(05-3-14)

Isolation and functional analysis of β-glucosidase involved in ginsenoside metabolism in *Panax ginsneg*

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Objectives

Cloning and characterization of ginseng β-glucosidase involved in ginsenoside metabolism

Materials and Methods

1. Material

Plant - *Panax ginseng*

2. Methods:

Four full-length cDNAs encoding β-glucosidase were cloned and introduced into E.coli vector pET21a. The recombinant β-glucosidase was expressed in *E.coli* BL21(DE3) CodonPlus RIL cells and purified. Purified proteins were characterized with artificial and natural substrates.

Results and Discussion

Ginseng saponins, ginsenosides, are considered to be the main active compounds of ginseng roots. More than thirty different ginsenosides have been isolated from natural and processed ginseng roots. Different ginsenosides, triterpene glycosides, have one or several monosaccharides to their triterpene aglycones. Some of the ginsenosides may be converted into another form through the cleavage of glucose, which is likely to be mediated by enzymes such as β-glycosidase. To isolate ginsenoside β-glycosidase, we isolated 12 putative β-glycosidase from the ginseng ESTs. Among them, we selected four cDNAs encoding proteins that are similar to previously isolated enzymes that modify triterpene saponin, furostanol glycoside and avenacoside. The selected cDNAs were expressed in *E.coli* and purified. β-glycosidase activity of the recombinant proteins were checked by hydrolyzing of the p-nitrophenyl-β-D-glycopyranoside. We confirmed that recombinant β-glycosidase, 15B04 can hydrolysis the glucoses on the ginsenoside Rb1. Transcripts of β-glycosidase were found most of tissue types except for 30C03 and especially, transcripts of 14G04 and 03H06 were relatively higher in stem and leaf tissue, whereas expression level of 15B04 was relatively higher in leaf tissue.

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