

Enzymatic Synthesis of Non-natural α -Glucosamine Chains by Thermostable Phosphorylase Catalysis

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Abstract

Phosphorylase is the enzyme that catalyzes phosphorolysis of α -(1 \rightarrow 4)-glucans at a nonreducing end, such as glycogen and amylose, giving α -D-glucose 1-phosphate (Glc-1-P). By means of the reversibility of the reaction, α -(1 \rightarrow 4)-glucans can be prepared by the phosphorylase-catalyzed α -glucosylation using Glc-1-P as a glycosyl donor and a maltooligosaccharide as a glycosyl acceptor [1]. Because of loose specificity for the recognition of substrates [2], phosphorylase recognizes several analogue substrates of Glc-1-P as glycosyl donors in α -glycosylations to give non-natural oligosaccharides. For example, we previously reported that α -D-glucosamine 1-phosphate (GlcN-1-P) could be used as a glycosyl donor in potato phosphorylase-catalyzed enzymatic α -glucosamylation to give oligosaccharides having a glucosamine (GlcN) residue at a nonreducing end [3]. Because it is known that thermostable phosphorylase differs in recognition ability of substrates from potato phosphorylase, in this study, we have examined the thermostable phosphorylase-catalyzed enzymatic α -glucosaminylation using GlcN-1-P (Figure 1). Consequently, we found that successive α -glucosaminylation occurred by thermostable phosphorylase catalysis to give non-natural α -glucosamine chains. When the enzymatic reaction was conducted in ammonia buffer containing Mg^{2+} ion, the α -glucosaminylation were accelerated owing to the precipitation of inorganic phosphate to produce the high molecular weight products.

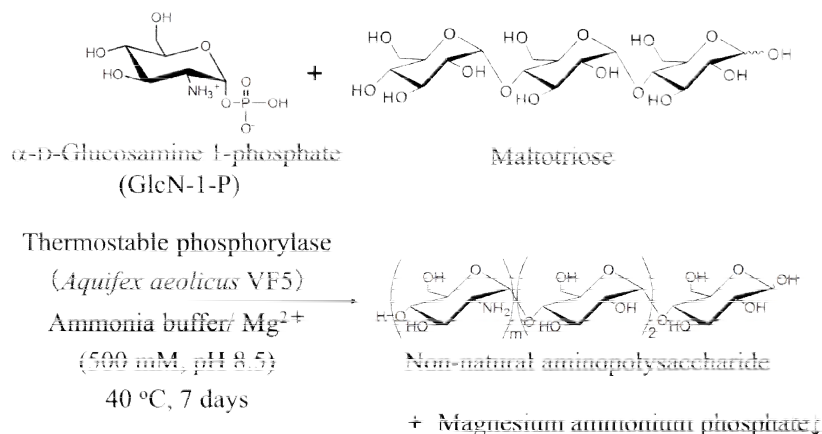


Figure 1. Thermostable phosphorylase-catalyzed successive α -glucosaminylation of maltotriose

References

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