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Insect Antifeedant Activity of Flavones and Chromones against *Spodoptera litura*

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The antifeedant polymethylated flavones 5-hydroxy-3,6,7,8,4′-heptamethoxyflavone, 5-hydroxy-3,6,7,8-tetramethoxyflavone, and 5,6-dihydroxy-3,7-dimethoxyflavone have been isolated from the cudweed, *Gnaphalium affine* D. Don (Compositae). These flavonoids and authentic analogues showed insect antifeedant activity against the common cutworm (*Spodoptera litura* F.). In a previous paper, it was suggested that there was no substituent on the B-ring of the flavonoid for the beneficial antifeedant activity against the common cutworm. These flavonoids having a phenyl group as the B-ring and the chromone as elimination of the B-ring from the flavonoids were used to test the hypothesis of the previously described B-ring effect. The known fact is that *Sculletaria baicarensis* (Rutaceae) produced the 2-phenyl flavone. Test compounds and their methylated derivatives were prepared from this material for the structure−activity relationship (SAR) study of insect antifeedant activity. In spite of the 2-phenyl flavonoids, some tested compounds did not show any insect antifeedant activity against the common cutworm, although these inactive flavonoids were deficient in the 6-substituent group on the A-ring of the flavonoid. This 6-position-substituted derivative almost showed strong insect antifeedant activity against common cutworm. Moreover, the tested flavonoids having a hydroxyl group as a substituent on any of the positions tended to increase the activity. These results suggested the importance of the 6-position substitution on the flavonoid; however, hydrophilic substituents decreased the activity. Baicalein (5,6,7-trihydroxyflavone) derivatives did not show any activity despite having the 6-substituent derivative. Although the activity of some chromones increased the activity of the flavone, the bulky B-ring was a disadvantage for the antifeedant activity. It was suggested that the charge on C(3) and C(5) of the flavonoid was important for the biological activity. Additionally, an adequate hydrogen bonding property, which is different from lipophilicity, was an advantage for the activity on the basis of a QSAR analysis.

KEYWORDS: Flavone; chromone; *Sculletaria baicarensis*; antifeedant; *Spodoptera litura*

INTRODUCTION

The widespread flavonoids act as various functional secondary metabolites in plants. These functions have been reported as the constituents for the ultraviolet defense system against sunlight, as an antioxidant in the plant body (*1*), and as an attractant for pollinators as flower pigments (*2*). On the other hand, we know some flavonoids have a repellency property against some phytophagous insects and a termite acting as an antifeedant (*3*, *4*). These various properties of the flavonoids can be applied to the development of functional food, medicinal, and agrochemical agents. The phytophagous insect overcoming the plant defense system can use the flavonoids for their host selection (*5*). In a previous paper, we isolated four antifeedants from the Japanese annual weed, *Gnaphalium affine*, and described their derivatives for insect antifeedant activity (*6*). In this study, we have further examined the role of the chemical structures of the flavonoid in a structure−activity relationsip (SAR) study of insect antifeedants. We regard the lack of substituents on the B-ring as benefitical to the insect antifeedant property. The known fact is that *Sculletaria baicarensis* (Rutaceae) produced the 2-phenyl chromone (*7*). We prepared the test compounds from this source and synthesized some chromones for the evaluation of this hypothesis.

MATERIALS AND METHODS

General. ^{1}H and ^{13}C NMR spectra were measured on a JEOL 270EX (270 MHz) spectrometer. Mass spectra were taken at 70 eV (probe) using a Shimadzu GCMS 9100-MK. The compounds were separated by column chromatography on silica gel, Fuji silysia chemical BW-127ZH and BW-300 (Fuji Silysia Chemical, Ltd.). TLC used silica gel plates with a fluorescent indicator (Merck silica gel 60 F_{254}, 0.25 mm thick).

Insect Rearing. Common cutworms (*Spodoptera litura* F.) were reared on an artificial diet (Insecta LF, Nihon Nosan Kogyo Co.) in a controlled room environment at 26.5 °C and 60% humidity.

