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The impact of thiamethoxam on bumble bee broods (*Bombus terrestris* L.) following drip application in covered tomato crops

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Abstract

The side effects of the neonicotinyl compound thiamethoxam on the brood of bumble bees (*Bombus terrestris* L.) were investigated on tomatoes in plastic tunnels and glasshouses.

Preceeding trials had revealed a strong contact and stomach activity of the compound by foliar application. A decisive improvement was obtained by replacing the foliar by drip irrigation of thiamethoxam at rates between 150 and 161 g ai/ha. The mortality figures of all bumble bee stages dropped to the level in the untreated control. No dead bumble bees were found in the thiamethoxam treated tunnels/glasshouses on the ground.

Thiamethoxam could be rated as being harmless to bumble bees with a single application through the irrigation system. No spilling of this compound during the application should occur to avoid intoxication of adult bumble bees by oral uptake or contact contamination during the cleaning process. If multiple applications via the irrigation system or under any hydroponic growing conditions is considered, further sequential testing of this mode of use is recommended.

1 Introduction

The development of rearing methods of the bumble bee species *Bombus terrestris* L. enabled growers to apply them to pollinating glasshouse crops on a large commercial scale 12 months per year (Duchateau and Velthuis, 1988; Van Heemert et al., 1990). Their use is a common practice nowadays in some 25 different covered crops world-wide (Griffiths and Roberts, 1996). Tomatoes are by far the most important crop where bumble bees are used for pollination (Van Ravenstijn and Van Der Sande, 1991). Since bumble bees are potentially exposed to pesticides, specific data on the effect on bumble bees are required. So far, data from the other important pollinator species, the honey bee (*Apis mellifera* L.), cannot be extrapolated to cover also the risk of pesticides to bumble bees (Lewis et al., 1998). There are two main routes of exposure under practical conditions: direct overspraying of adult bumble bees foraging on the glasshouse crop, and contamination via the uptake of treated pollen and nectar from the treated plants. An additional intoxication might occur by feeding bumble bee larvae with treated pollen or nectar.

Thiamethoxam belongs to the subclass of thianicotinyl compounds within the chemical group of neonicotinoids. It is highly active against aphids, whiteflies and some beetles (Senn et al., 1998; Maienfisch et al., 1998). It displays root-, leaf- and stem-systemic activity. In target insects it shows quick stomach and contact action.

A sequential testing procedure using laboratory, semi-field and field selectivity testing methods has been developed with the aim of evaluating all potential risks of pesticides to beneficial arthropods (Hassan, 1992). Thiamethoxam had been tested in the laboratory and under semi-field conditions (cages, tents) and had been proved to be toxic to bumble bee adults (Sechser et al., 2002). As the last step in the testing scheme, a modified approach of sequential testing against bumble bees (Sechser and Reber, 1998) was used in this study where thiamethoxam was applied via the irrigation system under realistic conditions in tunnels. The measurement of the impact of this type of treatment on bumble bee broods was the purpose of this study.

2 Materials and methods

Two separate experiments were conducted in 1998 in glasshouses and tunnels in collaboration with the Federal Agricultural Research Station, Changins, the Centre Horticole in Lullier, and in Troinex by the Office Technique de Culture Maraîchère de Gènève, Jussy, Switzerland.

2.1 Material

Thiamethoxam (ACTARA® WG 25) was used at rates between 150 and 161 g ai/ha via the irrigation system. Non-insecticidal tunnels served as negative control.

2.2 Evaluation

The hives were opened between 13 and 36 days after the application. The content of each hive was removed and all developmental stages examined for effects caused by the treatments.

2.3 Trial sites

2.3.1 Single drip application on tomatoes grown in a glasshouse/tunnel, Lullier, Switzerland, 1998

Thiamethoxam was applied via the irrigation system on tomato plants (cultivar DRW 3759), BBCH growth stage 601–610 (flowering) (Bleiholder et al., 1997) at the time of application, grown in stone wool, in a glasshouse (450 m^2). A non-insecticidal tunnel of the same size in the neighbourhood served as negative control.

29 g WP 25 (7.25 g) ai was dissolved in a 5 l stock solution and added continuously to a mixing tank of 20 l, of which the content was renewed permanently during the irrigation process of half an hour (the 7.25 g ai applied on 450 m^2 correspond to 161.1 g ai/ha). The application was done 27 days after the placement of a single colony in each treatment and bumble bees were monitored thereafter for 35 days (36 days in the untreated control).

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The glasshouse and tunnel were searched regularly for dead and paralyzed bumble bees. The hives were opened 35 days after application (36 days in the untreated control) for counting the alive and dead stages. The remaining food reserves were also evaluated.

2.3.2 Single drip application on tomatoes grown in tunnels, Troinex, Switzerland, 1998

This trial was done in the same season (1998) as the preceeding one and the trial outline is largely the same. Deviations in the outline are specially mentioned.

