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Evaluation of Biological Activity of Curcumin Extracted From Curcuma Longa L

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Abstract: Curcuma Longa (turmeric) has a long history of use in Ayurvedic medicine as a treatment for inflammatory conditions. Curcumin, a yellow pigment present in the Indian spice turmeric (associated with curry powder), has been linked with suppression of inflammation; angiogenesis; tumorigenesis; diabetes; diseases of the cardiovascular, pulmonary, and neurological systems, of skin, and of liver; loss of bone and muscle; depression; chronic fatigue; and neuropathic pain. . Because of the multiple therapeutic activities attributed to curcumin, the present study aimed to evaluate the anti-bacterial activity of aquatic and ethanolic extracts of Curcuma Longa against different bacterial isolates. In the present study in vitro, test confirmed the antibacterial activity of turmeric extract against five different bacterial species isolated from patients with chronic urinary tract infections, including (Escherichia coli, Staphylococcus aureus, Acinetobacter baumannii, Pseudomonas aeruginosa, and Klebsiella pneumonia). The antibacterial activity was measured by agar well diffusion method. The extract of Curcuma Longa L . showed antibacterial activity against all the test bacterial isolates. The results of the antibacterial activity of the ethanolic extract of Curcuma Longa rhizomes showed that the highest inhibition zone was observed in P. aeruginosa (16 mm), K. pneumonia (15 mm) S. aureus and E.coli (10 mm) , A. baumannii (8 mm), While The results of the aquatic extract showed that the highest inhibition zone was observed in P. aeruginosa (14 mm), K. pneumonia (13 mm) S. aureus and E.coli (8 mm) , A. baumannii (no result)

Introducation

The prevalence of antibiotic resistance towards microorganisms is progressively increasing around the globe, and resistance to antibacterial drugs is among the primary causes of therapeutic failure. To cope with this situation, an effective, safe, and economical natural product or phytochemical is required. From prehistoric times, therapies obtained from plants and their phytochemicals have been essential for health maintenance. Traditional medicines have been reported to have a significant influence on the treatment of pathogens by the regulation of diverse physiological processes in countless research studies based on experimental models and preclinical findings (1) Curcumin is a bioactive curcuminoid polyphenol that is isolated from the rhizomes of Curcuma longa. Chemically, curcumin is 1,7-bis(4-hydroxy-3-methoxyphenyl)-1,6- heptadiene-3,5-dione. It is also termed as diferuloyl methane(2). Curcuma longa is commonly known as turmeric, which belongs to the Zingiberaceae family. Turmeric is found abundantly and naturally in tropical areas, on the Indian subcontinent, and in South Asia. Turmeric is dark yellow in color due to the presence of a wide variety of polyphenolic curcuminoids. Curcuminoids such as curcumin, bisdemethoxycurcumin, and dimethoxy-curcumin are found in Curcuma longa (3). Turmeric has been used in the Ayurvedic medicine system for the management of various medical disorders such as jaundice, skin infections, wounds healing, flatulence, sprains, arthritis, and stomach disturbances since ancient times. Curcumin has also been established as an anti-asthmatic, antiarthritic, anti-inflammatory, antioxidant, antimicrobial, cardio-protective, 2 and immuno-modulatory agent (4). Curcumin targets several signaling molecules while illustrating cellular activity, supporting its numerous

health benefits. Curcumin supplements have been found to have potential nephroprotective, analgesic effects and to be useful in the management of metabolic syndromes because of its antioxidant effects (5)

Although curcumin has a broad spectrum of pharmacological properties, a critical challenge towards desirable therapeutic applications is its poor bioavailability, which is due to its poor intestinal absorption, hydrophobic character, and rapid metabolism. Its systemic bioavailability is very low after oral administration. However, studies have found that a small amount of systemically available curcumin has a marked therapeutic effect. Different agents were analyzed to better determine the bioavailability of curcumin . Curcumin is considered as a potential agent for the development of novel natural products, including nanocrystals and micro-particles, to improve its stability versus the identified factors, and to manipulate bioactivities (6).

