Cinnabar moth uses pyrrolizidine alkaloid composition and concentration as a cue for oviposition

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Introduction

- The **Cinnabar moth** (*Tyria jacobaeae*) is the main specialist herbivore of *Jacobeae vulgaris*

- **Pyrrolizidine Alkaloids (PAs)** are secondary metabolites which are used by the plant as a constitutive defense against herbivores and pathogens.

- PAs act as oviposition stimulants for *Tyria*. It has been confirmed by experiments using isolated PAs. However, no significant correlations between *Tyria*'s oviposition preference and PA variation in host plants were reported.

Fig. 1 **Cinnabar moth** *Tyria jacobaeae* adult

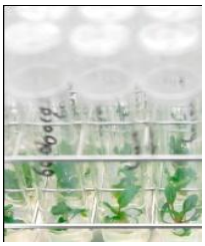

Fig. 3 *Jacobaea* hybrid plants kept in tissue culture

Fig. 2 Structure of two major PAs occurring in *Jacobaea* plants

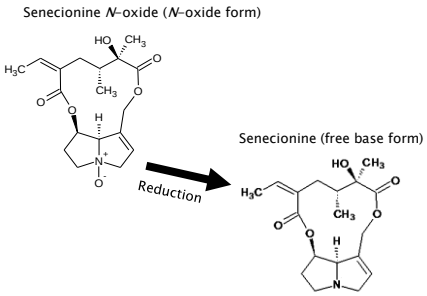

Fig. 4 Oviposition experiment in a greenhouse

Fig. 5 Eggs and egg batches laid by cinnabar moths on *Jacobaea* hybrid plants

Material and Methods

Experimental design

- 40 F_2 hybrid genotypes: (derived from a cross between *J. vulgaris* and *Jacobaea aquatica*), 6 replicates per genotype, 4 cages, 20 plants (>8 weeks old) in each cage, 30 male and female moths released in each cage (Fig4)
- Allow oviposition for 10 days
- Replicated 3 times

Measurements

- Fresh weight of plant shoots
- number of eggs per plant
- number of eggs batches per plant
- average number of eggs per egg batch

PA variation

- PA data were acquired by liquid chromatography–tandem mass spectrometry (LC–MS/MS) from an independent set of the same genotypes
- The 37 PAs detected in these genotypes are classified into four types (Table 1)

Data analysis

- **ANOVAs** to analyze genotype dependent oviposition preference using fresh weight as a covariate
- **Mutiple-regession** to find out which type of PAs related to oviposition preference
- **Correlation tests** to find out which individual PAs are related to oviposition preference

PAs in the shoots : 4 structural types

Senecionine-like PAs
Senecionine, Senecionine *N*-oxide
Intergerrimine, Intergerrimine *N*-oxide
Retrorsine, Retrorsine *N*-oxide
Usaramine, Usaramine *N*-oxide
Riddelliine, Riddelliine *N*-oxide
Seneciphylline, Seneciphylline *N*-oxide
Spartiodine, Spartiodine *N*-oxide
Acetylseneciphylline
Acetylseneciphylline *N*-oxide
Senecivernine
Jacobine-like PAs
Jacobine, Jacobine *N*-oxide
Jacoline, Jacoline *N*-oxide
Jaconine, Jaconine *N*-oxide
Jacozine, Jacozine *N*-oxide
Dehydrojaconine
Erucifoline-like PAs
Erucifoline, Erucifoline *N*-oxide
Acetylerucifoline
Acetylerucifoline *N*-oxide
Otosenine-like PAs
Senkirkine
Otosenine
Onetine
Desacetyldoronine
Florosenine
Floridanine
Doronine

Table 1: PAs detected in *Jacobaea* hybrid plants

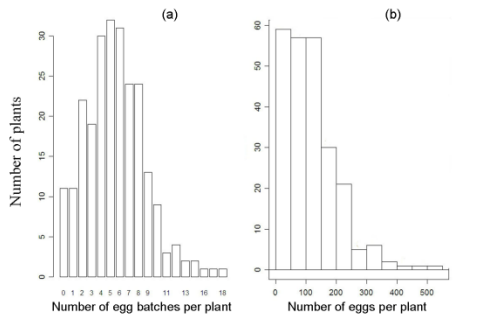

Fig.6 Frequency distribution of the number of egg batches per plant (a), number of eggs per plant (b) on the plants of 40 *Jacobaea* hybrid genotypes

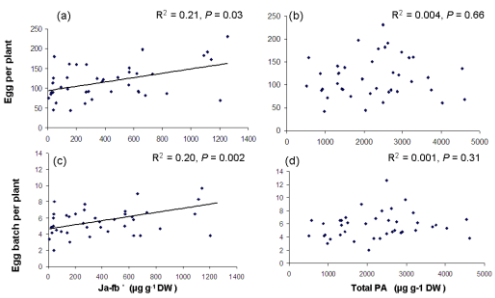

Fig.7 Scatter graphs of the number of eggs and egg batches per plant against the concentration of the free bases of the five jacobine-like PAs (jacobine, jacoline, jaconine, jacozine, dehydrojaconine)and total PA of 40 *Jacobaea* hybrid genotypes. Data are the genotypic mean values of 40 F2 genotypes

Results

- In total 28,323 eggs were laid in 1,375 egg batches on 240 plants (Fig.6).
- The number of eggs and egg batches depended on host plant genotype. (ANOVA for number of eggs, $F_{39,199}$ =1.58, P =0.02; for number of egg bathes, $F_{39,199}$ = 1.99, P = 0.001.).
- The free bases of the jacobine-like PAs were positively correlated with number of eggs and egg batches, but the corresponding *N*-oxides, other PAs or total PAs were not (Fig.7).

Conclusion

- Cinnabar moth oviposition preference depends on host plant genotype.
- Cinnabar moth oviposition preference is related to the PA variation in host plants: Oviposition of the cinnabar moth increases with the amount free bases of jacobine-like PAs in the host plants.
- Cinnabar moths can act as a selective agent on PA variation.

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