Indexed, Abstracted and Cited: Index Copernicus International (Poland), ISRA Journal Impact Factor, International Impact Factor Services (IIFS), Directory of Research Journals Indexing (DRJI), International Institute of Organized Research and Scientific Indexing Services, Cosmos Science Foundation (South-East Asia), International Innovative Journal Impact Factor, Einstein Institute for Scientific Information {EISI}, Directory of Open Access Scholarly Resources, Science Indexing Library (UAE), Swedish Scientific Publication (Sweden), citefactor.org journals indexing, Directory Indexing of International

World Journal of Biology and Medical Sciences

Published by Society for Advancement of Science®

ISSN 2349-0063 (Online/Electronic)

Volume 7, Issue-1&2, 84-91, January - June, 2020

Journal Impact Factor: 4.197

WJBMS 07/02/021/2020

A Double Blind Peer Reviewed Journal / Refereed Journal

www.sasjournals.com

wjbmedsc@gmail.com/wjbms.lko@gmail.com

RESEARCH PAPER

Received: 15/05/2020 Revised: 30/06/2020 Accepted: 30/06/2020

Pollen Grains as Antibacterial Agents – A New Approach for Therapy

Suvarna P. Patil, *Pranita P. Shuddhalwar,

***Poonam R. Bachale and *Vijay N. Charde**

Department of Botany, Taywade College, Mahadula-Koradi, Dist. Nagpur- 441 111, India

***Department of Microbiology, Taywade College, Mahadula-Koradi,**

Dist. Nagpur- 441 111, India

ABSTRACT

In ancient Indian literatures considered that all plant parts are potential sources of medicinal substances. Antibiotic resistance exhibited by pathogenic microbial infectious agent have led to the screening of several medicinal plants for their potential antimicrobial activity. Pollen as well as other apiculture products, has gained increased attention for its therapeutic properties as antibacterial, antifungicide, anti-caryogenic, antioxidant and immunomodulatory effects. Pollen finds its use in apitherapy and also as a functional food in the food industry due to their nutritional properties. Pollen is fine and light powder like material having nutritional value with proteins (25-30%), carbohydrate (30-55%), lipids, including fatty acids and sterol (1-20%), fats (1-13%), vitamins and mineral. The aim of this study was to investigate the antibacterial activity of pollen extracts of some common flowers from Vidharbha region against some pathogenic bacteria. Pollen samples showed a potential activity against the growth of both Gram positive and Gram negative bacteria. This would be a very interesting approach to control more dangerous species of microorganism in medical sciences.

Keywords: Antimicrobial Activity, Pollen grains, Common Flowers and Antibiotic Sensitivity.

INTRODUCTION

India has a large wealth of medicinal plant. The plant kingdom is an implicit profitable idea of potential drug targets and other active molecules waiting to be discovered. In the 70s and 80s, however, scientific and clinical reports began to come out of some European countries, especially Germany, indicating that the herbal remedies, which had never been discarded, had many substantial therapeutic and economic benefits for the living being [Tyler, 2000, Mowrey, 1990]. With this new knowledge and the rising widespread recognition of persistent troubles in treating disease with manufactured drugs, Americans began to demand herbal products [Tyler, 2000, Balch, 2002]. Indiscriminate usage of antibiotics was led to the appearance of drug resistant strains, which have a significant collision on patient's mortality and morbidity.

The effects of different plant part extracts on bacteria have been studied by very large number of researchers in different parts of the world. Here we studied the antimicrobial capacity of pollen and compare it with some standard antibiotics. Pollen as well as other apiculture products, has gained increased attention for its therapeutic properties as antibacterial [Garcia et al., 2001], antifungicide [Proestos, 2005], anti-caryogenic [Almas et al., 2001], antioxidant [Nikolaieva et al., 2019] and immunomodulatory [Gebara et al., 2002] effects. Other potential applications of pollen include its use in apitherapy and as a functional food in the food industry due to pollen nutritional properties.

Pollen grains have great commercial interest owing to its high nutritional quality and can be considered as potential source of energy and proteins for human consumption [Kroyer et al., 2007, Compos et al., 2003]. The old Egyptians described pollen as "a life giving dust pollen and its nutritional value is still surrounded by mysteries. It is called the only perfectly complete food [Marivalda et al., 2015].