Evaluation of Test Compounds with Choice Leaf Disk Bioassay. The experimental setup was described in a previous report (*6*). Evaluation of the test compounds used the ED_{50} value of the antifeedant value calculated from the rate of consumption (*8*). A straight line was fitted to the points obtained from the bioassay, and the ED_{50} was

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calculated as the dose corresponding to the midpoint between complete inhibition and no effect by a computer program.

Preparation of Test Compounds. The commercial folk drug (*Scuretallia baicarensis*; Japanese name, Oogon) was extracted with methanol. The concentrated yellowish oil was separated by silica gel (Fuji Silysia Chemical, BW-127ZH) column chromatography with hexane/ethyl acetate 3:1. The fraction including the flavones was rechromatographed on the silica gel with the same solvent system. The final purification of the flavones was performed by recrystallization. Two flavones, norwogonin (**6**) and oloxylin A (**9**), were isolated from this extract. These natural compounds were characterized by spectrum analyses. For the preparation of the flavonoid methyl ethers of appropriate molar concentration of methyl iodide corresponding to the number of phenols in the chemical structure of the parent compound, we used the reactions with acetone containing potassium carbonate. After completion of the reaction by thin-layer chromatography (TLC), water was added to the product. The solution was extracted with ethyl acetate and dried over anhydrous sodium sulfate. The solvent was removed by a rotary evaporator under reduced pressure. These derivatives were characterized by spectrum analyses: worgonin (**4**), isowogonin (**5**), and moslosooflavone (**7**), prepared from norwogonin by methylation. Nobiletin (**11**) was separated from the folk drug made from Japanese orange peel by silica gel column chromatography. The chromones were synthesized by the coupling reaction of the phenol and ketone. The phenol, cresol, 3,5-dimethoxyphenol, and 3,4,5-trimethoxyphenol were coupled with acetoacetic ethylate under base conditions. These coupling reactions produced 2,7-dimethylchromone (**19**), 2-methyl-5,7-dimethoxychromone (**20**), and 2-methyl-5,6,7-trimethoxychromone (**21**), respectively. Chromone **18** and 7-methoxychromone (**23**) were prepared by Claisen condensation from the phenol and alkyl ester following dehydroxygenation with strong acid. Chrysin (5,7-dihydroxyflavone, **1**), apigenin (5,7,4′-trihydroxyflavone, **2**), luteolin (5,7,3′,4′-tetrahydroxyflavone, **3**), baicarein (5,6,7-trihydroxyflavone, **8**), flavone **12**, 6-methylflavone (**13**), and 6-hydroxyflavone (**13**) were purchased from Sigma Chemical Co., Ltd.

Worgonin (5,7-dihydroxy-8-methoxyflavone, **4**): yellow powder (hexane); mp 190-193 °C (lit. 203 °C); UV (MeOH) λ_{max} 300 (sh), 260 nm; EIMS, *m*/*z* (relative intensity) 284 (M^+, 45.4%), 269 (97.0%), 241 (37.0%), 140 (100%), 69 (73.9%); ^{1}H NMR ($CDCl_3$) δ 12.50 (1H, s, Ar-OH), 7.89−7.95 (2H, m, Ar-H), 7.54−7.60 (3H, m, Ar-H), 6.69 (1H, s, Ar-H), 6.45, [1H, s, C═C(3)H], 4.04 (3H, s, Ar-OCH_3).

Isowogonin (5,8-dihydroxy-7-methoxyflavone, **5**): yellow needles (hexane); mp 162−164.7 °C; EIMS, *m*/*z* (relative intensity) 284 (M^+, 100%), 266 (31.3%), 255 (20.8%), 238 (54.8%), 153 (18.0%); ^{1}H NMR ($CDCl_3$) δ 12.49 (1H, s, Ar-OH), 7.87−7.92 (2H, m, Ar-H), 7.48−7.56 (3H, m, Ar-H), 6.68, (1H, s, Ar-H), 6.62 [1H, s, C═C(3)H], 4.01 (3H, s, Ar-OCH_3).