Thiamethoxam was applied via the irrigation system on tomato plants (cultivar Trust), BBCH growth stage 701–710 (development of fruit) at the time of application, in a tunnel (2300 m^2). Another tunnel (1800 m^2) with no insecticidal treatment (undefined cultivar of cherry tomatoes, BBCH growth stage 701–710) in the neighbourhood served as negative control. In both trial sites the tomatoes were grown in stone wool and each one had two bumble bee hives.

138 g 25 % WP (34.5 g ai) of thiamethoxam was dissolved in a stock solution and applied via the irrigation system in the whole tunnel within five minutes. The 34.5 g ai applied on 2300 m^2 correspond to 150 g ai/ha. The different rate is due to the difficult dosaging with this equipment. The application was done 16 days after the installation of the colony and bumble bees were monitored thereafter for 16 days (17 days in the untreated control).

The tunnels were searched regularly for dead and paralyzed bumble bees. The hives were opened 13 and 16 days after application in the treated tunnel, and after 16 and 17 days in the untreated one.

3 Results

3.1 Single drip application on tomatoes grown in a glasshouse/tunnel, Lullier, Switzerland, 1998

During the various and repeated evaluations in the glasshouse/tunnel neither paralyzed nor dead bumble bees could be found on the ground close to the plants or around the hives.

Table 1 gives the details of the hive analysis. In the thiamethoxam treated tunnel, the analyzed hive had a higher total number of cells of bumble bees (994) than the untreated one (834) in spite of a high number of calliphorid predators. Pollen/nectar cells had been occupied by larvae before, from which adults had hatched. Therefore they were added to the total number of cells occupied by bumble bees, as well as the empty cells which had not yet been filled with pollen or the regurgitated nectar/pollen mixture of the queen or workers. The untreated hive had more alive adults (109 versus 63) and less dead larvae, nymphs and adults (72 versus 119). The total number of alive larvae and nymphs is comparable in the two treatments (29 in the treated versus 34 in the untreated).

All the bumble bee food was consumed in the untreated hive, while some had remained in the treated one. On the other hand, the bumble bee population in the treated glasshouse had collected more nectar, which might explain that less bumble bee food had been consumed as compared to the population in the untreated tunnel. No or nearly no pollen was left in both treatments.

On the whole, it seems that thiamethoxam had no negative effect on bumble bee populations.

3.2 Single drip application on tomatoes grown in tunnels, Troinex, Switzerland, 1998

During the various and repeated evaluations in the tunnels, neither paralyzed nor dead bumble bees could be found on the ground close to the plants nor around the hives.

Table 2 gives the details of the final hive analysis. In both treatments there was one hive with about 1100 bumble bee cells and one with lower populations (390 and 647, respectively). The corresponding colonies in

Table 1. Final analysis of bumble bee hives 35 (treated) and 36 (untreated) days after a single application of thiamethoxam via the irrigation system. Centre Horticulture, Lullier, Switzerland, 1998.

Category/stages of development	Treated glasshouse		Untreated tunnel	
	Total n	%	Total n	%
Larvae L1	2	0.2	0	0
Larvae L2	11	1.1	8	1
Larvae L3	8	0.8	5	0.6
Larvae L4	3	0.3	5	0.6
White nymphs	2	0.2	9	1.1
Black nymphs	3	0.3	7	0.8
Unhatched adults	1	0.1	2	0.2
Empty cells	336	33.8	557	66.8
Pollen/Nectar cells	590	59.4	211	25.3
Cells preyed by calliphorid flies	38	3.8	30	3.6
Total cells with bumble bees	**994**	100 %	**834**	100 %
Adults	**63**		**109**	
Net weight of remaining bumble bee food 'BeeHappy' in ml (1500 ml at start)	**169.8**		0	
Net weight of remaining pollen in g	0		**10.2**	
Dead larvae, nymphs, adults	**119**		**72**	

Table 2. Final analysis of bumble bee hives 13–17 days after a single application of thiamethoxam. Troinex, Switzerland, 1998.

Category/stages of development	Treated tunnels				Untreated tunnels			
	Colony 1 n	%	Colony 2 n	%	Colony 3 n	%	Colony 4 n	%
Eggs	136	12	8	2	81	7	33	5
Larvae L1	108	10	9	2	82	7	5	1
Larvae L2	10	1	4	1	34	3	22	3
Larvae L3	65	6	7	2	33	3	30	5
Larvae L4	70	6	13	3	44	4	36	6
White nymphs	101	9	28	7	102	9	60	9
Black nymphs	70	6	21	5	84	7	29	4
Unhatched adults	19	2	11	3	27	2	12	2
Empty cells	155	14	30	8	132	12	23	4
Pollen/Nectar cells	361	33	217	56	419	37	187	29
Cells preyed by calliphorid flies	5	1	42	11	92	8	210	32
Total cells with bumble bees	**1100**	100 %	**390**	100 %	**1130**	100 %	**647**	100 %
Adults	**181**		**119**		**252**		**119**	
Queens	**1**		**1**		**1**		**1**	
Net weight of remaining bumble bee food 'BeeHappy' in ml (1500 ml at start)	**1114**		**1015**		**713**		**900**	
Net weight of remaining pollen in g	**49.3**		**3.9**		**45**		**7.6**	
Dead larvae, nymphs, adults	**26**		**317**		**42**		**209**	

the hives had also higher numbers of living adults and lower numbers of dead larvae, nymphs and adults, but total figures on all stages are comparable in both treatments. The figures for the adult offspring were very close in the treated and untreated hives. Because of the shorter interval between application and evaluation, more bumble bee food and pollen were remaining in all hives without a stringent difference between treated and untreated.

