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Curdlan Gum, Properties, Benefits and Applications

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Curdlan Gum, Properties, Benefits and Applications

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Abstract. Curdlan gum is a new polymer produced by the microbial fermentation of sugar raw materials. It has the unique property of forming a gel under heating conditions and is also called a thermal gel. In 1996, the FDA approved the application of curdlan gum as a food raw material in the food industry. Curdlan gum does not dissolve in water, but it will swell and disperse easily in cold water. It is heated to form a colloid, according to its properties can be divided into low colloid and high colloid. It is produced by heating to 55~65°C and cooling to about 40°C, forming low-grade colloid with thermal reflection. When the water dispersion of cordlan gum is heated to more than 80°C, a higher colloid is formed. The Curdlan gel can reduce the water loss rate of chilled meat products and extend their shelf life.

Keywords. Curdlan gum, Production source, Applications, Composition, Properties.

1. Agrobacterium

The *Agrobacterium* was discovered in 1853, and it is a group of bacteria that live naturally in the soil and was also found related to plants as well as mentioned by [1] . That in the year 1853 the first description was made of neoplastic diseases that affect different types of plants and the strains of some species of this bacteria infect the roots and stems of a large variety of dicotyledonous plants and some gymnosperms through wounds.

[2] The genus Agrobacterium is split into different species based on their pathogenicity, with A. Radiobacter (non-pathogenic), Agrobacterium tumefaciens (the cause of coronary gallbladder tumors), and Agrobacterium rhizogenes being the most well-known (the causative agent of the disease). *A. vitis* (the causative agent of coronary infection of grape roots).

The strains that cause coronary pathogens are classified as *A. tumefaciens*. They cause disease in dicotyledons. The disease causes the formation of tumor-like swellings and can generally be found on the crown of plants directly above the soil or in the roots. *Agrobacterium* causes coronary disease by transferring part From their DNA to plants [3].

2. Microbial Extracellular Polysaccharides

Polysaccharides are produced by plants, algae, fungi or bacteria, and they are bio-polymers of high molecular weight produced by microorganisms during the processes of demolition and building inside the cell. They are used industrially as thickeners, stabilizers and gelling agents in foodstuffs. It has also entered into the manufacture of biomedical materials that help heal wounds and adhesives used in surgical operations, antioxidant activity and biological antecedents are just two examples of how they might be useful in the body [4].

[5] stated that microbial polysaccharides are multiple substances produced outside the cell that have an important role in the metabolic processes of the microorganism. Polysaccharides can resist harsh conditions such as salinity and drought, and they are used in various industrial and agricultural applications, because they have unique properties such as vital activity, its ability to crystallize, as well as the presence of very large molecular weights that may reach several million.

Polysaccharides were produced from different sources such as plants, algae and fungi, but the increase in their production costs and their impact on seasonal climatic conditions and the increased demand for them due to their importance and applications in the fields of biomedicine, pharmaceuticals, nutrients and cosmetics, which made researchers in recent times to study, characterize and produce many microbial polysaccharides and study their properties. It is cheaper than other natural polysaccharides such as Arabic gum and Carrageenans produced by plants and seaweed, as well as the speed of their production compared to those produced from plants [4].

The Polysaccharides used commercially in the food industry are xanthan gum [6]. gellan gum, carrageenan, alginate, pullulan, manan, galactomannan, hyaluronic acid and uronic acid [7].

Microbial polysaccharides are widely used in various fields to improve food quality and have an important role in health through their antimicrobial, antioxidant and cholesterol-lowering effect, as well as their antiviral activity. And some of them have electrical charges that help to stabilize the cells in the suspension evenly, so the cells remain diffuse without collecting or precipitating [8].

