

Table 2 Number of insects (mean \pm SD) on tassels of dehiscent corn (*Zea mays*) grown from conventional seed treated with neonicotinoid insecticide or certified organic seed, Ontario 2013

Insect	Number insects counted (20 min)		Wilcoxon statistics
	Conventional	Organic	
<i>Bombus impatiens</i>	0.37 (0.76)	0.06 (0.36)	$Z = -1.10$, $P = 0.27$
Other <i>Bombus</i>	0.31 (0.54)	0.13 (0.37)	$Z = -1.05$, $P = 0.29$
<i>Apis mellifera</i>	0.06 (0.27)	0.44 (0.82)	$Z = 1.23$, $P = 0.22$
Solitary bees	4.25 (3.61)	1.44 (1.63)	$Z = -2.70$, $P = 0.007$
Other insects	9.00 (4.62)	8.50 (4.18)	$Z = 0.13$, $P = 0.89$

Table 3 Residues of thiamethoxam or clothianidin in pollen collected from corn (*Zea mays*) plants grown from conventional seed treated with neonicotinoid insecticide or certified organic seed, Ontario 2013

Field	Clothianidin (ng/g)	Thiamethoxam (ng/g)
Conventional 1	0.8	<0.1 ^a
Conventional 2	0.4	<0.1
Conventional 3	0.1	<0.1
Conventional 4	0.3	<0.1
Organic 1	<0.1	<0.1
Organic 2	<0.1	<0.1
Organic 3	<0.1	<0.1
Organic 4	<0.1	<0.1

^a Limit of detection = 0.1 ng/g

contained a healthy complement of food stores, brood, and adults.

In our observations of pollinators on corn plants, there was no difference among conventional or organic fields in the number of *B. impatiens*, other *Bombus*, or other insects (beetles, flies) on corn tassels. However, 3-fold more solitary bees (Andrenidae, Halictidae) were counted on dehiscent tassels at conventional fields (Table 2). Following the post-exposure period in corn, there was no difference ($Z = 0.29$, $P = 0.77$) among hives in the number of entries and exits of *B. impatiens* foragers over five minutes, whether hives were from conventional fields (9.4 ± 4.0 entries and exits) or organic fields (11.7 ± 10.3 entries and exits).

We did not detect thiamethoxam in any pollen samples and detected clothianidin in four of the eight samples, ranging from 0.1 to 0.8 ng/g (Table 3). All samples with positive detections were from conventional fields that contained neonicotinoid seed treatments. Pollen samples from organic fields contained no detectable thiamethoxam or clothianidin residues.

Table 4 Floral sources (species, genus, or family) used by commercial *Bombus impatiens* colonies when exposed during pollen shed to corn (*Zea mays*) grown from conventional seed treated with neonicotinoid insecticide or certified organic seed, Ontario 2013

Field	Pollen type ^a	Percentage total pollen
Conventional 1	<i>Lotus</i>	42.6
	cf. <i>Solanum dulcamara</i>	35.2
	<i>Coronilla</i>	9.6
	<i>Z. mays</i>	0.8
	Other (10)	11.8
Conventional 2	cf. <i>S. dulcamara</i>	38.4
	<i>Lotus</i>	34.0
	Type <i>Trifolium hybridum</i> ^b	11.2
	<i>Z. mays</i>	1.8
	Other (8)	14.6
Conventional 3	cf. <i>S. dulcamara</i>	96.4
	cf. <i>Hypericum</i>	1.2
	Type <i>Taraxacum</i> ^c	1.0
	<i>Z. mays</i>	0.0
	Other (4)	1.4
Conventional 4	cf. <i>S. dulcamara</i>	89.0
	cf. <i>Medicago sativa</i>	6.0
	Type <i>T. hybridum</i>	1.8
	<i>Z. mays</i>	0.0
	Other (7)	3.2
Organic 1	<i>Lotus</i>	31.4
	cf. <i>S. dulcamara</i>	23.4
	Type <i>Taraxacum</i>	12.4
	<i>Z. mays</i>	0.0
	Other (12)	32.8
Organic 2	cf. <i>S. dulcamara</i>	70.8
	Type <i>Taraxacum</i>	11.8
	<i>Arctium</i>	7.6
	<i>Z. mays</i>	0.0
	Other (4)	9.8
Organic 3	cf. <i>S. dulcamara</i>	67.6
	Type <i>Taraxacum</i>	25.6
	<i>Cirsium</i> or <i>Carduus</i>	2.2
	<i>Z. mays</i>	0.0
	Other (4)	4.6
Organic 4	cf. <i>S. dulcamara</i>	67.2
	Type <i>Taraxacum</i>	23.2
	<i>Z. mays</i>	2.6
	cf. <i>M. sativa</i>	2.2
	Other (6)	4.8

^a For brevity, only the top-three floral sources and portion of corn pollen detected in pollen samples are listed. Values in parentheses indicate the number of other pollen types found

^b May include (share the same palynological features) *T. hybridum*, *T. agrarium*, *T. arvense*, *T. repens*, and *Medicago lupulina*

^c May include (share the same palynological features) *Taraxacum*, *Arnoseris*, *Cichorium*, *Crepis*, *Hieracium*, *Hypochoeris*, *Lactuca*, *Lapsana*, *Leontodon*, *Picris*, *Sonchus*, and *Tragopogon*

Analysis of pollen types recovered from worker bees returning to hives found that a very low portion of corn pollen was collected. Corn pollen was recovered from bees at only two conventional sites and a single organic site, and never constituted more than 2.6 % of the total pollen collected (Table 4). Pollen samples recovered from bees at most sites were dominated by *Solanum dulcamara* (bittersweet nightshade), although 10–40 % of some samples consisted of Type *Taraxacum*, *Lotus* (e.g. bird's-foot trefoil, deervetches), Type *Trifolium hybridum*, or *Coronilla* (Table 4). Depending on the site, pollen from 4 to 12 other floral resources was found in lower amounts.

