# The Role of Endogenous Phenolics in Host Plant Resistance Among *Malus* Taxa to Japanese Beetles

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**Nature of Work:** Japanese beetles (JB), *Popillia japonica* Newman, are destructive, highly polyphagus herbivores that show a general preference for plants in the Rosaceae family. Although *Malus* taxa are often found to be susceptible to JB, there are substantial differences in resistance among *Malus* species and cultivars (1,2). In a prior study, total phenolic levels in Malus were not correlated with JB resistance (2). The objectives of this study were to compare natural resistance to JB among *Malus* taxa and to evaluate the role of individual phenolics in host plant resistance.

The choice feeding assay was conducted with field grown trees, n=3. Defoliation ratings were done on August 14, 1995 and were conducted by two independent observers. No-choice feeding assays were conducted using the methods given by Fulcher et al. (2). After a 24 hour feeding period, conducted in a growth chamber, leaf area consumption was calculated to document host plant resistance. Beetles for artificial diets were handled according to Fulcher et al. (2). Artificial diets were made of sucrose, cellulose, agar, and one of the eight test compounds: phloridzin, phloretin, naringenin, kaempferol, rutin, quercetin, catechin, and chlorogenic acid (Sigma Co., St. Louis, MO) in 0, 1.0, 3.2, 10.0, 31.6 and 100.0 mM. Ten replications of a compound at each concentration were used. One female beetle was placed in each petri dish with a plug of diet and was set randomly in the growth chamber. After 24 h beetles were removed and fecal matter was collected and dried for 24 h at 70°C (Isotemp Oven 665F Fischer Scientific, Pittsburgh, Pa.) and weighed.

Endogenous levels of individual phenolics were analyzed by a reversed phase HPLC method adapted from Hunter et al. (3). Standard curves were prepared of phloridzin, phloretin, kaempferol, naringenin, chlorogenic acid, and catechin from standards (Sigma Co., St. Louis, Mo.). An isocratic solvent system of 75 water:25 methanol and 0.1% phosphoric acid was used. The column was an adsorbospere C18, 250 x 4.6 mm, containing a 5 um C18 bonded phase (Alltech Associates Inc., Deerfield, Ill). Compounds were detected at 254 nm with a UV detector (Millipore Corp., Bedford, Mass.).

**Results and Discussion:**Feeding injury in the choice test varied from 0 to 73 percent defoliation (Table 1) Under these conditions 8 taxa were resistant with less than 10% defoliation. Feeding under no-choice conditions ranged from 1.0 $cm^2$ to 7.6 $cm^2$ (Table 1). Under this intense feeding pressure M. Golden Raindrops™, *M.* baccata 'Jackii', and *M.* Harvest Gold™ were highly resistant with less than 2 $cm^2$ leaf area consumption. Six taxa were intermediate and *M.* 'Radiant' was statistically the most susceptible with 7.6 $cm^2$ leaf area consumption.

Although total phenolic content can sometimes influence insect feeding, the presence and concentration of specific phenolic constituents can be more important than total phenolic content (4). Phloridzin, and its hydrolysis product phloretin, were highly effective at deterring JB when present in artificial diets. The effective dose to reduce feeding by 25% ($ED_{25}$) for phloridzin was 3.2 mM. Phloretin had an $ED_{25}$ of 2.9mM. Conversely, rutin increased feeding by 174% of the control at the 100mM concentration.

HPLC analysis revealed a wide range in the concentration of different endogenous phenolics among taxa. Using stepwise multiple regression analysis, phloridzin was the only endogenous phenolic significantly related to feeding from either the choice or no-choice feeding assays ($p<0.05$). These data implicate phloridzin as an important endogenous feeding deterrent for the JBs. Although some resistant plants had low total phenolic contents (e.g. *Malus* baccata 'Jackii'), they appeared to have adequate levels of phloridzin to deter beetles from feeding (Table 1).

**Significance to Industry:** This research documented a broad range of natural resistance to feeding by adult JBs among taxa of *Malus*. Greater use of pest resistance plants will reduce the need for chemical controls, reduce production and maintenance costs, and aid in the development of more sustainable landscapes. In addition, identification of resistant genotypes provides the basic information needed for breeding new plants with enhanced pest resistance.

## Literature Cited

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**Table 1.** Resistance to Japanese beetle as measured by leaf area consumption, percent defoliation, and endogenous levels of phloridzin among crabapples *(Malus* spp.).

| Taxon | No ChoiceTest Leaf area consumed ($cm^2$) | Choice Test Percent Defoliation | Phloridzin mM/kg fresh weight |
|---|---|---|---|
| M. x'Schmitcutleaf Golden Raindrops™ | 0 99 | 1 | 154 |
| *M. baccata* 'Jackii' | 1.07 | 0 | 144 |
| *M. x'Hargozam'* Harvest Gold™ | 1 83 | 1 | 143 |
| *M. x'Branzam'* Brandywine™ | 3.29 | 1 | 177 |
| *M. floribunda* | 3.61 | 0 | 146 |
| *M. x'Naragansett'* | 3.63 | 3 | 80 |
| *M. x 'Robinson'* | 4.19 | 2 | 170 |
| *M. x 'Red* Splendor' | 4.84 | 26 | 34 |
| *M. x'Baskatong'* | 5.05 | 9 | 96 |
| *M. x 'Radiant'* | 7.62 | 73 | 34 |
| $^{LSD}0.05$ | 2.01 | 10 | 36 |