Norwogonin (5,7,8-trihydroxyflavone, **6**): yellow powder (MeOH); mp 267−269.3 °C; EIMS, *m*/*z* (relative intensity) 270 (M^+, 100%), 168 (46.6%), 140 (17.9%), 69 (53.7%); ^{1}H NMR (acetone) δ 12.30 (1H, s, Ar-OH), 8.04−8.10 (2H, m, Ar-H), 7.56−7.64 (3H, m Ar-H), 6.78 (1H, s, Ar-H), 6.70 [1H, s, C═C(3)H]; ^{13}C NMR ($CDCl_3$) δ 186.9, 168.1, 157.3, 155.0, 151.4, 136.0, 135.9, 133.4, 133.3, 130.6, 108.9, 98.3.

Moslosooflavone (5-hydroxy-7,8-dimethoxyflavone, **7**): yellow needles (hexane); mp 181−183 °C; EIMS, *m*/*z* (relative intensity) 298 (M^+, 100%), 283 (92.4%), 269 (24.9%), 255 (62.4%), 153 (67.4%); ^{1}H NMR ($CDCl_3$) δ 12.68 (1H, s, Ar-OH), 7.89−7.92 (2H, m, Ar-H), 7.50−7.58 (3H, m, Ar-H), 6.69 (1H, s, Ar-H), 6.58 [1H, s, C═C(3)H], 3.98 (3H s, Ar-OCH_3), 3.93 (3H s, Ar-OCH_3).

Oloxylin A (5,7-dihydroxy-6-methoxyflavone, **9**): yellow powder (hexane); mp 184−186 °C (lit. 200−201, 231−232°C); UV (MeOH) λ_{max} 300 (sh), 256, 232 nm; EIMS, *m*/*z* (relative intensity) 284 (M^+, 85.5%), 269 (58.6%), 266 (42.4%), 69 (100%); ^{1}H NMR ($CDCl_3$) δ 13.10 (1H, s, Ar-OH), 7.85−7.92 (2H, m, Ar-H), 7.48−7.58 (3H, m, Ar-H), 6.78 (1H, s, Ar-H), 6.72 [1H, s, C═C(3)H], 4.15 (3H, s, Ar-OCH_3).

Mosloflavone (5-hydroxy-6,7-dimethoxyflavone, **10**): white needle (hexane); mp 146−148.7 °C; EIMS, *m*/*z* (relative intensity) 298 (M^+, 87.3%), 283 (86.6%), 269 (30.8%), 255 (79.5%), 153 (100%); ^{1}H NMR ($CDCl_3$) δ 12.68 (1H, s, Ar-OH), 7.86−7.93 (2H, m, Ar-H), 7.49−7.57 (3H, m, Ar-H), 6.69 (1H, s, Ar-H), 6.58 [1H, s, C═C(3)H], 3.98 (3H s, Ar-OCH_3), 3.93 (3H s, Ar-OCH_3).

Nobiletin (5,6,7,8,3′,4′-hexamethoxyflavone, **11**): yellow powder (hexane); mp 132−134 °C; EIMS, *m*/*z* (relative intensity) 402 (M^+, 24.3%), 387 (100%); ^{1}H NMR ($CDCl_3$) δ 7.48 (1H, dd, J = 2.5, 8.6 Hz, Ar-H), 7.34 (1H, d, J = 2.5 Hz, Ar-H), 6.92 (1H, d, J = 8.6 Hz, Ar-H), 6.57 [1H, s, C═C(3)H], 4.03 (3H, s, Ar-OCH_3), 3.96 (3H, s, Ar-OCH_3), 3.90 (3H, s, Ar-OCH_3), 3.89 (3H, s, Ar-OCH_3), 3.88 (6H, s, Ar-OCH_3 ×2); ^{13}C NMR ($CDCl_3$) δ 177.4, 161.1, 152.3, 151.9, 149.3, 148.3, 144.0, 130.8, 128.7, 123.9, 119.6, 114.7, 111.2, 108.6, 106.7, 62.2, 61.9, 61.7, 61.6, 56.0, 55.9.

6-Methoxyflavone (**15**): white needles (hexane); mp 162.7−164.3 °C; EIMS, *m*/*z* (relative intensity) 252 (M^+, 100%), 222 (32.0%), 150 (67.3%), 135 (20.8%), 122 (16.9%), 107 (41.9%); ^{1}H NMR ($CDCl_3$) δ 7.82−7.89 (2H, m, Ar-H), 7.42−7.49 (2H, m, ar-H), 7.53 (1H, d, J = 2.97 Hz, Ar-H), 7.22 (1H, dd, J = 9.23, 2.97 Hz, Ar-H), 6.72 [1H, s, C═C(3)H], 3.84 (3H, s, Ar-OCH_3); ^{13}C NMR ($CDCl_3$) δ 178.3, 163.2, 157.0, 151.1, 131.9, 131.5, 129.0, 126.2, 124.6, 123.8, 119.5, 106.8, 104.8.

