Toxicity of Isothiocyanates Produced by Glucosinolates in Brassicaceae Species to Black Vine Weevil Eggs

Vladimir Borek, Leslie R. Elberson,† Joseph P. McCaffrey, and Matthew J. Morra*

Department of Plant, Soil, and Entomological Sciences, University of Idaho, Moscow, Idaho 83844-2339

Control of the black vine weevil, *Otiorhynchus sulcatus* (F.), with allelochemicals produced from glucosinolates may be possible; however, plant-derived isothiocyanates are not readily available for bioassays. Our objective was to predict the toxicity of plant-derived isothiocyanates using a model developed with commercially available compounds. Contact toxicities of 12 organic isothiocyanates were determined by dipping black vine weevil eggs into isothiocyanate solutions. Quantitative relationships between the molecular structure of the isothiocyanates and their toxicities were estimated by regressing lethal concentrations against the compound's respective physiochemical parameters. Isothiocyanate polarity (log octanol/water partition coefficient) had the most significant effect on observed toxicities, whereas electronic and steric characteristics were unimportant. Using this linear structure−activity relationship, we predict that the highest contact toxicities to black vine weevil eggs will result from glucosinolates producing isothiocyanates with higher numbers of carbon atoms or those bearing sulfinyl, thio, or aromatic moieties.

Keywords: *Glucosinolates; isothiocyanates; allelochemicals; Brassica spp.; soil fumigation*

INTRODUCTION

Black vine weevil, *Otiorhynchus sulcatus* (F.) (Coleoptera: Curculionidae), is a serious polyphagous pest of high-value nursery, greenhouse, and field-cultivated crops. It has a reported host range of over 100 plant species and a distribution including many European countries, the eastern and western United States, Canada, Australia, New Zealand, Japan, and Chile (Essig, 1933; Masaki et al., 1984; Prado, 1988; Warner and Negley, 1976). Larval root feeding causes most of the damage to the host species. Early instars feed on small rootlets while later instars feed on larger roots or burrow into the crowns of plants (Moorhouse et al., 1992; Smith, 1932). Weevil infestations in Idaho have caused the removal of almost 1000 ha of hops from production during the past 10 years (Baird et al., 1992). Root weevils are Oregon's most significant ornamental pest and they are reported as an increasing problem in California nurseries (Capizzi, 1981, Parrella and Keil, 1984).

Soil fumigation is commonly used to control a wide array of soil-borne diseases, nematodes, insects, and weeds. However, the list of fumigants available for pest management is being reduced due to environmental and human health concerns, and prohibitive costs of reregistration. The most common fumigant, methyl bromide, will undergo a gradual phaseout starting in 1999 and will not be produced in or imported into the United States after the year 2005. Production of alternate fumigants, Telone and Vorlex, containing the active ingredients dichloropropene/dichloropropane and methyl isothiocyanate has already been canceled in California (White, 1994).

* Author to whom correspondence should be addressed (telephone, 208-885-6315; fax, 208-885-7760; e-mail, mmorra@uidaho.edu).

† Present address: USDA-ARS Western Regional Plant Introduction Station, WSU, Pullman, WA, 99164-6402.

These limitations have spurred interest in the development of alternative control strategies for soil-borne pests. One such approach is to use naturally occurring plant toxicants in integrated pest management strategies. Plants of the order Capparales, especially agriculturally important *Brassica* spp. of the Brassicaceae family, have shown insecticidal, fungicidal, nematicidal, and herbicidal properties (Lazzeri et al., 1993; Lichtenstein et al., 1964; McCloskey and Isman, 1993; Mojtahedi et al., 1991; Smolinska et al., 1997; Elberson et al., 1996; Brown and Morra, 1996; Borek et al., 1997). These toxic effects are linked to the biologically active degradation products of glucosinolates. Glucosinolates themselves possess limited biological activity; however, enzymatic degradation by myrosinase (thioglucoside glucohydrolase, EC 3.2.3.1) results in the formation of a number of allelochemicals (Chew, 1988; Underhill, 1980; Brown and Morra, 1997). More than 20 different aliphatic and aromatic isothiocyanates together with other potential allelochemicals have been identified among degradation products of glucosinolates originating from *Brassica napus*, *Brassica hirta*, *Brassica campestris*, *Brassica juncea*, and *Brassica nigra* (Brown et al., 1991; Spencer and Daxenbichler, 1980; Brown and Morra, 1996, 1995). Practical use of glucosinolate-containing plant tissues for pest management will require the determination of application rates necessary to obtain effective suppression of target pests. Unfortunately, most plant-derived isothiocyanates are not available for toxicity testing in bioassays.

