Bulletin of Environment, Pharmacology and Life Sciences
Bull. Env. Pharmacol. Life Sci., Special Issue [1]2022 : 1369-1372

Online ISSN 2277-1808
Journal's URL:http://www.bepls.com
CODEN: BEPLAD

ORIGINAL ARTICLE **OPEN ACCESS**

Antimicrobial Activity of Electrospun Nanofiber Mat Loaded *Acmella paniculata* (Wall. ex DC) R. K. Jansen.

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ABSTRACT

Acmella paniculata (Wall. ex DC) R. K. Jansen has attained substantial attention as a ethnopharmacological material in folk remedies mainly due to its wide use by tribal people. To exploit this, electrospun nanofiber mat were fabricated from poly (vinyl alcohol) loaded Acmella paniculata extract for characterization of antibacterial potential. Antibacterial activity of 8 wt. % poly (vinyl alcohol) (PVA) and 8 wt. % PVA/0.6 µg A. paniculata plant extract of electrospun nanofiber mat demonstrated excellent inhibition of Gram-positive Staphylococcus aureus and Streptococcus spp. and Gram-negative Escherichia coli and Pseudomonas spp. Fabricating 8 wt. % PVA and 8 wt. % PVA loaded A. paniculata to electrospun nanofiber mat is a novel attempt made by us and it may be useful for biomedical application such as antibacterial, wound dressing potential and so on. Further exploitation on A. paniculata would be great source for novel antibacterial biomedicinal discovery.

Keywords: *Electrospun nanofiber, Acmella paniculata, PVA, Antimicrobial activity*

Received 11.02.2022 Revised 19.03.2022 Accepted 24.04.2022

INTRODUCTION

Electrospinning technique is awell-developed, simple, and attractive electro-hydrodynamic method used to develop standard polymeric synthetic fibers from nanometers to micrometers and generate 3D network of porous webs with nonwoven, nano scale fiber-based matrix[1].According to [2] the use of electrospun nanofibers in several fields including biomedical applications such as drug delivery, medical implants, water filtration and scaffolds for tissue engineering. The potential use of electrospun nanofibers arises from their remarkable properties like easy surface functionalization, high pore interconnectivity and high surface area-to-volume ratio[3,4].Poly(vinyl) alcohol (PVA) is a biocompatible, water soluble, non-toxic, biodegradable synthetic polymer and have highly hydrophilic properties, better fiber forming and its fibers have been commercialized from1950's which can be widely used in the bio-medicinal field[5].The genus *Acmella* Rich. from the family Asteraceae which include 30 species that are spread in the tropical and sub-tropical regions around the world [6]. *A.paniculata* (Wall. ex DC) R. K. Jansen is commonly called as "Toothache" plant because its flowers are chewed to reduce toothache. It is an herb up to 65 cm. in height, branched stem and marigold eye flowers. The leaves of *Acmella paniculata* are eaten raw as a vegetable by many tribals from India. The crushed plant material have been used in rheumatism, Pulverization of kidney stone and gall stone remedy for stammering in children[7]. Hence it was intended to explore *A.paniculata* for antimicrobial compounds. In the present study electropsun nanofiber mat of0.6 µg *A.paniculata* extract loaded 8 wt. % PVA was prepared and confirmed the potentiality for antimicrobial nanofiber web as a new material. The antimicrobial effects were assessed by using Gram-positive *S. aureus*, *Streptococcus* spp. and Gram-negative *E.coli* and *Pseudomonas* spp. to form their colonies and their metabolic importance. An insight into the mode of antimicrobial potential8 wt. % PVA/0.6 µg *A. paniculata* electrospun nanofiber membranes was presented.

MATERIAL AND METHODS

Collection of plant material and preparation of plant extract

The selected fresh plant material was collected from campus of Shivaji University, Kolhapur, Maharashtra, India. The collected plant material waswashed, air dried and powdered using mechanical grinder. 1 gm. of powdered test plant material was mixed into 100 ml. of petroleum ether, this solution was used for Soxhlet extraction and obtained extract was used for preparation of electrospun nanofiber mat.

Preparation of electrospun nanofiber mat

Poly (vinyl alcohol) (PVA 99+ % hydrolyzed, 85-12.4 kDa) were provided by Otta chemie Pvt. Ltd. Mumbai, Maharashtra. The electrospinning solution was prepared by mixing 8 wt.% PVA in distilled water to dissolve 8 wt. % PVA in distilled water the solution was heated at 90^0 C using Magnetic stirrer hot plate with continuous stirring for 2 hours. For preparation of test samples of electrospinning solution,8 wt. % PVA and 8 wt. % PVA/0.6µg*A.paniculata*of plant extract was added at room temperature with constant stirring on magnetic stirrer, then solution was kept for overnight to avoid air bubbles and to settle the solution. Then the prepared electrospinning solution was loaded in 5 ml syringe with stainless Steel blunt tip needle. A positively charged High voltage power DC source was provided to needle tip. The negatively charged electrode was connected to a rotating drum collector at 400rpm with optimized condition of electrospinning unit.

