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Effective variables on production and structure of xanthan gum and its food applications: A review

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ABSTRACT

Xanthan gum is a microbial exo-polysaccharide produced industrially from carbon sources by fermentation using the Gram-negative bacterium *Xanthomonas campestris*. The yield and properties of xanthan gum production and its applications in food are influenced by several variables. Knowing these variables is now needed and will be useful for the scale up production of xanthan gum to achieve high-quality gum at a low price. There have been attempts to optimize variables in xanthan gum production by using various raw materials. This review represents comprehensive knowledge about optimization of effective variables on production and structure of high quality xanthan gum and the applications of xanthan gum for enhancing the quality of foods.

1. Introduction

Nowadays, many industries are searching for new environment friendly ingredients, or so-called "green" ones to use in their products (Shahhosseini et al., 2003; Khosravi-Darani et al., 2004; Mokhtari-Hosseini et al., 2009a, 2009b; Tanaka et al., 2011). Therefore, microbial biopolymers are now known as an important industrial resource to compete with herbal biopolymers or algae. Genetically engineered microorganisms are also reported for the production of new products with unique characteristics (Rosalam and England, 2006).

Xanthan gum is an extracellular heteropolysaccharide that is produced by *Xanthomonas* spp. such as *Xanthomonas* (*X.*) *campestris* (Farhadi et al., 2012), *Xanthomonas pelargonii* (Niknezhad et al., 2016), *Xanthomonas phaseoli* and *Xanthomonas malvacearum* during aerobic fermentation (Leela and Sharma, 2000). The microbial production of xanthan gum at an industrial scale is a non-continuous process (Khosravi-Darani et al., 2013).

This gum was authorized by the U.S. Food and Drug Administration for application as food additives (stabilizer and emulsifier) without any restrictions (Ghashghaei et al., 2016). The demand and production of xanthan gum from *X. campestris* has progressively increased, at an annual rate of 5–10% (Kongruang et al., 2005; Salah et al., 2010). It is estimated that 30000 t of this gum is produced per year (Li et al., 2016). The major producers of xanthan gum in the US are Merck, and Pfizer. In France, the major producers of xanthan gum are Rhone Poulenc, Mero-Rousselot-Santia, and Sanofi-Elf. In China, the major producer of xanthan gum is Saidy Chemical, and in Austria it is Jungbunzlauer (García-Ochoa et al., 2000). Xanthan gum when dispersed in water quickly produces a viscous, stable solution, even at low concentrations. Due to gum pseudoplasticity, its solution in water is a suitable thickener, stabilizer, and suspending agent in many foods (Niknezhad et al., 2015). Applications of xanthan gum in several kinds of food will be discussed separately.

Xanthan gum is the first natural biopolymer produced at an industrial scale. The safety of this gum has been investigated extensively. The acute toxicity of xanthan gum was evaluated orally in animal studies. There was no noticeable toxicity remarked in those studies for xanthan gum concentrations up to 20 g/kg body weight (Freitas et al., 2015). The digestibility and caloric availability tests indicated that xanthan was non-digestible in humans and improved passage of food through the upper gastrointestinal trace (Fan et al., 2008) The dermal irritation and sensitization potentials of xanthan gum were assayed in animal studies. Xanthan gum, up to 1%, was not irritating to rabbit skin (Bergfeld et al., 2012). Safety evaluation of xanthan gum by long-term feeding studies (xanthan gum in the diet at dosage levels of 0, 0.25, 0.50, and 1 g/kg body weight/day for 2-year studies on albino rats and at dosage levels of 0, 0.25, 0.37, and 1 g/kg body weight /day for 2-year studies on beagle dogs) showed no significant differences in the developmental parameters between test and controls (Freitas et al., 2015).

