

Modulation of Plumbagin Production in *Plumbago zeylanica* Using a Single-chain Variable Fragment Antibody Against Plumbagin

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Abstract

Introduction: Plumbagin (PL) is a naturally occurring yellow pigment mainly isolated from the genus *Plumbago* (Plumbaginaceae) and recently been attracted much attention due to their various pharmacological activities. In this study, production in hairy root of *Plumbago zeylanica* (*P. zeylanica*) was accelerated by an anti-PL single-chain variable fragment antibody (PL-scFv), which possesses highly specificity to PL¹⁾. **Method:** Recombinant *Agrobacterium rhizogenes* (ATCC 15834) containing a PL-scFv were obtained through triparental mating and transformed into *P. zeylanica* to induce PL-scFv protein in the hairy roots. **Results:** Up to 40 µg recombinant PL-scFv were expressed per milligram of soluble protein in transgenic *P. zeylanica* hairy root cultures. The mean PL content obtained from transgenic hairy roots (12.24 µg/100 mg dry weight) exhibited 2.2 times higher than those obtained from wild-type (5.48 µg/100 mg dry weight). When the expression level of PL-scFv and PL content were analyzed by indirect and indirect competitive enzyme-linked immunosorbent assay²⁾, they were positively correlated ($r^2=0.885$), which suggest that the PL biosynthesis pathway had been modulated by the expression of PL-scFv protein in the hairy roots of *P. zeylanica*. **Conclusion:** The metabolic engineering using scFv, as performed in this study, is useful technique because production of pharmacologically active compounds can be enhanced without producing a biosynthetic enzyme³⁾.

¹⁾ Sakamoto et al., *Biol. Pharm. Bull.*, 32 (2009) 434-439;

²⁾ Sakamoto et al., *Anal. Chim. Acta*, 607 (2008) 100-105;

³⁾ Sakamoto et al., *Plant Cell Rep.*, 31 (2012) 103-110

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