Antimicrobial activity

The disc diffusion assay was used to evaluate antimicrobial activity described by [8]. Antibacterial activity of 8 wt. % PVA and 8 wt. % PVA/0.6 µg *A.paniculata* was detected by observing growth response of Gram-positive *Staphylococcus aureus* and *Streptococcus* spp. and Gram-negative *Escherichia coli* and *Pseudomonas* spp. microorganisms. Electrospun nanofiber mat placed in contact with test microorganisms using agar diffusion method. The petri dishes were poured with liquidified agar medium to invariable thickness. The disc of electrospun nanofibers mats were placed on nutrient agar plate in contact with intended test bacteria. Antimicrobial potential of electrospinning nanofiber mats were recorded as zone of inhibition around the disc of electrospinning mat in millimeter.

RESULTS AND DISCUSSION

The antimicrobial activity of 8 wt. % PVA and 8 wt. % PVA loaded 0.6 µg *A.paniculata* electrospun nanofibers mat were evaluated by performing disc diffusion method against Gram-positive *Staphylococcus aureus* and *Streptococcus* spp. and Gram-negative *Escherichia coli* and *Pseudomonas* spp. Antimicrobial activity of electrospun nanofibers mat were measured as zone of inhibition around the disc in millimeter that were placed in contact with test bacteria. The 8 wt. % PVA used as control.The antimicrobial effect of with 8 wt. %PVA electrospun nanofibers showed their response for Gram-positive *Staphylococcus aureus* with 08mm.of inhibition zone, *Streptococcus* spp. 06mm.of inhibition zone and for Gram-negative *E. coli*it was 06mm.of inhibition zone, *Pseudomonas* spp. showed 07mm.of inhibition zone. It indicated that highest antimicrobial activity recorded against Gram-positive stains. The antimicrobial effect of test plant material with8 wt. % PVA/ *A.paniculata* showed its similar effect for both the Gram-positive bacteria (*S.aureus* and *Streptococcus* spp.)with 12 mm of inhibition zone, and for Gram-negative *E. coli*09mm. of inhibition zone and *Pseudomonas* spp. with 12 mm of inhibition zone. 8% wt. % PVA and 8 wt. % PVA/ *A.paniculata* showed its highest activity against Gram-positive bacteria with mean of 9.4 mm. of inhibition zone as compared to Gram-negative bacteria with 8.5 mm. of inhibition zone. Among the stain used *S.aureus* had prominent inhibition zone than *E. coli* this result are in agreement with [9,10].According to them difference between the both stains were probably a consequence the tendency of *S.aureus* to form cellular aggregates rather than a different response to surface hydrophicity.

The control used was 8 wt. % PVAwhich showed its effect against both Gram-positive and Gram-negative as a mean of 13.5 mm. inhibition zone where as8 wt. % PVA/ *A.paniculata* showed mean of 22.5 mm. of inhibition zone. The results are in agreement with[11]. According to[12,13] that petroleum ether, Chloroform, ethyl acetate, and methanolic extracts of *A.paniculata* comprised mostly with Alkaloids, Saponins, and Flavonoids. These compounds might be responsible for electrical depolarization of bacterial membrane.[14] stated that alkyl groups removed from the cell. Then, long hydrophobic alkyl chains and the charged nitrogen molecule mitigate with the phospholipid bilayer of cytoplasmic membranes of the cellthat caused a phase separation of the polar and hydrophobic regions. As a result, damage, leakage and weakness of the cytoplasmic membrane was caused by action of these compounds, which leads to a instability of the membrane and disruption of the permeability. Hence, it is clear that 8 wt. % PVA/ *A.paniculata* had a potential key role in inhibit the growth of test bacteria. Medicinally and traditionally used plants have been utilized as a main source for drug over many centuries in various developed and developing countries in the world.The medicinal properties of plant are due to presence of diverse phytochemical compounds as secondary metabolite, which played significant role in antimicrobial crude medicine as a new anti-inflectional agent[15].[16] stated that *A.paniculata* have been used to treat dysentery, against scurvy and as a fish poison. The methanolic extract of test plant showed its effect on blood pressure of dog and cat and it exhibited its potentiality against general amino modular properties when used internally, boosting production of leukocytes and antiviral interferon [12]. [17] reported four biologically active phytochemicals and their effectivity on antioxidant potential of

A.paniculata. Hence, purification and characterization of *A.paniculata* for its active principle followed by detailed investigations that are necessary prior to its use in biomedicinal field.

