frontiers | Frontiers in Bee Science

TYPE Original Research
PUBLISHED 06 January 2026
DOI 10.3389/frbee.2025.1644205

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OPEN ACCESS

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RECEIVED 10 June 2025
REVISED 23 October 2025
ACCEPTED 24 November 2025
PUBLISHED 06 January 2026

CITATION
Ramos JD, Santos GS, dos Santos CF, De Oliveira Kaminski T, Cione AP, Alves DA, Quenzer FCL, Campbell AJ, Pereira AM, Thompson H, Martins de Queiroz AC, Bento JMS and Menezes C (2026) Stingless bees in coffee: yield gains and assessing neonicotinoid impact. *Front. Bee Sci.* 3:1644205. doi: 10.3389/frbee.2025.1644205

Stingless bees in coffee: yield gains and assessing neonicotinoid impact

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Introduction: Coffee production depends heavily on pollination services, but the combined effects of managed pollinators and pesticide use on crop yield and pollinator health are still poorly understood. This study evaluated the contribution of supplemental pollination by the stingless bee *Scaptotrigona depilis* to coffee yield and assessed the impact of thiamethoxam, a neonicotinoid insecticide, on colony strength in Brazilian coffee farms.

Methods: Colonies of *S. depilis* were introduced into both conventional and organic coffee farms. Coffee yield was measured in branches located near and far from bee colonies. Colony strength parameters were monitored over time, and pesticide residues were quantified in plant tissues (leaves, nectar, pollen) and in bee-collected floral resources.

Results: Supplemental pollination by *S. depilis* significantly increased coffee yield by 67% in branches closer to the colonies. Low but detectable residues of thiamethoxam and its metabolite clothianidin were found in plant tissues and bee-collected resources. No significant negative effects were observed on brood production or brood mortality in colonies located in conventional farms compared to those in organic farms. Foraging activity differed between farm types before exposure to coffee bloom but normalized over time.

Discussion: Managed stingless bees can markedly enhance coffee production without experiencing measurable detrimental effects under current label-compliant neonicotinoid use. These findings offer practical insights for developing more sustainable coffee production strategies that align productivity with pollinator health and conservation.

KEYWORDS

crop pollination, integrated pest and pollinator management, native bees, sustainable agriculture, *Coffea arabica*, *Scaptorigona depilis*

1 Introduction

Coffee is a globally significant commodity, resulting in a record production forecast of 178.7 million bags for 2025/26—a 4.3 million bag increase over the previous year (USDA, 2025). Brazil is the world's leading producer and exporter of coffee, with a cultivated area of 2.25 million hectares in 2025, of which 1.84 million hectares, or 82%, are occupied by arabica coffee (Conab, 2025). *Coffea arabica* is a self-pollinating species, meaning it does not require pollen transfer between different plants to produce fruit. For this reason, farmers have traditionally believed that this variety does not rely on bee pollination. Indeed, its dependence on pollinators is relatively low compared to other agricultural crops. In other words, good productivity can be achieved even without bees (Klein et al., 2003a).

However, extensive research conducted worldwide has demonstrated that, although *C. arabica* does not rely on pollinators as much as other crops, it can still gain significant advantages from their presence (Ngo et al., 2011; Moreaux et al., 2022). When *C. arabica* flowers are isolated from visiting insects, the number of fruits formed decreases by an average of 28% compared to flowers that are open to naturally occurring pollinators in the field (Saturni et al., 2016). Furthermore, the better the conservation status of the landscape surrounding the crop, the greater the diversity of pollinators and the higher the productivity (Klein et al., 2003b; Moreaux et al., 2022).

Given the robustness of these scientific findings and the recent efforts to bring this knowledge to the farmers, we expect the relationship between coffee cultivation and the use of managed pollinators to strengthen over time. It is likely that bee rental will become a routine practice in the arabica coffee production system in Brazil, just as it already is for apples and melons (Freitas and Nunes-Silva, 2012), thus requiring more research to ensure the harmonious coexistence of pest management with the presence of introduced bees in the fields.

In intensively managed coffee agroecosystems, farmers often rely on chemical insecticides to reduce pest damage and protect crop quality. Depending on the mode of action and scale of exposure of bees to these insecticides, or their environmental metabolites, exposure under laboratory or similar controlled conditions has resulted in lethal or sublethal effects on a range of behavioural and physiological traits of social bees (Tosi et al., 2022). Reported effects include foraging performance, learning and memory, colony development, and disease resistance, which may ultimately compromise their health (Potts et al., 2016; Siviter et al., 2021; Tosi et al., 2022). Some chemical pesticides are commonly identified in food stores of colonies (Mitchell et al., 2017), thereby potentially extending the exposure of social bees.