Methyl isothiocyanate was a commonly used and effective active ingredient of commercial soil fumigants; however the relative toxicities of other isothiocyanates are poorly understood. It is difficult to predict the toxic effect of *Brassica* spp. soil amendments, even when the qualitative and quantitative composition of glucosinolates and their conversion rate to isothiocyanates are known, because the relationship of structure to activity

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Table 1. Physicochemical Qualities of Commercially Available Isothiocyanates Used in Bioassays of Acute Toxicity to Black Vine Weevil Eggs[a]

R group	FW	log *P*	MR	σ
methyl	73.1	0.43	21.33	0.00
ethyl	87.1	0.78	26.08	−0.10
allyl	99.2	1.17	30.49	0.23
propyl	101.2	1.25	30.60	−0.12
butyl	115.2	1.64	35.20	−0.13
pentyl	129.1	2.04	39.80	−0.16
phenyl	135.2	1.77	36.76	0.60
cyclohexyl	141.2	2.02	42.34	−0.15
benzyl	149.2	2.21	45.94	0.22
phenethyl	163.2	2.46	50.70	0.08
t-octyl	171.3	2.90	53.28	−0.06
2-naphthyl	185.3	3.12	57.56	−0.02

[a] Abbreviations: FW, formula weight; log *P*, hydrophobic parameter expressed as the log of the octanol/water partition coefficient; MR, parameter for steric effects as described using molecular refraction; σ, parameter for electronic effects.

has not been assessed. We previously studied the contact toxicity of six commercially available isothiocyanates to eggs of black vine weevil and determined that aromatic isothiocyanates are more toxic than aliphatic isothiocyanates (Borek et al., 1995). The effects of specific physicochemical characteristics of individual isothiocyanates could not be determined because of the relatively small number of compounds tested.

Our objective in the current work was to estimate the toxicity of naturally occurring isothiocyanates by developing quantitative structure−activity relationships (QSAR) using commercially available compounds (Lipnick, 1991). We chose black vine weevil eggs for our bioassays because this stage is available in large numbers, amenable to handling, and an appropriate target for weevil control. Toxicological studies of black vine weevil eggs with 12 commercially available isothiocyanates that possess a broad range of molecular structures were performed. Relative toxicities of the isothiocyanates were related to specific structural attributes to develop a structure versus toxicity model. Toxicities in the form of LC_{50} values can thus be predicted for isothiocyanates not commercially available, but present in Brassicaceae species. This model facilitates prediction of relative differences in pest control efficacy among plants with specific glucosinolate profiles. Such a model can be used in breeding programs to guide selection of cultivars having elevated concentrations of those glucosinolates producing the most toxic isothiocyanates. Utilization of these cultivars as a green manure crop would result in the potential slow release of isothiocyanates during growth and a flush of isothiocyanate after soil incorporation.

MATERIALS AND METHODS

Bioassay Chemicals. Isothiocyanates assayed for activity against black vine weevil eggs together with selected physicochemical characteristics are listed in Table 1. Isothiocyanates were obtained from Aldrich (Milwaukee, WI), except butyl and phenethyl isothiocyanate (Sigma, St. Louis, MO) and phenyl isothiocyanate (Baker, Phillipsburg, NJ). Purity of all isothiocyanates was at least 97%. Acetone solutions (HPLC grade, Fisher Scientific, Pittsburgh, PA) with isothiocyanate concentrations ranging between 0.01 and 100.0 mg/mL were assayed for contact toxicity. Solutions lower in concentration than 10 mg/mL were prepared by dilution of higher concentration stock solutions. Stock solutions were placed in 5-mL glass vials sealed with gastight PTFE-lined caps and stored at −20 °C.

