16 \times 10^{-2}$
Turmeric 3 100 micro liters	$9 \times 10^{-2}$	$5 \times 10^{-2}$
Turmeric 4 200 micro liters	$7 \times 10^{-2}$	$1 \times 10^{-2}$
Turmeric 5 400 micro liters	$2 \times 10^{-2}$	Nil
Turmeric 6 800 micro liters	Nil	Nil
Turmeric 7 1000 micro liters	Nil	Nil



Turmeric 8 2000 micro liters	Nil	Nil
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The results of this study reveal that the antiseptic property and inhibition effect of turmeric is fairly consistent on different batches of the turmeric laced agar plates. So we notice from the table above that the both species of bacteria grow normally on the control dishes, but we see the *Escherichia coli* strain K-12 can't grow with Turmeric 6 (800 micro liters), and *Salmonella typhi* can't grow with Turmeric 5 (400 micro liters) which is determine it as the maximum concentration of turmeric extraction which inhibit the *Escherichia coli* strain K-12 growth and *Salmonella typhi* respectively.

Based on the data presented in table, it can be concluded that turmeric has antiseptic property even at low concentrations of 50 micro liters.

### RECOMINDATIONS:

- 1- In future extension of the experiment, the four major molecules of turmeric (curcumin, demethoxycurcumin, bisdemethoxycurcumin, and 2, 5-xyleneol) will be isolated and the same experiment as described above will be repeated for each of the molecules to establish the most effective molecule and its concentration against *Escherichia coli* and *Salmonella typhi*.
- 2- The turmeric can be used as a food and meat preservative because it has the antiseptic effect, anti-inflammation effect, antibacterial effect, antiviral effect and antifungal effect.
- 3- The turmeric based pharmaceutical drugs can be prescribed to patients where all other antibiotics have failed.

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