

Drug delivery applications and future prospects of microbial exopolysaccharides

Tioluwani Ibukun Adegbolagun^{1,B,D–E}, Olubusola Ayoola Odeniyi^{2,A,D–F}, Michael Ayodele Odeniyi^{1,A,D–F}

¹ Department of Pharmaceutics & Industrial Pharmacy, University of Ibadan, Nigeria

² Department of Microbiology, University of Ibadan, Nigeria

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

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Address for correspondence

Michael Ayodele Odeniyi

E-mail: deleodeniya@gmail.com

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Abstract

Over the years, exopolysaccharides (EPSs) have been utilized in various areas of research, including health, industry, environment, and agriculture, due to their flexible physical, chemical and structural properties that can be readily modified to suit desired purposes. Current research trends have shown that EPS production is dependent on numerous factors which can be combined to varying extent to optimize production yields. Although the majority of research is directed towards their industrial and medicinal uses, these chemical substances possess peculiar characteristics which are also exploited for biomedical research, where they are being used as drug delivery systems, some of which include their abundance in nature, biocompatibility, biodegradability, non-toxicity, and ability to efficiently encapsulate sensitive bioactive agents. However, despite the numerous beneficial prospects of microbial EPSs in drug delivery, there are limitations to the commercial production and industrial applications of these biopolymers. These limitations have inspired revolutionary research into the cost-effective production of safe EPSs polymers. In this review, we classify EPSs and discuss their methods of extraction and characterization. We also summarized current drug delivery applications and discussed limitations to extensive industrial commercialization of EPSs, while highlighting prospects for the utilization of microbial EPSs and implications for research.

Key words: exopolysaccharides, drug delivery, biopolymers

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Introduction

Exopolysaccharides

Exopolysaccharides (EPS) are extracellular metabolic by-products excreted by bacteria, yeast, fungi, etc.¹ They are high-molecular-weight polymeric biomolecules with glycosidic linkages connecting monosaccharide units that are synthesized by microbial cells and secreted to the outside of the cells, where they perform beneficial functions, including protection from extreme pH and temperature.² Exopolysaccharides are crucial components that determine the structural and functional integrity of biofilms, which provide microorganisms with protection from harsh environments.^{3,4} For example, xanthan, alginate and cellulose form a biofilm on bacterial cell surfaces that serves as a protective cover.⁵ Along with these biological functions, EPSs have distinct chemical and physical properties that make them suitable industrial raw materials.⁶

In agriculture, EPSs have been used to improve the flow of pesticides and uniformly disperse solid components in formulations; the improved rheology of the formulation also ensures surface cling.⁷ Exopolysaccharides are also used as gelling agents for culture media, paints and ink, and detergents.

Pharmacologically, EPSs have shown peculiar biological bioactivities which have found therapeutic applications in animal and human medicine.⁸ These activities have demonstrated potential for use in prophylactic and antibiotic therapies. Some of them include; anti-inflammatory activities, antimicrobial and anti-tumor activities, hypocholesterolemic activities, immunomodulatory activities, and anti-diabetic activities.^{9,10} Through technological advancement in immunological studies, EPSs have been used to develop capsular polysaccharide-based vaccines for infectious illnesses.¹¹ Microbial EPSs have been extensively studied for their applications in vaccine development. They show prospects as antigen delivery systems and as antigenic materials (as adjuvants) to provide more robust immune responses.¹²

Due to their non-toxic nature, EPSs are being used to re-fashion international treatment models by modifying drug therapy, surgical therapy and disease diagnosis; this forms the basis of their biomedical applications.¹³ They are versatile biomedical applications because they have flexible physical, chemical and structural properties that can be readily modified to suit desired purposes. Furthermore, they have published and standardized methods of fermentation and isolation, which makes them suitable to meet global demands. As a result, EPSs have found numerous applications in tissue engineering and drug delivery. They have also been used as surgical sealants and coating materials for medical devices.¹⁴

The most notable biomedical application of EPSs is in the delivery of sensitive drug molecules and biomolecules. Exopolysaccharides have shown potential as drug delivery materials due to their capacity to effectively entrap

bioactive agents. Through this application, they are used to improve the therapeutic efficacy of drugs and increase the drug shelf life and duration of pharmacological action.^{14,15} The versatile applications of EPSs in drug delivery applications are owed to their abundance in nature, biocompatibility, biodegradability, and non-toxicity.¹

Classifications and structure of exopolysaccharides

Due to differences in their chemical components which influence their physical, biological, chemical and functional properties, EPSs are a diverse and complex class of biomolecules.⁶ Based on their chemical composition, EPSs are primarily categorized according to their monomeric content into homopolysaccharides (HoPs), which contain one type of sugar monomer unit, and (HePs) which have several recurring subunits of different sugars from disaccharides to heptasaccharides, monosaccharide derivatives and/or substitutes.^{16,17} This classification is summarized in Fig. 1.

To make further distinctions within the major categorizations, EPSs are subclassified based on the nature of their monomeric units, linkages and the degree of branching.¹⁶

Homopolysaccharides

Homopolysaccharides are identified based on monosaccharide units and classified as glucans, fructans and galactans, according to the presence of sugars (D-glucose, D-fructose and D-galactose) in their backbones.^{17,18} Glucans exhibit varying degrees of branching and monomeric linkages; they are further classified into 1) α -D-glucans (alternan, reuteran, dextran, and mutan), where D-glucose residues are linearly or non-linearly interlinked by α -1-2, α -1-3, α -1-4 and/or α -1-6 glycosidic bonds; and 2) β -D-glucans (cellulose and curdlan), comprising of D-glucose connected by β -1-3 bonds and β -1-2 branching.⁶ Due to these different linkage patterns, glucans exhibit varying degrees of water solubility and viscosity. Fructans are water-soluble and are grouped into 1) levan with β -2-6 bonds and 2) inulin with β -2-1 bonds.¹⁹ Galactans, although less abundant, are also water-soluble and consist of α -1-6 linked galactose monomers.¹⁷

Heteropolysaccharides

Heteropolysaccharides comprise repeating sugar units at varying ratios which may include D-xylose, D-ribose and D-arabinose, which are pentose sugars, and D-galactose, D-mannose and D-glucose, which are hexose sugars. The HePs also include uronic acids such as D-galacturonic acids and N-glucuronic acid. Other HePs are acetylated monosaccharide units, including N-acetyl-glucosamine and N-acetyl-galactosamine.^{20,21} They may also be branched or unbranched, and contain non-sugar substituents (organic or inorganic) such as phosphate, acetyl and glycerol.²²

