

## **The regulation of *Bacillus intermedius* glutamyl endopeptidase biosynthesis in the recombinant *Bacillus subtilis* strain AJ73 during sporulation**

Chastukhina I., Sharipova M., Gabdrakhmanova L., Balaban N., Safina D., Kostrov S., Rudenskaya G., Leshchinskaya I.

*Kazan Federal University, 420008, Kremlevskaya 18, Kazan, Russia*

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### **Abstract**

The growth of the recombinant *Bacillus subtilis* strain AJ73 carrying the *Bacillus intermedius* 3-19 glutamyl endopeptidase gene on a multicopy plasmid and the effect of some nutrients on the efficiency of extracellular glutamyl endopeptidase production in the stationary growth phase were studied. In this phase, the concentration of glutamyl endopeptidase in the culture liquid peaked at the 48th and 78th h of cultivation and depended on the composition of the cultivation medium. Unlike the synthesis of glutamyl endopeptidase in the trophophase (i.e., during vegetative growth), which was suppressed by glucose, the synthesis of this enzyme during sporulation was resistant to glucose present in the cultivation medium. A multifactorial experimental design allowed optimal proportions between the concentrations of major nutrients (peptone and inorganic phosphate) to be determined. Inorganic phosphate and ammonium ions augmented the production of glutamyl endopeptidase by 30-150%, and complex organic substrates, such as casein and gelatin, enhanced the production of glutamyl endopeptidase by 50-100%. During sporulation, the production of glutamyl endopeptidase was stimulated by some bivalent cations ( $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ , and  $\text{Co}^{2+}$ ) and inhibited by others ( $\text{Zn}^{2+}$ ,  $\text{Fe}^{2+}$ , and  $\text{Cu}^{2+}$ ). The inference is drawn that the regulatory mechanisms of glutamyl endopeptidase synthesis during vegetative growth and sporulation are different.

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### **Keywords**

Glutamyl endopeptidase, Growth conditions, Recombinant strain, Regulation of biosynthesis, Sporulation