Materials and Methods

Tools and devices

The tools and devices utilized in the current study are displayed in Table 1.

Table (1): Equipment and Apparatuses.

Equipment and Apparatuses	Manufacture Company	Origin
Autoclave	Gallenkamp	UK
Disposable Petri-dishes	Meheco	China
Distillator	Kottermann	Germany
Incubator	Memert	Germany
Micropipette	Volac	England
Oven	Memaret	Germany
PH-Meter	Radiometer	Denmark
Refrigerator	Indesit	Turkey
Rotary Evaporator	Memmert	Germany
Sensitive Balance	Sartorius	Germany
Soxhlet	Sci-plus	UK
Ultrasound		Germany
Vortex	Hermal	Germany
Whattman Filter paper No. 1	GE Healthcare	China

Chemical and Biological Materials

The organic and inorganic substances utilized in the current study are displayed in Table 2

Table (2): Chemical and biological materials.

Materials	Manufacture Company	Origin
Eethanol (C2H6O)	Chem-Lab	Belgium
Mueller Hinton Agar	Hi-media	India
Nutrient agar	Oxoid	England
Nutrient broth	Oxoid	England

The media that were ready for use underwent sterilization through autoclaving at a temperature of 121ºC and a pressure of 15 pounds per square inch for a duration of 15 minutes. Additionally, the pH level was modified using either a 0.1 N solution of NaOH or HCL.

Method

Bacterial isolates

Five isolates were obtained from the Micrological Laboratory at the Biology Department/ college of science / university of Diyala including Escherichia coli, Staphylococcus aureus, Acinetobacter baumannii, Pseudomonas aeruginosa, and Klebsiella pneumonia.

Curcuma longa L. Extracts Preparation

Plant material

Curcuma longa L. rhizomes were obtained from a local herbal market in Diyala province, Iraq. These rhizomes were authenticated by a botanist at the Iraqi National Herbarium, To prepare the rhizomes for further use, any impurities were removed, then processed into a fine powder using an electric mill. Finally, the powdered were stored in airtight bottles to maintain their quality and preserve their properties.

Prepare aqueous extrac

Grounded rhizome (25g) was soaked for 3 days in D.W (500 ml) with constant stirring and filtered with the help of filter paper. The extraction procedure was repeated three times and the filtrates were pooled. The filtrate was evaporated by rotary evaporator (45 °C) producing 3.5g (14% w/w) extract.

Prepare alcholic extract

An alcoholic extract of the plant was prepared by taking 50 grams of plant powder and placing it in a thimble within a Soxhlet apparatus. A solution of 100 milliliters of 70% ethanol was used as the extracting solvent, and the extraction process lasted for five hours. The resulting extract was then concentrated using a rotary evaporator. Subsequently, the extract was dried in an electric oven at a temperature of 40°C. To ensure preservation, the dried extract was stored in opaque glass bottles in a refrigerator until it was ready for use (Fernandez-Botran et al., 2000). The extract was kept at -4°C in a tightly sealed container, away from both heat and light, to prevent the evaporation of the solvent and the entry of water.

Determination of the Anti-bacterial Activities of Plant Extract

The agar-well diffusion method was employed according to Mahmood et al. (1989), with slight modifications as follows: ♣ The surface of Muller Hinton Agar medium was inoculated with a sterilized swab of the bacterial suspension containing (1.5X10^8) cells/ml with turbidity equivalent to 0.5 McFarland, respectively,

using a densichek instrument. The plates were then left for 10 minutes to dry at room temperature (Graf et al., 2000).

- Two wells with a diameter of 5mm were created using a sterilized Cork Borer in the middle of the cultured plates.
- After sterilizing the extract with a syringe filter unit with a diameter of 0.22µm (Shneider and Ermel, 1986), 0.1ml (100µl) of the extract was added to each well using a micropipette, and the control well was filled with sterile distilled water.
- To allow the extract to diffuse through the medium, the plates were kept in the refrigerator for 30 minutes at 4°C, as described by Crespo et al. (1990). Subsequently, the plates were incubated at 37ºC for 24 hours.