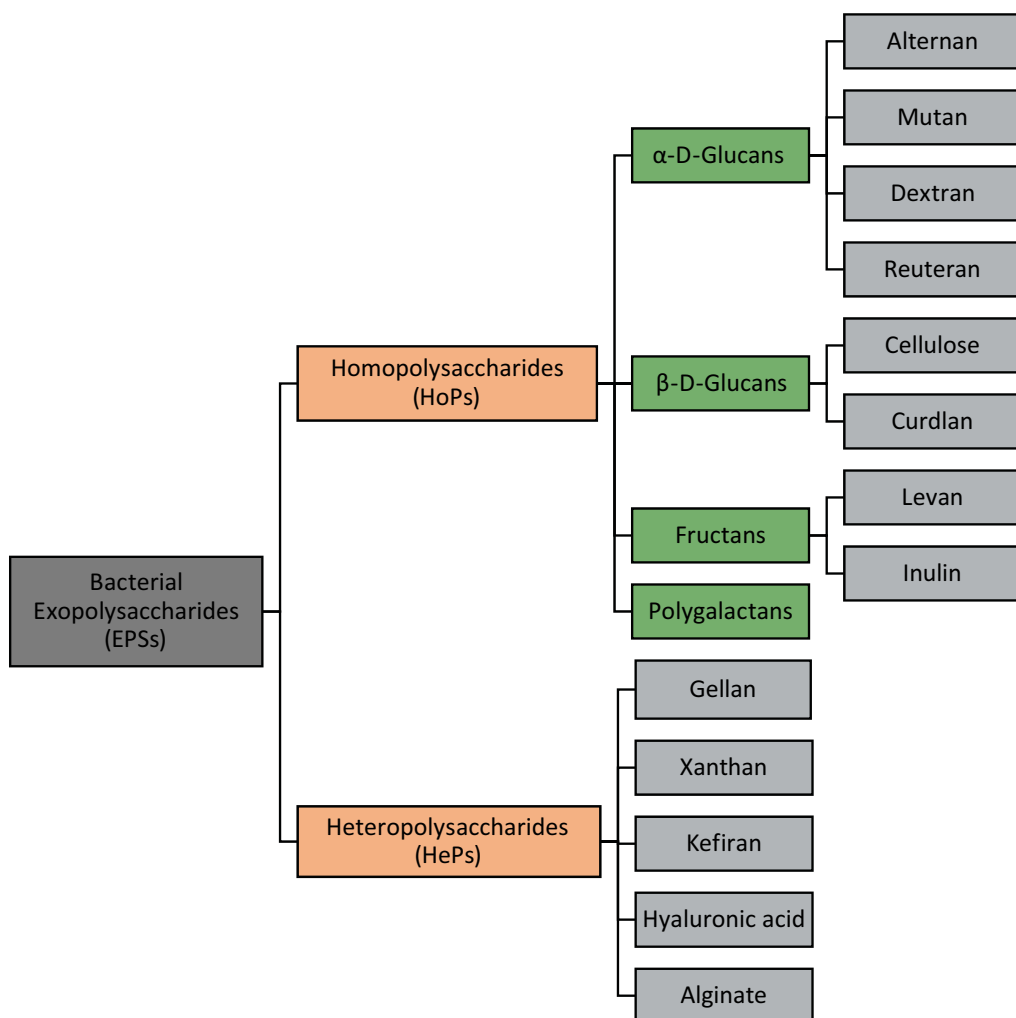


Fig. 1. Classifications and examples of bacterial exopolysaccharides¹⁴

The linkages between monomeric units in polymers are rigid 1,2- α - or 1,6- α - and 1,4- β - or 1,3- β - linkages, which are more flexible.¹⁶ Examples include gellan, xanthan, kefiran, hyaluronic acid, and alginate.^{1,18}

Exopolysaccharides can also be identified by their degree of association with the microbial cell wall and where their synthesis occurs. Based on the degree of attachment, they can be broadly grouped as: 1) capsular EPSs, which are firmly attached to the cell surface, creating a resistance mechanism against attacks from phagocytes and bacteriophages, and protection against excessive water loss and osmotic stress; and 2) slime EPSs, which are loosely associated with the cell surface and can be easily isolated. Depending on the location of synthesis, extracellular EPSs are produced outside the cell envelope, whereas intracellular polysaccharides are accumulated inside the cells and released outside the cells.^{1,23}

Microorganisms producing exopolysaccharides

Microorganisms that produce EPSs are abundant in nature and have been identified in numerous environments in the ecosystem, especially those with increased

carbon–nitrogen ratios.^{24,25} Exopolysaccharides can therefore be extracted from bacterial, fungal and archaeobacterial sources (such as algae).^{13,26}

Some bacteria genera that produce EPSs are *Acetobacter* (cellulose), *Agrobacterium* (curdlan), *Bacillus* (levan), *Brenneria*, *Geobacillus*, *Gluconacetobacter* (cellulose, levan), *Halomonas* (levan), *Lactobacillus* (dextran, kefiran), *Rhizobium* (curdlan), *Saccharomyces*, *Sarcina*, *Pseudomonas* (alginate, gellan, cellulose, gellan), *Streptococcus* (levan, hyaluronic acid), *Xanthomonas* (xanthan), and *Zymomonas* (levan).^{18,27} Some probiotic bacteria, such as *Lactobacillus* (dextran), *Leuconostoc* (dextran), *Lactococcus*, *Bifidobacterium* (hyaluronic acid), *Streptococcus* (levan, hyaluronic acid), and *Enterococcus* have also been used to synthesize EPSs for numerous medical and biomedical applications.^{1,14} Other bacterial sources include marine bacteria such as *Vibrio diabolicus* (hyaluronian-like EPS) and *Arthrospira platensis*, and extremophiles such as *Bacillus thermantarcticus* and *Geobacillus sthermodenitrificans*.^{13,16}

Due to their cellular and structural complexity, fungi are considered to be major non-bacterial sources of EPSs. Major fungi from which EPSs are isolated include *Aspergillus fumigatus* (galactosaminogalactan), *Candida albicans*

(chitosan, chitin) and *Zygosaccharomyces rouxii* (chitosan, chitin). Other examples of EPS-producing fungi are *Botryosphaeria* sp. (botryosphaeran), *Pleurotus ostreatus* (pleuran), *Aureobasidium pullulans* (pullulan), *Schizophyllum commune* (schizophyllan), *Phellinus linteus*, *Ganoderma lucidum*, *Fusarium* sp., and *Inonotus obliquus*.¹³ Exopolysaccharides have also been extracted from surface biofilms of archaea that grow at extreme temperatures and salt concentrations: *Thermococcus*, *Sulfolobus*, *Archaeoglobus fulgidus*, and *Thermococcus litoralis*.²⁸

Production of exopolysaccharides

Biosynthesis of exopolysaccharides

Among various species of EPS-producing microorganisms, the biosynthetic pathways involved in EPS biosynthesis are notably similar, regardless of their different characteristics.⁶ In bacteria, the synthesis of EPS occurs through intricate pathways that consist of specialized enzymes, carriers and transporter proteins²⁹: They are the intracellular and extracellular pathways.¹ Depending on their natures, bacteria EPSs can either be produced entirely outside by extracellular bacterial enzymes, or synthesized inside the cells and released into the cell surrounding.³⁰

Through the intracellular biosynthetic pathway, extracellular sugar residues are taken into the cells, broken down into various monomers, polymerized and transported out

of the cells through a membrane-bound lipid carrier protein.³¹ The extracellular biosynthetic pathway occurs outside the cell when sugar molecules are lysed into uniform monomeric units which are assembled, with glycosyl transferases enzymes or fructosyl transferases, into polymers that are secreted into the surrounding environment.^{1,19} Although the extracellular pathway is majorly utilized in the production of HoPs, either of both biosynthetic pathways are involved in the synthesis of HePs.^{32,33}

In general, there are 4 mechanisms involved in the biosynthesis of EPSs in bacterial cells¹⁴: the Wzx/Wzy-dependent pathway; the ABC transporter-dependent pathway; the synthase-dependent pathway; and extracellular biosynthesis by sucrose protein.^{34–36} These mechanisms are illustrated in Fig. 2.