Egg Collection and Preconditioning. Adult black vine weevils were collected in early June from a hop yard in Greenleaf, ID. Adults were maintained on strawberry foliage in a laboratory colony. Eggs were collected at 2-day intervals. Details of egg pretreatment procedures were previously described (Borek et al., 1995). Eggs were kept for 3 days above a saturated solution of sodium chloride, 1 day on wet filter paper, and again for 1 day above a saturated solution of sodium chloride. Only eggs with a melanized chorion, turning yellow-brown during the preconditioning period, were used in the toxicity assay since nonmelanized eggs are not viable (Moorhouse et al., 1992).

Treatments. The procedure for egg exposure to the isothiocyanates was as described by Borek et al. (1995). Eggs, placed in modified plastic syringe tips with inserted porous bottoms, were treated for 2 min (± 5 s) with acetone solutions containing a single isothiocyanate. Syringe tips containing the treated eggs were held vertically and spaced 2 cm apart. Eggs were incubated at 24 ± 2 °C and maintained at 100% relative humidity by placing them above deionized water in a closed plastic box. After a 14-day incubation period, we counted the number of unhatched eggs in each replicate.

Unequal solubility of isothiocyanates in water, instability of their water emulsions, and the toxic effect of emulsifiers required that acetone be used as a solvent. In preliminary studies without acetone we found that to obtain a stable water emulsion (10 min) of those isothiocyanates with higher molecular masses (2-naphthyl, phenethyl) up to 5% of the surfactant Triton X-100 (Sigma, St. Louis, MO) was required. Such a high concentration of surfactant produced mortality of eggs in the control greater than 20%. Two-minute treatment of eggs with pure acetone resulted in an acceptable mortality rate within a range of 5 to 10%. This rate is consistent with egg mortality obtained after a 2-min treatment of eggs with deionized water and is comparable to rates found in other black vine weevil studies (Maier, 1981; Mason, 1960; Moorhouse, 1990). Longer exposure times increased background mortality to levels prohibiting the accurate calculation of isothiocyanate toxicities. Acetone treatment caused a higher vulnerability of eggs to fungal infections, most likely because of epicuticular wax disruption and removal. We minimized such infections by frequently autoclaving our equipment. In agreement with other observations, we found humidity as the most critical environmental factor controlling egg viability (Shanks and Finnigan, 1973; Montgomery and Nielsen, 1979). Relative humidity of the posttreatment environment lower than approximately 90% significantly reduced viability and increased egg mortality in control tests to rates higher than 20%. However, even with relative humidity kept near 100%, we observed high variability in natural viability of eggs and in their responses to treatments. Variability in egg viability has been observed in other studies (Garth and Shanks, 1978; Maier, 1981), possibly being caused by periodic cycles in female oviposition activity (Nielsen and Dunlap, 1981; Stenseth, 1979).

Experimental Design. At least six concentrations of 12 isothiocyanates were replicated seven times to complete the bioassays. Each isothiocyanate concentration was tested on an experimental unit containing approximately 10 black vine weevil eggs. The toxicity assay was broken into uniform blocks as follows: seven replicates of three different isothiocyanates at two concentration levels and an acetone control were placed in random pattern in a 7 × 7 grid. Data were accumulated until each isothiocyanate was tested at a minimum of six concentrations, resulting in mortality responses between 0 and 100%. Black vine weevil eggs and predetermined isothiocyanate concentrations were selected randomly and solutions were freshly prepared for each block (Robertson and Preisler, 1992).