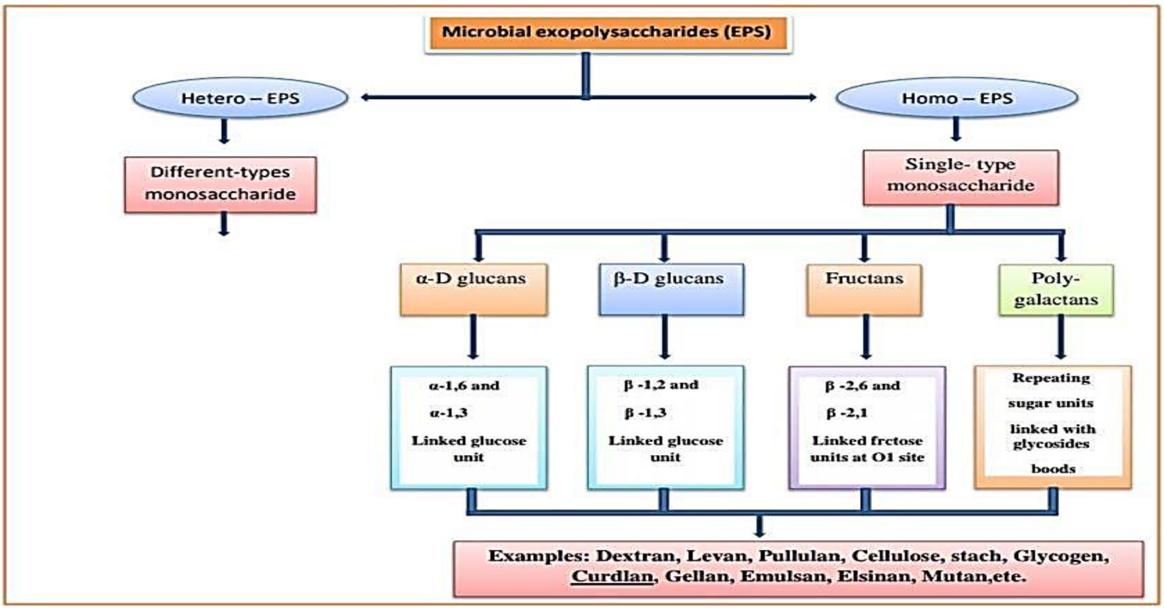

Figure 1. Homologous polysaccharides that can be produced by microorganisms Stewart-Tull, D E S (1980).

3. Curdlan Gum

Curdlan is a resinous substance resulting from various properties of microorganisms and the reason for its unique properties for its tolerance of high temperatures is called thermal gel. The basic building block of curdlan is glucose and the units are bound mediated by (1-3) β-glucose which is a linear, molecular homopolymer of $5.3 \times 10^4 – 2.0 \times 10^6$ Daltons. [9].

Many previous studies have shown that Curdlan is synthesized in the medium by the enzyme Uridine-diphosphate-glucose phosphorylase to form Uridine-diphosphate-glucose, as it is a pioneer to the biosynthesis of Curdlan. The presence of the carbon source in the growth medium is a factor that stimulates bacteria to secrete special enzymes in production, so the enzymes decompose sugars and convert them to curdlan [10].

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3.1. A brief History of the Production of the Curdlan

Started the idea of producing Curdlan, which is one of the polysaccharides, after increasing interest in its use as a food additive, and it has several functional properties. Therefore, it is used as a stabilizer, supporter, and thickener. The researcher Harada was the first to discover it in 1966 and it was called Curdlan. Its capacity to "coagulate" in a water bath is where the term comes from [11].

In 1996, Curdlan (then known as Pure Glucan) received FDA approval for commercial usage. Curdlan's physical and chemical features set it apart from other polysaccharides like starch, garnering significant interest from the food and non-food industries. Cordlan is odorless, tasteless, and insoluble in water and alcohol but soluble in alkaline solutions, giving it excellent stability in industrial sterilization, frying, and freezing processes [12].

In 1996, Curdlan was granted registration with the FDA in the United States. It is approved for use in the European Union as a stabilizer under the symbol E424 and its formula $(C_6H_{10}O_5)n$ [13].

3.2. Curdlan Production Source

The genus *Agrobacterium* is the most productive of linear curdlan, mainly (3-1)-Glucan, which may have a few chains, producing the branched sugar Glucan (1→3,1→6) [14]. It was discovered for the first time in *Alcaligenes faecalis* and was produced with other extracellular exogenous sugars [15]. The table below shows the most important microorganisms producing Curdlan sugar.

Table 1. The most important microorganisms producing curdlan gum.