- The efficacy of the extract was determined by measuring the diameter of the inhibition zone around each hole

Results and Discussion:

preparation of aquatic and alcoholic extracts of Curcuma longa L. Prepare alcoholic and aqueous extracts from dried Curcuma longaL. rhizomes using a Soxhlet apparatus for three hours at 50°C. and by maceration respectively , The result of the extraction is a yellow color (aqueous extract) and dark black orange colour(alcoholic extract). Store the extracts in opaque glass bottles in the refrigerator until ready for use.

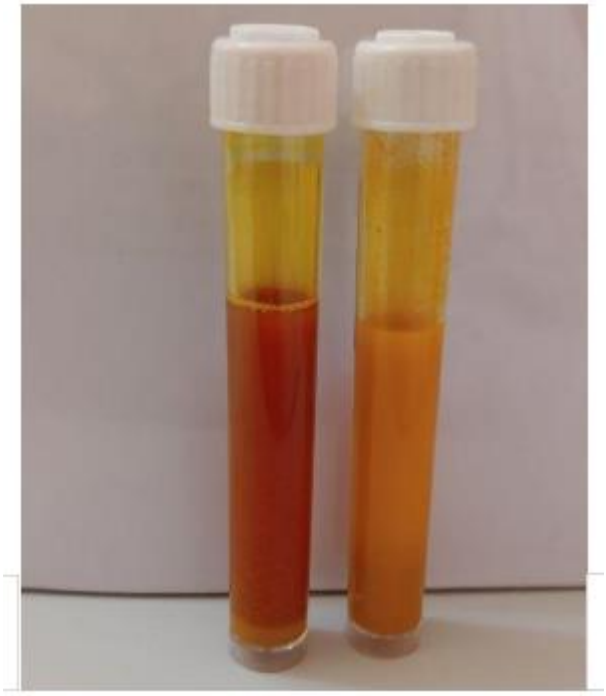

Figure 4.1 :Aquatic and ethanolic extracts of Curcuma longaL.

Study the Effect of alcoholic and aquatic extract of Curcuma Longa

rhizomes on some Bacterial spp.

The results of the antibacterial activity of the ethanolic extract of Curcuma Longa rhizomes showed that the highest inhibition zone was observed in P. aeruginosa (16 mm), K. pneumonia (15 mm) S. aureus and E.coli (10 mm) , A. baumannii (8 mm), While The results of the aquatic extract showed that the highest inhibition zone was observed in P. aeruginosa (14 mm), K. pneumonia (13 mm) S. aureus and E.coli (18 mm) , A. baumannii (no result), Table(1) figures (2) .

The natural dye powder was active against E. coli and vibrio cholera. Chandrana, et al., (2005) who studied antimicrobial activity of turmeric reported that it was effective against E. coli, B. subtilis and S. aureus and suggested that the activity is due to the presence of curcuminoid, a phenolic compound. The antimicrobial property of turmeric has been attributed to the presence of essential oil, an alkaloid, curcumin and other curcuminoids, turmeric oil, turmerol and veleric acid (Cikrikci, et al., 2008).

Table (1): Inhibition Effect of crude ethanolic and aquatic extracts of the rhizomes of Curcuma longa L.on Some Bacterial Species at 37ºC for 24 Hrs.

Type of Bacteria	Inhibition zone (mm) for Ethanolic Extract	Inhibition zone (mm) for aquatic Extract
Escherichia coli	10	8
Staphylococcus aureus	10	-
Acinetobacter baumannii	8	8