Pollen contains nutritional compounds like carbohydrates, proteins, amino acids, lipids, vitamins and minerals [Serra Bonvehni et al., 2001]. Moreover, it also contains number of macro and micro elements like zinc, copper, phosphorus, magnesium, calcium, selenium, iron and potassium [Kieliszek et al., 2018]. In addition to these, pollen contains significant amounts of polyphenolic substances, which provide distinct color of pollen (black, purple, red, brown, yellow etc.) and their organoleptic feature is bitter to taste [Zuluaga et al., 2016]. Polyphenols contributed in prevention of cancer [Sobral et al., 2017], type 2 diabetic treatment [Forato Anhe et al., 2013] and also in dementia disease [Hugel et al., 2015]. By recognizing the relationship between nutrition and health, these apicultural products are included in human diet by keeping out of mind only taste, aroma and general nutrition of food. It energizes, restores destabilized nervous system, and helps recover nervous disorder and emotional conditions into young and old.

The present study was undertaken to investigate the antimicrobial capacity of pollen extract of six flower bearing plants *Rosa rubiginosa (L.), Tecoma stans (L.), Calendula officinalis (L.), Nerium oleander (L.) Millingtonia hortensis* (L.f.) and *Hibiscus rosa-sinensis* (L.) against some pathogenic bacteria and compare it with some standard antibiotics.

PLANT PROFILES

***Rosa rubiginosa(L.) (*Rose, gulab)** belongs to family *Rosaceae* have been used for commercial perfumery. Rose water, made as a byproduct of rose oil production. Rose syrup is used to make rose scones and marshmallows.

Tecoma stans(L.) (Yellow bells, ghantichful) belongs to family *Bignoniaceae* has been used for a variety of purposes in herbal medicine. Its primary applications have been in treating

diabetes and digestive problems. Extracts from *Tecoma stans* leaves have been found to inhibit the growth of the yeast infection, *Candida albicans*.

Calendula officinalis (L.) (Marigold, Zendu) belongs to family *Asteraceae* possesses anti-inflammatory, antifungal, antiviral and antiseptic properties. Its ointments and lotions are used for soothing irritated or cracked skin, to treat eczema, sunburn and vaginal infections. The *calendula* flowers showed good effect in treating bed sores, varicose veins, gum inflammations, bruises, conjunctivitis, stomach ulcers, jaundice, breast cancer and said to relieve HIV symptoms.

Nerium oleander (L.) (Oleander, Kahner) belongs to family *Apocynaceae* used traditionally by herbalists as a folk remedy for a wide variety of maladies and conditions, including dermatitis, abscesses, eczema, psoriasis, sores, warts, corns, ringworm, scabies, herpes, skin cancer, asthma, dysmenorrheal, epilepsy, malaria, abortifacient, emetics, heart tonics, tumors, loss of hair, syphilis, gingivitis and as a nose drop for children.

Millingtonia hortensis (L.f.) (Indian cork tree, Akash Nim) belongs to family *Bignoniaceae* known as Indian cork tree. The leaves of ***Millingtonia hortensis*** constituting mainly flavanoids, tannins and alkaloids and are used as antipyretic, sinusitis, and tonic in folklore medicine.

Hibiscus rosasinensis(L.) **(China rose, Jaswanda)** belongs to family *Malvaceae* the young leaves and flowers are used in inducing abortion, to cure hepatitis C and as a cure for headache, for hair care.

MATERIALS AND METHODS

Antibiotics

Amikacin, Amoxicillin, Ampicillin, Cefazolin, Chloramphenicol, Co-Trimoxole, Gentamycin, Minocyclin, Norfloxacin, Ofloxacin, Penicillin G, Piperacillin, Rifampicin, Streptomycin, Tetracycline, Tobramycin, Vancomycin.

Test Organisms

Escherichia coli (ATCC-8739), *Staphylococcus aureus* (ATCC-6017)*, Salmonella typhi* (ATCC-6538)*, Bacillus subtilis* (ATCC-6633).

Flowers selected for pollen grains

Flowers of *Rosa rubiginosa (L.), Tecoma stans (L.), Calendula officinalis (L.), Nerium oleander (L.), Millingtonia hortensis* (*L.f*.) and *Hibiscus rosa-sinensis* (*L*.).

