



Studies On The Antibacterial Activities Of Mushroom

Lakshmi Priya .J

Microbiology department ,Vivekanandha college of Arts and Science for women,
Tiruchengode/ Periyar University, India

Abstract:

Mushrooms have been cultivated worldwide for commercial purposes. However, little research has been done to ascertain the antibacterial properties of indigenous edible mushrooms. Four species of cultivated and wild varieties were taken for the present study. Among the cultivated species Ganoderma shows inhibitory activity against Bacillus sp., Staphylococcus aureus., Escherichia coli, Klebsiella sp.,and all other cultivated species like Pleurotus ostreatus, Lentinus edodes, Agaricus bisporus shown no inhibitory activity until 48 hours of incubation . In the wild species strain 1 shows activity against Bacillus sp., and Staphylococcus aureus and other strains didn't show any such activity. The results obtained in this study suggest that the cultivated varieties doesn't show any antibacterial activity due to alteration of genes and loss of microbial activity by continuous sub culturing from mother spawn and the wild varieties shows some activity confirms that they posses such character in a given natural population.

Key words: Antibacterial, Bacteria, cultivated strains , mushroom, wild strains.

1.Introduction

Mushroom is a macro fungus with a distinctive fruiting body that is large enough to be seen by the naked eyes. It includes both edible and non edible species. Some mushrooms serve as food because of their nutrient contents while some have been used extensively in traditional medicine (Stamets, 2000). Of the hundreds of known mushroom varieties, several have been studied for their ability to enhance the human immune system and fight infections. They are known to possess all essential aminoacids, minerals, vitamins recommended as a food especially to the under developed countries where the protein malnutrition has become a real threat to human health. In fact , mushrooms have a definite primary and secondary physiological effects on the human immune modulating, antibacterial, antiviral, antitumor, antiparasitic ,cardiovascular effects. Intake of some varieties can reduce total cholesterol level and also affect glyceimic levels and inflammatory conditions. In the 20th century, a study in Japan found three kinds of antibacterial substances in Shiitake mushrooms that were effective against *Streptococcus* sp., *Actinomyces* sp., *Lactibacillus* sp., *prevotella* sp., *Porphyromonas* sp., of oral origin. In the present study a few strains of mushrooms were tested for their antibacterial activities against known bacterial cultures.

2.Materials And Methods

2.1.Sources Of Mushrooms

Some quantities of four different mushroom species such as *Ganoderma*, *Pleurotus ostreatus*, *Lentinus edodes*, *Agaricus bisporus* were purchased from local markets and four different wild strains were collected from field sides.

2.2.Extraction Of Mushroom

Fresh mushrooms were thoroughly washed with clean water and alcohol for surface sterilization cut into pieces and air-dried. Each of the different air-dried mushroom samples were respectively collected and boiled with distilled water and the extracts were separated (crude extract) stored (4°C) in a clean sterile container for further use.

2.3.Glassware

Glassware used in present work were thoroughly washed and dried. They were done sterilized at 180c for one hour in a hot air oven.

2.4.Sources Of Microorganisms

Pure culture of *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus sp.*, were obtained from bacteriology laboratory and was sub-cultured on nutrient agar to ensure the purity of the culture and the pure isolate identified and used for present study.

