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Scleroglucan Production by Microbes and Downstream Processing

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Abstract

Polymer based synthetic petroleum and plant natural polysaccharides do have the drawback of limited sources, as well as the latter's non-biodegradability. Eco-friendly, low-cost, and standardised microbial polysaccharides, on the other hand, offer a viable solution to this problem. They drew international recognition due to their original and distinctive physical and chemical propertiesas well as a diverse spectrum of industrial applications, the majority of which are rapidly becoming economically competitive. Scleroglucan, a 1, 3-beta-1, 6-glucan secreted by Sclerotium fungus, has a great economic potential and can have a variety of branching frequencies, side-chain lengths, and molecular weights dependent on the generating strains and cultivation circumstances. Scleroglucan's viscosifying ability, water solubility, and pH, wide temperature, and salt concentrations stabilisation make it viable for just a variety of bioengineering (food additives, improve oil recovery, cosmetic, drug delivery biocompatible materials, and pharmaceutical products, and so on) and biomedical, immunotherapy, antitumor, and so on application areas. It could be generated in large quantities at a bioreactor scale under standardised circumstances, with a high exopolysaccharide proportion governing performance improvement.

Keywords: Sclerglucan, Downstream process, Polymers, Production, Culture, Microbes

INTRODUCTION

Structural Formula and Conformational Characteristics of Scleroglucan

Scleroglucan is a branched glucan that is non-ionic and has a high molecular weight. Rinaudo and Vincendon describe it as having a backbone of (1, 3) β-linked D-glucopyranosyl residues with a single (1, 6) β-linked D-glucopyranosyl unit every three sugar residues in the main chain. The degree of branching of this repeated unit is roughly 0.33 due to its structure. Apart from being a common characteristic of most physiologically active (1, 3) β-glucans, is considered to be responsible for the polysaccharide's high solubility in water. Scleroglucan is thought to acquire a highly organised, triple helical tertiary structure and rigid when dissolved in water at room temperature and low alkali concentrations, usually less than 0.15 M NaOH. Protruding (1, 6)-glycosidic lateral splitting hinders the intermolecular approach via substantial H-bonding, that would otherwise contribute to aggregation formations and precipitate, throughout this complex molecular shape. At the triplex's core, though, crosslinks hydrophobic interactions preserve the macromolecular structural arrangement [1–7]. Furthermore, at greater NaOH concentrations, when severe viscosity changes were typical, the triple-strand helices are likely to undergo hydroxyl group ionisation, which destroys hydrogen bonding and leads to polysaccharide decomposition [8].

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Microbial Polysaccharide

The word scleroglucan is often used in the field of

microbial polysaccharides to refer to a group of EPSs with a structure similar that are mostly (but not solely) generated by filamentous fungi of the genus Sclerotium. Only D-glucose is liberated from this water-soluble homopolysaccharide after full hydrolysis [9]. Polymers are created at a rate of around 180 million tonnes per year, and they play an important part in our modern civilization. Petroleum-based polymers and plant-based polymers both have a finite availability of materials, especially additional towards the former's well-known environmental consequences. Furthermore, exopolysaccharides (EPSs) derived from microbial sources typically have shorter production durations, the ability to use industrial wastes, no interference with productive lands, and a high rate of restoration [10].

Properties and Applications of Scleroglucan

Scleroglucan has a number of unique physico-chemical features that set it apart from other polysaccharides, particularly in the production of specific processes and products. Nevertheless, the polymer assembly lines, and also downstream processing, are all depending on the culture supernatant, factual information which may modify the molecular weight, conformational parameters, DP, DB, or polymer purity grade, and therefore analyze its final conceivable benefits, minor to big differences in these features might be detected. Concentration (e.g., 2 g/L) of pure (90–98 percent EPS) S. rolfsii ATCC 201126 scleroglucan in water can give very viscous solutions with non-Newtonian, non-thixotropic, as well as pseudoplastic behaviour, for example [11–15].

Scleroglucan concentrations are relatively unchanged at temperatures up to 100–120°C and throughout a wide pH range. Furthermore, the neutral nature of EPS permits it to maintain pseudoplasticity though in the presence of salts ions such as NaCl, KCl, $CaCl_2$, $MgCl_2$, and $MnCl_2$. When subjected to alkali, high temperatures, or salts, slightly refined solutions (2 g/L) of commercial scleroglucans and crude polymer isolates from fermentation broths yield lower viscosity solutions with a decreased ability to keep stable rheological properties. There were additional variations among Biopolymer CS6 (60–70 percent scleroglucan) and Biopolymer based on the EPS purification level. Because of a triple-helix inter-iingn pathway that interacts with molecules, scleroglucan as triplex has a tendency to create thermo-reversible gels at low temperatures (near to 7°C). Scleroglucan triple helices, from the other hand, could be influenced by denaturing circumstances (e.g., pH 13) [16], fragmentation occurs when the interstrand H-bonding is destabilised into single stranded randomized spirals, similar to the behaviour of other strongly linked (1-3)β-D-glucans [17].