1. Wzy/Wzy-dependent pathway: At the inner membrane, imported monomeric sugar molecules are converted into sugar nucleotides and linked to a lipid carrier.³⁷ Via the addition of more sugar units by glycosyltransferases, the sugar chain is extended and transported, as an oligosaccharide unit, through the membrane by Wzx flippase.³⁸ The unit is then enzymatically modified, and the Wzy protein polymerizes the altered oligosaccharide into polysaccharides, which are released into the cell exterior by outer membrane polysaccharide transporters.^{14,39}

2. ABC transporter dependent pathway: This pathway is employed in the synthesis of capsular EPSs⁴⁰ and it is similar to the Wzy/Wzy-pathway but excludes the activities

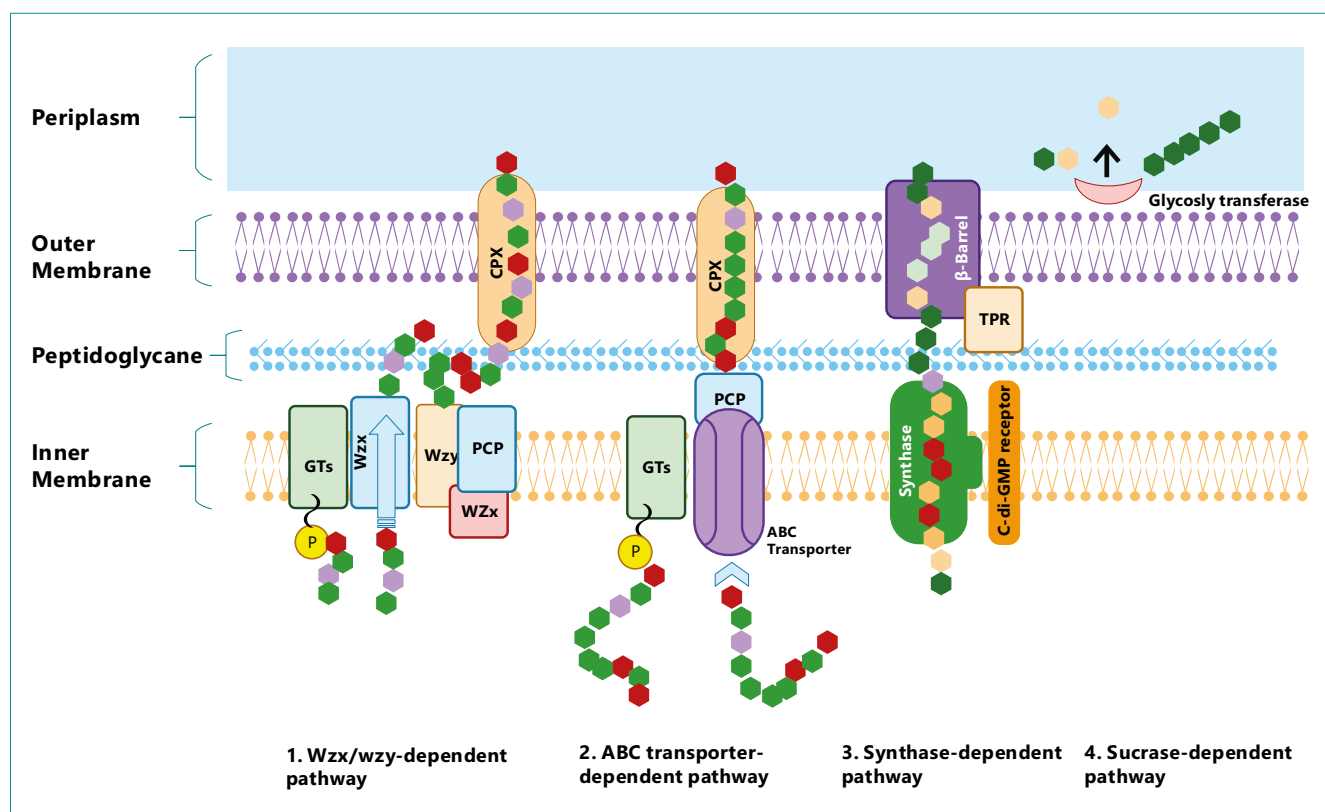


Fig. 2. Four major biosynthetic pathways of bacterial exopolysaccharides¹⁴

PCP – polysaccharide co-polymerase; OPX – outer membrane polysaccharide export; TPR – tetratricopeptide repeat protein; GTs – glycosyltransferase.

Table 1. Optimized exopolysaccharide production medium and incubation conditions for some of the microorganisms and the methods of optimization employed

Microorganism	Production medium	Incubation condition			Method of optimization	Reference
		temp. [°C]	pH	time		
<i>Alkalibacillus</i> sp. w3	Horikoshi I broth media	35	10	7 days	OFAT PBD	44
<i>Lactobacillus acidophilus</i>	MRS	30	6	48 h	PBD BBD	45
<i>Lactobacillus plantarum</i> R301	MRS medium	27	7.4	24 h	OFAT PBD CCD	46
<i>Enterobacter cloacae</i> MBB8	modified minimal medium	28	7.2	24 h	PBD	48
<i>Tuber borchii</i>	yeast extract medium	25	7	28 days	OFAT	49
<i>Fusarium proliferatum</i>	malt extract broth	25	6	9 days	PBD	50
<i>Pseudomonas aeruginosa</i> MTCC 1688	sterile nutrient broth medium	32	6	96 h	OFAT	51

OFAT – one-factor-at-a-time; PBD – Plackett-Burman design; BBD – Box-Behnken design; CCD – central composite design; MRS – De Man–Rogosa–Sharpe medium.

of Wzx flippase and Wzy protein. Polysaccharide chains synthesized by glycosyltransferases from nucleotide sugar units are transported across the inner membrane via a tripartite efflux pump complex – the (ATP)-binding cassette (ABC) transporter.⁴¹

3. Synthase-dependent pathway: In this pathway, same-type monosaccharide units are arranged into undecaprenyl phosphate glucose units by synthase protein located in the membrane.⁴²

4. Extracellular synthesis by sucrose enzyme: This extracellular pathway occurs in the periplasm where sucrose is converted, outside the outer membrane, into its monosaccharide components (glucose or fructose) by sucrose enzymes. The monomer units are polymerized (with varying degrees of branching) by glycosyltransferases to form glucans or fructans – glucose and fructose units, respectively.^{14,43}

Heteropolysaccharides are mainly produced through the ABC transporter-dependent and Wzx/Wzy-dependent pathways, while HoPs – via the extracellular production and synthase-based pathways.¹

Fermentation of microorganisms: Production media and incubation conditions

The quality and quantity of EPSs are determined by several factors; bacteria strains, nutrient level and makeup of the growth/production media (salt concentration and the carbon:nitrogen composition) and incubation conditions (such as time, pH and temperature).^{14,44} Consequently, EPS production is subject to numerous non-microbial factors which can be combined and permuted to varying degrees to achieve different production yields and characteristics of EPSs. This complicates the selection process of production media and incubation

conditions for EPS production, inspiring the research into predicting the best combinations and arrangement of culture conditions (variables) to maximize production yields using one-factor-at-a-time (OFAT) optimizations and statistical experiment designs such as response surface methodology (RSM).^{44,45} The OFAT optimizations involve testing each variable, one at a time while holding other variables constant, to determine which variable has the most effect on EPS production. The RSM statistically analyzes several independent variables to predict their relationship with each other and their effects on production yield⁴⁵; it is being extensively employed by researchers to optimize the production of EPSs in microbes using experimental designs including Plackett–Burman Design (PBD), Box–Behnken Design (BBD) and Central Composite Design (CCD).^{46,47} Table 1 summarizes some findings of recent research works on the optimization of production factors to improve EPS production from microbial sources.^{44–46,48–51}

Extraction and recovery of exopolysaccharides

The recovery process of EPSs includes concentration, isolation and purification. After EPSs are concentrated in the fermentation broths, they are isolated from microorganisms by centrifugation.⁴⁵ More extreme methods are boiling in saline solution, sonication and autoclaving. Isolated EPSs are mostly extracted from isolated mixtures by solvent precipitation via the addition of a polar organic (such as ethanol) that reduces the solubility of EPSs in water.^{45,46} Desalting and deproteinization treatments may be further carried out to purify precipitated polysaccharides. The resultant precipitate is collected by centrifugation, filtration or sedimentation and dried under pressure.^{24,25}

