ABSTRACTS (MASTER THESIS)

Analysis of lipid transport machinery using *Lithospermum erythrorhizon*

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Higher plants produce a variety of lipophilic compounds. Representatives are cutin and wax that are primary metabolites, whereas there are a number of hydrophobic secondary metabolites, for example monoterpenes and furanocoumarins as well as several alkaloids. Many of these compounds are secreted out from particular cells at specialized tissues like glandular trichomes, oil glands, and also from epidermal cells [1] [2]. To date, however, secretory processes of lipophilic secondary metabolites from plant cells are largely unknown especially at the molecular level [3].

An herbal medicinal plant *Lithospermum erythrorhizon* Sieb. et Zucc. produces shikonin derivatives, red naphthoquinone pigments in the root bark. These pigments are highly lipophilic, and almost exclusively secreted out of the cells. This study aims to elucidate the secretion mechanisms of lipophilic metabolites using shikonin as a model due to several reasons, i.e. high productivity, visibility as a red pigment, and clear reversible regulation of its production by light and medium compositions [4].

By use of inhibitors of vesicle transport, cytochalasin D, an inhibitor of actin filament polymerization and Brefeldin A, an inhibitor of the adenosine diphosphate (ADP)-ribosylation factor/guanine nucleotide exchange factor (ARF/GEF) protein system, we find that these inhibitors strongly inhibit shikonin secretion without suppressing its biosynthetic activity. These data suggest that secretion of shikonin derivatives into the apoplast utilizes, at least partly, the pathways common to the ARF/GEF system and actin filament polymerization [5].

From our current data, we estimate that shikonin derivatives are dissolved in common lipids. In order to identify the lipids co-localized with shikonin, we have analyzed lipid extracts from cultured cells using LC-TOF-MS. We expect to summarize these results as a publication in near future.

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