Type of glucan	Bacterial Source	Sources
(1→3)-β-D-glucan,linear	Agrobacterium sp. 10C3 and derivatives	[16]
	Agrobacterium sp. ATCC 31749 and derivatives	[17]
	FO12607,12665,13127,13256 A.radiobacter	[16]
	A. rhizogenes IFO13259	[16]
	Rhizobium trifolii J60	[18]
	Rhizobium sp. TISTR 64B	[19]
	Cellulomonas spp.	[20]
	C. flavigena KU	[20]
(1→3,1→6)-β-Dglucan,cyclic	Bradyrhizobium japonicum USDA 110	[21]
	R. loti NZP 2309	[22]
	Azorhizobium caulinodans HAMBI 216	[23]
	A. brasilense ATCC29710	[24]
(1→3)-β-D-glucan,cyclic	Bradyrhizobium japonicum ndvC mutants	[25]
	Sinorhizobium meliloti ndvB mutant with the B. japonicum ndv locus	[25]
(1→3,1→2)-β-Dglucan, side-chainbranched	Streptococcus pneumoniae type 37	[26]

3.3. Curdlan Gum Composition

Curdlan is a homopolymer. The basic building unit is glucose. The units are related by a β-1,3-glucan glycosidic bond as shown in Figure 1. Some of its types contain β-(1→3, 1→2) glucose subunits. Their molecular weight is up to 5.3×10^4 - 2.0×10^6 dalton and is approved for use in the European Union as a stabilizer under the symbol E424 and formula $(C_6H_{10}O_5)n$ [27].

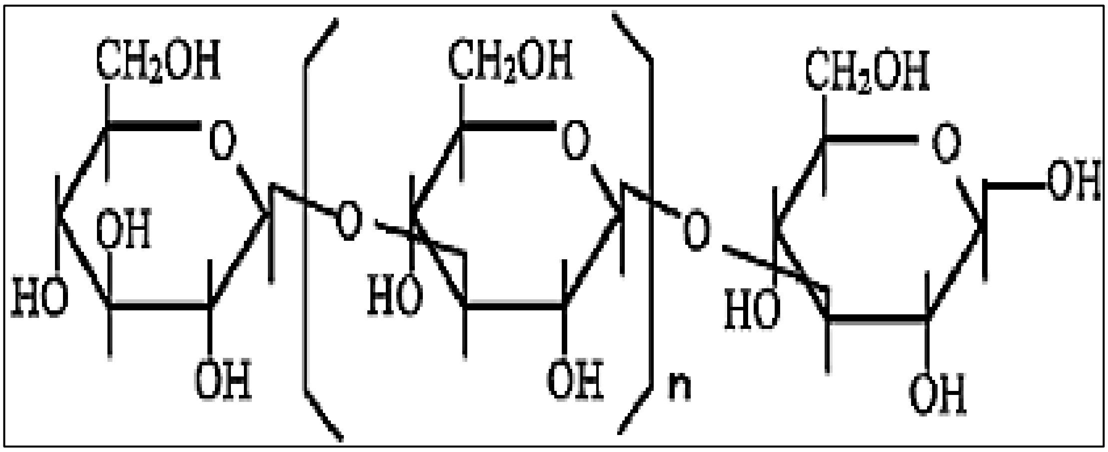

Figure 2. Curdlan Structure Produced by Bacteria. [28]

Curdlan consists of a carbohydrate concentration of at least 90% and a maximum water content of 10%. Curdlan lacks acidic components despite the abundance of glucose subunits linked by beta bonds between the ring's first and third carbon atoms [28]. The D-glucose that curdlan is made of is similar to cellulose, however the bonds are different. Curdlan is insoluble in water although it can be dissolved in aqueous sodium hydroxide due to the ionization of hydrogen bonds. When hydrated NaOH is introduced, the bonds break due to swelling and the granular structure of Curdlan is lost [29].

3.4. Curdlan Gum Properties

Curdlan is characterized by many important properties. It is a resinous substance characterized by its tolerance to high temperatures and the behavior of crystallization at high temperature. It is colorless, tasteless, and odorless. It is insoluble in water, alcohol, or acidic solutions, but it is soluble in alkaline solutions (pH12), as sodium hydroxide and trisodium phosphate. Although it is insoluble in water, it is able to form gels after heating, and it is a biodegradable, highly polar, non-toxic polymer. It can be easily decomposed into carbon dioxide and water.It can be used as an edible film which has good elongation when broken and the ability to form heat-resistant gel at the same time. It is considered a dietary fiber and characterized by its ability to form hard and flexible gels that cannot be thermally reversed when heated in aqueous suspensions at temperatures of 80 ° C or higher. 130 °C and is not affected by freezing and thawing processes [11,30].