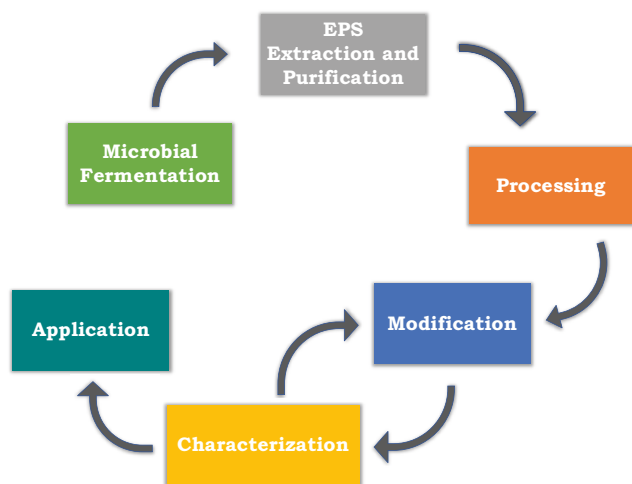


Fig. 3. Schematic process flowchart of microbial exopolysaccharides

To further separate sugars and minute proteins, the collected precipitate may undergo membrane filtration after dissolution in water. The filtrate is then freeze-dried to get the EPS powder which is packaged for use.^{17,52,53} Anion exchange chromatography and size exclusion anion exchange are additional steps of purification employed in advance separation processes.^{54,55}

Oftentimes, extracted EPSs are utilized in the same form as they are extracted (native form). They may also be physically or chemically treated before their use.²⁴ These treatments are applied as a means to modify the characteristics of these polysaccharides, thereby optimizing their suitability for pharmaceutical, food and biomedical purposes (Fig. 3).

Characterization of exopolysaccharides

Extensive characterization of extracted EPSs requires the evaluation of monosaccharide content, ring formations, extent of branching, and molecular weight. However, because these parameters cannot be accurately evaluated with singular methods, multiple methods and techniques are employed in combination.⁶ Some of these parameters and respective determination methods are summarized in Table 2.^{34,44,45,48,49,56,57}

Table 2. Some of the parameters used to characterize exopolysaccharides and methods of determination

Parameter	Determination methods	Reference
Molecular weight	gel permeation chromatography	49
	ion exclusion chromatography	56
Monosaccharide composition	high-performance liquid chromatography-refractive index	34
	high-performance ion exchange chromatography-pulse amperometry detection	45
	gas chromatography-mass spectrometry	48
Bond types and chemical groups	Fourier transform infrared spectroscopy	44
	X-ray diffraction	49
Glycosidic linkages and ring and anomeric formation	nuclear magnetic resonance	57
Extraction yield	phenol-sulfuric acid method	44
	gravimetric analysis	45

Drug delivery applications of exopolysaccharides

The EPSs have shown potential as drug delivery materials for the delivery of sensitive drug molecules and biomolecules due to their bioactive function and capacity to hold bioactive agents effectively. Although they have similar functions as drug delivery systems, they are easier to apply as drug carriers than to produce as biological scaffolds of viable cells. This is because they can be readily modified to improve therapeutic efficacy by controlling and targeting the release of drugs in body tissues and protecting drug molecules from harsh physical and biological environments to increase the shelf life and duration of pharmacological action.^{14,15} In addition to their abundance in nature, biocompatibility, biodegradability and non-toxicity are the main factors responsible for the ubiquitous drug delivery applications of EPSs.¹

Nano- and micro-delivery systems

The development of nano- and microparticles is arguably the most significant application of EPSs in the delivery of drugs. The benefits of these include increased efficacy and decreased toxicity of encapsulated drugs. Levan nanoparticles exhibit decreased intestinal toxicity of selenium, iron and cobalt.⁵⁸ Serum bovine encapsulated by Sezer et al. exhibited zero-order drug release kinetics.⁵⁹ Hydrogel nanoparticles of curdlan derivatives have shown potential in the delivery of anticancer drugs.⁶⁰ Because EPSs are bio-compatible and non-toxic, they offer more benefits in the green synthesis of silver nanoparticles compared to synthetic polymers.⁶¹

Transdermal drug delivery systems

Exopolysaccharides are also utilized in the development of transdermal drug delivery systems (TDDS). Researchers postulate hyaluronic acid is the most commonly used marine EPS drug delivery via the transdermal route.⁶² Alginate is also frequently applied in TDDS to prepare microneedles (which have been used to deliver vaccines, bovine serum

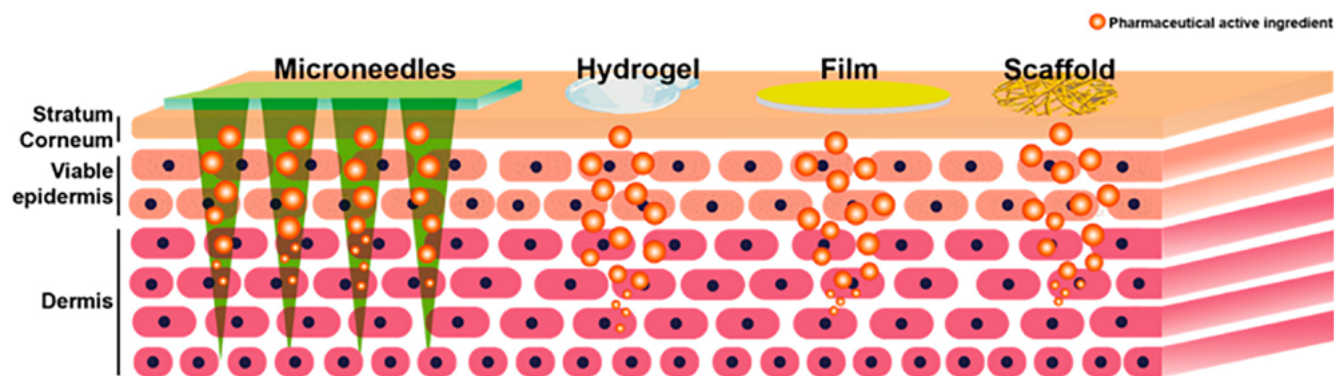


Fig. 4. Four types of recent transdermal drug delivery vehicles⁶⁶

proteins and insulin) and nanoparticles (in combination with chitosan) for target antibiotic therapy of cutaneous pathogens.^{63–65} Transdermal drug delivery systems made with EPSs are constantly evolving. Recent TDDSs which are illustrated in Fig. 4 vehicles include⁶⁶:

1. Hydrogels: These are hydrophilic, polymeric networks which are 3-dimensional and have large water-holding capacities. A notable use is “smart” hydrogels that regulate molecule release by responding to changes in external stimuli.⁶⁶

2. Films: These are thought to be a good substitute for topical patches and traditional topical dosage forms due to their flexibility, transparency, non-occlusivity, and ease of use because they are non-invasive. They also demonstrate higher drug loading capacity and prolonged drug release and drug retention on the skin.^{67,68} The film-forming ability of EPSs makes them particularly suitable candidates in the formulation of these delivery vehicles.