Data Analyses. We calculated descriptive statistics and conducted probit analysis (normal model) using SAS for Windows software (SAS, 1993). We discarded entire replication blocks in cases where egg mortality in the control exceeded 20% and when any fungal infection was observed. Lethal concentrations were estimated from data of cumulative rep-

Table 2. Contact Toxicity of Twelve Organic Isothiocyanates to Eggs of Black Vine Weevil[a]

isothiocyanate	relative potency (95% FL)	*n*	NR ± SE	log-(slope ± SE)	LC_{50} (μmol/mL) (95% FL)	χ^2	prob> (χ^2)	pLC_{50} (mL/mmol)[b] observed	pLC_{50} (mL/mmol)[b] calculated
methyl	1.00 (0.84−1.51)	245	0.218 ± 0.085	5.965 ± 1.523	88.41 (58.62−105.03)	2.63	0.6220	1.1	1.08
ethyl	2.63 (2.15−3.78)	252	0.095 ± 0.077	3.610 ± 0.713	33.62 (23.37−41.15)	6.14	0.1886	1.5	1.37
propyl	4.30 (3.48−6.30)	256	0.092 ± 0.082	3.401 ± 0.674	20.58 (14.03−25.44)	1.68	0.7939	1.7	1.85
allyl	4.97 (3.99−7.40)	243	0.133 ± 0.092	3.784 ± 0.724	17.79 (11.95−22.18)	2.52	0.6415	1.7	1.77
butyl	17.93 (15.62−21.51)	258	0.058 ± 0.042	3.957 ± 0.544	4.930 (4.11−5.66)	2.61	0.6248	2.3	2.25
phenyl	24.65 (17.76−50.49)	265	0.009 ± 0.159	2.993 ± 0.646	3.587 (1.75−4.97)	6.00	0.1993	2.4	2.38
benzyl	28.21 (17.86−99.40)	265	0.001 ± 0.246	2.764 ± 0.711	3.134 (0.88−4.94)	5.23	0.2648	2.5	2.83
pentyl	44.25 (33.58−76.68)	258	0.022 ± 0.122	3.104 ± 0.626	1.998 (1.15−2.63)	3.48	0.4808	2.7	2.65
phenethyl	53.65 (36.50−137.71)	265	0.062 ± 0.185	2.872 ± 0.683	1.648 (0.64−2.42)	5.99	0.2000	2.8	3.08
cyclohexyl	57.04 (34.67−401.86)	258	0.066 ± 0.246	2.505 ± 0.794	1.550 (0.22−2.55)	1.21	0.8768	2.8	2.63
t-octyl	485.24 (336.67−1302.06)	265	0.022 ± 0.143	2.306 ± 0.611	0.182 (0.068−0.26)	7.46	0.1136	3.7	3.53
2-naphthyl	701.28 (461.41−1968.16)	265	0.047 ± 0.116	1.181 ± 0.459	0.126 (0.045−0.19)	7.37	0.1174	3.9	3.75

[a] Abbreviations: Relative potency, LC_{50} of methyl isothiocyanate divided by LC_{50} of respective compound; FL, fiducial limits; *n*, number eggs used in assay (excluding control); NR, natural response of eggs in control; LC_{50}, concentrations lethal to 50% of the tested population; χ^2, chi−square value; pLC_{50}, log of the reciprocal value of median molar lethal concentration. For an explanation of the statistical parameters see Finney (1971) and Russell at al. (1977). [b] Observed, values obtained from bioassay results; calculated, values obtained from model predictions.

licates. A pLC_{50} value represented by the log of the reciprocal value of median molar lethal concentration, $\log(1/LC_{50})$, was used as an insecticidal activity index (Hansch et al., 1973).

Toxicity Model. Hansch's equation was used to study the relationship between the toxicity of isothiocyanates and their physicochemical parameters (Hansch et al., 1973):

$$pLC_{50} = a \log P + bE_s + c\sigma + d \quad (1)$$

In this equation $\log P$ is the hydrophobic parameter expressed as the log of the octanol/water partition coefficient; E_s is the parameter for steric effects; σ is the parameter for electronic effects; and a, b, c, and d are coefficients of the regression equation (Table 1). Values of hydrophobic and steric parameters for particular isothiocyanates were calculated using HyperChem and ChemPlus v. 3.1 software (HyperCube, Waterloo, Ontario, Canada). We used molecular refraction as the parameter for describing steric effects (Hansch et al., 1973) and obtained values for the electronic parameters of substituents from published data (Martin, 1978). Regression analysis using the parameters in Table 1 was performed using SAS (1993).

