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RESEARCH PAPER

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Evaluation of Antimicrobial Activity of Methanolic Extract of Turmeric (*Curcuma longa*) and Pepper (*Piper nigrum*)

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ABSTRACT

Plant extracts are used in food processing technologies as natural additives. The aim of our study is to evaluate the antimicrobial activity of the two spices "Pepper (Piper nigrum)"and "Turmeric (Curcuma longa)", which can be used in different herbal medicines and therapies. Both spices were assayed for the in vitro antimicrobial property against gram positive, Bacillussubtilis and Staphylococcus aureus and gram-negative, Escherichia coli and Pseudomonas aeroginosa. The antimicrobial activities of the selected spices were carried out by using agar well diffusion technique.The results showed more antimicrobial effect of methanolic extract of turmeric against gram positive as compared to gram negative, Pseudomonas aeroginosa gave more effect as compared to Escherichia coli. While, methanolic extract of black pepper against Bacillussubtilis gave more effectible zone of inhibition as compared to Staphylococcus aureus and gram negative bacteria.

Keywords: Antimicrobial activity, Spices, Methanolic extract, Pepper and Turmeric.

INTRODUCTION

Spices were some of the most valuable items of trade in the ancient and medieval world. Herbalist and folk practitioners have used plant remedies for centuries, but only recently

have scientist begun to study the powers of common herbs and spices. Spices are rich in phytonutrients and other active ingredients that protect against disease and promote healing. In worldwide studies, spices have been linked to the prevention and treatment of chronic conditions such as heart disease, cancer, Type II diabetes, and Alzheimer's. Unlike pharmaceutical drugs, spices can be used long- term without concern for side effects. In short, spices are among the great gifts nature has bestowed upon us (Articles/The healing power of spices). Many naturally occurring compounds found in edible and medicinal plants, herbs and spices have been shown to possess antimicrobial function and could serve as a source of antimicrobial agents against food pathogens (Lai P., and Roy J., 2004). The spices have a unique aroma and flavor which are derived from compounds known as phytochemicals or secondary metabolites (Avato P, et al., 2000).The phytochemicals are antimicrobial substances present in the spices which are capable of attracting benefits and repel harmful organisms; they also serve as photoprotectants and responds to environmental changes. Numerous classes of phytochemicals including the isoflavones, anthocyanins and flavonoids are found associated with the spices (Chang HM, and Butt PPH., 1988). Black pepper (*Piper nigrum*) bears the royal pedigree, "King of Spices." In early times, it was more valuable than gold. Only the wealthy could afford it; dowries were endowed with it and many bribes for special favors were paid with it.The sharp flavor and healing prowess come from piperine and other volatile oils in the pepper. It's the piperine that zaps the taste buds, often triggering a sneeze when it hits the nerve endings inside your nose. Studies are finding that piperine can be effective in treating a vast array of conditions, including cancer, digestive disorders, heart disease, high blood pressure, loss of hearing, quitting smoking, and more. The Latin name for turmeric is "Curcuma Longa", which comes from the Arabic name for the plant, "Kurkum" (Stavric B., 1994). It is called "Jiang Huang" in Chinese, and "Haldi" ("Yellow") in Hindi, the most common name in India (Blumenthal M., 1998). The healing properties of turmeric lie in the golden fingerlike stalk, or rhizome, the same part that is used to flavor, color, and preserve food. Turmeric is commonly found in Indian curries, giving the food a golden orange color (Stavric B., 1994). Turmeric (Curcuma longa) has been used for 4,000 years to treat a variety of conditions. Studies show that turmeric may help fight infections and some cancers, reduce inflammation, and treat digestive problems, and it has gotten a lot of press lately (Turmeric pdf). Turmeric owes its preventive and curative characteristics to its active ingredient curcumin, a compound so diverse and powerfully rich in antioxidant and anti-inflammatory actions that thousands of studies have shown that it protects and improves the health of virtually every organ in the body.

The aim of this study is to evaluate the antimicrobial activity of methanolic extract of turmeric and black pepper.

MATERIALS AND METHODS

Collection of Samples

Two plants were used Pepper (*piper nigrum*) and Turmeric (*curcuma longa*) were purchased from a local market in Karachi, Pakistan and evaluated for their microbial activity against 4 organisms (obtained from the culture bank, department of Microbiology, Jinnah University). The plants were brought to the laboratory and thoroughly washed in distilled water and dried in an oven at 50°C for 48 hours followed by a grinding into a fine powder. The powdered material was stored in an air tight jar in refrigerator at 4°C.

Methanolic extract preparation

Plant materials were finely grinded to powder by using a blender. Twenty-five grams of each plant material was dissolved in enough methanols to make 100ml of methanolic extract (25% w/v). The mixture was kept undisturbed at room temperature for 24 hours in a sterile flask covered with aluminium foil to avoid evaporation and subjected to filtration through Whatman No. 1 filter paper. After filtration, the extract was evaporated in water bath until 25ml extract was left in the flask. Extract thus obtained were immediately evaluated for antibacterial activities.

Antimicrobial Activity of Plant Extract against bacterial isolates by Agar well diffusion method

Take the respective organism culture and make lawn on MHA and NA with the help of a borer make a well of 8mm size in the center of both the agar plates. Mark the plates with a single organism. Pour 100ul of both the extract in the respective agar plate with the help of a juster. The plates were left at room temperature for 10 minutes allowing the diffusion of the extract into the agar. Incubate the plates for 24 hours at 37c.Observe the colonies on the plate. Measure the zone of inhibition around the well with the help of a ruler. Record the results.

