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ORIGINAL RESEARCH ARTICLE

In Vitro Study of Antibacterial Activity of γ- irradiated and Unirradiated Leguminous Seed Coats

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ABSTRACT

Antimicrobial activity of seed coats contents of Vigna Mungo, Cajanus Cajan and Vigna Unguiculata was evaluated against Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Bacillus subtilis and Klebsiella pneumonia.Gamma irradiated and unirradiated seed coats extracts were screened for antibacterial activity, which involved the determination of inhibition zone in mm. Norfloxacin was used as standared reference. Gamma unirradiated seed coat extracts showed a high antibacterial activity against Escherichia coli, Staphylococcus aureus, Pseudomonas aerunginosa Bacillus Subtilis and Klebsiella pneumonia as compared to gamma irradiated seed coats extracts.

Key Words: Antibacterial Activity, Vigna Mungo, Cajanus Cajan, Vigna Unguiculata

INTRODUCTION

Natural products are preferred for biologically screening based on ethnomedical use of plants, because many infectious diseases are known to have been treated with herbal remedies throughout the history of mankind. Antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world[1, 2]. Drug resistance to pathogenic microorganisms has been reported in literature[3]. It is suggested that antimicrobials may have a significant clinical value in treatment of resistant microbial strains[4]. The antimicrobial activities of plants have been investigated by a number of researchers worldwide. The Cajanus Cajan, Vigna Unguiculata and Vigna Mungo are members of Leguminosea family. The Pigeon Pea (Cajanus Cajan L.) seeds contain anti-nutritional substances which affect their utilization in poultry feeding especially the raw seeds[5]. Leaves are also used for toothache, mouthwash[6], hepatitis, diabetes, urinary infections, yellow fever, vermifuge, vulnerary etc. Scorched seed are added to coffee, to alleviate headache and vertigo. Chemical component of Cajanus Cajan are 2'-2'methylcajanone, 2'-hydroxygenistein, 5,7,2'trihyroxyisofalvone, betamryin, betasitosterol, cajaflavanone, cajaisoflavone, cajanin, cajanone, cajaquinone, concajanin, ferreirin, genistein, lupeol, phenylalanine, stigmasterol. Cowpea (Vigna Unguiculata L.) seeds contain small amounts of b-carotene equivalents, thiamin, riboflavin, vitamin A, niacin, folic acid and ascorbic acid. Cowpea seed is a nutritious component in the human diet[7], as well as a nutritious livestock feed. Blackgram (Vigna Mungo L.) is very rich in phosphoric acid, nutritious and is recommended for control of diabetics[8]. Chemical compounds found in seed coats of legumes generally contain tannins, lignin and non-tannin polyphenolic compounds which comprise several groups of phenolic compounds[9]. Literature survey revealed that the seed coats extract have yet not been screened for its traditional claim of antibacterial activity. Therefore, the objective of this work was to explore the antibacterial properties of seed coats of leguminous plants.

MATERIALS AND METHODS

The seed coats of three leguminous plants namely Cajanus Cajan, Vigna Unguiculata, Vigna Mungo were collected from Multai (Madhya Pradesh)

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India and authenticated by Department of Botany, Rashtrasant Tukadoji Maharaj Nagpur University. Seed coats were separated from cotyledons, dried and ground to a flour, stored in a screw cap bottles and used for isolation. It was divided in two parts: (i) The seed coats were irradiated with gamma radiations (^{60}Co source GC-900 at a dose rate of 0.8 kGy/hr for 48 hr -72 hr) and (ii) The seed coats were used without exposure to radiation. Both material were defatted with petroleum ether (60-80 ^{0}C) followed by water-ethanol mixture (1:1) using Soxhlet extraction process. The solvent was evaporated under reduced pressure at 70 ^{0}C and the extract thus obtained was used directly for the assessment of antibacterial activity through *in vitro* method.

ANTIBACTERIAL ACTIVITY

The gamma irradiated and unirradiated seed coats extract from the Cajanus Cajan, Vigna Unguiculata and Vigna Mungo were investigated for their antibacterial activity against Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Bacillus subtilis and Klebsiella pneumonia. For antibacterial activity Norfloxacin was used as a standard. The agar dilution method was performed using Mueller-Hinton agar (Hi-Media) medium. This method depends on the diffusion[10]of drug from bore through the solidified agar layer of a petri dish to an extent such that growth of the inoculated microorganism is prevented entirely in a circular area "zone" around the cup containing the solution of the compound under test.