Collection of Pollens and Preparation of Pollen Extracts

Fresh flowers were plucked out from plants and brought to the laboratory. The pollen grains were dislodged from an anther either manually or by using forceps and carefully collected in petridish and 1g of each pollens were taken in three different test tubes containing 10ml 90% ethanol, 70% methanol and distilled water respectively. Crushed it with smooth flatten glass rod for 10-15 minutes and kept at 70^0C for 30 minutes in water bath. The extract was filtered through sterile muslin cloth. This extract was used for antibacterial study.

Preparation of Inoculum

Nutrient broth was inoculated with freshly sub-cultured bacteria and incubated at 37^0C for 6-8 hours corresponds to the turbidity of 0.5 MacFarland standards (1.5×10^8 CFU/ml). Such prepared inoculum was used to spread onto Hi-sensitivity test agar (Himedia Laboratories Ltd., India).

Antimicrobial activity by well diffusion method

100 µl of inoculum was transferred to Hi-sensitivity agar petri plate with the help of micropipette under aseptic conditions. Culture was spread uniformly on the agar medium

using sterile spreader. Three equidistant wells per plate were cut with the help of sterile metal borer having diameter of 5mm. Each well was labelled for different extract, 100 µl of each extract was transferred to respective well with the help of micropipette. Plates were kept undisturbed in a refrigerator for 1 hour for diffusion of substances into the media and then incubated at 37^0C for 24 hours. Zone of inhibition was noted and susceptibility of the test organisms against antimicrobials was categorized as Resistant, Intermediate and Sensitive. Antimicrobial activity was evaluated on the basis of diameter of zone of inhibition according to Table 1 [Johnson and Case, 1995].

Table 1.Zone Evaluation Table [Johnson, T and C. Case, 1995].

Category	Diameter of zone of inhibition (mm)
Resistant	10 or less
Intermediate	11 or 15
Sensitive	16 or more

Antibiotic Sensitivity Test by Kirby Bauer Method

Antibiotic sensitivity of test organisms against standard antibiotics was carried out using Kirby Bauer method and zone of inhibition was measured in mm and the result were noted down as sensitive, intermediate or resistance [Bauer and Kirby, 1966].

RESULTS AND DISCUSSION

In present study, the pollen extract of 6 different flowers were tested against the test organisms i.e., *Escherichia coli* (ATCC-8739), *Staphylococcus aureus (*ATCC-6538), *Salmonella typhi* (ATCC-6017), *Bacillus subtilis* (ATCC-6633). The results of anti-microbial activity of Pollen grains with various solvent extracts are given in Table 2. Sensitivity of test organisms was tested against standard antibiotics and results were interpreted as per CLSI standards [CLSI, 2007] as shown in Table 3.

B. subtilis was found to be susceptible to ethanol, methanolic and distilled water extract of *Calendula officinalis (L)* whereas only distilled water extract of *Rosa rubiginosa (L.)* was found to be effective against *B. subtilis* remaining extracts were either moderately effective or ineffective against it.

S. aureus was found to be susceptible to ethanol extract of *Calendula officinalis (L.),* and *Hibiscus rosa-sinensis (L.),* methanol extract of *Calendula officinalis (L.)* and distilled water extract of *Millingtonia hortensis (L.f.), Hibiscus rosa-sinensis (L.),* whereas other pollen extract were failed to inhibit the growth of *S. aureus.*

E. coli was found to be susceptible to ethanol extract of *Rosa rubiginosa (L.), Tecoma stans (L.) and Millingtonia hortensis (Lf.)* methanol extract *Rosa rubiginosa (L.)* and distilled water extract of *Hibiscus rosa-sinensis (L.)* remaining extracts were either moderately effective or ineffective against *E. coli.*

Salmonella typhi was found to be susceptible to ethanol extract of *Rosa rubiginosa (L.), Calendula officinalis (L.), Millingtonia hortensis (L.f.) and Hibiscus rosa-sinensis (L.),* methanol extract of *Rosa rubiginosa (L.), Calendula officinalis (L.), Millingtonia hortensis (L.f.)* whereas it was moderately sensitive to ethanol extract of *Tecoma stans (L.),* methanol extract of *Nerium oleander (L.)* and distilled water extract of *Calendula officinalis (L.).* Other pollen extracts were failed to inhibit the growth of *Salmonella typhi.*

Table 2. Anti-Microbial Activity of Pollen Grains with Various Extracts.