7-Methoxyflavone (**17**): pearl pink solid (hexane); mp 103−105 °C; EIMS, *m*/*z* (relative intensity) 252 (M^+, 100%), 224 (49.6%), 209 (49.2%), 150 (36.4%), 122 (35.8%); ^{1}H NMR ($CDCl_3$) δ 8.14 (1H, d, J = 9.23 Hz, Ar-H), 7.89−7.97 (2H, m, Ar-H), 7.45−7.53 (3H, m, Ar-H), 6.99 (1H, d, J = 9.23 Hz, Ar-H), 6.97 (1H, s, Ar-H), 6.76 [1H, s, C═C(3)H], 3.94 (3H, s, Ar-OCH_3); ^{13}C NMR ($CDCl_3$) δ 177.9, 164.2, 163.0, 158.0, 131.9, 131.4, 129.0, 127.1, 126.2, 117.8, 114.4, 107.5, 100.4.

Chromone (**18**): white powder (hexane); mp 38.7−40.1 °C; EIMS, *m*/*z* (relative intensity) 146 (M^+, 100%), 118 (63%), 92 (46.4%), 63 (46.3%); ^{1}H NMR ($CDCl_3$) δ 8.20 (1H, dd, J = 7.92, 1.65 Hz, Ar-H), 7.86 [1H, d, J = 5.94 Hz, C(2)H═C], 7.67 (1H, ddd, J = 7.92, 6.92, 1.65 Hz, Ar-H), 7.44 (1H, d, J = 7.92 Hz, Ar-H), 7.38 (1H, ddd, J = 7.92, 6.92, 1.65 Hz, Ar-H), 6.33 [1H, d, J = 5.94 Hz, C═C(3)H]; ^{13}C NMR ($CDCl_3$) δ 177.4, 156.4, 155.2, 133.6, 125.6, 125.1, 124.7, 118.0, 112.8.

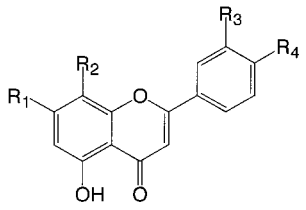
2,7-Dimethylchromone (**19**): white solid (hexane); mp 95.7−98 °C; EIMS, *m*/*z* (relative intensity) 174 (M^+, 100%), 146 (45.1%), 134 (33.7%), 106 (22.3%); ^{1}H NMR ($CDCl_3$) δ 7.96 (1H, d, J = 8.25 Hz, Ar-H), 7.01 (1H, s, Ar-H), 7.09 (1H, d, J = 8.25 Hz, Ar-H), 6.04 [1H, s, C═C(3)H], 2.37 (3H, s, Ar-CH_3), 2.27 [3H, s, C═C(2)-CH_3]; ^{13}C NMR ($CDCl_3$) δ 178.2, 165.8, 156.5, 144.6, 126.3, 125.2, 121.1, 117.4, 110.3, 21.7, 20.5.

2-Methyl-5,7-dimethoxychromone (**20**): pearl pink solid (hexane); mp 168.3−171.3 °C; EIMS, *m*/*z* (relative intensity) 220 (M^+, 87.8%), 192 (100%), 177 (87.3%), 149 (21.2%); ^{1}H NMR ($CDCl_3$) δ 6.43 (1H, d, J = 2.31 Hz, Ar-H), 6.29 (1H, d, J = 2.31 Hz, Ar-H), 5.95 [1H, s, C═C(3)H], 3.86 (3H, s, Ar-OCH_3), 3.85 (3H, s, Ar-OCH_3), 2.53 [3H, s, C═C(2)-CH_3]; ^{13}C NMR ($CDCl_3$) δ 162.7, 161.1, 159.1, 156.9, 154.5, 111.3, 104.8, 95.4, 93.3, 55.8, 55.7.