It has the ability to absorb water, bind it and prevent separation, so it is used in many food industries as a support material, such as the cream and water ice cream industry, the meat industry of all kinds, and the pasta industry. Curdlan was used as a catalyst for prebiotic bacteria in therapeutic food and beverages, as it showed great stability during industrial processes such as frying and freezing and solubility [31].

[32] Describe how curdlan can soak up water at a rate greater than 100 times its own weight. At 60 degrees Celsius, a low-density, thermally reversible gel forms, while at 80 degrees Celsius, a flexible, high-density gel is produced.

3.5. Curdlan Gum Synthesis

By sequencing the genome of ATCC 31749, we were able to determine regulatory mechanisms for curdlan biosynthesis and localize the genes involved in this process to two distinct genomic locations. In one location, you'll find the crdR gene, which encodes a positive transcriptional regulatory protein, and two other genes, crdS (which is homologous to glycosyltransferases like cellulose synthase), crdA (whose function is unclear), and crdC (whose function is also unclear) [33,34].

Curdlan gum synthesis requires the presence of the enzyme Hexokinase to form glucose-6-phosphate via glucose phosphorylation. The process takes place inside the cell after glucose enters the cytoplasm by active transport, UDP-glucose phosphorylase synthesizes UDP glucose from uridine triphosphate (UTP) as the initiator of Curdlan biosynthesis. A polymerization reaction catalyzed by a polymerase enzyme leads to the formation of Glucose bonds and the release of the polymer into the extracellular environment after chain elongation, after which the transferase binds glucose to a fat molecule, isoprenoid fat phosphate, releasing UDP. Thereafter, UTP is synthesized from UDP by UDP kinase, and the curdlan production cycle is resumed [35].

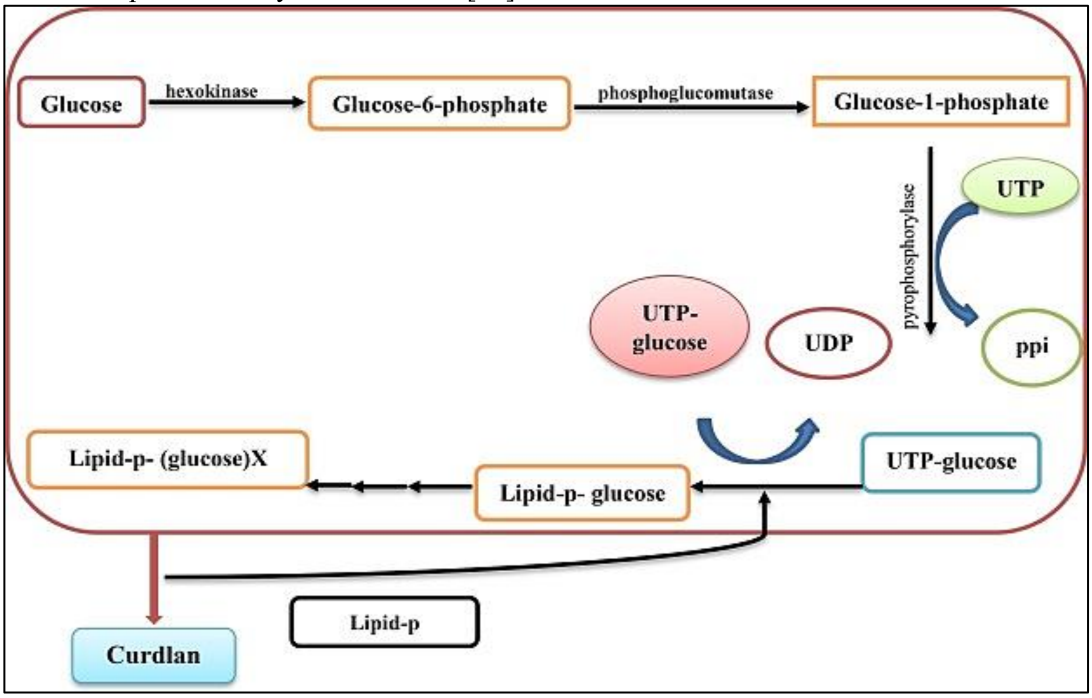

Figure 3. The metabolic pathway for the production of Curdlan from bacteria [36]

4. Optimal Conditions for the Production of Curdlan Gum

The study development conditions of the microorganisms producing Curdlan were studied in order to find the best conditions that lead to an increase in the quantities of Curdlan produced and thus reduce production costs. It was found that the processes of producing curdlan gum from microorganisms are affected by many factors such as carbon source, temperature, ventilation, pH, type of fermentation, and the size of the bacterial inoculum [37,38].