Neonicotinoid insecticides, such as thiamethoxam, are efficient in controlling economically important crop pest populations. For example, thiamethoxam is commonly used to control some key coffee pests, such as the coffee leaf miner *Leucoptera coffeella*, one of the major threats to coffee production, and the fungus *Hemileia vastatrix*, a pathogen that causes the devastating disease coffee leaf rust. However, there have been concerns raised about their potential harmful effects on pollinators due to their widespread use and high systemicity, meaning that they are absorbed by the treated plant and translocated to all plant organs, reaching nectar and pollen in flowers (Simon-Delso et al., 2015). Most ecotoxicological studies for pesticide regulation are performed with honeybees and for first tier risk assessment are focused on individual mortality, although study guidelines also require sublethal effects to be recorded. Subsequent higher tier studies required to refine understanding of potential risks are performed in semi-field and field studies which incorporate any behavioural effects of sublethal exposure on colony growth and functioning of social bees (Thompson and Maus, 2007). Although several studies have been published on the effects of insecticides, including thiamethoxam, on stingless bees in the laboratory, effects on stingless bee colonies under real-use field conditions, have received far less attention (Bogo et al., 2025).

The stingless bee *Scaptotrigona depilis* is a promising alternative pollinator for Brazilian coffee, offering high resilience to management, ease of large-scale multiplication, naturally large colonies, attraction to coffee flowers, and a distribution overlapping major coffee-producing regions (Menezes et al., 2013).

This species uses tree cavities to establish their nests, which are built with a mixture of wax and plant resins (i.e., cerumen) that inhibit the growth of multiple bacteria and fungi due to their antimicrobial properties (Paula et al., 2021). Nests are essentially composed of an entrance tube, egg-shaped pots to store pollen and honey and multilayered horizontal brood comb with same-sized cells to rear workers and males, while queens are reared in larger royal cells (Bueno et al., 2023). All brood cells are constructed, mass-provisioned with liquid larval food immediately before the queen lays her egg on top of it, and then sealed by workers (Bueno et al., 2023). In addition to larval food, *S. depilis* larvae consume *Zygosaccharomyces* fungus that grows inside brood cells and provides steroid precursors necessary for brood survival and metamorphosis (Paula et al., 2021). The colonies typically contain around 10,000 adult workers headed by a mother queen who lays around 300 eggs daily (Menezes et al., 2013).

This study addressed two key questions: (1) Does supplemental pollination with *S. depilis* significantly enhance coffee yields? and (2) Does commercial application of thiamethoxam affect *S. depilis* colony strength in conventional coffee farming systems? We investigated these questions by comparing coffee yields and bee colony strength metrics (brood production, mortality, and foraging activity) on conventional and organic farms in southeastern Brazil, with and without nearby *S. depilis* colonies. This study helps fill a key knowledge gap by evaluating both the benefits of *S. depilis* pollination and the risks posed by thiamethoxam under real farming conditions. The findings aim to support more sustainable coffee production by balancing pest control with pollinator health in Brazil's arabica systems.

2 Material and methods

2.1 Study sites

Our study was conducted between 2022 and 2023 on various coffee farms located in the states of Minas Gerais and São Paulo in Southeastern Brazil. This region is the country's most traditional hub for arabica coffee production, encompassing a mix of intensively managed conventional farms that utilize chemical pesticides and certified organic farms. All the selected farms cultivated arabica coffee under full-sun conditions.

2.2 Installation of stingless bee colonies

Healthy bee colonies were carefully selected one month prior to the coffee flowering season, based on the presence of an active egg-laying queen, the number of combs with brood, food stores, and worker population size. At least 15 days before the blooming season (2022), we supplemented six conventional farms with managed colonies of the stingless bee *S. depilis* (CS1–CS6; Supplementary Table 1) at ten colonies per hectare. Colonies of *S. depilis* are morphologically and behaviourally similar to *S.* aff. *postica*, whose average colony size was estimated at approximately 7,400 ± 1,391 adult individuals (range: 5,898–10,036) (Leão et al., 2024). Based on these findings, we assume that colonies used in this study contained a comparable number of workers. Workers of *Scaptotrigona* species typically forage within a radius of 500 to 1,500 m from their nests, depending on landscape structure and floral resource availability. For each farm, we assessed coffee yield at two distinct treatment areas: (1) at a distance of < 50 m from the colonies ('close to bee colonies') and (2) a more distant area from the bee colonies (mean distance: 250 m; range: 200–300 m, 'far from bee colonies') (Almeida-Dias et al., 2025).