RESULTS AND DISCUSSION

Isothiocyanate Toxicities. We used an average of 257.9 ± 2.3 eggs (mean ± SE) in each isothiocyanate assay. An irregular number of eggs was used because of fluctuating female oviposition activity and discarded data. The average mortality response of eggs to the 2-min control treatment in acetone was 6.9 ± 1.8% (mean ± SE).

The results of probit analysis of mortality responses of black vine weevil eggs to concentrations of the 12 isothiocyanates are presented in Table 2. Fiducial intervals of effective concentrations were calculated for the 95% level of probability. Methyl isothiocyanate was the least toxic and 2-naphthyl isothiocyanate the most toxic of the compounds tested. Relative toxicities of these compounds with respect to methyl isothiocyanate are numerically expressed as relative potency in Table 2 (Finney, 1971). We found the following ascending order of relative toxicities in the set of tested isothiocyanates: methyl < ethyl < propyl < allyl < butyl < phenyl < benzyl < pentyl < phenethyl < cyclohexyl < *tert*-octyl < 2-naphthyl.

These results do not support our original assumption that the presence of an aromatic ring in a molecule is essential for an increase in toxicity (Borek et al., 1995). For example, nonaromatic *tert*-octyl isothiocyanate has a relatively high toxicity and aliphatic cyclohexyl isothiocyanate has a comparable toxicity with aromatic phenethyl isothiocyanate. In fact our data suggest that isothiocyanate contact toxicity increases with molecular weight (Table 1).

The toxicity of organic isothiocyanates has not been studied extensively or comprehensively in the past. However, insecticidal activities of single isothiocyanates (methyl, allyl, benzyl, phenethyl) have been demonstrated (Lichtenstein et al., 1962, 1964; Lowe et al., 1971; Ahman, 1986; Seo and Tang, 1982; Toba, 1984; Williams et al., 1993). The order of toxicity, isopropyl > methyl > ethyl isothiocyanate as fumigants to the rice weevil (Roark and Cotton, 1930), is consistent with results reported here. Lehman (1933, 1942) found allyl isothiocyanate more toxic than ethyl isothiocyanate when tested as fumigants for wireworms.

Toxicity Model. To assess quantitatively the dependence of isothiocyanate toxicity on structure, data from black vine weevil bioassays was modeled using regression analysis. Three physicochemical parameters were applied, $\log P$, E_s, and σ, in the form of Hansch's equation to the toxicity data reported in Table 2 with the following results:

$$pLC_{50} = 1.207(\pm 0.511) + 1.600(\pm 0.475) \log P - 0.260(\pm 0.232)\sigma - 0.043(\pm 0.034)E_s \quad (2)$$

The parameters used in or obtained from this model include: $n = 12$, $r = 0.969$, $s = 0.172$, and $F = 85.69$. The E_s and σ terms were not significantly different from zero ($P > |T| = 0.248$ for σ and 0.295 for E_s). We tested linear and quadratic forms with one to four variables to find the most suitable model. The simplest model yielding a significant relationship was a linear one containing the hydrophobic parameter $\log P$ as a single variable. Regression analysis applied to the isothiocyanates listed in Table 2 using only $\log P$ values resulted in the development of the following relationship (see Figure 1):

$$pLC_{50} = 0.582(\pm 0.131) + 1.014(\pm 0.066) \log P \quad (3)$$

The hydrophobic nature of the specific isothiocyanate is most important in determining contact toxicity against eggs of black vine weevil, with $\log P$ values accounting for approximately 95% of the variability in the regression equation ($n = 12$, $r = 0.9591$, $s = 0.179$, and $F =$