2-Methyl-5,6,7-trimethoxychromone (**21**): yellow powder (hexane); mp 107.3−110.7 °C; EIMS, *m*/*z* (relative intensity) 250 (M^+, 100%), 235 (91.0%), 207 (56.3%), 179 (18.7%), 164 (23.2%), 149 (23.6%); ^{1}H NMR ($CDCl_3$) δ 6.65 (1H, s, Ar-H), 6.04 [1H, s, C═C(3)H], 3.96 (3H, s, Ar-OCH_3), 3.92 (3H, s, Ar-OCH_3), 3.86 (3H, s, Ar-OCH_3), 2.56 [3H, s, C═C(2)-CH_3]; ^{13}C NMR ($CDCl_3$) δ 160.9, 156.2, 153.5, 151.6, 151.3, 139.2, 113.0, 108.0, 96.2, 61.3, 60.9, 56.2, 22.9.

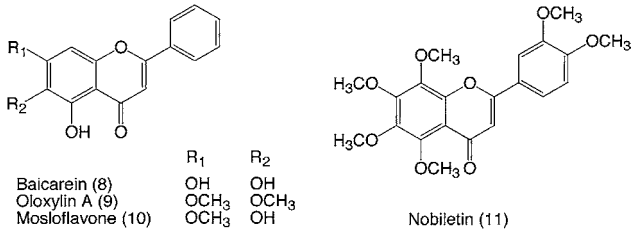
2-Methyl-7-methoxychromone (**22**): pearl pink powder (hexane); mp 158−160.7 °C; EIMS, *m*/*z* (relative intensity) 190 (M^+, 100%), 162 (74.7%), 147 (94.5%), 91 (25.5%); ^{1}H NMR ($CDCl_3$) δ 7.50 (1H, d, J = 8.91 Hz, Ar-H), 6.86 (1H, dd, J = 8.58, 2.31 Hz, Ar-H), 6.82 (1H, d, J = 2.31 Hz, Ar-H), 6.14 (1H, s, Ar-H), 6.04 [1H, s, C═C(3)H], 3.87 (3H, s, Ar-OCH_3), 2.40 [3H, s, C(2)-CH_3]; ^{13}C NMR ($CDCl_3$) δ 162.7, 161.3, 155.3, 152.5, 125.5, 113.6, 112.3, 112.0, 100.9, 55.7, 18.6.

7-Methoxychromone (**23**): white powder (isopropyl alcohol); mp 107−109 °C; EIMS, *m*/*z* (relative intensity) 176 (M^+, 100%), 148 (29.5%), 133 (40.4%), 122 (23.4%); ^{1}H NMR ($CDCl_3$) δ 8.09 (1H, d, J = 8.91 Hz, Ar-H), 7.78 [1H, d, J = 5.94 Hz, HC(2)═C], 6.96 (1H, dd, J = 2.31, 8.91 Hz, Ar-H), 6.82 (1H, d, J = 2.31 Hz, Ar-H), 6.27 [1H, d, J = 5.94 Hz, C═C(3)H], 3.89 (3H, s, Ar-OCH_3); ^{13}C NMR ($CDCl_3$) δ 176.9, 164.0, 158.1, 154.8, 127.0, 118.6, 114.4, 112.8, 100.2, 55.7.



	R_1	R_2	R_3	R_4
Chrysin (1)	OH	H	H	H
Apigenin (2)	OH	H	H	OH
Luteolin (3)	OH	H	OH	OH
Wogonin (4)	OH	OCH_3	H	H
Isowogonin (5)	OCH_3	OH	H	H
Norwogonin (6)	OH	OH	H	H
Moslosooflavone (7)	OCH_3	OCH_3	H	H

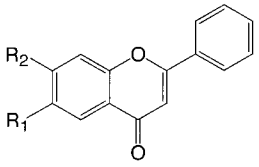
Figure 1. 6-Unsubstituted flavone tested for antifeedant bioassay against *S. litura*.



	R_1	R_2
Baicarein (8)	OH	OH
Oloxylin A (9)	OCH_3	OCH_3
Mosloflavone (10)	OCH_3	OH

Nobiletin (11)

Figure 2. 6-Methoxyflavones tested for antifeedant bioassay against *S. litura*.