3. Microneedles: These are efficient, minimally invasive vehicles of transdermal drug delivery, which typically create micro-channels through the layers of the skin and release the drugs, thereby localizing drug delivery and drug effects.⁶⁹ Biodegradable polymers such as hyaluronic acid, alginate and cellulose are more beneficial as microneedle components than silicon and metals because they are non-biodegradable.⁷⁰

4. Tissue scaffolds: Polysaccharides are used to enhance the mechanism of drug release and improve the performance of tissue scaffolds. Hydrogels are frequently used as tissue engineering scaffolds, and those based on alginate exhibit better mechanical strength and controlled drug release.⁷¹

Modifications to improve drug delivery

Bacterial EPSs can be modified in a variety of ways to customize drug delivery properties and enhance the functionality of the delivery systems. Altering the side chain and functional group composition via chemical reactions of cross-linking reactions is one way of modification.⁷² Partial sulfation of curdlan at the O-6 portion improves the aqueous solubility of curdlan hydrogel nanoparticles; however, the extent of substitution affects the thermal stability and ability to cross-link with polycytidylic acid.^{60,73}

Exopolysaccharides are also combined with other polysaccharides, biopolymers or synthetic polymers to optimize desirable material characteristics of these polymers and improve upon their limitations. This combination results in polymer blends with high-performance characteristics capable of being tailored toward various novel drug delivery applications. A study by Inoue et al. demonstrated the ability of cellulose membrane to increase the release of chlorhexidine (which complexed with β -cyclodextrin) by 10-fold; this was postulated to be a result of rigid chemical interactions between cellulose and the chlorhexidine- β -cyclodextrin complex.⁷⁴ In vitro studies of ciprofloxacin microspheres demonstrated adequate gastric protection of ciprofloxacin by kefirin-alginate polymer blends.⁷⁵ Similarly, xanthan hydrogels modified with succinic anhydride extended the release of gentamycin over 9 days.⁷⁶

Future prospects for the use of microbial exopolysaccharides

The spectrum of future EPS applications is as wide as the extent to which their functionality can be improved. Consequently, EPS-based drug delivery systems show promise in revolutionizing the paradigm of drug delivery.¹² Diverse works of research have been dedicated to unearthing the untapped drug delivery potentials of EPSs.

Temperature- and pH-sensitive formulations

In transdermal delivery systems, they are capable of being modified with nanotechnology to prepare “smart” formulations which release entrapped drug molecules in response to temperature and pH changes at target delivery sites.⁶⁶ Skin conditions induced by pH disparity have been treated with isoliquiritigenin-loaded hydroxyethyl cellulose – hyaluronic acid pH-sensitive hydrogels.⁷⁷ In combination, hydrogels sensitive to temperature and pH have been developed; gallic acid loaded within Pluronic® F-127 bi-responsive hydrogels of chitosan oligosaccharide lactate and hyaluronic acid nano-conjugates have been used to resolve atopic dermatitis.⁷⁸ In addition to the potential of improved clinical therapy in preventing infections and

improving wound healing, these “smart” formulations have the benefit of being cost-effective.⁶⁶

Simultaneously, in systemic delivery systems, EPSs show promise as substrates for “Intelligent Drug Delivery Systems”.⁷⁹ The fungal EPS pullulan which, due to its recurring hydroxyl groups, is especially amenable to derivatization through chemical means, has been grafted on the backbones of poly-(N-isopropyl-acrylamide)-co-acrylamide and succinic carboxyl ether groups in modulating the release of lysozyme protein using pH and temperature.⁸⁰ Similarly, carboxy-methyl pullulan modified with polyetheramine has demonstrated sol–gel transition at physiological temperature, providing assurance for thermally regulated sustained drug delivery.⁸¹

From current research in drug delivery, microbial EPSs show potential for application in targeted gastrointestinal delivery to the intestines. A typically useful feature of EPSs is their swelling and shrinking tendencies in response to changes in their environment; swelling increases the porosity of the polymers, thus triggering the release of contained drugs. Due to this sensitivity, restriction of drug release to the upper section of the small intestine is possible. This is because the majority of EPSs that are sensitive to pH shrink in mediums with low pH and swell in environments with high pH.⁸² Therefore, such oral delivery systems withhold the release of the drugs till the upper intestine, where swelling occurs due to the high pH.⁸³ Exopolysaccharides are also being modified to improve target drug release even in an acidic medium. Similarly, non-starch EPSs that are highly stable and resistant to digestion all through the gastrointestinal tract (GIT) remain intact till the colon, where the colonic bacterial flora degrades them. This implies a possibility for target drug delivery to the colon.⁸⁴

Interpenetrating polymer network

Exopolysaccharide hydrogels may also have the potential for molecular-scale penetration and drug delivery.¹² This is demonstrated by the entrapment and the extended-release properties shown by the interpenetrating polymer network (IPN) of alginate and polymers with an anti-inflammatory agent (indomethacin), an antibiotic (gatifloxacin), anticancer agent (5-fluorouracil), and an anticoagulant (heparin).⁸⁵ Steroid hormones are bioactive agents that stand to benefit significantly from this potential, as they are hardly mentioned in recent EPS research. Micro- and nano-transdermal delivery systems can bypass the side effects associated with their oral administration to improve their therapeutic efficacy.¹²

Discussion

Despite the numerous beneficial prospects of microbial EPSs and their composites in drug delivery, there are limitations to the commercial production and industrial

applications. The most apparent of these limitations is the high cost associated with production and extraction, which is non-commensurate with their commercial value.¹⁶ This is consequent to the complexities of their cost-intensive fermentation, isolation, purification, and characterization processes; reconciliation of these with the often-minimal production output of EPSs shows the poor economic feasibility and sustainability of large-scale EPS production. Another limitation is the relative paucity of knowledge about the interconnection between structure and biological functions.⁹ This is particularly complicated for bacterial EPS by the realization that the structure and biological effects of EPSs vary with every strain of bacteria.¹⁴ There is also the concern of health risks associated with certain microbial sources. For instance, the most studied microbial source of alginate is *Pseudomonas aeruginosa*, a bacteria linked to numerous infections with severity ranging from mild to severe. Although genetically engineered *P. aeruginosa* strains that are not pathogenic have been developed by researchers such as Valentine et al.,⁸⁶ there is still a significant proportion of un-engineered microbial sources. Bacterial capsular EPSs have been found to possess surface antigens that elicit immunogenic responses. This contributes to the pathogenicity risks of microbial EPSs.⁸⁷ Therefore, it would not be quixotic to envision a revolution in the production processes of EPSs as a prospect of microbial EPSs. The main objective of the revolution would be to enhance commercialization by optimizing the production yield and quality of microbial EPSs while minimizing their cost of production. There should be more research studies on approaches to achieve this, such as methods to optimize industrial-scale fermentation and extraction processes,¹⁴ as well as on developing methods of artificial chemical synthesis to aid the modification and enhancement of structural and functional properties of EPSs,⁸⁸ utilization of less expensive production materials (living and non-living), development of manipulation methods (mutagenesis, genetic and metabolic) of producing microbial strains with higher yields,³⁸ and less pathogenicity. All these will ultimately result in a significant improvement in the commercial value of these biomaterials and, consequently, their availability for various industrial and biomedical applications.


Specifically, these developments promise a significant impact in Africa by improving the current perspective on the utilization potentials of EPSs. A random preliminary study by Osemwegie et al. to evaluate the course of global research on EPSs over a period of 18 years was used to provide an overview of African research on EPS applications. The review showed that Africa ranked low among the continents with prominent research in microbial EPS research and application. These authors related the small number of publications to the low interest of African countries in the numerous industrial benefits of EPSs despite the possibility of Africa being one of the best repositories of EPSs in the world.¹³


Conclusions


As the commercial value and industrial applications of microbial EPSs increase, there is sure to be a corresponding paradigm shift in African research on the potential applications and benefits of these biomaterials.