3.Used Media

3.1.Nutrient Agar

- Ingredients Gms / Litre

Peptic digest of animal tissue 5.000

Sodium chloride 5.000

Beef extract 1.500

Yeast extract 1.500

Agar 15.000

Final pH (at 25°C) 7.4±0.2

3.2.Mannitol Salt Agar

- Ingredients Gms / Litre

Proteose peptone 10.000

Beef extract 1.000

Sodium chloride 75.000

D-Mannitol 10.000

Phenol red 0.025

Agar 15.000

Final pH (at 25°C) 7.4±0.2

3.3.Endo Agar

- Ingredients Gms / Litre

Peptic digest of animal tissue 10.000

Lactose 10.000

Dipotassium phosphate 3.500

Sodium sulphite 2.500

Basic fuchsin 0.500

Agar 15.000

Final pH (at 25°C) 7.5±0.2

3.4. Centrimide Agar

- Ingredients Gms / Litre

Pancreatic digest of gelatin 20.000

Magnesium chloride 1.400

Potassium sulphate 10.000

Cetrimide 0.300

Agar 15.000

Final pH (at 25°C) 7.2±0.2

3.5. Cystine Lactose Electrolyte Deficient Agar (Cled)

- Ingredients Gms / Litre

Peptic digest of animal tissue 4.000

Casein enzymic hydrolysate 4.000

Beef extract 3.000

Lactose 10.000

L-Cystine 0.128

Agar 15.000

Final pH (at 25°C) 7.3±0.2

4. Methodology

Medically important bacteria such as *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus sp.*, were swabbed in the selective media under sterile conditions.

Sterile disc was dipped in the crude extract of mushrooms was placed in the media containing bacteria. After over night incubation the plates were observed for the appearance of zones. The results were noted.

5.Result And Discussion

5.1.Experiment No : 1

In the analysis the Ganoderma strain were tested against five bacterial species. It shows medium inhibitory action against Bacillus sp., on Nutrient agar and Staphylococcus aureus on Mannitol salt agar during the incubation period of 24 hours and low inhibitory action against Escherichia coli on Endo agar and Klebsiella pneumoniae on CLED during the incubation period of 24 hours respectively. But against Pseudomonas aeruginosa on Cetrimide agar no zone was observed, it shows that the organism is resistant against the strain(Table 1)

5.2.Experiment No : 2

In the analysis the Wild strain 1 was tested against five bacterial species. It shows medium inhibitory action against Bacillus sp., on Nutrient agar and Staphylococcus aureus on Mannitol salt agar during the incubation period of 24 hours respectively. But against Escherichia coli on Endo agar, Klebsiella pneumoniae on CLED and Pseudomonas aeruginosa on Cetrimide agar no zone was observed, it shows that the organism is resistant against the strain(Table 2)

5.3.Experiment No : 3

In the analysis the Wild strain(2,3,4) were tested against five bacterial species such as Bacillus sp., on Nutrient agar shows medium inhibitory activity. But against Staphylococcus aureus on Mannitol salt agar, Escherichia coli on Endo agar and Klebsiella pneumoniae on CLED and Pseudomonas aeruginosa on Cetrimide agar no zone were observed until 48 hours of incubation. it shows these species were resistant against the strain(Table 3)

5.4.Experiment No : 4

In the analysis the Lentinus edodes strain were tested against five bacterial species such as Bacillus sp., on Nutrient agar, Staphylococcus aureus on Mannitol salt agar, Escherichia coli on Endo agar and Klebsiella pneumoniae on CLED and Pseudomonas aeruginosa on Cetrimide agar. No zone were observed until 48 hours of incubation. it shows these species were resistant against the strain(Table 4)

5.5.Experiment No : 5

In the analysis the *Agaricus biporus* strain were tested against five bacterial species such as *Bacillus sp.*, on Nutrient agar, *Staphylococcus aureus* on Mannitol salt agar , *Escherichia coli* on Endo agar and *Klebsiella pneumoniae* on CLED and *Pseudomonas aeruginosa* on Cetrimide agar . No zone were observed until 48 hours of incubation. it shows these species were resistant against the strain(Table 5)

5.6.Experiment No : 6

In the analysis the *Pleurotus ostreatus* strain were tested against five bacterial species such as *Bacillus sp.*, on Nutrient agar, *Staphylococcus aureus* on Mannitol salt agar , *Escherichia coli* on Endo agar and *Klebsiella pneumoniae* on CLED and *Pseudomonas aeruginosa* on Cetrimide agar . No zone were observed until 48 hours of incubation. it shows these species were resistant against the strain(Table 6)

| Organisms | Media | Incubation Period | Inhibitory Activity |
|------------------------|--------------|--------------------------|----------------------------|
| Bacillus sp | NA | 24 hours | ++ |
| Staphylococcus aureus | MSA | 24 hours | ++ |
| Escherichia coli | ENDO | 24 hours | + |
| Pseudomonas aeruginosa | CETRIMIDE | 24 hours | - |
| Klebsiella pneumoniae | CLED | 24 hours | + |

