

## Extra-cellular Production of Recombinant Fungal Phytase in Sesame Hairy Root Cultures

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### Abstract

Sesame (*Sesamum indicum* L.) hairy roots were transformed with a fungal (*Aspergillus*) phytase and their culture conditions were surveyed for the extra-cellular production of the recombinant phytase protein in shake flasks. Kanamycin resistance of sesame hairy roots was observed at 50  $\mu\text{g ml}^{-1}$  kanamycin sulfate and Southern hybridization analysis confirmed the existence of the phytase gene in the hairy root genomic DNA. The continuous dark condition was more effective for both the root growth and phytase production than light. Slightly higher root growth was determined at 30 °C than 26 °C in Murashige & Skoog (MS) medium supplemented with 3% sucrose, while the final phytase production was greatest in MS medium with 5% or 3% sucrose at both temperatures of 26 °C and at 30 °C. Among the culture media used, full-strength MS medium was exclusively efficient for production of the recombinant phytase. Most rapid increase rates in both the root growth and phytase production were detected at the fourth week of the culture periods and thereafter their rates began to decrease. Our results indicated that 5- to 6-week culture periods may be necessary for the maximal phytase production. Western analysis revealed that even though the phytase proteins expressed were measured with greater activities in the liquid medium than in the root tissues, they were still retained in the tissues.

### References

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