The best conditions for the production of curdlan gum are from *Agrobacterium sp. ATCC* was using different sources of carbon and nitrogen when using sucrose as a carbon source by 140 g/L, was obtained 32 g/L curdlan, and when using a nitrogen source, Using urea (28.16 and 9.58 g/L), ATCC 31749 was obtained, while using NH_4Cl resulted in (15.17 and 6.25 g/L) and incubation time of (95 hours) [39]. [40], found that the best optimum conditions for the production of curdlan gum from *Agrobacterium sp.* are the use of sucrose 140 g / l, NH_4Cl 2.4 g / l, KH_2PO_4 1.0 g / l and $MgSO_4$•$7H_2O$ 0.5 g / l at a temperature of is 30 °C, the optimal pH is 7, the incubation period is 96 hours, and by using the vibrating incubator at a speed of 150 rpm, the highest production rate of curdlan gum reached 5.2 g / liter.

Researchers [41] investigated the most effective carbon sources for curdlan formation by Agrobacterium sp., which include sugars like glucose, sucrose, and maltose. A specific group of the three sources were examined from 5 to 25% to obtain maximum yield The results showed 38.1 g/L at 15% sucrose, 27.3 g/L at 20% glucose and 37.4 g/L at 20% maltose, and the maximum yield of Curdlan was obtained using sucrose under limited nitrogen concentration. [11], indicated that Curdlan can be produced from bacteria using soluble condensed corn syrup as a source of carbon and nitrogen. As the soluble condensed corn juice material concentration rose from 50 g/L to 400 g/L, the Curdlan concentration rose as well, though not in direct proportion. After 120 hours, the strain produced the maximum concentration of Curdlan when fed 400 g/L soluble condensed corn juice. Its concentration exceeds that seen in curdlan formation that relies on glucose. Similarly, ATCC 31749's biomass production was greater after 120 hours of culture on 400 g/L soluble condensed corn juice than it was after growing on glucose.

[42] studied the effect of nitrogen sources on cordlan production, potassium nitrate, sodium nitrate, ammonium nitrate, ammonium acetate, ammonium citrate, ammonium chloride, ammonium sulfate, and ammonium acetate. Based on the observations, a maximum achieved was yield of 4.8 g/L Curdlan. They [43] indicated that the optimum acidity function for the production of Curdlan is 5.5 and to obtain an increase in production, it is the optimum nutrition for microorganisms to obtain 60 g/L Curdlan. [33], was able to obtain a maximum Cordlan yield of 47.97 ± 0.57 g/L from *Agrobacterium sp. ATCC 31749* using optimized medium containing 60 g/L sucrose, 6 g/L yeast, 2 g/L KH_2PO_4, 0.4 g/L $MgSO$• $7H^2O$, 2 g/L $CaCo_3$, 0.1 g/L $FeSO_4$• $7H_2O$, 0.04 g /L $MnSO_4$, and 0.02 g/L $ZnCl_2$ at 30 °C and 280 rpm after 96 hours of fermentation.

5. The Use of Alternatives in the Production of Curdlan Gum

Curdlan gum and other biopolymers have gained a lot of attention in recent years due to their useful physical and chemical properties and wide range of applications. The chemical, culinary, petroleum, medical, and biotech industries are just a few of the many that can benefit from microbial sugars. Many researchers have resorted to using agricultural waste as an alternative carbon source for sucrose, thus reducing production costs. Sugars constitute only a small part of the current polymer market due to their high cost production and recovery. For this reason a lot of effort has been put into developing cost-effective production processes using cheaper fermentation substrates [44].

Found [41] that the effect of different carbon sources on biomass production and Curdlan was distinguished by using sucrose among other sugar sources (glucose, fructose, maltose, xylose, mannose) at the pH level 7 after 72 hours of a period of time. Fermentation at 30 °C on a rocking incubator at a shaking rate of 100 rpm with 50 mL of modified fermentation medium. Explain [45], the use of sugar beet molasse in the production of curdlan gum from *Agrobacterium sp.* It gave good yield when incubating for 120 hours using the incubator at a speed of 150 rpm at a temperature of 30°C and the pH function was 7, and the yield was 42 g/liter of molasses as a carbon source instead of sucrose.

