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EFFECT OF TEA METABOLITES ON DEVELOPMENT AND BEHAVIOUR OF SELECTED INSECTS AND THE TRANSFORMATION OF STEROIDS BY *MONACROSPORIUM AMBROSIUM*

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Effect of Tea Metabolites on Development and Behaviour of Selected Insects and The Transformation of Steroids by *Monacrosporium ambrosium*

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Part I is a ten generation study on the effect of secondary metabolites, on the shot hole-borer beetle, *Xyleborus fornicatus* (Coleoptera: Scolytidae) of tea, *Camellia sinensis* L. The uninhibited growth of the beetle in a sterol free medium indicated the invalidity of the hypothesis, which claimed that the resistance of tea clones is due to high saponin content reducing availability of tea sterols to the beetle. Progeny production was almost completely inhibited in media containing 100 ppm caffeine but the effect was reversible if exposure was for less than 48 hours. No significant differences were observed from control which tea saponins were introduced.

In behavioural studies in an olfactometer, females showed a density dependent attraction turning into a strong repulsion against increasing numbers of females. Males were attracted to non-mated females but not to mated individuals. GC analysis of tea bark showed its main constituents to be β-pinene, terpenolene, hexanol, linalool oxide, linalool, geraniol, methyl salicylate and eugenol. Olfactometer studies showed the beetle to be significantly attracted towards ethanol and mixtures of ethanol with α-pinene, eugenol and hexanol.

When both non-infested and infested tea stems were offered to beetles, a higher number entered the non-infested stems of both clones in the first hour while entry to infested TRI2025 stems was significantly delayed by 2 - 3 hours and to infested TRI2023 stems by over 24 hours, indicating an induced resistance

on infestation. Caffeine is known to be the main antifungal compound in the tea plant. A quantitative analysis using HPLC showed that caffeine content varied depending on the part of the plant and the infestive stage. Non-infested TRI2025 stem contain more caffeine than non-infested TRI2023 stem, while bark contained more than stem and inter-nodal bark more than nodal bark. The caffeine content of both clones was significantly higher in infested plants with the increase being greater in TRI2025 than in TRI2023.

Part II describes olfactometry studies on the social behaviour and host discrimination of the cowpea aphid, *Aphis craccivora* Koch (Homoptera:Aphididae). The occurrence of density dependent pheromones and odour responses to its host plant *Vigna unguiculata* were studied. Apterae responded with positive anemotaxis to air passed over both apterae and alatae in groups of less than ten individuals, but negatively to air that passed over groups greater than twenty. Alatae responded similarly to groups of apterae but were repelled by alatae. Both were able to distinguish between other hosts and their original host, cowpea. When attacked by aphids, plants responded with a temporary increase in attraction that reached a maximum in 48 hours.

In Part III, the insecticidal activity of *Vernonia anthelmintica* (Family: Asteraceae) seeds is described. The dichloromethane extract showed activity against *Aedes aegypti* second instar larvae. In the residual film bioassay the extract was active against *Callosobruchus maculatus* adult. Activity guided fractionation gave a crystalline substance identified as a mixture of angelicin and psoralen whose toxicity to *A. aegypti* was highest in the presence of sunlight. In the absence of sunlight, 100% growth inhibition was observed. With *C. maculatus* the substance showed 40% moribundancy in 24 hours in the residual film bioassay and 90% reduction of ovulation and 0% adult emergence in seed treatment bioassays.

When separated neither were as active as the original mixture and when combined, none of the combinations showed the activity of the isolated mixture.

Part IV of the thesis describes the biotransformation of steroids by *Monacrosporium ambrosium*, the fungus associated with the shot hole borer

beetle *X. fornicatus*. 5α-Androstan-17β-ol, 5α-androstan-17-one, 5α-androstan-3-one and 5α-androstan-3β-ol were prepared from 17β-hydroxy-5α-androstan-3-one and incubated with the fungus. Results suggest that *M. ambrosium* is not specific in its microbial reactions effecting hydroxylations, oxidations and reductions often leading to mixtures of products. Of the transformations observed, the 11α-hydroxylation of 3-oxygenated steroids appears to have some potential.