Eco. Env. & Cons. 30 (January Suppl. Issue) : 2024; pp. (S179-S181)

ISSN 0971–765X

DOI No.: http://doi.org/10.53550/EEC.2024.v30i01s.034

Comparative Study of Antimicrobial Effect of Ginger, Garlic and Turmeric on Penicillin Resistant Bacteria isolated from Cattle Farm Soil

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(Received 2 June, 2023; Accepted 20 August, 2023)

ABSTRACT

Antibiotics are medicines used to treat or prevent infections caused by bacteria. They work by inhibiting the growth of or destroying the bacteria. Antibiotic resistant bacteria are rapidly increasing, causing a problem in the treatment of disease caused by them. The present study focused on the effect of local spices on the antibiotic resistant bacterial isolates obtained from the soil sample of the local cattle farms. The use of antibiotics in the feed of farm animals has given rise to antibiotic resistance of microorganisms in natural habitat. Soil samples were collected from different cattle fields of the Palghar district and isolation of resistant bacteria against penicillin was done using Nutrient and Mueller-Hinton agar. The isolates obtained were subject to varying increased antibiotic concentration of penicillin and found to be resistant for concentration up to 25 µg/ml. The effect of natural herbs/spices was studied. While the soil sample showed the presence of an increased amount of penicillin resistant bacteria, the effect of aqueous and methanolic extracts of spices like ginger, garlic and turmeric were studied against the penicillin resistant isolates.

***Key words**: Antibiotic resistance, Penicillin, Local spices, Cattle farms, Palghar*

Introduction

Antibiotics are medicines used to treat or prevent infections caused by bacteria. They work by inhibiting the growth of or destroying the bacteria. They do this in various ways, such as destroying the bacterial cell wall or inhibiting the generation of energy from glucose within the bacterial cell. Antibiotic resistant bacteria are rapidly increasing, causing a problem in the treatment of disease caused by them. Antibiotic resistance happens when bacteria are genetically altered and become resistant to the antibiotic used to treat the infection they cause. Drug resistant bacteria reach humans through food, the environment (water, soil, air) or by direct human-animal contact, leading to a growing number of infections as antibiotics used to treat them become less effective and also cause chronic problems. Antibiotics are given to food producing animals and crops. Cattles can also contribute to the antibiotic resistance organism as antibiotics such as penicillin, tetracycline, tilmicosin etc are fed to cattles to prevent and cure their diseases. Animals develop drug resistant bacteria in their gut. As these cattle are free for grazing, they move to the cattle farm areas, when the cattle excrete or urinate, which may give rise to the increase in antibiotic resistance in various bacteria present in soil. Hence the soil sample was collected from the local cattle farms in the Palghar region. Penicillin are a group of antibiotics originally obtained from Peni-

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cillium molds, principally *P. chrysogenum* and *P.ruben.* Penicillin was among the medications to be effective against many bacterial infections caused by Staphylococci and Streptococci. They are still widely used today for different bacterial infections though many types of bacteria have developed resistance following extensive use. Thus, in light of the evidence of rapid global spread of resistant clinical isolates, the need to find new antimicrobial agents is of paramount importance. Traditionally used medicinal plants produce a variety of compounds of known therapeutic properties. There has also been a considerable effort to discover plant derived antibacterial activity against resistant bacteria. Herbs and spices have been used as a medicine to treat various diseases since ancient times. The presence of secondary metabolites in plants such as flavonoids, tannins, saponins, phenolic compounds, aromatic compounds etc., has antibacterial activity. As the world enters into the modern days, the usage of these herbs is declining.

This study focuses on investigating the antimicrobial activity of methanolic and aqueous extract of herbs like garlic, ginger, turmeric on Penicillin resistant bacteria isolated from cattle farm soil. All these spices and herbs have anti-microbial, anti-inflammatory, anti-oxidant, antifungal and antiseptic properties which are said to be effective against these resistant bacteria. So the hypothesis was made to study the effect of local spices on these organisms.

Materials and Methods

Screening and isolation of penicillin resistant bacteria

For Isolation of bacteria, soil samples were collected from the cattle farm of Mahagaon, Boisar and Gothanpura areas of the Palghar region in Maharashtra. Samples were collected and brought to the laboratory in plastic bags. Samples collected were serially diluted and isolated by the surface spread plate method. The sample dilutions were also spread on the selective media, i.e. Macconkey (selective media for Gram negative bacterial species) and Mannitol Salt Agar (for isolation of *S. aureus)* agar plate. Incubation was done at 37 ^{o}C for 24 hours.

Screening of penicillin resistant bacteria was done by agar cup method. Subsequently increasing concentrations of penicillin from 1µg, 5µg, 10µg, 15µg, 20µg and 25µg was used in Mueller Hinton Agar. After the incubation period, zones of exhibition were observed indicating the presence of penicillin resistant bacteria in the soil samples. From this initial screening, five random isolates from the highest concentration plate were studied further.

Morphological study

Morphological study was done by Gram staining of isolated resistant bacteria and colony characteristics for each were noted. All the five random isolates were found to be Gram negative in nature.

Crude extraction of herbs

Aqueous and Methanolic extracts of spices (i.e ginger, garlic and turmeric) were prepared. The spices were obtained from the local markets of Palghar. Use of mortar and pestle and filter method was used to obtain the crude extracts.