Figures do not indicate a negative effect of thiamethoxam on bumble bee populations.

4 Discussion

The healthy state of a hive population is a prerequisite for the development of strong adult bumble bee populations for the pollination of tomato crops, which should not be affected by pesticide use in covered crops.

The appropriate recommendations for use of thiamethoxam with regard to bumble bees is based on the findings as described here. Since thiamethoxam has been proven harmful in contact and feeding tests by foliar application, strict recommendations have to be communicated to glasshouse growers in order to exclude risks caused by careless spillage of thiamethoxam in the course of a drip application.

Although bumble bees must achieve water balance for themselves and their brood entirely from the water contained in nectar (Prys-Jones and Corbet, 1987), there remains the risk of contact of the tarsi with spilled water. This could lead to lethal effects when bumble bees leak contaminated tarsi during the cleaning process. Adequate food stores, a particular worker/larva ratio, chemical cues, nest temperature stability and bee density in the nest, are probably all influential factors. *B. terrestris* belongs to the group of 'complex' species (Brian, 1980), where the queen can, probably with the aid of pheromones, stop the workers feeding the larvae with sufficient quantity and/or frequency for them to develop into queens, even at high worker/larva ratios when ample food is available. Thus, complex species can delay queen production, allowing the build-up of large worker populations. Pollen and nectar collection outside the hive is dictated by the needs of the nest, i.e. the size and good health of the brood. A small and weakened offspring in the hive diminishes the foraging activity of the adults (Koppert, personal communication). Therefore it is of utmost importance that pesticides do not have a negative effect on the developmental stages within a hive.

B. terrestris is known for its nectar robbing behaviour (Matheson, 1995). Nectar is not just an energy source. Bumble bees must achieve water balance for themselves and their brood entirely from the water contained in nectar. Pollen and nectar are stored in empty pupal cocoons and specially constructed wax cells, from where the larvae are fed. The presence of these filled cases in the hives in the thiamethoxam treated glasshouse/tunnels is a sign of firstly the successful hatching of adults and secondly the good sampling performance of the workers, which is a prerequisite for numerous offspring.

Preceeding trials have proven the safety of thiamethoxam with regard to flight and pollination activity and mortality of all bumble bee stages when applied as a drench or via drip irrigation at 100 to 170 g ai/ha (Sechser et al., 2002). The analysis of hives in the two tests described in this paper prove again that thiamethoxam is harmless to bumble bees in a single application via drip irrigation when applied once via the irrigation system at a rate of up to 161 g ai/ha. If multiple application during a growing season is practiced under any form of hydroponic growing conditions, further equivalent testing may be required for proper management of supposed risks.

5 Conclusions

Thiamethoxam at rates up to 161 g ai/ha is safe for bumble bees pollinating tomato plants when it is used in

a single application via the irrigation water in a hydroponic system. Attention has to be paid not to spill any contaminated water because of the potential oral and contact toxicity of the compound through the cleaning process of bumble bee adults.

Zusammenfassung

Die Nebenwirkungen der Neonicotinyl Verbindung Thiamethoxam auf Hummeln (*Bombus terrestris* L.) wurden in Gewächshäusern und Plastiktunnels untersucht. Prüfungen mit direkter Applikation der Substanz auf das Blatt hatten eine zu hohe Toxizität für Hummeln ergeben.Eine entscheidende Verbesserung wurde erzielt, indem man die Behandlung der Substanz auf die Blätter durch die Tröpfchenberegnung ersetzte. Die Aufwandmengen von Thiamethoxam in den zwei Prüfungen betrugen 150 bzw. 161 g Aktivsubstanz pro ha. Bei dieser Anwendungsmethode fiel die Sterblichkeit der Hummeln auf das gleiche Niveau wie in der unbehandelten Kontrolle.Thiamethoxam kann als harmlos eingestuft werden bei einer Einzelanwendung in einer Hydrokultur wie z. B. via Tröpfchenberegnung. Ein Überlaufen der Substanz während der Anwendung sollte vermieden werden, um eine Vergiftung der adulten Hummeln durch orale Aufnahme oder Kontaktwirkung beim Putzprozess zu vermeiden. Wenn eine mehrmalige Anwendung über das Bewässerungssystem vorgesehen wird, empfiehlt sich eine weitere Prüfung unter diesen Bedingungen.

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