

## Abstract

Sesame (*Sesamum indicum* L.) seed oil contains high level of both oleic acid (OA, C18:1,  $\omega$ -9) and linoleic acid (LA, C18:2,  $\omega$ -6) but lowest level of  $\alpha$ -linolenic acid (ALA, C18:3,  $\omega$ -3). The combination of  $\alpha$ -linolenic acid deficiency and a skewed ratio of  $\omega$ -6: $\omega$ -3 fatty acids in the diet is a major explanation for the prevalence of cardiovascular and autoimmune diseases. The sesame seed oil can be developed as highly nutritious vegetable oil for human with a balanced proportion of  $\omega$ -6: $\omega$ -3 fatty acids. The conventional breeding has certain limitation to develop the suitable sesame cultivar with the desired trait due to the lack of specific research and low seed yield. In the present study, a germplasm collection of 54 sesame cultivars were analyzed for the seed fatty acid profiles. The fatty acid compositions were found to vary among the cultivars and Var-9 was found to contain high  $\alpha$ -linolenic acid content. The germplasm analysis thus served as groundwork for identifying potential targets for genetic intervention as well as for identifying the potential target cultivar for such genetic engineering approaches in sesame. In order to express the gene of interest in a seed-specific manner, two novel promoter elements, viz. sesame *2S albumin* and soybean *fatty acid desaturase-3* were isolated and characterized. The sesame *2S albumin* promoter showed higher seed-specific activity, and thus was selected for expression of the candidate gene for improving the seed oil quality in sesame. The over-expression of soybean (*Glycine max*) *fatty acid desaturase-3* gene led to enhancement of the  $\alpha$ -linolenic acid content from 1.30% to 6.21% of total seed fatty acid in transgenic sesame. In plant system, nuclear gene *fatty acid desaturase-7* is responsible for the production of trienoic fatty acids, namely C16:3 (hexadecatrienoic acid) and C18:3 ( $\alpha$ -linolenic acid) in chloroplast, which in turn participate in plastid membrane lipid formation. However, it does not accumulate in seed lipid. To find out the utility of the endogenous *fatty acid desaturase-7* gene of sesame in increasing the  $\alpha$ -linolenic acid content in seed oil, it was tested in the transgenic tobacco system. Here, we replaced the chloroplastidial signal peptide from the upstream of *fatty acid desaturase-7* gene by the endoplasmic reticulum signal peptide and an endoplasmic reticulum retention signal was placed at the C-terminal of the gene. The  $\alpha$ -linolenic acid content in the seeds of transgenic tobacco lines was found to increase from 0.83% to 5.37% of total seed lipid after expressing the modified *fatty acid desaturase-7* gene. The study opens up possibility to identify potential target cultivars for genetic engineering or breeding

purpose in general. The newly characterized *2S albumin* promoter could be useful to drive expression of transgene(s) involved in seed fatty acid modification. Further, this study creates an opportunity to use the endogenous *fatty acid desaturase-7* gene as a replacement for the heterologous *fatty acid desaturase-3* gene.

**Keywords:** *Sesamum indicum*,  $\alpha$ -linolenic acid, *2S albumin*, *fatty acid desaturase-3*,  $\omega$ -3 fatty acid desaturase.