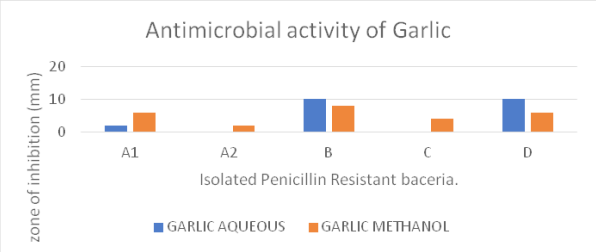
D) Agar cup method was used to study the effect of the crude extracts on the bacterial isolates obtained on 25 µg/ml Mueller Hinton agar plates. The isolates were swabbed separately on MH agar plates and aqueous and methanolic extracts were added to individual wells. Methanol was used as a comparative control in one of the wells. On incubation at 37 ^{0}C for 24 hours, plates were observed the next day for zones of inhibition.

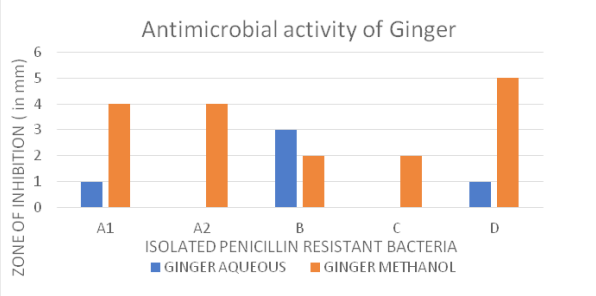
Results

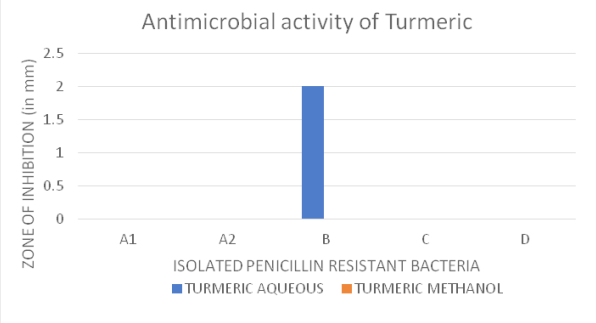
The isolation of antibiotic resistance bacteria was carried out by using cattle farm soil as a sample on nutrient agar and Mueller Hinton agar plate. Five different organisms were isolated and named as A_1,A_2,B,C and D which were all Gram negative in nature. Further these isolated colonies were used as test samples to perform antibiotic sensitivity tests against methanolic and aqueous extraction of herbs with garlic, ginger and turmeric. Antibiotic sensitivity test was performed using agar well diffusion method. Zone of inhibition of penicillin resistant bacteria was observed against methanolic and aqueous herbs extract. Comparative study showed that aqueous garlic extract showed more susceptibility to isolated culture sample D resistance bacteria, where zone of inhibition by aqueous extract was 10mm and by methanolic extract was 6 mm (Fig. 1). In case of other isolates (A1, A2, B and C) methanolic extracts and aqueous extracts of ginger, garlic and turmeric showed inhibition zones as mentioned tabu-

Table 1. Zones of inhibition observed against the test isolates from cattle farm soil.

Extract	Zone of inhibition					
	A_1	A_2	B	C	D	
Garlic	aqueous	2mm	No inhibition	10mm	2 mm	10 mm
	methanol	6mm	2mm	8mm	4mm	6mm
Ginger	aqueous	1mm	No inhibition	3mm	No inhibition	1mm
	methanol	4mm	4mm	2mm	2mm	5mm
Turmeric	aqueous	No inhibition	No inhibition	2mm	No inhibition	1 mm
	methanol	1-2 mm	1-2 mm	1-2 mm	1-2 mm	No inhibition







lated as in Table 1.

Conclusion

Antibiotic resistance is accelerated by the misuse and overuse of antibiotics. In this study, penicillin resistant bacteria from cattle farm soil samples have shown resistance against penicillin (25 µg/ml). The herbs which are used in present study showed promising antibacterial activity against the resistant bacterial isolates. The aqueous and methanolic extracts of garlic, ginger and turmeric show varying degrees of inhibition but prove to be effective against the resistant strains as they are all Gram negative in nature. These herbs and spices are a daily incorporation in Indian cuisines. Hence the study suggests that these plants can be used against common infections which are caused due to their presence in natural environments and that can be knowingly or unknowingly consumed by humans through agricultural products. Ailments that had penicillin and other antibiotics as a common treatment are becoming more and more difficult to treat due to the indiscriminate use of the antibiotics in animal feed and agricultural land. Use of easily available herbs and spices can be promising home remedies with less side effects compared to the chemically synthesized drug. These also prove to be cost-effective treatments accessible to even the people at the lowest economical strata.

Further biochemical tests and research can be done to identify the resistant organisms obtained as isolates and their strain. Further exploration can be done by finding out the phytochemical constituents of the natural remedies and extracting them to render as pure medicines.

This study also opens up new opportunities to explore different plant extracts that are easily available. As per the studies, allopathic drugs are becoming more resistant. Hence propagation of Ayurveda can now be scientifically supported through such research data.

References

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