Substrate Specificity of Fatty Acyl-coA Reductases (FARs) from Mouse, Arabidopsis and *Simmondsia chinensis*

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Fatty acyl-coA reductases (FARs) catalyze the formation of fatty alcohols and/or fatty aldehydes from fatty acyl-CoAs. Alcohol-forming FARs carry out a four-electron reduction of fatty acyl-CoA and use NADPH as a cofactor to accomplish this reduction. The reaction proceeds through an aldehyde intermediate but a free aldehyde is not released. Alcohol-forming FARs are thought to be membrane associated proteins with the molecular mass in the range of 56-58 kDa.

Three FAR genes encoding Arabidopsis FAR5, jojoba (*Simmondsia chinensis*) FAR and mouse FAR1 were expressed heterologously in *Saccharomyces cerevisiae*. The *in vivo* substrate specifity of FAR enzymes was investigated by measuring the accumulation of fatty alcohols in the yeast cells and in the culture medium.

The expression of jojoba FAR resulted in accumulation of C16:0, C18:0 and C18:1 alcohols and the expression of mouse FAR1 resulted in accumulation of C16:0 alcohols and small amounts of C18:0 alcohols. The yeast expressing Arabidopsis FAR5 produced C16:0 and C18:0 alcohols with preference to the latter ones. In liquid cultures considerable amount of produced fatty alcohols were secreted into the medium.

In vitro assays using a [14C]16:0-CoA or a nonradioactive 16:0-CoA as a substrate showed very low enzyme activity in whole cell extracts and in the microsomal fractions isolated from the yeast expressing tested FARs. However, the high activity of palmitoyl-CoA hydrolases and elongases as well as the activities of other enzymes involved in lipid metabolism were detected in the analyzed microsomal fractions. The optimization of *in vitro* assay conditions is currently under study.

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