ORCID iDs

Tioluwani Ibukun Adegbolagun

 <https://orcid.org/0000-0002-9442-2639>

Olubusola Ayoola Odeniyi  <https://orcid.org/0000-0002-0826-791X>

Michael Ayodele Odeniyi  <https://orcid.org/0000-0002-9918-4377>

References

1. Angelin J, Kavitha M. Exopolysaccharides from probiotic bacteria and their health potential. *Int J Biol Macromol*. 2020;162:853–865. doi:10.1016/j.ijbiomac.2020.06.190
2. Manjanna K, Shivakumar B, Pramodkumar T. Natural exopolysaccharides as novel excipients in drug delivery: A review. *Arch Appl Sci Res*. 2009;1(2):230–253. [https://sphinxnsai.com/sphinxnsaiVol_2No.1/ChemTech_Vol_2No.1/ChemTech_Vol_2No.1.PDF/CT=82%20\(509-525\).pdf](https://sphinxnsai.com/sphinxnsaiVol_2No.1/ChemTech_Vol_2No.1/ChemTech_Vol_2No.1.PDF/CT=82%20(509-525).pdf). Accessed September 15, 2023
3. Flemming H, Wingender J, Griebe T, Mayer C. Physico-chemical properties of biofilms. In: Evans LV, ed. *Biofilms: Recent Advances in Their Study and Control*. Boca Raton, USA: CRC Press; 2000:19–34. doi:10.1201/9781482284157
4. Fulaz S, Vitale S, Quinn L, Casey E. Nanoparticle–biofilm interactions: The role of the EPS matrix. *Trends Microbiol*. 2019;27(11):915–926. doi:10.1016/j.tim.2019.07.004
5. Kodali VP, Das S, Sen R. An exopolysaccharide from a probiotic: Biosynthesis dynamics, composition and emulsifying activity. *Food Res Int*. 2009;42(5–6):695–699. doi:10.1016/j.foodres.2009.02.007
6. Jurášková D, Ribeiro SC, Silva CCG. Exopolysaccharides produced by actin acid bacteria: From biosynthesis to health-promoting properties. *Foods*. 2022;11(2):156. doi:10.3390/foods11020156
7. Majee S, Avlani D, Biswas G. Rheological behavior and pharmaceutical applications of bacterial exopolysaccharides. *J App Pharm Sci*. 2017;7(9):224–232. doi:10.7324/JAPS.2017.70931
8. Lima Santos F, Modesto De Amorim G. Biotechnological challenges and perspectives of using exopolysaccharides. *J Anal Pharm Sci*. 2018;7(3):264–266. doi:10.15406/japlr.2018.07.00236
9. Hussain A, Zia KM, Tabasum S, et al. Blends and composites of exopolysaccharides: Properties and applications. A review. *Int J Biol Macromol*. 2017;94:10–27. doi:10.1016/j.ijbiomac.2016.09.104
10. Patel S, Majumder A, Goyal A. Potentials of exopolysaccharides from lactic acid bacteria. *Indian J Microbiol*. 2012;52(1):3–12. doi:10.1007/s12088-011-0148-8
11. Flemming HC, Neu TR, Wozniak DJ. The EPS matrix: The “house of biofilm cells.” *J Bacteriol*. 2007;189(22):7945–7947. doi:10.1128/JB.00858-07
12. Moscovici M. Present and future medical applications of microbial exopolysaccharides. *Front Microbiol*. 2015;6:1012. doi:10.3389/fmicb.2015.01012
13. Osemwegie OO, Adetunji CO, Ayeni EA, et al. Exopolysaccharides from bacteria and fungi: Current status and perspectives in Africa. *Heliyon*. 2020;6(6):e04205. doi:10.1016/j.heliyon.2020.e04205
14. Mohd Nadzir M, Nurhayati RW, Idris FN, Nguyen MH. Biomedical applications of bacterial exopolysaccharides: A review. *Polymers (Basel)*. 2021;13(4):530. doi:10.3390/polym13040530
15. Rahbar Saadat Y, Yari Khosroushahi A, Pourghassem Gargari B. A comprehensive review of anticancer, immunomodulatory and health beneficial effects of the lactic acid bacteria exopolysaccharides. *Carbohydr Polym*. 2019;217:79–89. doi:10.1016/j.carbpol.2019.04.025
16. Nwodo U, Green E, Okoh A. Bacterial exopolysaccharides: Functionality and prospects. *Int J Mol Sci*. 2012;13(12):14002–14015. doi:10.3390/ijms131114002
17. Torino MI, Font De Valdez G, Mozzi F. Biopolymers from lactic acid bacteria: Novel applications in foods and beverages. *Front Microbiol*. 2015;6:834. doi:10.3389/fmicb.2015.00834
18. Chaisuwan W, Jantanasakulwong K, Wangtueai S, et al. Microbial exopolysaccharides for immune enhancement: Fermentation, modifications and bioactivities. *Food Biosci*. 2020;35:100564. doi:10.1016/j.fbio.2020.100564
19. Zannini E, Waters DM, Coffey A, Arendt EK. Production, properties, and industrial food application of lactic acid bacteria-derived exopolysaccharides. *Appl Microbiol Biotechnol*. 2016;100(3):1121–1135. doi:10.1007/s00253-015-7172-2
20. Daba GM, Elnahas MO, Elkhateeb WA. Contributions of exopolysaccharides from lactic acid bacteria as biotechnological tools in food, pharmaceutical, and medical applications. *Int J Biol Macromol*. 2021;173:79–89. doi:10.1016/j.ijbiomac.2021.01.110
21. Mozzi F, Vaningelgem F, Hébert EM, et al. Diversity of heteropolysaccharide-producing lactic acid bacterium strains and their biopolymers. *Appl Environ Microbiol*. 2006;72(6):4431–4435. doi:10.1128/AEM.02780-05
22. Ruas-Madiedo P, Hugenholtz J, Zoon P. An overview of the functionality of exopolysaccharides produced by lactic acid bacteria. *Int Dairy J*. 2002;12(2–3):163–171. doi:10.1016/S0958-6946(01)00160-1
23. Ruas-Madiedo P, De Los Reyes-Gavilán CG. Invited review: Methods for the screening, isolation, and characterization of exopolysaccharides produced by lactic acid bacteria. *J Dairy Sci*. 2005;88(3):843–856. doi:10.3168/jds.S0022-0302(05)72750-8
24. Suresh Kumar A, Mody K, Jha B. Bacterial exopolysaccharides: A perception. *J Basic Microbiol*. 2007;47(2):103–117. doi:10.1002/jobm.200610203
25. Morin A. Screening of polysaccharide-producing microorganisms, factors influencing the production and recovery of microbial polysaccharides. In: Dumitriu S, ed. *Polysaccharides: Structural Diversity and Functional Versatility*. 2nd ed. Boca Raton, USA: CRC Press; 2004:275–296. doi:10.1201/9781420030822
26. Rodríguez-Carmona E, Villaverde A. Nanostructured bacterial materials for innovative medicines. *Trends Microbiol*. 2010;18(9):423–430. doi:10.1016/j.tim.2010.06.007
27. Yildiz H, Karatas N. Microbial exopolysaccharides: Resources and bioactive properties. *Process Biochem*. 2018;72:41–46. doi:10.1016/j.procbio.2018.06.009
28. Poli A, Di Donato P, Abbamondi GR, Nicolaus B. Synthesis, production, and biotechnological applications of exopolysaccharides and polyhydroxyalkanoates by archaea. *Archaea*. 2011;2011:693253. doi:10.1155/2011/693253
29. Patten DA, Laws AP. *Lactobacillus*-produced exopolysaccharides and their potential health benefits: Z review. *Beneficial Microbes*. 2015;6(4):457–471. doi:10.3920/BM2014.0117
30. Lynch KM, Coffey A, Arendt EK. Exopolysaccharide producing lactic acid bacteria: Their techno-functional role and potential application in gluten-free bread products. *Food Res Int*. 2018;110:52–61. doi:10.1016/j.foodres.2017.03.012
31. Madhuri K, Prabhakar K. Microbial exopolysaccharides: Biosynthesis and potential applications. *Orient J Chem*. 2014;30(3):1401–1410. doi:10.13005/ojc/300362
32. Yother J. Capsules of *Streptococcus pneumoniae* and other bacteria: Paradigms for polysaccharide biosynthesis and regulation. *Annu Rev Microbiol*. 2011;65(1):563–581. doi:10.1146/annurev.micro.62.081307.162944
33. Li J, Wang N. The *gpsX* gene encoding a glycosyltransferase is important for polysaccharide production and required for full virulence in *Xanthomonas citri* subsp. *citri*. *BMC Microbiol*. 2012;12(1):31. doi:10.1186/1471-2180-12-31
34. Rana S, Upadhyay LSB. Microbial exopolysaccharides: Synthesis pathways, types and their commercial applications. *Int J Biol Macromol*. 2020;157:577–583. doi:10.1016/j.ijbiomac.2020.04.084
35. Ates O. Systems biology of microbial exopolysaccharides production. *Front Bioeng Biotechnol*. 2015;3:200. doi:10.3389/fbioe.2015.00200
36. Schmid J. Recent insights in microbial exopolysaccharide biosynthesis and engineering strategies. *Curr Opin Biotechnol*. 2018;53:130–136. doi:10.1016/j.cobio.2018.01.005
37. De Vuyst L, De Vin F, Vaningelgem F, Degeest B. Recent developments in the biosynthesis and applications of heteropolysaccharides from lactic acid bacteria. *Int Dairy J*. 2001;11(9):687–707. doi:10.1016/S0958-6946(01)00114-5