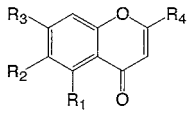
	R_1	R_2
Flavone (12)	H	H
6-Methylflavone (13)	CH_3	H
6-Hydroxyflavone (14)	OH	H
6-Methoxyflavone (15)	OCH_3	H
7-Hydroxyflavone (16)	H	OH
7-Methoxyflavone (17)	H	OCH_3

Figure 3. Monosubstituted flavones tested for antifeedant bioassay against *S. litura*.

Computational Methods. The antifeedant flavonoids including those in the previous paper were compared with respect to formal charge on the benzopyranone and the biological activity by computational analysis and other physicochemical parameters. The geometry of the structures has been optimized on minimum energy using the AM1 model Hamiltonian including WinMopac software. We had a training set of the 19 antifeedant flavonoids and chromones by QREG software (*9*). As a result, the biological activity was not related to the HOMO and LUMO parameters. On the other hand, the formal charge on the structure and hydrogen bonding parameter (HB) calculated from the R_f value on the Si-OH TLC related to the biological activity. We suggested a primitive formula for the expected value of the insect antifeedant activity against the common cutworm.

RESULTS AND DISCUSSION

We evaluated the 6-unsubstituted flavones (**Figure 1**), the 6-substituted flavones (**Figure 2**), the monosubstituted in the A-ring the flavones (**Figure 3**), and chromones (**Figure 4**) for insect antifeedant activities against the common cutworm. From the choice leaf-disk bioassay, we recognized the B-ring of the flavonoid effect based on the data from the previous paper and nobiletin (**11**) and the chromones (**20**) in this study. Nobiletin (**11**) showed decreased activity by 1 order or magnitude toward tangeretin (0.28) and 5-hydroxyauranetin (0.11) (**Table 1**). These



	R_1	R_2	R_3	R_4
Chromone (18)	H	H	H	H
2,7-Dimethylchromone (19)	H	H	CH_3	CH_3
2-Methyl-5,7-dimethoxychromone (20)	OCH_3	H	OCH_3	CH_3
2-Methyl-5,6,7-trimethoxychromone (21)	OCH_3	OCH_3	OCH_3	CH_3
2-Methyl-7-methoxychromone (22)	H	H	OCH_3	CH_3
7-Methoxychromone (23)	H	H	OCH_3	H

Figure 4. Chromones tested for antifeedant bioassay against *S. litura*.

results suggested that the insect antifeedant activity decreased due to the 2-position bulky substituents. In this study, the flavones had a phenyl group as the B-ring, and the chromones as derivatives of the elimination of the B-ring were used for the evaluation of the previous hypothesis; some of them inhibited common cutworm feeding. On the other hand, there were some compounds that showed decreased insect antifeedant activity in spite of haivng the phenyl group as the B-ring. These results suggested that the biological activity required not only the important 2-position substituent but also the substituted pattern on the A-ring of these insect antifeedants, flavonoids, and chromones. Most of the test compounds having a hydroxyl group did not show the antifeedant activity. Additionally, there was no active compound having a substituent on the 6-position of the benzopyranone in the oxygenated derivatives.

There were some reports that the insect taste sensitivity was dominated by $GABA_A$ receptors and active compounds against one of the receptors acted as the insect antifeedant (*10*). There were some reports about antifeedants having effective $GABA_A$ receptors of pictotoxinin and strychnine (*11*). Originally, these compounds were used in the insecticidal studies. Recently, there was a report that some of our tested flavones bind to the benzodiazepine receptor (*12*), and insecticidal natural compounds having the same target site (*13*) were reported. Ai et al. reported that the most active compound was 6-methylflavone (*12*), but the flavones have hydroxyl groups with high binding activity against the benzodiazepine receptor. In our case, the flavones having a hydroxyl group did not show any insect antifeedant activity, because there was a difference between the receptor assay and topical application. Commonly, the receptor assay canceled the biological barrier, for example, penetrability or transition. We believe the data did not correlate with the benzodiazepine receptor binding data because the penetrability was significantly related.