Sr. No.	Pollen Extracts	*B. subtilis*			*S. aureus*			*E. coli*			*S. typhi*		
		E	M	DW	E	M	DW	E	M	DW	E	M	DW
1	*Rosa rubiginosa*	-	14 (I)	22 (S)	-	-	-	17 (S)	20 (S)	-	16 (S)	19 (S)	-
2	*Tecoma stans*	-	-	-	-	-	-	16 (S)	10 (R)	-	15 (I)	-	-
3	*Calendula officinalis*	18 (S)	28 (S)	17 (S)	20 (S)	22 (S)	-	-	-	-	19 (S)	22 (S)	14 (I)
4	*Nerium oleander*	-	-	12 (I)	-	-	-	-	-	-	-	13 (I)	-
5	*Millingtonia hortensis*	-	-	-	-	-	18 (S)	18 (S)	-	-	35 (S)	18 (S)	-
6	*Hibiscus rosasinensis*	-	-	-	17 (S)	-	24 (S)	-	-	24 (S)	17 (S)	-	-

Note- E-Ethanol extract, M- Methanol extract, DW-Distilled Water extract, S- Sensitive, I- Intermediate, R- Resistant

Table 3. Standard Antibiotics against Organisms.

Sr. No.	Antibiotics	*Bacillus subtilis*	*S. aureus*	*E. coli*	*Salmonella typhi*
1	Amikacin (Ak)	-	-	21(S)	19(S)
2	Ampicillin (A)	-	-	-	-
3	Amoxicillin (Ac)	-	-	-	-
4	Co-Trimoxazole (COT)	-	18(S)	-	-
5	Chloramphenicol (C)	19(S)	20(S)	-	25(S)
6	Gentamycin (G)	-	-	23(S)	22(S)
7	Minocyclin (MI)	17(I)	15(I)	-	-
8	Norfloxacin (NX)	-	-	22(S)	28(S)
9	Ofloxacin (OF)	13(I)	28(S)	20(S)	27(S)
10	Penicillin (G)	10(R)	20(R)	-	-
11	Piperacillin(PC)	-	-	10(R)	11(R)
12	Rifampicin (R)	13(S)	10(S)	-	16(S)
13	Streptomycin (S)	10(R)	21(S)	-	-
14	Tetracycline (TE)	11(R)	21(I)	15(I)	19(S)
15	Tobramycin (TOB)	19(S)	19(S)	18(S)	20(S)
16	Vancomycin (VA)	22(S)	19(S)	-	-
17	Cefazolin (CZ)	-	-	10(R)	18(S)

Note- S- Sensitive, I- Intermediate, R- Resistant

Bacillus subtilis was found to be susceptible to Chloramphenicol (C), Rifampicin (R), Tobramycin (TOB) and Vancomycin (VA). It was moderately susceptible to Minocyclin (MI), Ofloxacin (OF). The antimicrobial activity of pollen grains of *Rosa rubiginosa (L.), Calendula officinalis (L.),* and were same or more than Chloramphenicol (C), Rifampicin (R), Tobramycin (TOB) and Vancomycin (VA).

S. aureus was found to be susceptible to Co-Trimoxazole (COT), Chlorophenicol (C), Ofloxacin (OF), Rifampicin (R), Streptomycin (S), Tobramycin (TOB), Vancomycin (VA). It was moderately susceptible to Minocyclin (MI), Tetracycline (TE). Whereas, the antimicrobial activity of pollen grains of *Calendula officinalis (L.), Millingtonia hortensis (L.f.) and Hibiscus rosa-sinensis (L.),* were same or more than Co-Trimoxazole (COT), Chlorophenicol (C), Ofloxacin (OF), Rifampicin (R), Streptomycin (S), Tobramycin (TOB), Vancomycin (VA).

E. coli was found to be susceptible to Amikacin (Ak), Gentamycin (G), Norfloxacin (NX), Ofloxacin (OF) and Tobramycin (TOB). It was moderately susceptible to Tetracycline (TE) and resistant to Piperacillin (PC) and Cefazolin (CZ). The antimicrobial activity of pollen grains of *Rosa rubiginosa(L.), Tecoma stans (L.), Millingtonia hortensis (L.f.), Hibiscus rosa-sinensis (L.)* were same or more than the Amikacin (Ak), Gentamycin (G), Norfloxacin (NX), Ofloxacin (OF), Tobramycin (TOB).