38. Rehm BHA. Bacterial polymers: Biosynthesis, modifications and applications. *Nat Rev Microbiol*. 2010;8(8):578–592. doi:10.1038/nrmicro2354
39. Islam ST, Lam JS. Synthesis of bacterial polysaccharides via the Wzx/Wzy-dependent pathway. *Can J Microbiol*. 2014;60(11):697–716. doi:10.1139/cjm-2014-0595
40. Huszczyński SM, Hao Y, Lam JS, Khursigara CM. Identification of the *Pseudomonas aeruginosa* O17 and O15 O-specific antigen biosynthesis loci reveals an ABC transporter-dependent synthesis pathway and mechanisms of genetic diversity. *J Bacteriol*. 2020;202(19):e00347-20. doi:10.1128/JB.00347-20
41. Cuthbertson L, Kos V, Whitfield C. ABC transporters involved in export of cell surface glycoconjugates. *Microbiol Mol Biol Rev*. 2010;74(3):341–362. doi:10.1128/MMBR.00009-10
42. Krasteva PV, Bernal-Bayard J, Travier L, et al. Insights into the structure and assembly of a bacterial cellulose secretion system. *Nat Commun*. 2017;8(1):2065. doi:10.1038/s41467-017-01523-2
43. Ua-Arak T, Jakob F, Vogel RF. Fermentation pH modulates the size distributions and functional properties of *Gluconobacter albidus* TMW 2.1191 levan. *Front Microbiol*. 2017;8:807. doi:10.3389/fmicb.2017.00807
44. Arayas MA, Mabrouk MEM, Sabry SA, Abdella B. Exopolysaccharide production from *Alkalibacillus* sp. w3: Statistical optimization and biological activity. *Biologia*. 2022;78(1):229–240. doi:10.1007/s11756-022-01233-1
45. Abo-Zaid GA, Kenawy AM, El-Deeb NM, Al-Madboly LA. Improvement and enhancement of oligosaccharide production from *Lactobacillus acidophilus* using statistical experimental designs and its inhibitory effect on colon cancer. *Microb Cell Fact*. 2023;22(1):148. doi:10.1186/s12934-023-02153-8
46. Wang J, Zhang J, Guo H, et al. Optimization of exopolysaccharide produced by *Lactobacillus plantarum* R301 and its antioxidant and anti-inflammatory activities. *Foods*. 2023;12(13):2481. doi:10.3390/foods12132481
47. Sharma S, Roy T, Kashyap Y, et al. Characterizing and demonstrating the role of *Klebsiella* SSN1 exopolysaccharide in osmotic stress tolerance using neutron radiography. *Sci Rep*. 2023;13(1):10052. doi:10.1038/s41598-023-37133-w
48. Prakash Shyam K, Rajkumar P, Ramya V, Sivabalan S, Kings AJ, Miriam LRM. Exopolysaccharide production by optimized medium using novel marine *Enterobacter cloacae* MBB8 isolate and its antioxidant potential. *Carbohydr Polym Technol Appl*. 2021;2:100070. doi:10.1016/j.carpta.2021.100070
49. Chen CC, Nargotra P, Kuo CH, Liu YC. High-molecular-weight exopolysaccharides production from *Tuber borchii* cultivated by submerged fermentation. *Int J Mol Sci*. 2023;24(5):4875. doi:10.3390/ijms24054875
50. Behera S, Kumari Panda A, Kumar Behera S, Gupta N. Media optimization, extraction, purification and characterization of exopolysaccharide from *Fusarium proliferatum*: A novel source bioactive polysaccharide. *Results Chem*. 2023;5:100923. doi:10.1016/j.rechem.2023.100923
51. Chug R, Mathur S, Kothari SL, Harish, Gour VS. Maximizing EPS production from *Pseudomonas aeruginosa* and its application in Cr and Ni sequestration. *Biochem Biophys Rep*. 2021;26:100972. doi:10.1016/j.bbrep.2021.100972
52. Petry S, Furlan S, Crepeau MJ, Cerning J, Desmazeaud M. Factors affecting exocellular polysaccharide production by *Lactobacillus delbrueckii* subsp. *bulgaricus* grown in a chemically defined medium. *Appl Environ Microbiol*. 2000;66(8):3427–3431. doi:10.1128/AEM.66.8.3427-3431.2000
53. Robitaille G, Moineau S, St-Gelais D, Vadeboncoeur C, Britten M. Detection and quantification of capsular exopolysaccharides from *Streptococcus thermophilus* using lectin probes. *J Dairy Sci*. 2006;89(11):4156–4162. doi:10.3168/jds.S0022-0302(06)72460-2
54. Barcelos MCS, Vespermann KAC, Pelissari FM, Molina G. Current status of biotechnological production and applications of microbial exopolysaccharides. *Crit Rev Food Sci Nutr*. 2020;60(9):1475–1495. doi:10.1080/10408398.2019.1575791
55. Wu J, Zhang Y, Ye L, Wang C. The anti-cancer effects and mechanisms of lactic acid bacteria exopolysaccharides in vitro: A review. *Carbohydr Polym*. 2021;253:117308. doi:10.1016/j.carbpol.2020.117308
56. Loeffler M, Hilbig J, Velasco L, Weiss J. Usage of in situ exopolysaccharide-forming lactic acid bacteria in food production. Meat products: A new field of application? *Comp Rev Food Sci Food Safe*. 2020;19(6):2932–2954. doi:10.1111/1541-4337.12615
57. Zhang G, Zhang W, Sun L, et al. Preparation screening, production optimization and characterization of exopolysaccharides produced by *Lactobacillus sanfranciscensis* Ls-1001 isolated from Chinese traditional sourdough. *Int J Biol Macromol*. 2019;139:1295–1303. doi:10.1016/j.ijbiomac.2019.08.077
58. Bondarenko OM, Ivask A, Kahru A, et al. Bacterial polysaccharide levan as stabilizing, non-toxic and functional coating material for microelement-nanoparticles. *Carbohydr Polym*. 2016;136:710–720. doi:10.1016/j.carbpol.2015.09.093
59. Sezer AD, Kazak H, Öner ET, Akbuğa J. Levan-based nanocarrier system for peptide and protein drug delivery: Optimization and influence of experimental parameters on the nanoparticle characteristics. *Carbohydr Polym*. 2011;84(1):358–363. doi:10.1016/j.carbpol.2010.11.046
60. Bhatia S. Microbial polysaccharides as advance nanomaterials. In: Bhatia S. *Systems for Drug Delivery*. Cham, Switzerland: Springer International Publishing; 2016:29–54. doi:10.1007/978-3-319-41926-8_2
61. Kanmani P, Lim ST. Synthesis and structural characterization of silver nanoparticles using bacterial exopolysaccharide and its antimicrobial activity against food and multidrug resistant pathogens. *Process Biochem*. 2013;48(7):1099–1106. doi:10.1016/j.procbio.2013.05.011
62. Kim KS, Kim H, Park Y, et al. Noninvasive transdermal vaccination using hyaluronan nanocarriers and laser adjuvant. *Adv Funct Mater*. 2016;26(15):2512–2522. doi:10.1002/adfm.201504879
63. Wu M, Zhang Y, Huang H, et al. Assisted 3D printing of microneedle patches for minimally invasive glucose control in diabetes. *Mater Sci Eng C Mater Biol Appl*. 2020;117:111299. doi:10.1016/j.msec.2020.111299
64. Arshad MS, Fatima S, Nazari K, et al. Engineering and characterisation of BCG-loaded polymeric microneedles. *J Drug Target*. 2020;28(5):525–532. doi:10.1080/1061186X.2019.1693577
65. Friedman AJ, Phan J, Schairer DO, et al. Antimicrobial and anti-inflammatory activity of chitosan–alginate nanoparticles: A targeted therapy for cutaneous pathogens. *J Invest Dermatol*. 2013;133(5):1231–1239. doi:10.1038/jid.2012.399
66. Li J, Xiang H, Zhang Q, Miao X. Polysaccharide-based transdermal drug delivery. *Pharmaceuticals (Basel)*. 2022;15(5):602. doi:10.3390/ph15050602
67. Chen C, Zhou P, Huang C, et al. Photothermal-promoted multi-functional dual network polysaccharide hydrogel adhesive for infected and susceptible wound healing. *Carbohydr Polym*. 2021;273:118557. doi:10.1016/j.carbpol.2021.118557
68. Tripathi K, Singh A. Chitin, chitosan and their pharmacological activities: A review. *Int J Pharm Sci Res*. 2018;9(7):2626–2635. doi:10.13040/IJPSR.0975-8232.9(7).2626-35
69. Serrano-Castañeda P, Escobar-Chavez JJ, Rodriguez-Cruz IM, Melgoza LM, Martinez-Hernandez J. Microneedles as enhancer of drug absorption through the skin and applications in medicine and cosmetology. *J Pharm Pharm Sci*. 2018;21:73–93. doi:10.18433/jpps29610
70. Shelke NB, James R, Laurencin CT, Kumbhar SG. Polysaccharide biomaterials for drug delivery and regenerative engineering. *Polym Adv Technol*. 2014;25(5):448–460. doi:10.1002/pat.3266
71. Zare P, Pezeshki-Modaress M, Davachi SM, et al. Alginate sulfate-based hydrogel/nanofiber composite scaffold with controlled Kargogenin delivery for tissue engineering. *Carbohydr Polym*. 2021;266:118123. doi:10.1016/j.carbpol.2021.118123
72. Freitas F, Alves VD, Torres CAV, et al. Fucose-containing exopolysaccharide produced by the newly isolated *Enterobacter* strain A47 DSM 23139. *Carbohydr Polym*. 2011;83(1):159–165. doi:10.1016/j.carbpol.2010.07.034
73. Koumoto K, Umeda M, Numata M, et al. Low Mw sulfated curdlan with improved water solubility forms macromolecular complexes with polycytidylic acid. *Carbohydr Res*. 2004;339(1):161–167. doi:10.1016/j.carres.2003.09.022
74. Inoue BS, Streit S, Dos Santos Schneider AL, Meier MM. Bioactive bacterial cellulose membrane with prolonged release of chlorhexidine for dental medical application. *Int J Biol Macromol*. 2020;148:1098–1108. doi:10.1016/j.ijbiomac.2020.01.036
75. Blandón LM, Nosedá MD, Islan GA, et al. Optimization of culture conditions for kefir production in whey: The structural and biocidal properties of the resulting polysaccharide. *Bioact Carbohydr Diet Fibre*. 2018;16:14–21. doi:10.1016/j.bcdf.2018.02.001