In respect to scleroglucan bioactive components, it was discovered that giving the substance to animals through various methods did not result in toxicity, tissue diseases, or blood abnormalities. Rabbits, pigs and humans showed no signs of eye or skin discomfort. Additionally, scleroglucan has indeed been demonstrated to act as an immune stimulant and a non-digestible dietary fibre in humans. A variety of physicochemical, nutritive, and biological characteristics have already been thoroughly discussed in the literature and are undoubtedly worth mentioning [18–27].

Preservation of Strain

While it may appear to be a small issue, good strain preservation is unquestionably recognised as a critical method for ensuring long-term survival and the retention of fungal characteristics. Scleroglucan-producing strain preservation was initially accomplished through transference on PDA or PDY slants monthly. Even though this is a widely used procedure, another option involves preserving mycelium in deionized water [28]. These and other approaches are used to analyse several S. rolfsii strains found in the wild. Early observations suggested that sub-culturing on diverse culture media on a regular basis, followed by storage at a low temperature (4–7°C), could result in a loss of viability and a significant reduction in scleroglucan synthesising capacity [29, 30].

Inoculum Standardization

The quantity of inoculation and its purity, that must be standardised, are also important factors in the effectiveness of scleroglucan synthesis in bioprocesses. The creation of a standard inoculum is known to be difficult due to the lack of spores produced by the species Sclerotium as well as the non-homogeneous character of mycelial suspensions [31–33]. The Sclerotium species, on the other hand, has the ability to produce sclerotia, which can be employed for strains reactivation and inocula production. A homogeneity phase using mycelium-covered agar plugs suspended in the appropriate quantity of growth media is yet another strategy that substantially aids in the creation of homogeneous inocula. Standardization quantities must be used to ensure reproducibility, and inoculum procedures could be done securely with the help of a hand mixer at a regulated velocity so for a set amount of time, within sterile condition [34].

Nutritional Requirements

According to several studies, the nature of carbon and nitrogen sources, and also it's the initial metal ion concentrations in the fermentation medium may affect the quantities of fungal EPS in the broth. It was highlighted that possessing the greatest biomass often does not imply producing the most EPS. To boost EPS production in S. rolfsii ATCC 201126, as with other scleroglucans, a high carbon to nitrogen ratio Concentration when compared in the growth medium is necessary. In terms of nutritional resources, higher polymer quantities would've been related to a preference use of sucrose as $NaNO_3$ and a nitrogen and carbon source. And from the other hand, N-sources like $(NH_4)_2SO_4$ as well as other NH_3-based N-sources caused a significant reduction in scleroglucan formation, which would be likely related to ammonium's negative metabolic regulation of the EPS biosynthesis mechanism. The C-source fraction has traditionally had a significant impact on scleroglucan synthesis when it comes to growth media characteristics [35]. As previously stated, a high C:N ratio favours EPS formation, and various theories have been proposed to explain this phenomenon. One possibility would be that the C-source is favoured for making a carbonaceous product (polysaccharide) with lower osmotic pressure than that of the initial sugar substrate (sucrose), which would have been available for development in future famine conditions. A rise in scleroglucan synthesis growing during high-osmotic pressure circumstances, but from the other hand [36].

Conditions of Culture

Controlling and maintaining (or modulating) operative parameters is critical for increasing efficiency. Some characteristics unique to the situation of scleroglucan synthesis. Many researchers and engineers have experimented with various methods to improve EPS synthesis and purifying over the history of scleroglucan studies, and these would be reviewed again [7, 11, 37].

Temperature Effect

Both culture growth and polysaccharide production are often affected by this parameter. However, it has been shown that maximal EPS biosynthesis is attained in batch cultures at temperatures slightly lower than those required for optimal growth rate. When the development rate of an organism is slowed by lowering the cultivation temperature, the availability of isoprenoid lipid carriers for non-growth tasks increases, promoting polysaccharide formation. Temperatures above 28°C promote biomass production, while temperatures below 28°C promote scleroglucan growth. Instead, below 28°C, by-product creation increases gradually, until acid production exceeds biomass and EPS biosynthesis at 20°C. The production method for S. rolfsii ATCC 201126 is generally carried out at 30°C, resulting in excellent EPS outputs [38].

CONCLUSION

With the development of modern biotechnology, a whole new world of possibilities for the utilisation of fungi as novel product producers has opened up. Scleroglucan biopolymer is one of these distinctive fungal products, and because of its adaptability and unique qualities, it might be used in a wide range of industries, including oil, cosmetics, food, and pharmaceuticals. Scleroglucan

production by bacteria, both in the lab and in industry, is one of the most complex systems presently understood. The financial prosperity and efficiency of this growth will be ensured by optimising scleroglucan manufacturing and removing related impediments. This will necessitate combining data from a variety of fields, including biochemical engineering, microbiology, process engineering, genetics, and statistics, among others.

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