PURIFICATION AND CHARACTERIZATION OF A NOVEL ALGINATE LYASE FROM THE MARINE BACTERIUM *BACILLUS* SP. ALG07

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Alginate is the most abundant carbohydrate in brown algae, accounting for up to 10–45% of the dry weight of brown algae. As a linear homopolymer, alginate is composed of (1–4)-linked α-L-guluronic acid (G) and its C5 epimer β-D-mannuronic acid (M), in blocks of poly-α-L-guluronate (polyG), poly-β-D-mannuronate (polyM), and random heteropolymeric sequences(polyMG). Alginate oligosaccharides with various bioactivities can be prepared through the specific degradation of alginate by alginate lyases. Therefore, alginate lyases which can degrade alginate under mild conditions, have drawn great interest. Although many alginate lyases have been discovered and characterized, few can be applied in industrial production of alginate oligosaccharides.

In this study, a novel marine strain with efficient degradation ability toward brown algae was isolated and classified to *Bacillus* sp. A novel alginate lyase, named as AlgA, with high specific activity was purified from the culture medium of this strain. AlgA had a molecular weight of approximately 60 kDa, wtih the optimal temperature and pH of 40°C and 7.5,respectively. The activity of AlgA was dependent on sodium chloride and could be considerably enhanced by Mg^{2+} or Ca^{2+}. Under optimal conditions, the activity of AlgA reached up to 8306.7 U/mg, which is the highest activity recorded for alginate lyases. Moreover, the enzyme was stable over a broad pH range (5.0–10.0), and its activity negligibly changed after 24 h of incubation at 40 °C. AlgA exhibited high activity and affinity toward poly-β-D-mannuronate (polyM). These characteristics suggested that AlgA is an endolytic polyM-specific alginate lyase (EC 4.2.2.3). The degradation products of alginate and polyM by AlgA were purified and identified through fast protein liquid chromatography and electrospray ionization mass spectrometry, revealing that AlgA mainly produced disaccharides, trisaccharides, and tetrasaccharide from alginate and disaccharides and trisaccharides from polyM. Therefore, the novel alginate lyase AlgA has potential applications in the production of mannuronic oligosaccharides and poly-α-L-guluronate blocks from alginate.