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The main chain of the molecule is based on a linear backbone of 1,4-linked β-D-glucose; which has a charged trisaccharide side chain containing a glucuronic acid residue between 2 mannose units at the C(3) position of every alternate glucose residue. The terminal β-D-mannose is linked via β−1,4 to the glucuronic acid which, in turn, is linked via α-(1,2) to the α-D-mannose (Wang et al., 2017). On approximately 1/2 of the terminal mannose residues, a pyruvic acid moiety is joined by a ketal linkage to the O(4) and O(6) positions. Acetate groups are present as substituents at the O(6) position of the non-terminal mannose. Typically, acetate groups can be found on 60–70% of the internal mannose residues whereas 30–40% of the terminal mannose residues contain pyruvate residues (Abbaszadeh et al., 2015).

A major concern in the xanthan commercial production is the cost of the fermentation medium. Therefore, recent research studies have focused on low-cost materials and effective solutions for the cost reduction of fermentation process (Kongruang et al., 2005; Salah et al., 2010; Khosravi-Darani et al., 2013; Li et al., 2017).

2. Optimization of culture medium composition for xanthan production

The production of xanthan gum at an industrial scale is carried out mainly in a submerged fermentation system by monitoring several process variables, such as *Xanthomonas* strain, carbon and nitrogen sources, batch or fed-batch process, pH, temperature, inoculum size, airflow rate, agitation, and duration of fermentation (Peters et al., 1989; García-Ochoa et al., 1992; Esgalhado et al., 1995; Umashankar et al., 1996; Liakopoulou-Kyriakides et al., 1999; Casas et al., 2000; Leela and Sharma, 2000; Lopez et al., 2001; Kalogiannis et al., 2003; Kurbanoglu and Kurbanoglu, 2007; Psomas et al., 2007; Mesomo et al., 2009; Silva et al., 2009; Salah et al., 2010, 2011; Gilani et al., 2011; Mirik et al., 2011; Moshaf et al., 2015; Farhadi et al., 2012; Murugesan et al., 2012; El Enshasy et al., 2013; Khosravi-Darani et al., 2013; Niknezhad et al., 2015; Li et al., 2016; Ghashghaei et al., 2016) (Table 1).

In one study, some isolates of *Xanthomonas* strains had been collected from various sources. All strains were evaluated in terms of morphology, final gum concentration, final viscosity, and antibiotic resistance (Table 2). There was no relation between colony diameter and gum viscosity in the final product. Also report shows differences in gum production and viscosity among different strains. The results suggested that the sensitivity to penicillin may be a good indicator for the screening of high-quality xanthan gum producers (Torrestiana et al., 1990).

Glucose (Peters et al., 1989; Esgalhado et al., 1995; Leela and Sharma, 2000) and sucrose (García-Ochoa et al., 1992; Casas et al., 2000; Leela and Sharma, 2000) have been used as carbon sources in the commercial production of xanthan gum. Leela and Sharma (2000) reported influence of using different carbon sources on the production of xanthan gum (at 2% concentration or 2 g sugar per 100 g medium). The maximum efficiency was achieved with glucose, sucrose, maltose, and starch for xanthan production while polyol sugars like inositol and sorbitol were not good carbon sources for xanthan gum production (Table 3). Glucose concentrations of less than 2–5% are not effective for maximum cell growth. Also, high concentration of glucose has no significant effect on growth and broth with 50 g/kg inhibiting cell growth and xanthan gum production (Umashankar et al., 1996; Amanullah et al., 1998; Leela and Sharma, 2000; Niknezhad et al., 2015).

In the commercial production of xanthan gum, the cost of the fermentation medium is seen as a value opportunity. For this reason, recent studies have focused on the use of industrial residues as low-cost natural alternatives to serve as substrates in the production of xanthan gum. Different sources were evaluated for this purpose: sugar beet pulp (Yoo and Harcum, 1999), olive mill wastewaters (Lopez et al., 2001), agricultural wastes, acid hydrolysates (López et al., 2004), unmodified starch (Rosalam and England, 2006), ram horn hydrolysate (Kurbanoglu and Kurbanoglu, 2007), cheese whey (Mesomo et al., 2009; Silva et al., 2009; Zabot et al., 2011; Niknezhad et al., 2015), sugarcane molasses (El-Salam et al., 1994; Murugesan et al., 2012), coconut juice and sugar cane (Kongruang et al., 2005; Faria et al., 2009), molasses (Kalogiannis et al., 2003; Gilani et al., 2011), date extract (Farhadi et al., 2012; Khosravi-Darani et al., 2013), waste date juice (Moshaf et al., 2015), kitchen waste (Li et al., 2016), grape juice concentrate (Ghashghaei et al., 2016) and palm date juice (Salah et al., 2010). Using molasses is a good alternative for synthetic media (El-Salam et al., 1994; Kalogiannis et al., 2003; Gilani et al., 2011; Murugesan et al., 2012), but its use as a sole carbon source leads to prolonged lag phase in cell growth and a delayed viscosity increase (De Vuyst and Vermeire, 1994).