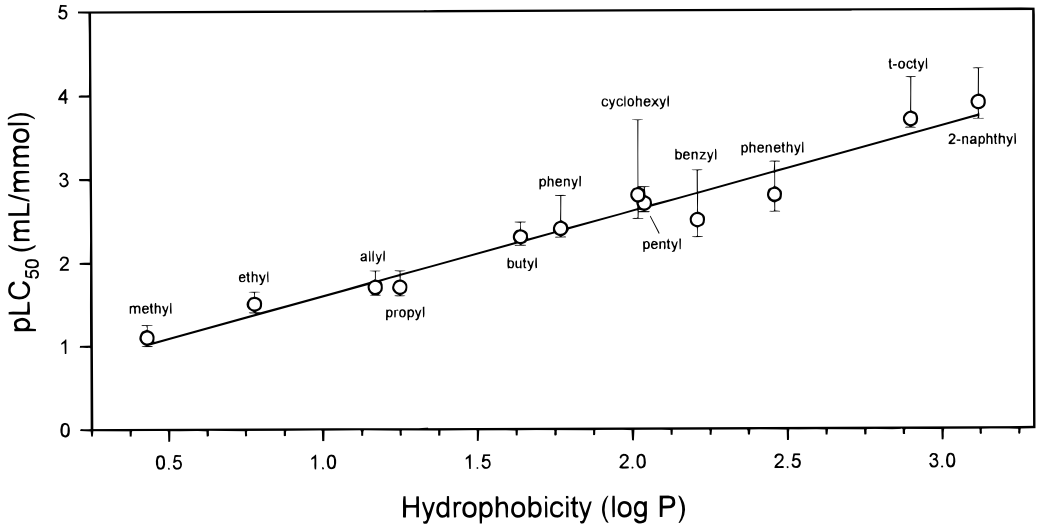

Figure 1. Regression of relative toxicity of isothiocyanates determined using pLC_{50} values (log $1/LC_{50}$, mL/mmol) and isothiocyanate hydrophobicities. The model is described by $pLC_{50} = 0.582(\pm 0.131) + 1.014(\pm 0.066)$ log P; $n = 12$, $r = 0.9591$, $s = 0.179$, and $F = 234.57$. Vertical bars represent standard errors for each respective mean.

234.57). Neither electronic nor steric effects of substituents had a significant effect on observed isothiocyanate toxicities.

Our results are in agreement with the assumed mode of isothiocyanate toxicity in that isothiocyanates are relatively reactive compounds. They interact with nucleophilic compounds, forming dithiocarbamic esters with −SH groups, thiourea derivatives with $-NH_2$ groups, and N-monosubstituted thiocarbamic esters with −OH groups (Duus, 1979). Thus electrophiles such as isothiocyanates are capable of reacting and forming a covalent bond with −SH, $-NH_2$, and −OH groups on enzymes and other critical biological macromolecules. This results in biochemical damage and, at sufficiently high doses, an overall effect sufficient to cause death (Lipnick, 1991). Because the rates of these reactions are not the same, and reactions with thiol groups are significantly faster than reactions with hydroxyl groups, isothiocyanates are hydrolytically stable. Reaction of isothiocyanates with −SH groups of proteins contained in soil-borne organisms such as black vine weevil is thus probable even in a soil environment containing substantial amounts of water-filled pore space.

The lack of significant electronic and steric effects with respect to model results is also consistent with known physicochemical properties of isothiocyanates. Within the group of naturally occurring isothiocyanates and synthetic analogues used here, all substituents are attached to the −NCS group with a σ carbon−nitrogen bond. This configuration tends to isolate the isothiocyanate group from the remaining portion of the molecule, minimizing any associated electronic interactions. Since substituents do not significantly alter electron densities on the carbon atom of the isothiocyanate group, the site of nucleophilic attack, reactivity of all isothiocyanates is quite uniform. Steric effects are insignificant because (1) there are only small differences in molecular dimensions among the tested isothiocyanates, (2) the −NCS group is terminally located and creates little steric interference, and (3) a precise three-dimensional orientation of the reactant is unnecessary since the reaction is nonspecific. A specific binding site need not be identified and only a hydrophobic parameter is required to describe isothiocyanate toxicity. A single-term linear relationship between hydrophobicity and biological activity is found quite frequently in QSAR analyses, with approximately 135 such relationships reported by Hansch and Dunn (1972).