Table no. 1- Antimicrobial activity of electrospun nanoifiber mat loaded *A.paniculata.*

Sr. No	Sample Name	Zone of Inhibition (mm)	
		Gram Positive	
		Staphylococcus aureus	***Streptococcus spp.***
1	8 wt. % PVA	8	6
2	8 wt. % PVA + 0.6 µg *A. paniculata*	12	12
		Gram negative	
		Escherichia coli	***Pseudomonas spp.***
1	8 wt. % PVA	6	7
2	8 wt. % PVA + 0.6 µg *A.paniculata*	9	12

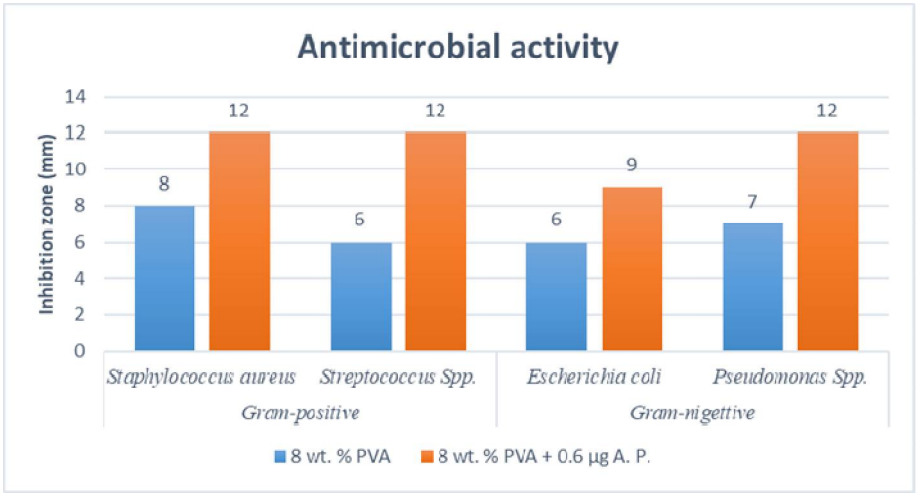

Figure no. 1- Histogram of Antimicrobial activity of electrospun nanoifiber mat loaded *A.paniculata.*

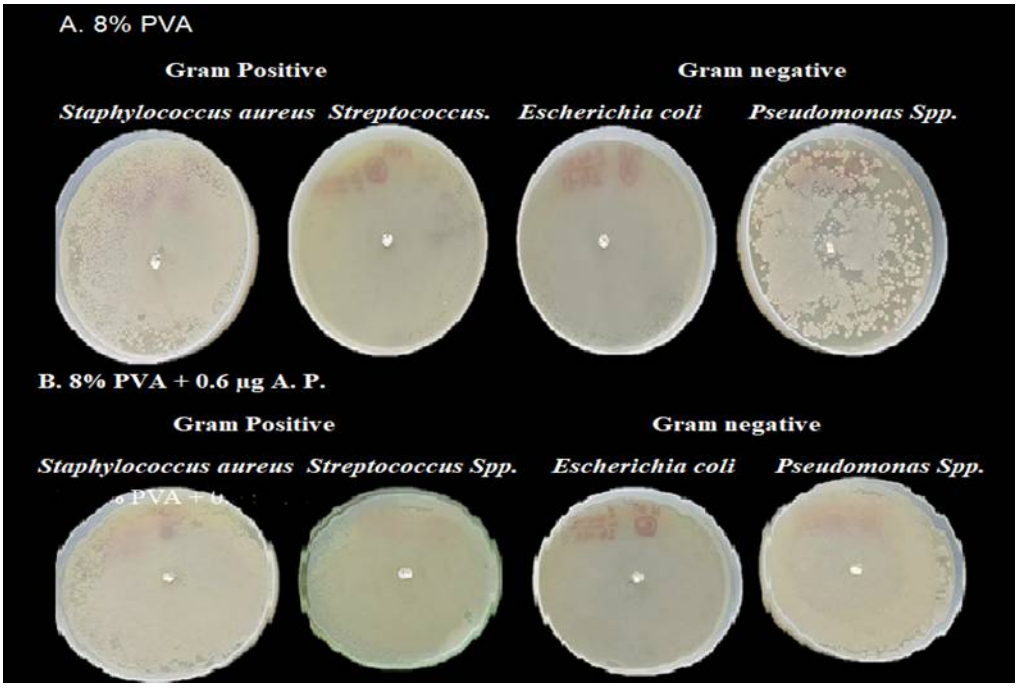

Photo plate no.1- Disc diffusion assay of antimicrobial activity of electrospun nanoifiber mat loaded *A.paniculata.*

CONCLUSION

In present investigation we successfully fabricated 8 wt. % PVAand 8 wt. % PVA/0.6 µg *A.paniculata* loaded electrospun nanofiber mat for its antimicrobial potential.The results showed that antimicrobial activity for Gram-positive *S.aureus* and *Streptococcus* spp. and Gram-negative *E.coli* and *Pseudomonas* spp. of electrospun nanofibers ceased growth of test bacteria. Hence, it is suggested that *A.paniculata* could be used as potential source for antibacterial activity. However, further studies are required for purification and characterization of active principles and detailed investigation necessary for prior to its medical application.

ACKNOWLEDGEMENT

The authors are thankful to Principal and Head Department of Botany, Balwant College, Vita for providing necessary facilities. S. A. Patole is thankful to Dr. Babasaheb Ambedkar Research and Training Institute (BARTI), Pune for financial assistance.

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CITATION OF THIS ARTICLE

S. A. Patole and C. J. Khilare. Antimicrobial Activity of Electrospun Nanofiber Mat Loaded *Acmella paniculata* (Wall. ex DC) R. K. Jansen. Bull. Env.Pharmacol. Life Sci., Spl Issue [1] 2022 : 1369-1372