Original Article

Journal of Microbiology and Related Research
Volume 10 Number 2, July - December 2024
DOI: http://dx.doi.org/10.21088/jmrr.2395.6623.10224.3

Isolation of Exopolysaccharides Producing Microorganisms from Garden Soils

Kalavati Prajapati[1], Sreeja Bopin[2]

How to cite this article:

Kalavati Prajapati, Sreeja Bopin, Isolation of Exopolysaccharides-Producing Microorganisms from Garden Soils. J Microbiol Relat Res. 2024;10(2):69–72.

Abstract

Diverse structural, functional, and valuable polysaccharides are synthesized by bacteria of all taxa and secreted into the external environment. These polysaccharides are referred to as exopolysaccharides (EPS) and they may either be homopolymeric or heteropolymeric in composition. So, the study aimed to isolate exopolysaccharide-producing microorganisms from the various samples. A total of 7 different exopolysaccharide-producing microorganisms were isolated from the garden soil samples. Among them, two are Gram-positive and five Gram-negative bacterial isolates. These EPS-producing isolates were further studied for their colonical, morphological, and biochemical characteristics and various enzyme productions. The ability of EPS producers in various raw carbon sources i. e. Rice flour, wheat starch, horse gram starch, maize starch, and potato extract were also checked. These EPS producers were further studied for their EPS production ability as well as the characterization and quantification of EPS.

Keywords: Exopolysaccharides; Garden soil; Raw carbon sources; Physio chemical properties.

INTRODUCTION

Exopolysaccharides (EPS) are polysaccharides produced by bacteria and secreted into the external environment. EPS, natural polymers and having differential physical and chemical structures and properties. They have different prospective applications in food, pharmacy, cosmetics, agricultural, and having the advantages of being eco-friendly, nontoxic, and biodegradable. EPS in normal condition is thought to act as an important function like microbial cell desiccation, phagocytosis, phage attack, antibiotics or toxic compounds, predation by protozoans, osmotic stress, adhesion to solid surfaces, and in cellular recognition. In the food industry, they are used as viscosifiers, stabilizing or emulsifying agents, and gelling and texturizers[1]. EPS got more attention in the current era due to their unique structure and properties and the context in various fields of science, industry, medicine, and technology [2–7]. The term exopolysaccharide was first introduced by Sutherland in 1972[8] for high molecular weight polyhydroxy compounds produced by microorganisms. Bacterial exopolysaccharides

Author's Affiliation: [1]Assistant Professor, Department of Microbiology, Shri PHG Municipal Arts & Science College, Kalol, Gujarat 382721, India, [2]Lecturer, Department of Microbiology, HVHP Institute of Post Graduate Studies and Research, KSV Kadi, Gujarat, India.

Corresponding Author: Kalavati Prajapati, Assistant Professor, Department of Microbiology, Shri PHG Municipal Arts & Science College, Kalol, Gujarat 382721, India.

E-mail: kalavatiprajapati@gmail. com

Received on: 13-12-2024 **Accepted on:** 21-01-2025

have a diversified structure and are excreted by an extensive bacterium[9]. The site of location from where the EPS production take place, are divided into capsular polysaccharides that are correlated with the cell surface or extracellular environment [4,10]. EPSs synthesized by bacteria exploit over those from plants (cellulose, starch, and pectin), and animals (glycogen and chitin). Using the different optimizing conditions, and genetic and metabolic engineering, it is possible to modulate the yield as well as the structural and functional properties of bacterial EPSs.[10-13] Bacterial EPS are characterized by the presence of functional groups (hydroxyl, carboxyl, carbonyl, acetate, etc.), that enable them to alter their molecules to impart new valuable properties.[14] Therefore, recent interesting reviews by Aditya *et al.* and Aziz *et al.*, have presented the alteration of bacterial cellulose (BC) by chemical and physical methods to obtain nanocomposites and change materials with improved functionality for biomedical applications [14, 15]. Generally, EPS are classified into two types: homopolysaccharides (unbranched or branched) and composed of single-type monosaccharides (glucose and fructose linked through glycosidic bonds) and heteropolysaccharides, that includes two or more units of different monosaccharides (glucose, fructose, galactose, mannose, rhamnose, fucose, N-acetylglucosamine, and uronic acids).[5,9] Homopolysaccharides are divided into α-D-glucans (dextran, alternan, and reuteran), β-D-glucans (bacterial cellulose), fructans (levan and inulin), and polygalactans. Heteropolysaccharides (xanthan, alginate, hyaluronic acid, kefiran, and gellan).[5,16]

MATERIALS AND METHODS

Sample Collection

The soil samples were collected from the garden soils such as soils from citrus canker-affected plants as well as the ripened fruits.

Isolation and Screening

Ooze canker was performed from citrus cancer leaves, inoculated on Nutrient agar supplemented with 2% Glucose and 1% Calcium carbonate and Potato Dextrose Agar (PDA) plate, and incubated at 37 °C for 48 to 72 hours.

