

ABSTRACT

BIOTRANSFORMATION OF STEMODANE DITERPENOID

BY

RHIZOPUS ORYZAE ATCC 11145

AND

PHYTOCHEMICAL ANALYSIS OF *SALVIA COCCINEA*

Glenroy Dean Anthony Martin

Described in this dissertation are the isolation of terpenoids from *Stemodia maritima* (Scrophulariaceae) and their structural modification utilizing chemical and microbial sources. This work was undertaken with the aim of developing an active site model of a cytochrome P450 enzyme system within the fungus *Rhizopus oryzae* ATCC 11145, (formerly *Rhizopus arrhizus*). Included also is the phytochemical analysis of *Salvia coccinea* (Lamiaceae).

A review of the bioconversion of steroids, terpenes, esters, octalin and hydrindenone derivatives, thioethers, prostaglandins, aromatic and other miscellaneous compounds by *Rhizopus oryzae* is outlined (Chapter 1).

The isolation of betulinic acid, stemarin, stemodin and the stemodinosides from *Stemodia maritima* (Scrophulariaceae) was carried out (Chapter 2). A new stemodinoside, stemodin- α -L-arabinofuranoside, was isolated from the plant. Stemodin, stemodinone and betulinic acid were chemically transformed to new analogues. A number of these analogues were biotransformed by *Rhizopus* to provide new metabolites (Chapter 3). The results from the fermentations provided useful information about the relationship of the functional groups associated with the substrates and their products of bioconversion.

A predictive active site model of a cytochrome P450 enzyme system of the fungus was generated from the data obtained in our laboratory and that reported in the literature (Chapter 4). The predictive nature of the model was affirmed with the incubation of 15(13 $\rightarrow$ 12) *abeo*-13 β -hydroxystemaran-2-one, 15(8 $\rightarrow$ 9) *abeo*-8 β (H)-12 β -hydroxystachan-2-one, methyl betulinate and betulin. The predicted sites of hydroxylation were observed for the rearranged stemodanes while no transformations were noted for the triterpene derivatives.

A phytochemical analysis of the plant *Salvia coccinea* was carried out (Chapter 5). Squalene, betulinic acid, betulin, β -sitosterol, and the glycoside 3 β -O-[tetra-O-acetyl- β D-glucopyranosyloxy]sitosterol were obtained.

Keywords: Glenroy Dean Anthony Martin, Natural Products Isolation, Lamiaceae, Terpenoids, Stemodin, Organic Chemical Synthesis, Biotransformation, *Rhizopus oryzae*, Active Site Model, Cytochrome P450 enzymes.