Pseudomonas aeruginosa	16	14
Klebsiella pneumonia	15	13

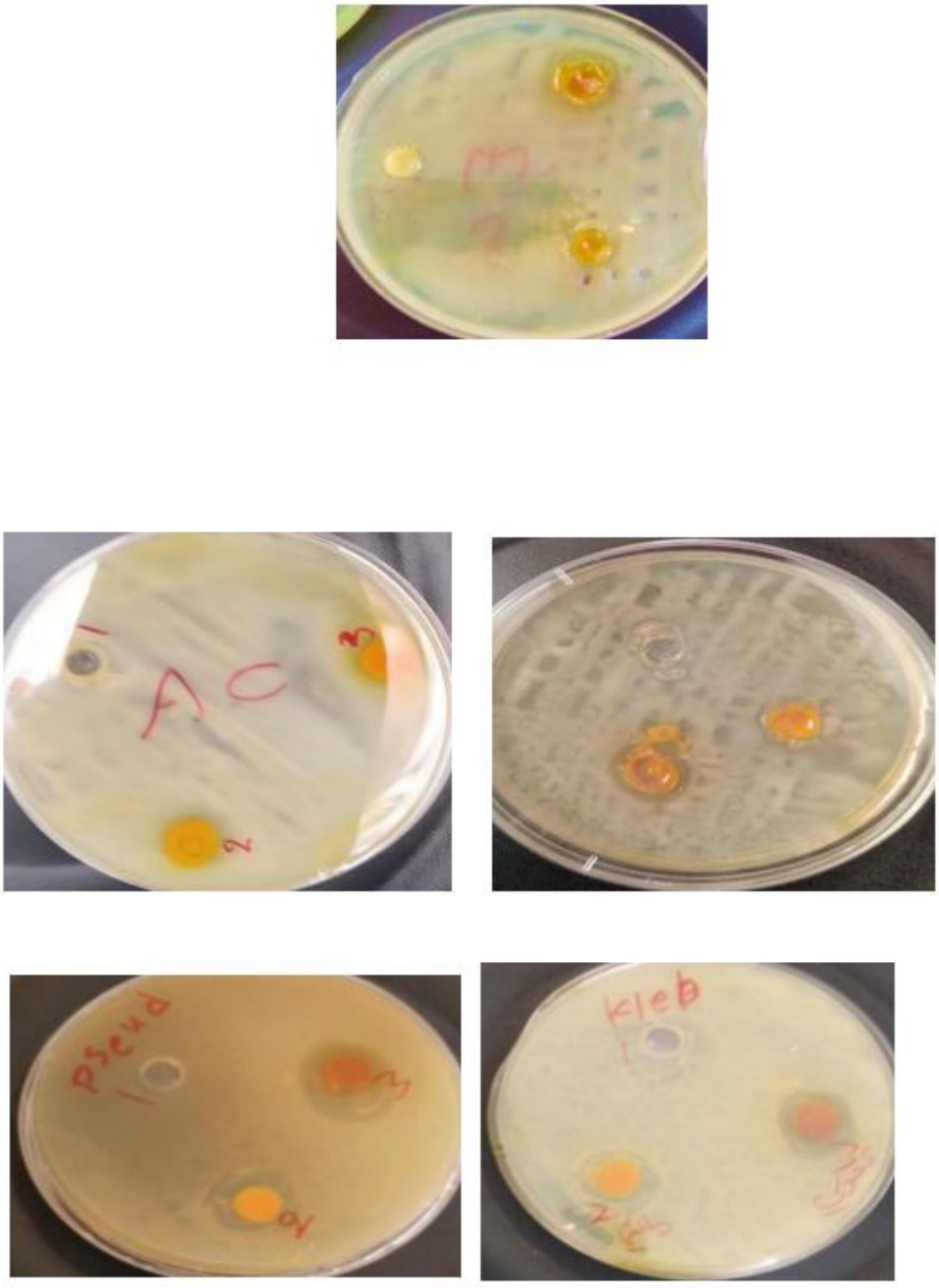

Figure (2)Antibacterial inhibitiobn zones of alcoholic and aquatic extract of Curcuma Longa rhizomes on some Bacterial spp

Conclusions

The Ethanolic and aquatic extracts of Curcuma longa L rhizomes exhibited the strongest antibacterial efficacy when tested against P.aeruginosa bacteria. And exhibited the lowest antibacterial efficacy when tested against A. boumannii and S. aureus

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