Discussion

Our field study suggests that exposure to corn grown from neonicotinoid-treated seed during pollen shed poses low risk to *B. impatiens*. This is significant given that neonicotinoid insecticides have been suggested as possible culprits in ongoing bumble bee declines, as they have been suggested as a cause of failing honey bee colonies. Several recent laboratory-based studies have indeed shown that feeding bumble bees food contaminated with neonicotinoids can adversely affect individual bees and colony development (Mommaerts et al. 2010; Cresswell et al. 2012; Gill et al. 2012; Laycock et al. 2012; Whitehorn et al. 2012; Feltham et al. 2014). These controlled experiments are important for the risk assessment process and highlight the potential hazard (but not risk) of neonicotinoids to bumble bees and other pollinators.

However, just as field studies have limitations, laboratory-based experiments have inherent uncertainties that limit their use in risk assessment (OCSPP 2012). Perhaps most important is the uncertainty in the accuracy of feeding in the laboratory to reflect foraging in the field. Even if it is readily accessible in the field, a bee may not choose to forage upon a particular crop depending on characteristics or constraints of floral anatomy, or if the pollen is of poor nutritional value (Winston 1987; Somerville 2001; Heinrich 2004; Willmer 2011). On the other hand, a crop may be an adequate source of forage for a pollinator, but some other floral resource in close proximity may be more favored, or competition with other pollinators could change foraging patterns (Heinrich 2004). Either scenario could reduce the exposure of bees to crop pollen and nectar.

Our results showed that although our bumble bee hives were directly next to corn fields during pollen shed that provided easy access to an abundance of pollen, very little (0–2.6 %; mean of 8 samples = 0.6 %) of the pollen collected off returning forager bees was corn pollen. This indicates that even if corn pollen does contain pesticide residues, as it did in our study, there is a low probability

that foragers or bees in the nest (queens, drones, workers and brood) will be exposed to the pesticide via this route. Foragers we collected returning to hives carried pollen from a large number floral sources that were in the landscape. Although corn is often considered not a nutritious pollen source for bees (Somerville 2001), some varieties of maize can produce pollen with relatively high levels of protein (one of the best indicators of nutritional quality) with a good spectrum of essential amino acids (Hoecherl et al. 2012), and corn/maize pollen is used by honey bees (Somerville 2001; Nguyen et al. 2009; Hoecherl et al. 2012; Krupke et al. 2012). It is possible that where there is a dearth of alternative pollen, bumble bees would increase their use of corn pollen, which would increase exposure to pesticides if they were present in the pollen.

Measured concentrations of neonicotinoids in pollen and nectar from crops grown with treated seeds have recently been summarized by the US EPA (OCSPP 2012) and show that mean concentrations of clothianidin in corn pollen range from 2.9 to 3.9 ng/g, whereas residues of thiamethoxam in corn pollen average 1.7 ng/g (Krupke et al. 2012). The levels of clothianidin we detected in corn pollen from conventional sites (0.1–0.8 ng/g; mean of 4 samples = 0.4 ng/g) were similar to, albeit several-fold lower than, levels previously reported from corn grown from treated seed. It is not unusual for there to be high variability in levels of neonicotinoid detected in pollen of a particular crop. For example, Bonmatin et al. (2005) collected samples throughout France from 2000 to 2003 and reported an average concentration of imidacloprid in corn pollen of 2.1 µg/kg (ng/g), with a range of less than 0.1 µg/kg up to 33.6 µg/kg. We did not detect thiamethoxam in pollen samples from conventional sites CV3 and CV4, which were planted with seeds treated with both clothianidin and thiamethoxam. It is probable that the tassels we collected in the field were from those plants treated with clothianidin rather than thiamethoxam. It is also possible that thiamethoxam on treated seed was metabolized to clothianidin, as may occur with foliar sprays or irrigation treatments of thiamethoxam (Dively and Kamel 2012) or on corn seeds treated with thiamethoxam (Pilling et al. 2013). This second possibility seems less likely because in a previous study where thiamethoxam was used as the corn seed treatment the metabolite (clothianidin) was detected in lower levels than the parent compound (Pilling et al. 2013). As expected, we did not detect clothianidin or thiamethoxam in pollen from tassels of organic corn that contained no pesticide treatments.

During the study, all hives qualitatively appeared active and healthy, and the quantitative data support this assessment. The only statistically significant effects we found in our study were that: (1) more solitary bees were observed on tassels in conventional than organic fields; and (2) fewer