Salmonella typhi was found to be susceptible to Amikacin (Ak), Chlorophenicol (C), Gentamycin (G), Norfloxacin (NX), and Ofloxacin (OF), Rifampicin (R), Tetracycline (TE), Tobramycin (TOB), and Cefazolin (CZ). The antimicrobial activity of pollen grains of *Rosa rubiginosa(L.), Calendula officinalis (L.), Millingtonia hortensis (L.f.), Hibiscus rosa-sinensis(L.),* were same or more than Amikacin (Ak), Chlorophenicol (C), Gentamycin (G), Norfloxacin (NX), Ofloxacin (OF), Rifampicin (R), Tetracycline (TE) and Tobramycin (TOB). It was resistant to Piperacillin (PC) and *Nerium oleander (L.).*

Very scanty literature is available on antimicrobial activity of pollen grains. Some researchers had studied the antibacterial activity of bee pollens. Carpes et al. had studied antioxidant and antibacterial activity of Bee-pollen. They observed antibacterial activity of ethanolic extract of Bee pollen against *S. aureus, B. subtilis, Pseudomonas aeruginosa* and *Klebsiella sp.* [Carpes et al, 2007]. Abonda et al. 2011 studied antibacterial activity of Bee bread and Bee pollen against pathogenic bacteria. Gram positive bacteria were found to be more sensitive to bee bread and bee pollen than Gram negative bacteria [Abonda et al*.,* 2011]. Similarly, Didaras et al. demonstrated antimicrobial activity of Bee Collected Pollen (BCP) and Bee Bread (BB) higher against Gram positive bacteria compared to Gram negative bacteria. He also concluded that there is high variation in antimicrobial activity of BCP and BB [Didaras et. al., 2020]. The present study also reveals the antibacterial activity of pollens derived from six different flower bearing plants.

CONCLUSION

Pollen samples showed a potential activity against the growth of both Gram positive and Gram-negative bacteria. This would be a very interesting approach to control more dangerous species of microorganism in medical sciences. Because of development of resistance by the micro-organisms to common antibiotics, it has become necessary to search for an alternative approach dealing with this situation. It has been suggested that natural products are preferable. So, the pollen extract of these flowers can be used as a medicine against the infection of *S. typhi, E. coli, S. aureus, B. subtilis.* However, more detail investigation is needed to exploit the use of pollen grains as therapeutic agent against pathogenic micro-organisms. According to our results pollen seems to have interesting antimicrobial properties. Other studies will be necessary for a better understanding of the functional properties of pollen.

COMPETING INTERESTS

The authors declare that they have no conflict of interests.

REFERENCES

Abonda, Z., Zerdani, I., Kalalou, I., Faid, M. and Ahami, M.T. (2011). Study the Antimicrobial activity of Moroccan Bee Bread and Bee-pollen against pathogenic bacteria, *Research Journal of Microbiology* 6 (4):376-384.

Almas, K., Mahmoud, A. and Dahlan, A. (2001).A comparative study of propolis and saline application on human dentin: a SEM study. *Indian Journal Dentist Research*, New Delhi, v. 12, p. 21-70.

Balch, P.A. (2002). *Prescription for herbal healing*. New York: Penguin Putnam, Inc., p. 4, 40, 49, 52, 64, 65, 109, 110, 141.

Bauer, A.W., Kirby, W.M., Sherris, J.C. and Turck, M. (1966). Antibiotic Susceptibility Testing by a Standardized Single Disc Method, *Am. J. Clin. Path.,* 45 (Vol. 4):493-496.

Carpes, S.T. (2007). Study of preparations of Bee pollen extracts, Antioxidant and Antimicrobial activity, *Larvas*, V. 31, n. 6,p.1818-1825nov./dez.

CLSI (2007). Performance standards for antimicrobial susceptibility testing: 17th Informational supplement. Approved standard M100-S17, Wayne, USA: Clinical and Laboratory Standards Institute.

Compos, M.G., R.F. Webby, K.R. Markham, K.A. Mitchell and A.P. da Cunha, (2003). Age induced dimination of free radical scavenging capacity in bee pollens and the contribution of constituent flavonoids. *J. Agric. Food chem.,* 51; 742-745.

Forato Anhe, F., Desjardins, Y., Pilon, G., Dudonne S., Genovese, M., Lajolo, F. and Marette, A. (2013). Polyphenols and type 2 diabetes: A prospective review. *Pharma Nutrition*, 1, 105–114.