All the operations were carried out under aseptic conditions. Sterile medium was melted on water bath and kept at 45 ^{0}C in constant temperature water bath. In each sterile petri dish, 25 ml of molten medium was added (thickness: approx 4-5 mm) and 10^7 /ml of sub cultured organism under study was inoculated. The inoculated dishes were allowed to set for 30 min at room temperature. Three cups of 8 mm diameter were then made with the help of sterile stainless steel borer. 100 µl were added to each cup. Petri dishes were kept in refrigerator for 30 minutes so as to allow diffusion of the solutions in the medium, then incubated for about 24 h at 37 ^{0}C, Zones of inhibition produced by test compounds were measured in mm on antibiotic zone reader (Expo Industrial Corporation Cat. No. E/C41) in various axis and average reading was considered. The activity index was calculated against the standard.

RESULTS AND DISCUSSION

Antibacterial activity of aqueous- ethanol extracted products of gamma irradiated and unirradiated seed coats from the Cajanus Cajan, Vigna Unguiculata and Vigna Mungo were examined and found to exhibit a good antibacterial activity at 1 ml against most of the studied gram positive and gram negative organisms (**Table 1**).

Table 1: In vitro, Antibacterial activity of γ-irradiated and unirradiated Cajanus Cajan, Vigna Unguiculata and Vigna Mungo seed coats

Microorganism	Diameter of zone of Inhibition in (mm)						
	γ -unirradiated seedcoats			γ-irradiated seed coats			Norfloxacin
	Cajanus Cajan	Vigna Unguiculata	Vigna Mungo	Cajanus Cajan	Vigna Unguiculata	Vigna Mungo	
Staphylococcus aureus	12.1	15.3	14.7	6.8	10.5	8.2	27.1
Escherichia coli	14.3	14.6	13.9	6.2	12.0	4.3	23.8
Pseudomonas aeruginosa	15.1	14.4	17.2	7.0	13.6	4.2	28.4
Bacillus subtilis	13.2	14.2	15.0	6.4	11.1	8.5	24.4
Klebsiella pneumonia	14.6	14.9	16.6	6.8	12.5	5.6	26.7

The aqueous -ethanol extracts of γ-unirradiated seed coats was found to have more antibacterial activity against Escherichia coli, Staphylococcus aureus and Pseudomonas aeruginosa, Bacillus subtilis and Klebsiella pneumonia , when compared to the γ- irradiated seed coats extracts but less than that of the standard drug Norfloxacin. Among the pathogenic bacteria, Pseudomonas aeruginosa was highly susceptible to γ-unirradiated Cajanus Cajan, Vigna Mungo and γ-irradiated Cajanus Cajan, Vigna Unguiculata seed coats extract, while

Staphylococcus aureus showed high sensitivity to γ-unirradiated Vigna Unguiculata and γ-irradiated Vigna Mungo seed coats extract. The present study reveals that unirradiated seed coats extracts prepared in aqueous-ethanol solvent posses good antibacterial activity. The phytochemicals though not fit to be used as food preservative may be developed as disinfectants. Further work is still under progress to explore the chemical nature of the active constituents.

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REFERENCES

1. Saxena K. Antimicrobial Screening of Selected Medicinal Plants from India. J of Ethnopharmacology 1997; 58 (2):75-83.
2. Nimri LF, Meqdam MM, Alkofahi A. Antibacterial activity of Jordanian medicinal Plants. Pharmacological Biology 1999; 37 (3): 196-201.
3. Davis J. Inactivation of antibiotic and dissemination of resistance genes. Science 1994; 264: 375–382.
4. Eloff J N. Which extract and should be used for the screening and isolation of antimicrobial components from plants? J Ethnopharmacology 1988; 60: 1-8.
5. Singh SK. Food legumes: distribution, adaptability and biology of yield In:FAO plant production and protection paper 3. Rome: FAO; 1977.
6. Parrotta JA. Healing plants of Peninsular India. Wallingford, UK and New York: CABI Publishing; 2001.
7. Karakaya S, El SN, and Tas AA. Antioxidant activity of some foods containing phenolic compounds. Intern. J Food Sci Nutri 2001; 52:501-508.
8. Nadkarni MK. Bombay: Indian Material Medica. Popular Prakashan; 2000.
9. Carmona A, Seidl SD and Faffe WG. Comparison of extraction methods and assay procedures for the determination of the apparent tannin content of common beans. J of the Science of Food and Agriculture 1991; 56: 291-301.
10. Bauer RK, Kirby MDK, Sherris JC and Turck M. Antibiotic susceptibility testing by standard single disc diffusion method. Asian J Tropic Agric Sci 1966; 18: 115-121.