Silva et al. (2009) have evaluated different cheese whey: sucrose ratios (40:40, 60:20, and 80:0 v:w) in xanthan gum production. They reported that there were no significant differences ($p < 0.05$) between these ratios to the final gum yield in comparison to cheese whey as sole carbon source.

El-Salam et al. (1994) evaluated impact of different organic and inorganic nitrogen sources on the production of xanthan gum by *X. campestris* E-NRC-3. The findings have shown that the organic nitrogen sources such as peptone, yeast extract, and corn steep liquor (as a complex organic and cheap nitrogen source) increased the efficiency of *X. campestris* E-NRC-3 in gum production. Inorganic nitrogen sources (600 mg nitrogen per L) displayed different effects on the efficiency of xanthan gum production by *X. campestris* E-NRC-3 after 5 days of incubation (Table 3). Among mentioned nitrogen sources, ammonium chloride causes the highest efficiency and also application of 2400 mg nitrogen per L of ammonium chloride leads to the production of 66.3 g/L of xanthan gum.

Xanthan gum production by *X. campestris* NCIM 2961, using different nitrogen sources, was reported in both batch and fed-batch fermentations. Production of xanthan gum was affected by the nitrogen sources used. In batch and fed-batch fermentations, the production of xanthan was 3.6 g/L and 5.2 g/L, respectively. Xanthan gum production by urid dhal, toor dhal, green gram, soybean meal, and yeast extract were 2.8, 2.1, 2.4, 2.5, and 3.6 g/L, respectively. Yeast extract was selected as the most effective nitrogen source for xanthan gum production (Palaniraj et al., 2011).

Xanthan gum production enhanced with carbon concentration while nitrogen source has a negative impact. Enhanced concentration of nitrogen source causes increase in biomass concentration. In fact, a high concentration of nitrogen source is not suitable for xanthan production because it does not have a participating role in this polysaccharide structure. The available nitrogen source is mainly used for cell growth and enzyme production for the catabolic and anabolic pathways of bacterial cells (García-Ochoa et al., 1992; Casas et al., 2000; García-Ochoa et al., 2000; Kalogiannis et al., 2003; Psomas et al., 2007; Salah et al., 2010; Gilani et al., 2011; Moshaf et al., 2015; Farhadi et al., 2012; Khosravi-Darani et al., 2013). Some reports indicate that higher xanthan gum concentrations can be achieved by controlling the carbon:nitrogen (C:N) ratio in both the trophophase (cell growth phase) and the gum production phase of the fermentation. However, nitrogen is required for growth but not for xanthan production. So, it is probable to restrict its accessibility in the production phase and thereby shift the metabolic pathway into the direction of xanthan production rather than cell growth. It is normal that a low C:N ratio is recommended in the trophophase to obtain both high cell concentration and high specific growth rate, which could eventually lead to further xanthan production (De Vuyst et al., 1987; Xueming et al., 1992; Lo et al., 1997; Khosravi-Darani et al., 2013). De Vuyst et al. (1987) suggested that a 2-step fermentation process with a low C:N ratio in the cell growth phase and a high C:N ratio in the gum production phase enables xanthan gum production up to 30 g/L. Xueming et al. (1992) reported that 18 g/L xanthan gum was attainable from 24 g/L glucose in a batch fermenta-