These results showed an unclear correlation between the chemical structures and biological activity. In the QSAR analysis of the benzodiazepine binding activity, the activity was important for the molecule refractivity (MR) and lipophilicity (π) in the 6-position of the flavone (*14*). On the basis of our results of the computational analysis with electric charge on the benzopyranone and HB parameter, we concluded that the formal charge on the benzopyranone calculated at the AM1 level was more important than the values of HOMO (−0.129), LUMO (0.07), and ΔH-L (0.121) (correlation coefficient toward pED_{50}) (*9*). The best relation to the biological activity was the melting point parameter (−0.575) (correlation coefficient toward pED_{50}). None of the compounds having a melting point >250 °C showed any insect antifeedant activity in this test. The parameters most related with biological activity were the electric charges at C(3) and C(5). We adopted four descriptors: the dipole moment, the electric charges of both the 3- and 5-carbons (δ3, 5), and a parameter relating the hydrogen bond on the Si−OH TLC (HB). The HB parameter is defined by an adequate solvent system;

Table 1. Structural Features and Evaluation of Insect Antifeedant Activity for Flavones and Chromones Tested against *S. litura* F.

test compound	A-ring 5	A-ring 6	A-ring 7	A-ring 8	B-ring 3′	B-ring 4′	antifeedant activity, ED_{50}[a] (μmol/cm^2)
flavones							
chrysin (**1**)	OH	H	OH	H	H	H	2.50
apigenin (**2**)	OH	H	OH	H	H	OH	inactive
luteolin (**3**)	OH	H	OH	H	OH	OH	inactive
wogonin (**4**)	OH	H	OH	OCH_3	H	H	2.00
isowogonin (**5**)	OH	H	OCH_3	OH	H	H	inactive
norwogonin (**6**)	OH	H	OH	OH	H	H	1.52
moslosooflavone (**7**)	OH	H	OCH_3	OCH_3	H		1.30
baicalein (**8**)	OH	OH	OH	H	H	H	0.96
oloxylin A (**9**)	OH	OCH_3	OH	H	H	H	inactive
mosloflavone (**10**)	OH	OCH_3	OCH_3	H	H	H	inactive
nobiletin (**11**)	OCH_3	OCH_3	OCH_3	OCH_3	OCH_3	OCH_3	5.60
flavone (**12**)	H	H	H	H	H	H	0.11
6-methylflavone (**13**)	H	CH_3	H	H	H	H	0.035
6-hydroxyflavone (**14**)	H	OH	H	H	H	H	inactive
6-methoxyflavone (**15**)	H	OCH_3	H	H	H	H	0.15
7-hydroxyflavone (**16**)	H	H	OH	H	H	H	inactive
7-methoxyflavone (**17**)	H	H	OCH_3	H	H	H	0.059
chromones							
chromone (**18**)	H	H	H	H			0.168
2,7-dimethylchromone (**19**)	H	H	CH_3	H			0.102
2-methyl-5,7-dimethoxychromone (**20**)	OCH_3	H	OCH_3	H			inactive
2-methyl5,6,7-trymethoxychromone (**21**)	OCH_3	OCH_3	OCH_3	H			0.41
2-methyl-7-metrhoxychromone (**22**)	H	H	OCH_3	H			inactive
7-methoxychromone (**23**)	H	H	OCH_3	H			0.096

[a] ED_{50} value given >10 μmol/cm^2 showed no activity in this table.

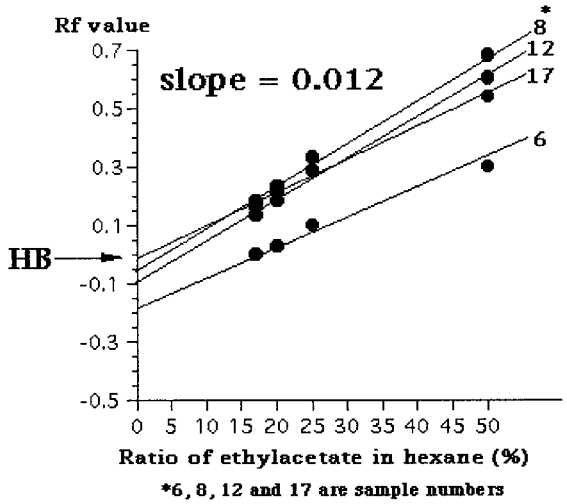


Figure 5. Conception of hydrogen bonding parameter (HB).

