Mouse acidic mammalian chitinase expressed in Escherichia coli possesses chitinase functions comparable to CHO-expressed protein

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Outline

AMCase has attracted considerable attention due to its increased expression under pathological conditions. The biochemical characterization of chitinases requires large quantities of purified protein. Currently, efforts to characterize these proteins structurally and biochemically rely on mammalian and insect cell expression systems. To understand its pathophysiological roles, we expressed the mouse AMCase in *E. coli* as a fusion Protein (Figure 1). The chitinolytic activity of the recombinant AMCase against 4-nitrophenyl N,N'-diacetyl- β -D-chitobioside was comparable to the CHO-expressed AMCase (Figure 2; see also Figure 1). Furthermore, the recombinant AMCase bound to chitin beads (Figure 3), cleaved colloidal chitin and released mainly N,N'-diacetylchitobiose fragments (Figure 4).

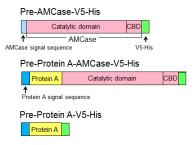


Figure 1. The CHO- and *E. coli*-expressed mouse AMCase fusion proteins

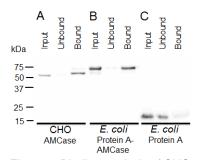


Figure 3. Binding analysis of CHO-expressed or *E. coli*-produced AMCase to chitin beads.

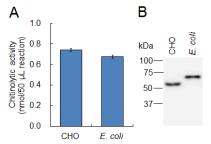


Figure 2 . The chitinolytic properties of murine AMCase prepared from *E. coli* with the enzyme from CHO cells.

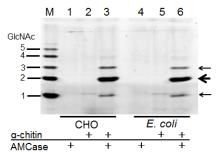


Figure 4. Degradation products of colloidal chitin by CHO- and *E. coli*-expressed mouse AMCase.

Novelty

The *E. coli*-expressed Protein A-mouse AMCase-V5-His fusion protein possesses chitinase functions comparable to the CHO-expressed AMCase.

Application

This recombinant protein can be used to elucidate detailed biomedical functions of the mouse AMCase.

Related information

- Original paper: Kashimura, A., Okawa K., Ishikawa, K., Kida, Y., Iwabuchi, K., Matsushima, Y., Sakaguchi, M.,
 Sugahara Y. and Oyama F. (2013) Protein A-mouse acidic mammalian chitinase-V5-His
 expressed in periplasmic space of *Escherichia coli* possesses chitinase functions comparable
 to CHO-expressed protein. *PLoS ONE* 8: e78669.
- URL: http://www.plosone.org/article/info:doi/10.1371/journal.pone.0078669