Table 1 : Ganoderma

++ - *Medium inhibition*

+ - *Low inhibition*

- - *No inhibition*

| Organisms | Media | Incubation Period | Inhibitory Activity |
|------------------------|-----------|-------------------|---------------------|
| Bacillus sp | NA | 24 hours | ++ |
| Staphylococcus aureus | MSA | 24 hours | ++ |
| Escherichia coli | ENDO | 24 hours | - |
| Pseudomonas aeruginosa | CETRIMIDE | 24 hours | - |
| Klebsiella pneumoniae | CLED | 24 hours | - |

TABLE 2: Wild Strain 1

++ - Medium inhibition

- - No inhibition

| Organisms | Media | Incubation Period | Inhibitory Activity Of 2,3,4 |
|------------------------|-----------|-------------------|------------------------------|
| Bacillus sp | NA | 24 hours | + |
| Staphylococcus aureus | MSA | 24 hours | - |
| Escherichia coli | ENDO | 24 hours | - |
| Pseudomonas aeruginosa | CETRIMIDE | 24 hours | - |
| Klebsiella pneumoniae | CLED | 24 hours | - |

Table 3: Wild Strain 2,3,4

+ - *Low Inhibition*

- - *No Inhibition*

| Organisms | Media | Incubation Period | Inhibitory Activity |
|------------------------|--------------|--------------------------|----------------------------|
| Bacillus sp | NA | 24 hours | – |
| Staphylococcus aureus | MSA | 24 hours | – |
| Escherichia coli | ENDO | 24 hours | – |
| Pseudomonas aeruginosa | CETRIMIDE | 24 hours | – |
| Klebsiella pneumoniae | CLED | 24 hours | – |

Table 4: Lentinus Edodes

– - No Inhibition

| Organisms | Media | Incubation Period | Inhibitory Activity |
|------------------------|--------------|--------------------------|----------------------------|
| Bacillus sp | NA | 24 hours | – |
| Staphylococcus aureus | MSA | 24 hours | – |
| Escherichia coli | ENDO | 24 hours | – |
| Pseudomonas aeruginosa | CETRIMIDE | 24 hours | – |
| Klebsiella pneumoniae | CLED | 24 hours | – |

Table 5: Agaricus Bisporus

– - No inhibition

| Organisms | Media | Incubation Period | Inhibitory Activity |
|------------------------|-----------|-------------------|---------------------|
| Bacillus sp | NA | 24 hours | – |
| Staphylococcus aureus | MSA | 24 hours | – |
| Escherichia coli | ENDO | 24 hours | – |
| Pseudomonas aeruginosa | CETRIMIDE | 24 hours | – |
| Klebsiella pneumoniae | CLED | 24 hours | – |

*Table 6: Pleurotus Ostreat
– - No Inhibition*

6. Conclusion

On the whole, the mushrooms studied were found to be a good source of protein, fibre and minerals. For the present study four species of cultivated and wild varieties were taken to assess their antibacterial activities. Among which the cultivated species *Ganoderma* shows inhibitory activity against *Bacillus* sp., *Staphylococcus aureus*., *Escherichia coli*, *Klebsiella* sp., and all other cultivated species like *Pleurotus ostreatus*, *Lentinus edodes*, *Agaricus bisporus* shown no inhibitory activity until 48 hours of incubation . In the wild species strain 1 shows activity against *Bacillus* sp., and *Staphylococcus aureus* and other strains didn't show any such activity. The results obtained in this study suggest that the cultivated varieties doesn't show any antibacterial activity due to alteration of genes and loss of microbial activity by continuous sub culturing from mother spawn and the wild varieties shows some activity confirms that they possess such character in a given natural population. Boiling or cooking did not dilute or reduce the medicinal properties. Hence, it is necessary to identify the biological and pharmacological potential of mushrooms especially edible mushrooms that are collected indigenously and cultivated locally or sold in local and international market. The production and marketing of mushrooms and their products is vital for an economic importance. Therefore, it is also necessary to

intensify research in identifying and isolating different varieties of mushrooms having nutraceutical and medicinal properties and to commercialize their production and marketing, which will boost the food industry and create employment especially in villages.