Garcia, M., Perez-Arquillue, C., Juan, T., Juan, M.I. and Herrera, A. (2001). Note: pollen analysis and antibacterial activity of Spanish honeys. *Food Science Technology International*, Cambridge, V. 7, n.2, p.155-158.

Gebara, E.C.E., Lima, L.A.and Mayer, M.P.A. (2002). Propolis antimicrobial activity against periodontopathic bacteria. *Brazillian Journal Microbiology*, Sao Paulo, V. 33, n. 4, p. 365-369.

Hugel, H.and Jackson, N. (2015). Polyphenols for the prevention and treatment of dementia diseases. *Neural Regen. Res.*, 10.

Johnson, T. and C. Case, (1995). Chemical methods of control, adapted from laboratory experiments in microbiology, Brief Edition, 4th ed. Redwood city CA: Benjamin/Cummings Publishing Co: Available online from The National Health Museum, Access Excellence activities. Exchange (accessed September 11, 2006) http://www.accessexcellence.Org/AE/AEC/CC/Chance activity.html.

Kieliszek, M., Piwowarek, K., Kot, A., Błaz˙ejak, S., Chlebowska-S´migiel, A. and Wolska, I. (2018). Pollen and bee bread as new health-oriented products: A review. *Trends Food Sci. Technol.* 71, 170–180.

Kroyer, G. and N. Hegedus (2007). Evaluation of bioactive properties of pollen extracts as functional diatory food supplement. Innov. *Food Science Emerg. Tech.* 2; 171-174.

Marivalda, S.B., Machado, C.S., Sodré, G.S., Dias, L.G., Estevinho Leticia, M. and de Carvalho C.A.L. (2015). Microbiological Assessment, Nutritional Characterization and Phenolic Compounds of Bee Pollen from *Melliponamandacaia* Smith, 1983. *Molecules 20* (7): 12525-12544.

Mowrey, D.B. (1990). *Next Generation herbal medicine, 2nd Ed.* New Canaan: Keats Publishing, Inc., (5).

NataliiaNikolaieva, Miroslava Kacaniova, Jacinta Collado Gonzalez , Olga Grygorieva and Janka Nozkova (2019). Determination of microbiological contamination, antibacterial and antioxidant activities of natural plant hazelnut (*Corylusavellana* L.) pollen. *Journal of Environmental Science and Health* Part B; 54 (6):525-532.

Nikos Asoutis Didaras, Katerina Karatasou, Tilemachos G. Dimitriou, Grigoris D. Amoutzias and Dimitris Mossialos (2020). Antimicrobial Activity of Bee-Collected Pollen and Beebread: State of the Art and Future Perspectives. *Antibiotics*, *9* (11), 811.

Proestos, C., Chorianopoulos, N., Nichas, G.J.E. and Komaitis, M. (2005). HPLC analysis of the phenolic compounds of plant extracts: investigation of their antioxidant capacity and antimicrobial activity. *Journal Agricultural Food Chemistry*, Easton, V. 53, n. 4, p. 1190-1195.

Serra Bonvehi, J., SolivaTorrento M. and Centelles Lorente E. (2001). Evaluation of polyphenolic and flavonoid compounds in honeybee-collected pollen produced in spain, *J. Agric. Food Chem.*, 49, 4, 1848–1853.

Sobral, F., Calhelha, R.C., Barros, L., Duenas, M., Tomas, A., Santos-Buelga, C., Vilas-Boasand M. Ferreira (2017). I.C. Flavonoid Composition and Antitumor Activity of Bee Bread Collected in Northeast Portugal. *Molecules*, 22, 248.

Tyler, V.E. (2000). Herbal medicine: from the past to the future. *Public Health Nutrition*, *3* (4A), 447-452.

Zuluaga, C., Martinez, A., Fernandez, J., Lopez-Baldo, J., Quiles, A. and Rodrigo, D. (2016). Effect of High Pressure Processing on Carotenoid and Phenolic Compounds, Antioxidant Capacity, and microbial counts of bee-pollen paste and bee-pollen-based beverage. *Innov. Food Sci. Emerg. Technol.*, 37.

Corresponding author: Dr. Suvarna P. Patil, Department of Botany, Taywade College, Mahadula-Koradi, Dist. Nagpur- 441 111, India
Email: patil.suvarna26@gmail.com