

## Studies on the relationships between host-plant acceptability and plant constituents in host selection by a swallowtail butterfly, *Papilio polytes*

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*Papilio polytes*, a Rutaceae feeder, exploits only a few plant species as larval food plants in nature. To clarify the physiochemical factors underlying its narrow host range, we examined the acceptability of four rutaceous plants, *Citrus depressa* (a host plant) and other potential hosts, *Fagara ailanthoides*, *Evodia meliifolia*, and *Orixa japonica* to ovipositing females of the butterfly. We identified active compounds present in foliage of *C. depressa* as stimulants of egg-laying and two groups of compounds system to evoke oviposition activity in the foliage of *F. ailanthoides*.

Female responses to the foliage, methanol extracts and partitioned fractions prepared from these plants were assayed for the presence of oviposition stimulants and/or deterrents. Larval survivorship on these plants was also recorded as an estimate of fitness. The foliage and methanol extracts of *C. depressa* and *F. ailanthoides* readily released egg-laying, while those of *E. meliifolia* evoked weak responses from females. In contrast, ovipositing females never accepted *O. japonica*. Further experiments with fractions derived from respective plants revealed that *C. depressa* and *F. ailanthoides* contained potent oviposition stimulant(s) and *E. meliifolia*, both moderate stimulant(s) and weak deterrent(s). Negative oviposition response to *O. japonica* proved to be ascribed to the presence of weak deterrent(s) and the lack of any appreciable stimulant(s). In good coincidence with female oviposition responses, both neonate and 5th instar larvae performed well on *C. depressa* and *F. ailanthoides*, while larval performance was a little worse on *E. meliifolia*. On the other hand, the survivorships of two larval instars on *O. japonica* differed greatly from each other: whereas the mortality of neonates was very high, 5th instar larvae grew into pupae as well or better on this plant as on *E. meliifolia*. Our results, when taken together, suggest that the four rutaceous plants are arrayed as follows in decreasing order of acceptability: *C. depressa* = *F. ailanthoides* > *E. meliifolia* >> *O. japonica*.

We identified active compounds present in *Citrus depressa*, a host plant of *P. polytes*, involved in stimulation of egg-laying. Strong ovipositional activity was found to reside in aqueous fraction obtained from methanolic extract of *C. depressa*. The oviposition response of *P. polyte* was evoked by synergistic action of constituents of acidic, neutral and amphoteric and basic fraction separated from aqueous extract. Five major compounds, D-(-)-quinic acid, (-)-synephrine and choline as basic substances, L-(-)-proline and L-(-)-stachidrine as amphoteric ones, were identified as oviposition sutimulants and characterized the chemical composition of leaves of *C. depressa*. A mixture of five compounds showed noticeable oviposition-stimulatory activity, although each compound was inactive alone.

Two groups of compounds system contributed to evoke oviposition activity in the foliage of *F. ailanthoides*. Strong ovipositional activity was found in aqueous fraction obtained from

methanolic extract of *F. ailanthoides*. The oviposition response of *P. polyte* was evoked by synergistic action of constituents of two groups, neutral, amphoteric and basic fraction from Fa-3-3 and acidic, neutral and amphoteric fraction from Fa-3-1 and Fa-3-2. Such an oviposition stimulants system have not been known in Papilionidae butterfly.

Furthermore, using knowledges and experiences on the study about butterfly, we developed teaching materials and processes to enhance investigation learning for science in primary and secondary education. Method and pathway of feeding and bioassay were established to apply “butterfly” to learning science, especially biology and chemistry.

**Key word:** host selection, oviposition stimulant, *Papilio polytes*, Lepidoptera, Papilionidae, Rutaceae, *Citrus depressa*, *Fagara ailanthoides*, science education, teaching material