R_f values of most compounds strictly rely on the constitution of the eluents developing the TLC analysis. The differential condition of the TLC analysis defining the hexane/ethyl aceate eluent system was able to normalize the coefficient (HB) by the R_f value $= 0.12x +$ HB ($x =$ concentration of ethyl acetate, %) (**Figure 5**). Improvement of the correlation was achieved using this HB parameter (**Figures 6** and **7**).

$$pED_{50} = -0.195 \text{ dipole} + 6.487\ \delta 3 - 5.155\ \delta 5 - 0.119 \log P + 8.091$$

$$(n = 19;\ r^2 = 0.73) \qquad \textbf{(Figure 6)}$$

$$pED_{50} = -0.129 \text{ dipole} + 6.434\ \delta 3 - 5.290\ \delta 5 - 0.486 \text{ HB} + 8.085$$

$$(n = 19;\ r^2 = 0.79) \qquad \textbf{(Figure 7)}$$

In summary, various flavones and chromones were evaluated for the insect antifeedant activity against common cutworms.

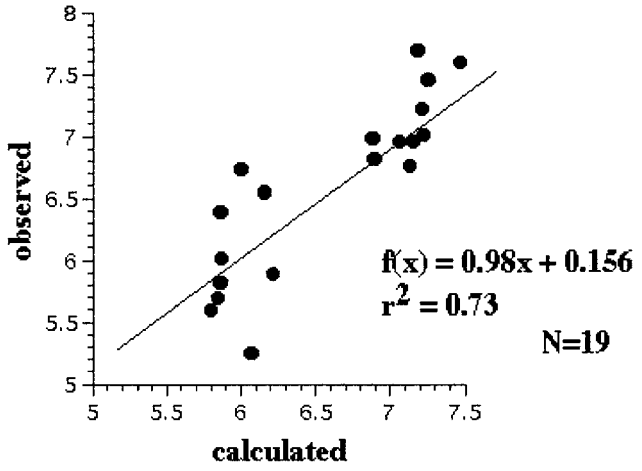


Figure 6. Correlation between observed and calculated biological activities of flavonoids and chromones.

The results suggested that the introduction of a bulky substituent to the 2-position decreased this activity. On the other hand, the introduction of a substituent to the 6- or 7-position increased the activity. We suggested that our prediction of the antifeedant activity requires a positive charge at the C(3) and a negative charge at the C(5) nuclei and a high hydrogen bonding property. In other words, increasing the insect antifeedant activity required the introduction of an electron donor substituent to the 6- or 2-position of benzopyranone. We showed that there was a good correlation between the insect antifeedant activity and R_f value from the TLC analysis in the same eluent for the dihydrobenzofurans (*15*). However, we are subsequently analyzing the QSAR by considering the other parameters.

Although the decrease in the activity and number of hydroxyl groups correlated, this was attributable to the penetrability toward the receptor in the insect taste-sensitive organ. The reason

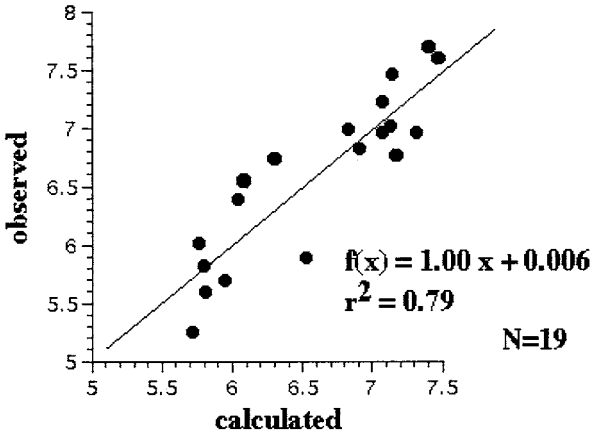


Figure 7. Correlation between observed and calculated biological activities of flavonoids and chromones.

for the reduction of the activity is the poor solubility of the flavonoids as the hydroxyl groups increase, thus facilitating the formation of crystals.

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