RESULTS AND DISCUSSION

The effects of the methanolic extract of plants on the organisms were summarized in Table 1, 2, 3 and 4. The results showed more antimicrobial effect of methanolic extract of turmeric against gram positive bacteria as compared to gram negative; *Pseudomonas aeroginosa* gave more effect as compared to *Escherichia coli* with a zone of 26mm. While, methanolic extract of black pepper against *Bacillussubtilis* gave more effectible zone of inhibition of 40mm in both MHA and NA as compared to *Staphylococcus aureus* 25mm in MHA and 15mm in NA. Gram negative bacteria i.e. *Escherichia coli* in MHA gave no zone of inhibition and *Pseudomonas aeroginosa* in NA and MHA gave 35-40mm zone of inhibition. There has been increasing consumer demand for foods free or with low, if any, added synthetic preservatives because synthetic preservatives could be toxic to humans (C. Bedin, et al., 1999).The data supports the hypothesis that some spices have an inhibitory effect on the growth borne pathogens (Shamsuddeen U., et al., 2009).

Medicinal plants, since dawn of civilization, have been used in virtually all cultures as a source of medicine. There is growing interest in medicinal plants as a re-emerging health aid has due to the rising costs of prescription drugs in the maintenance of personal health and well-being, and the bioprospecting of new plant-derived drugs (Hoareau, et al., 1999).

Wide research shows that turmeric, taken as supplemental curcumin, is as effective and, in some cases, even more effective than pharmaceutical drugs — without their side effects. Recently, my colleagues and I compared curcumin with anti-inflammatory and pain-killing medications. And we compared curcumin with cancer drugs, testing those agents for their effectiveness in reducing inflammation and stopping the proliferation of cancer cells. Curcumin proved to be more effective at reducing inflammation than over-the-counter aspirin and ibuprofen, and as effective as the more powerful prescription drug Celebrex. It also proved as effective in thwarting breast cancer cells as tamoxifen, a drug widely used to stop the spread or recurrence of breast cancer. These results are nothing less than astounding (Article/ Healing spices).Although it is often claimed that exotic spices were sought as valuable food preservatives, this is not correct. Thus, simple pickling with common-place vinegar, garlic and mustard can preserve and flavor food almost as well as

dehydrating and salting can. Honey and strong sugar solutions can also be used as food preservatives. There is little evidence that pepper, cloves, nutmegs, ginger and other expensive spices were used as alternatives to garlic, etc. to preserve food or to delay the spoilage of cooked dishes. Their use in their countries of origin is not related to spices serving as an alternative torefrigeration, since they are usually added to fresh foods as flavors. In particular, they add zest to a bland diet based on rice and other high-carbohydrate vegetable staples. Indeed, the concentrations of spices that would be needed to significantly retard food spoilage by microorganisms would result in an overwhelming flavor that may be worse than that of the decaying food (Al-Zubaydi, et al., 2009).Medicinal plants play an important role for the management of different microbial infections because overmedication and long-term side effects of synthetic drugs have assumed alarming range (Mahmud, et al., 2009) (Cowan, and M. M., 1999).

Table1. Diameter of zone of inhibition on MHA by Turmeric.

Organisms	Zone of inhibition	Results
Escherichia coli	30mm	Sensitive
Pseudomonas aeroginosa	25mm	Sensitive
Staphylococcus aureus	15mm	Sensitive
Bacillus subtilis	30mm	Sensitive

Table 2. Diameter of zone of inhibition on NA by Turmeric.

Organisms	Zone of inhibition	Results
Escherichia coli	22mm	Sensitive
Pseudomonas aeroginosa	26mm	Sensitive
Staphylococcus aureus	35mm	Sensitive
Bacillus subtilis	23mm	Sensitive

Table 3. Diameter of zone of inhibition on MHA by Black Pepper.

Organisms	Zone of inhibition	Results
Escherichia coli	No zone	Resistant
Pseudomonas aeroginosa	35mm	Sensitive
Staphylococcus aureus	25mm	Sensitive
Bacillus subtilis	40mm	Sensitive

Table 4. Diameter of zone of inhibition on NA by Black Pepper.

Organisms	Zone of inhibition	Results
Escherichia coli	No zone	Resistant
Pseudomonas aeroginosa	40mm	Sensitive
Staphylococcus aureus	15mm	Sensitive
Bacillus subtilis	40mm	Sensitive

As we talk about the standard results of gram positive bacteria like *Staphylococcus aureus* gave 28mm zone of inhibition in streptomycin and 38mm in cefamezin antibiotics. But in our results *Staphylococcus aureus* is sensitive to a certain point by turmeric and black pepper. *Bacillus subtilis* 25 mm in cefamezin, 10mm in chloramphenicol and 29mm in streptomycin, while in this study *Bacillus subtilis* gave more effective zone of 40mm which indicates that the organism is sensitive to the both spices (Adwan G, and Mhanna M., 2008) (Cronin, et al., 2008).

The standard result of gram negative bacteria, *Escherichia coli* gave 35mm zone of cefamezin and 26mm of streptomycin, in our observational study, *Escherichia coli*gave no zone of inhibition of black pepper extract but in turmeric it gave effective zones. Pseudomonas aeroginosa gave 25mm on streptomycin, but it gave more effective zone of black pepper as compared to turmeric (Cox, et al., 1998).

We used methanol for extraction instead of water because the methanol has the ability to dissolve the volatile oils or compounds that are present in the spices and water gave no affectivity because of the undissolvement of the spices volatile oils (Burt, and S.,2004).

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