

**STUDIES ON THE ROLE OF THE GLUCOSINOLATE-MYROSINASE SYSTEM IN
RESISTANCE OF OILSEED RAPE TO
*Sclerotinia sclerotiorum***

by

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Summary

The glucosinolate-myrosinase (GSL-M) system in oilseed rape and other members of the family Brassicaceae produces toxic products which can limit fungal pathogen attacks on the host. The role of this system in resistance of oilseed rape to *Sclerotinia sclerotiorum*, causal agent of stem rot, was investigated.

Mustard powder was used as a GSL and myrosinase source in bioassays. The effect of toxic volatiles derived from hydrolysis of glucosinolates was observed as inhibition of fungal growth. Oxalic acid, a pathogenicity factor of the pathogen, did not affect production of toxic volatiles and inhibition occurred only at very acidic pH levels, regardless of the presence of oxalic acid. This indicated that oxalic acid at physiological concentrations and pH did not affect the GSL-M defense system.

Exposure of *S. sclerotiorum* colonies to inoculated leaves or leaf discs of host species or cultivars revealed that volatiles derived from infected leaf tissues have a toxic effect. This suggested that the GSL-M system is activated during infection of leaves and disease development. Freeze-dried powders of shoot parts of brassica species and cultivars including leaf, petiole, and stem demonstrated significant differences in producing toxic volatiles through their inhibitory effects on *S. sclerotiorum* mycelial growth *in vitro*, indicating that GSL contents in brassica species and even cultivars have different potentials for toxic products.

Tolerance of *S. sclerotiorum* to toxic volatiles derived from mustard powder and also synthetic isothiocyanates developed during repeated exposure of mycelium to these biocidal chemicals. Applying sublethal concentrations of mustard powder toxic volatiles and allyl isothiocyanate upregulated three putative glutathione-S-transferase enzyme encoding genes after 1h exposure. This is the first report of *S. sclerotiorum* response to isothiocyanates. There was significantly higher GST enzyme activity in treated mycelium compared with the control. GSTs may be involved in detoxification of isothiocyanates.

The resistance of oilseed rape varieties and brassica species was evaluated with leaf disc and intact plant inoculations under controlled conditions. No significant differences between varieties were observed in the leaf disc evaluations. In contrast, intact plant inoculations resulted in significant differences between genotypes and an oxalic acid assay followed the same pattern as for fungal inoculations. AV-Sapphire and AG-Castle among the oilseed rape cultivars were the most resistant and susceptible genotypes, respectively. The evaluated brassicas responded to the disease independently of their potential to produce toxic

volatiles derived from GSL hydrolysis. Moreover, there was no correlation between quantity of epicuticular wax and resistance.

Histopathological studies also revealed that the main sites of the GSL-M system in leaves of brassica plants were invaded without any appreciable signs of toxicity to the fungus of ITCs, which are produced upon cellular damage.

The study established that toxic volatiles, particularly isothiocyanates, released during hydrolysis of glucosinolates in brassica plants and oilseed rape, despite their toxic effect on *Sclerotinia sclerotiorum in vitro*, are detoxified by the pathogen. Therefore, these preformed antimicrobial products do not prevent the disease during host-pathogen interactions.

Declaration

This thesis contains the results of studies conducted at the Plant Pathology Research Laboratories in the discipline of Botany, School of Environmental and Rural Science at the University of New England.

I certify that all the work reported in this thesis is original and my own research work. The text of this thesis does not contain material which has been accepted as part of the requirements for any other degree in any university or any material previously published unless due reference is made.

Signed

A solid black rectangular box redacting the signature of the author.

Siamak Rahmanpour Ozan

Dedication

Dedicated to
my wife, *Sarira*, spring of kindness
and
my son, *Radin*, my shining little star

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