7. Acknowledgement

The gracious deeds of the ALMIGHTY are beyond the words of compare". With a warm heart, I thank God for his blessing showered upon me to complete my project work.

I express my whole hearted thanks to Dr. V. SRINIVASAN, M.Sc., Ph.D., Lecturer, Department of Microbiology, Vivekanandha College of Arts and Science for women for his keen interest, meticulous support, valuable, enthusiastic guidance and brilliant suggestions throughout my project work.

I owe my true and profound sense of gratitude to thank Dr. B.T. SURESH KUMAR M.Sc., Head, Department of Microbiology, for his valuable suggestion and successful completion of my project.

8.Reference

1. Anke(1989) (In paul Stamens) the extracellular secretion by the mycelium are exquisitely designed to combat a wide range of bacteria and other micro organisms
2. Beltran – Garcia, M.J., Escarron- Espinosa M.V. and Ogura T. (1997). Volatile compounds secreted by the Oyster Mushroom (*Pleurotus ostreatus*) and the antibacterial activities.J. Agri food chem.,(45): 4049-4052
3. Gunde-Cimerman.N., and Cimerman,A., (1995). *Pleurotus* fruiting bodies contain the inhibitor of 3- hydroxyl, 3- methelglutaryl, Coenzyme A reductase- lovastain. Exp.Mycol.,(19), 1-6.
4. Hirasama M, T. Neta, N. Shuji, T.K. Fukushima and K.Takada(1999) Three kinds of antibacterial substances from *Lentinus edodes* (Berk) sing (*Shiitake*, an edible mushrooms) International Journal of antimicrobial agents Feb: 11 (2) : 151-157
5. Hsu, Hong-Yen et al (1986). *Oriental Materia Medica* , a concise guide .Long Beach: Oriental healing arts institute
6. Jonathan, S. G. and Fasidi, I. O. 2003. Antimicrobial activities of two Nigerian edible macrofungi- *Lycoperdon pusillum* and *Lycoperdon giganteus*. Afri. J. Biom. Res., 6: 84 – 90.
7. Jong, S.C. and J.M. Birmingham, (1992) Medical effects of the mushroom *Ganoderma* , Adv. Appl. Microbiol (37): 101-134
8. Lakshmi, B., Tilak, J. C., Adhikri, S., Devasayagam, T. P. A., Janardhanan, K. K. 2004. Evaluation of antioxidant activity of selected Indian mushrooms. *Pharma. Biol.*, 42: 179 –185
9. Lillian, B., Ricardo, C., Josiana, A., Isabel, C. F., Ferreira, R., Paula B. and Leticia, M. E. 2006. Antimicrobial activity and bioactive compounds of Portuguese edible mushrooms methanolic extracts. *J. Euro. Food Res. Technol.*, 225 (2): 151 – 156.
10. Lindequist, U., Niedermeyer, T. H. J., Julich, W. 2005. The pharmacological potentials of mushrooms. *Ecam.*, 2: 285 –299
11. Mizuno T. (1995) Ed; *Mushrooms: The versatile fungus food and medicinal properties in Food Review International (II)* no.1.Marcal Dekkar , Inc ., New York.

12. Perch M.H. (1990) In vitro interactions between *Armillaria Luteobubalina* and other wood decay fungi *Mycol.Res.*(94), 753-761
13. Schwietzer,C.H.(1970) Mushroom poisons and poisoning. *Muechan Med Wuchenschr* (112): 1085-1090
14. Stamets, P. 2000. Growing gourmet and medicinal mushroom. Berkeley Ten Speed press. Pp 45-49.
15. Stewart R.L. (1972) Species of *Ganoderma* and related genera mainly of the Bogor of Leiden Herbaria. *Persoonia* (7) 55-118
16. Wasser S.P. and Weis A.L. (1999) Medicinal properties of substances occurring in higher Basidiomycetes Mushrooms: current perspectives (review) *Int. J. Med Mushr* (1) 31-62