Colonially and morphologically distinct yeast isolates were selected for further study. Gram staining and Capsule staining were performed for all the isolates.

Enzyme spectrum

Selected organism isolates were studied for the production of various enzymes, such as Amylase, Lipase, Gelatinase, and Catalase.

Amylase, Protease, Lipase, and Gelatinase were studied by plate assay using Starch agar, Casein Agar, tributyrin agar, and Gelatin Agar, respectively.

Sugar Fermentation tests

Various Sugar i. e. Glucose, Galactose, Fructose, Maltose, Mannitol, Xylose, and Sucrose fermentation tests were studied. Nutrient sugar broth with Durham's Vial and Andrade's Indicator were inoculated and incubated at 37° C for 48 to 72 hours.

Exopolysaccharide Production

Exopolysaccharide production was checked by using Glucose yeast extract broth supplemented with 10% Glucose and 1% $CaCO_3$.

RESULTS AND DISCUSSION

Colonically and morphologically distinct exopolysaccharides were selected from the total 20 isolates and coded as EPS-1 to EPS-7.

Among 7 EPS producers, 2 isolates were Gram-positive bacilli and 5 were Gram-negative Short rods. Isolates EPS-1 and EPS-2 gave maximum viscous colonies on the PDA Agar plate.

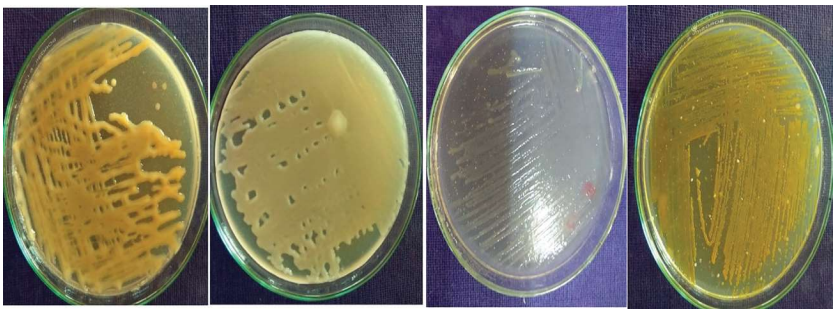

Fig. 1: Colony characters of EPS on the PDA plate

EPS-01	EPS-02	EPS-03	EPS-04

Table 1: Enzyme spectrum of EPS Producers

Enzyme	EPS-1	EPS-2	EPS-3	EPS-4	EPS-5	EPS-6	EPS-7
Amylase	+	+	+	+	+	+	+
Protease	+	+	_	_	_	+	+
Lipase	_	_	_	+	_	_	_

Table 2: Sugar Fermentation Test of EPS Producers

Sugar	EPS-1	EPS-2	EPS-3	EPS-4	EPS-5	EPS-6	EPS-7
Glucose	Acid gas	Acid gas	-	Acid	Acid gas	Acid gas	-
Fructose	Acid gas	Acid gas	Acid	Acid	Acid gas	Acid gas	-
Maltose	Acid gas	Acid gas	-	Acid	Acid gas	Acid gas	-
Sucrose	Acid gas	Acid gas	-	Acid	Acid gas	Acid gas	Acid
Xylose	Acid gas	Acid gas	-	Acid	Acid gas	-	-
Galactose	Acid gas	Acid gas	-	Acid	Acid gas	-	-
Mannitol	Acid gas	Acid gas	-	Acid	Acid gas	-	-

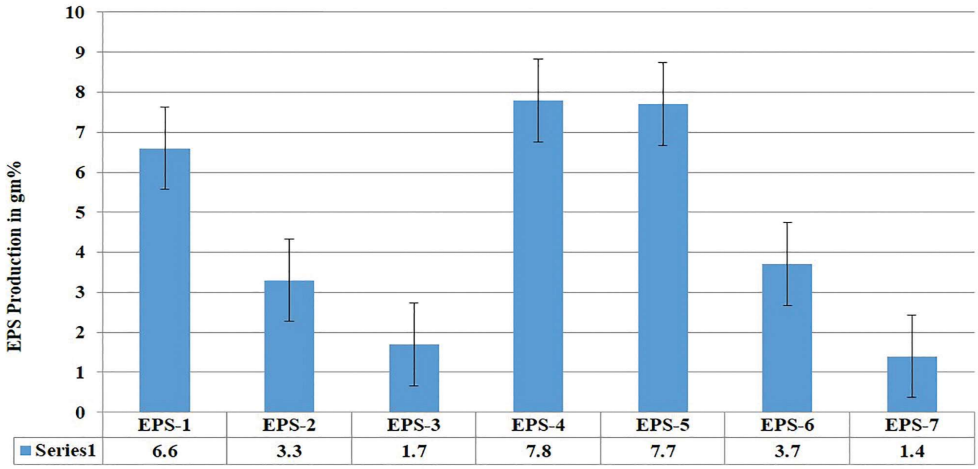

Fig. 2: EPS Production in Glucose Yeast Extract+ CaCo3 Broth

CONCLUSION

A total of 7 EPS Producers were selected from 20 isolates. Among them, 2 were Gram Positive and 5 were Gram Positive. All EPS Producers produce Amylase and Protease Enzyme. These isolate and fertilize the various types of sugars. Among all EPS producers Isolate EPS-4 and EPS-5 give higher production of EPS production by using glucose as a sole source of carbon. The further various types of raw carbon sources like rice flour, gram flour, maize flour, potato infusion, etc. would be studied.