76. Wang B, Han Y, Lin Q, et al. In vitro and in vivo evaluation of xanthan gum–succinic anhydride hydrogels for the ionic strength-sensitive release of antibacterial agents. *J Mater Chem B*. 2016;4(10):1853–1861. doi:10.1039/C5TB02046H
77. Kwon SS, Kong BJ, Park SN. Physicochemical properties of pH-sensitive hydrogels based on hydroxyethyl cellulose–hyaluronic acid and for applications as transdermal delivery systems for skin lesions. *Eur J Pharm Biopharm*. 2015;92:146–154. doi:10.1016/j.ejpb.2015.02.025
78. Chatterjee S, Hui PCL, Wat E, Kan CW, Leung PC, Wang W. Drug delivery system of dual-responsive PF127 hydrogel with polysaccharide-based nano-conjugate for textile-based transdermal therapy. *Carbohydr Polym*. 2020;236:116074. doi:10.1016/j.carbpol.2020.116074
79. Patel D, Patel D, Patel B, Patel C. An overview on intelligent drug delivery systems. *Int J Adv Pharm Rev*. 2011;2:57–63.
80. Fundueanu G, Constantin M, Ascenzi P. Preparation and characterization of pH- and temperature-sensitive pullulan microspheres for controlled release of drugs. *Biomaterials*. 2008;29(18):2767–2775. doi:10.1016/j.biomaterials.2008.03.025
81. Dulong V, Mocanu G, Picton L, Le Cerf D. Amphiphilic and thermo-sensitive copolymers based on pullulan and Jeffamine®: Synthesis, characterization and physicochemical properties. *Carbohydr Polym*. 2012;87(2):1522–1531. doi:10.1016/j.carbpol.2011.09.049
82. Andhare P, Kishor C, Margi D, Hilor P. Chapter 0311. Microbial exopolysaccharides: Advances in applications and future prospects. In: Tewari R, ed. *Biotechnology Volume 3: Microbial Biotechnology*. New Delhi, India: Studium Press LLC; 2014:25–51. doi:10.13140/RG.2.1.3518.4484
83. Manjanna K, Kumar T, Shivakumar B. Natural polysaccharide hydrogels as novel excipients for modified drug delivery systems: A review. *Int J Chemtech Res*. 2010;2(1):509–525. https://www.researchgate.net/profile/Dr-Manjanna-Km/publication/215538020_Natural_polysaccharide_hydrogels_as_novel_excipients_for_modified_drug_delivery_systems_A_review/links/551bf1960cf2909047b98045/Natural-polysaccharide-hydrogels-as-novel-excipients-for-modified-drug-delivery-systems-A-review.pdf. Accessed August 10, 2023.
84. Chavhan S, Shinde S, Sapkal S, Shrikhande V. Herbal excipients in novel drug delivery systems. *Asian J Pharm Res*. 2017;7(2):111. doi:10.5958/2231-5691.2017.00019.3
85. Matricardi P, Di Meo C, Coviello T, Hennink WE, Alhaique F. Interpenetrating Polymer Networks polysaccharide hydrogels for drug delivery and tissue engineering. *Adv Drug Deliv Rev*. 2013;65(9):1172–1187. doi:10.1016/j.addr.2013.04.002
86. Valentine ME, Kirby BD, Withers TR, et al. Generation of a highly attenuated strain of *Pseudomonas aeruginosa* for commercial production of alginate. *Microb Biotechnol*. 2020;13(1):162–175. doi:10.1111/1751-7915.13411
87. Looijesteijn PJ, Van Casteren WHM, Tuinier R, Doeswijk-Voragen CHL, Hugenholtz J. Influence of different substrate limitations on the yield, composition and molecular mass of exopolysaccharides produced by *Lactococcus lactis* subsp. *cremoris* in continuous cultures. *J Appl Microbiol*. 2000;89(1):116–122. doi:10.1046/j.1365-2672.2000.01082.x
88. Oerlemans MMP, Akkerman R, Ferrari M, Walvoort MTC, De Vos P. Benefits of bacteria-derived exopolysaccharides on gastrointestinal microbiota, immunity and health. *J Funct Foods*. 2021;76:104289. doi:10.1016/j.jff.2020.104289