Table 3. Isothiocyanates Potentially Produced from Glucosinolates in Members of the Brassicaceae and Their Calculated Toxicities[a]

R group	FW	log P[b]	LC_{50} (μmol/mL)	pLC_{50} (mL/mmol)
3,4-epithiobutyl	145.24	0.66	55.9	1.25
methylthiopropyl	147.25	0.76	44.3	1.35
4,5-epithiopentyl	159.26	1.11	19.6	1.71
methylthiobutyl	161.28	1.22	15.1	1.82
methylthiobuten-3-yl	159.26	1.35	11.2	1.95
3-butenyl	113.18	1.43	9.26	2.03
methylthiopentyl	175.31	1.61	6.08	2.22
3,4-methoxybenzyl	209.26	1.71	4.81	2.32
methylthiopenten-4-yl	173.29	1.74	4.49	2.35
4-pentenyl	127.20	1.82	3.72	2.43
4-hydroxybenzyl	165.21	1.93	2.88	2.54
3-methoxybenzyl	179.24	1.96	2.68	2.57
4-methoxybenzyl	179.24	1.96	2.68	2.57
4-methylsulfinylbutyl	177.28	2.80	0.38	3.42
4-methylsulfinylbuten-3-yl	175.26	2.93	0.28	3.56
4-methylsulfinylpentyl	191.31	3.19	0.15	3.82
4-methylsulfinylpenten-3-yl	189.29	3.33	0.11	3.96

[a] Abbreviations: FW, formula weight; log P, hydrophobic parameter expressed as the log of the octanol/water partition coefficient; LC_{50}, concentrations lethal to 50% of the tested population; pLC_{50}, log of the reciprocal value of median molar lethal concentration. [b] Values obtained using HyperChem and ChemPlus v. 3.1 software (HyperCube, Waterloo, Ontario, Canada).

We used the observed toxicities for commercially available isothiocyanates and the developed relationship to predict the toxicity of naturally occurring isothiocyanates (Table 3). Prominent isothiocyanates potentially produced from glucosinolates known to occur in members of the Brassicaceae family were selected. Toxicities in the form of LC_{50} values were predicted using the developed model and log P values obtained from HyperChem and ChemPlus software. The calculated lethal concentrations for black vine weevil eggs suggest that highest contact toxicities will result from the most hydrophobic or lipophilic isothiocyanates. In the case of naturally produced isothiocyanates, specific toxic compounds include those with higher numbers of

carbon atoms or those bearing sulfinyl, thio, or aromatic moieties (Table 3).

It must be remembered that specific conclusions and estimated lethal concentrations of isothiocyanates made with the current set of compounds are dependent on the biological test organism and bioassay method. Care must be taken in generalization or extrapolation of our results to estimate the effect of isothiocyanates on other organisms or a different life stage of the same organism. Similarly, bioassay conditions must be considered when making toxicity predictions for a different system. For example, isothiocyanate toxicities in aqueous environments will vary from those reported here since acetone was used as the solvent. The same absolute toxicities are not expected with aqueous solutions, where the effective concentrations of the isothiocyanates with higher molecule mass will be limited by their lower solubility. The use of acetone solutions, however, should not alter relative toxicities and the same order of isothiocyanate toxicity as observed with acetone should occur when using water emulsions.

Since the chemical and physical parameters that have the greatest effect on the toxicity of molecules are solubility, reactivity, and stability, an even more complicated situation exists in heterogeneous soil environments. Structure−activity relationships of isothiocyanates in soil will be governed by the distribution of isothiocyanates among solid, liquid, and gas phases; isothiocyanate volatility; rate of bacterial degradation; and the mode of toxicity (contact/respiratory). The combined effect of all variables makes complete assessment of isothiocyanate toxicity in soil difficult.

Conclusions. Glucosinolate degradation in soil produces similar products as does its decomposition in buffered solution (Borek et al., 1994). From commonly planted cultivars of *B. napus*, *B. hirta*, *B. campestris*, *B. juncea*, or *B. nigra* we can expect the release of aliphatic unsaturated isomers and their thio derivatives with three to six carbons and aromatic isothiocyanates such as benzyl and 2-phenethyl (Brown and Morra, 1997). Our results suggest that soil amendments of tissues from members of the Brassicaceae will have insecticidal activity against black vine weevil eggs and that plant tissues containing higher molecular mass isothiocyanates or those of a greater lipophilicity may have greater insecticidal potential. QSAR can be used to direct Brassicaceae breeding efforts and cultivar selection in order to enhance the control of soil-borne plant pests such as the black vine weevil.

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