ACKNOWLEDGEMENT

The research work is supported by the Department of Microbiology, HVHP Institute of Postgraduate Studies and Research, Kadi. We are

very thankful to our principal and director of the college.

Disclaimer (Artificial Intelligence)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (Chat GPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

REFERENCES

1. Sutherland I.W. (1994). Structure-function relationships in microbial exopolysaccharides. Biotechnol. Adv. 12: 393-448.
2. Ahuja, V.; Bhatt, A.K.; Banu, J.R.; Kumar, V.; Kumar, G.; Yang, Y.H.; Bhatia, S. K. Microbial Exopolysaccharide Composites in Biomedicine and Healthcare: Trends and Advances. Polymers 2023, 15, 1801. [CrossRef] [PubMed].
3. Kaur, N.; Dey, P. Bacterial Exopolysaccharides as Emerging Bioactive Macromolecules: From Fundamentals to Applications. Res. Microbiol. 2022, 29, 104024. [CrossRef] [PubMed].
4. Mahmoud, Y.A.G.; El-Naggar, M.E.; Abdel-Megeed, A.; El-Newehy, M. Recent Advancements in Microbial Polysaccharides: Synthesis and Applications. Polymers 2021, 13, 4136. [CrossRef] [PubMed].
5. Mohd Nadzir, M.; Nurhayati, R. W.; Idris, F. N.; Nguyen, M. H. Biomedical Applications of Bacterial Exopolysaccharides: A Review. Polymers 2021, 13, 530. [CrossRef] [PubMed].
6. Rana, S.; Upadhyay, L.S.B. Microbial exopolysaccharides: Synthesis pathways, types and their commercial applications. Int. J. Biol. Macromol. 2020, 157, 577–583. [CrossRef] [PubMed].
7. Barcelos, M.C.S.; Vespermann, K.A.C.; Pelissari, F.M.; Molina, G. Current status of biotechnological production and applications of microbial exopolysaccharides. Crit. Rev. Food Sci. Nutr. 2019, 60, 1475–1495. [CrossRef].
8. Osemwegie, O.O.; Adetunji, C.O.; Ayeni, E.A.; Adejobi, O.I.; Arise, R.O.; Nwonuma, C.O.; Oghenekaro, A.O. Exopolysaccharides from bacteria and fungi: Current status and perspectives in Africa. Heliyon 2020, 6, e04205. [CrossRef].
9. Sutherland, I.W. Bacterial exopolysaccharides. Adv. Microb. Physiol. 1972, 8, 143–213. [CrossRef].
10. Xiao, M.; Ren, X.; Yu, Y.; Gao, W.; Zhu, C.; Sun, H.; Kong, Q.; Fu, X.; Mou, H. Fucose-containing bacterial exopolysaccharides: Sources, biological activities, and food applications. Food Chem. X 2022, 13, 100233. [CrossRef].
11. Schmid, J.; Sieber, V.; Rehm, B. Bacterial exopolysaccharides: Biosynthesis pathways and engineering strategies. Front. Microbiol. 2015, 6, 496. [CrossRef].
12. Pereira, S.B.; Sousa, A.; Santos, M.; Araújo, M.; Serôdio, F.; Granja, P.; Tamagnini, P. Strategies to obtain designer polymers based on cyanobacterial extracellular polymeric substances (EPS). Int. J. Mol. Sci. 2019, 20, 5693. [CrossRef] [PubMed].
13. Manan, S.; Wajid Ullah, M.; Ul-Islam, M.; Shi, Z.; Gauthier, M.; Yang, G. Bacterial cellulose: Molecular regulation of biosynthesis, supramolecular assembly, and tailored structural and functional properties. Prog. Mater. Sci. 2022, 129, 100972. [CrossRef].
14. Aditya, T.; Allain, J.P.; Jaramillo, C.; Restrepo, A. M. Surface Modification of Bacterial Cellulose for Biomedical Applications. Int. J. Mol. Sci. 2022, 23, 610. [CrossRef] [PubMed].
15. Aziz, T.; Farid, A.; Haq, F.; Kiran, M.; Ullah, A.; Zhang, K.; Li, C.; Ghazanfar, S.; Sun, H.; Ullah, R.; *et al.* A Review on the Modification of Cellulose and Its Applications. Polymers 2022, 14, 3206. [CrossRef] [PubMed].
16. Angelin, J.; Kavitha, M. Exopolysaccharides from probiotic bacteria and their health potential. Int. J. Biol. Macromol. 2020, 162, 853–865. [CrossRef] [PubMed].