6. Curdlan Gum Purification

Biotechnology is divided, depending on the nature of the microorganism, into two main groups, the first of which is easy to separate, which is the biological products outside the cell, compared with the second group whose products are inside the cell and requires a number of steps to obtain them in pure form [46]. [47], the possibility of purifying the Curdlan produced from *Agrobacterium* by adding one ml of the bacterial medium with two ml of sodium hydroxide (1N), the mixture was kept for one hour at room temperature, then the mixture was expelled at 4000 rpm to remove the bacterial cells, then mixed The filtrate was mixed with hydrogen chloride (3N) until the pH reached 5-7 and the mixture was kept at 2-8 m overnight. To obtain the curdlan, a centrifuge was carried out at 4000 rpm. In order to get rid of the salts, the residues were washed with distilled water several times and then washed using acetone. To provide a dry product and store at room temperature until use.

[48] showed that the agrobacterium-derived curdlan was purified by the following steps: dissolving the curdlan in 5 ml of fermentation medium containing 100 ml of sodium hydroxide 0.5 M at 60 °C for one hour; centrifuging the mixture at 10,000 g for 10 minutes; adding 100 mL of hydrogen chloride 1 M to the supernatant; centrifuging the suspension for 30 minutes; washing three times with distilled water.

7. Description and Diagnosis of Curdlan Gum

The product obtained during fermentation processes must know its physical and chemical composition in order to characterize the compound, so some researchers used modern characterization techniques that determine the physical and chemical properties, molecular mass, molecular structure, shape, thermal and mechanical properties and some other chemical properties, some researchers do not consider characterization and diagnosis results Multiple chemical assays and bimoleculars are clear [49].

8. Curdlan Gum Applications

There is currently a growing interest in microbial sugars as an important source of new biopolymers for industrial use. One of the industrially important microbial sugars is curdlan, which has many uses due to its physical and chemical properties that make it important in food, industrial and pharmaceutical applications. It is safe and non-toxic and is used industrially as thickeners, stabilizers and agents. Crystallization in foodstuffs is also used for its biological functions, such as its antioxidant and prebiotic activity. [50].

Dissect this [40] As -glucan is consumed as an edible fiber, curdlan has been utilized as a stabilizer in jelly foods, a bio-thickener for pasta, and in other applications. Its unusual qualities [tasteless, odorless, and colorless] and its capacity to contain water make it useful as a textile material in the meat, dairy, and baking sectors.

Explain [51] the possibility of using curdlan gum in foods as a thickener, stabilizer, flavor enhancer, and in other therapeutic materials. On the structure of milk, which encouraged a decrease in the synergy of milk compared with milk without curdlan, the curdlan which also contributed to a decrease in the growth of lactic acid bacteria during the storage period, which and helps the gradual conversion of lactose to lactic acid by the initiator.

As shown in [52], curdlan sulfate has potential as both a standalone immunotherapeutic agent and as an adjuvant to an antiviral vaccination for the treatment of hepatitis infection. Curdlan, when combined with activated carbon, can be used to purge heavy metals from herbs used in the creation of oriental medicine, making it a valuable industrial use.

[33] studied that curdlan was used to determine the quality of pasta and texture frequently, as a strong gluten network tended to enhance the firmness and malleability of the pasta, and that the preparation of pasta with curdlan improved the eating quality of both raw and cooked pasta, which showed a gelatinous strength. High resulting in excellent edible quality noodles. Also, the tensile properties of the noodles changed, the rupture strength and distance, and the percentage of breaking of curdlan-containing noodles was much higher than that of the no-curdlan pasta which led to a significant improvement in the eating quality of raw and cooked pasta.

Conclusions

Curdlan has been studied for over fifty years due to its distinct gel formation, poor water solubility, mild processing conditions, indigestibility, and process ability. Researchers have only recently begun to investigate its applications in the food, health, and biomedical sectors. It has a nutritional benefit because it has several functional properties, so it is used as a stabilizer, fortifier, and thickener, and it has health benefits from its prebiotic use. It can be introduced into many food industries. Agricultural waste was used as a carbon source, such as date juice, molasses, grape juice, and whey protein, to reduce the economic cost of curdlan production. In addition to the disposal of agricultural waste that causes environmental damage, The yield of curdlan varies according to the type of strain and the culture media used.

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