2.3 Coffee yield

On six farms supplemented with stingless bees, coffee berries were manually harvested between June and August 2023. In each site, we established a 10-m transect from stingless bee colonies, and we chose 10 branches at chest level from 10 different coffee bushes, totalling 360 branches (10 bushes × 3 sites × 2 treatment areas × 6 farms). Ripe berries were then collected, counted, and weighed to calculate the coffee yield (kg branch^{-1}) for each site.

2.4 Effects of thiamethoxam-based products on bee colony strength

We selected six conventionally (CS1–CS6; Supplementary Table 1) and two organically (OS1–OS2) managed coffee-producing farms, the latter as a chemical pesticide-free control. Based on the pest control efficiency (i.e., agronomic efficiency) in conventional arabica coffee farms, the commercially available thiamethoxam-based products Verdadero 600 WG and/or Actara 250 WG were applied in 2021 season, by soil drenching, during the berry expansion stage according to label recommendations (Table 1).

The stingless bee colonies were introduced in the subsequent season, approximately 10–18 days before coffee blooming in September-October 2022, to ensure that the potential effects of thiamethoxam residues from the previous season's application (November 2021) on colony strength could be accurately assessed (5 and 10 colonies on each conventional and organic farm, respectively). Among the temporal dynamics components of a bee colony, we assessed three colony performance traits according to international standardised protocols to assess bee colony strength (OEPP/EPPO, Guideline No. 170 (4) (EPPO Bulletin, 2010) and OECD No. 75 (2014)): (1) brood production, by counting brood cells in the pre-provisioning and oviposition stages (Bueno et al., 2023); (2) brood mortality, by calculating the percentage of empty pupal brood cells in a brood comb, representing brood removed by workers; and (3) foraging activity, by counting foragers leaving the colony during a 3-minute period between 9:00 and 12:00 h. These parameters were measured at five time points: 5–7 days before coffee blooming (pre-b), 5–7 days after blooming (post-b), and 45, 75, and 105 days post-exposure. Pre-b and post-b assessments were conducted on the coffee farms; subsequent assessments were performed at a stingless bee apiary located in a 30-ha forest patch at Embrapa-

TABLE 1 Field rates and application dates of thiamethoxam-based products applied by soil drenching on conventionally managed arabica coffee farms.

Farm	Farming system	Field rates (kg ha^{-1}) [a,b]		Application dates	
		Verdadero 600® WG [c]	Actara 250® WG [c]	Verdadero 600® WG	Actara 250® WG
CS1	Conventional	1.00	1.00	Nov 2021	Feb 2022
CS2	Conventional	1.00	not applied	Nov 2021	–
CS3	Conventional	1.00	not applied	Nov 2021	–
CS4	Conventional	1.00	not applied	Nov 2021	–
CS5	Conventional	not applied	1.00	–	Dec 2021
CS6	Conventional	1.00	1.00	Nov 2021	Jan 2022
OS1	Organic	not applied	not applied	–	–
OS1	Organic	not applied	not applied	–	–

[a]Active ingredient: 30%w/w and 25%w/w thiamethoxam in formulated products Verdadero and Actara, respectively.
[b]Field rates based on the efficiency in coffee pest control.
[c]Label recommendations: 0.7–1.0 kg ha^{-1} and 1.4–2.0 kg ha^{-1} for Verdadero and Actara, respectively.

Environment (Jaguariúna, São Paulo State), over 90 km south of the southernmost coffee farm. Colonies were housed at this apiary before and during the assessments of the field post-exposure phase.

2.5 Pesticide residue analyses

Thiamethoxam and clothianidin residues were analysed in coffee leaves (at least 10 leaves from different bushes were sampled per farm) and in flower resources collected by *S. depilis* foragers at the six conventional farms during blooming period (CS1–CS6; Supplementary Table 1). To assess them, in late afternoon, we sampled recently collected pollen (mean ± SD = 0.502 ± 0.219 g/sample, n = 17 pollen samples) nectar (1.578 ± 0.689 g/sample, n = 17 nectar samples) directly from open food storage pots within 3–5 stingless bee nests for each farm, as described in Menezes et al. (2012). All samples were stored at –20°C for pesticide residue analyses performed at Eurofins Agroscience Services (Indaiatuba, São Paulo State).

To screen for thiamethoxam and its metabolite clothianidin, each pollen (50 ± 10 mg sample^{-1} farm^{-1}) and nectar sample (150 mg sample^{-1} farm^{-1}) were placed in a centrifuge tube. Water, acetonitrile and a mixture of salts were added and then vortexed. For pollen, after centrifugation, the supernatant was transferred to a vial and frozen, and a portion of extract was transferred to a vial with combination of salts and C18 for the clean-up step, followed by evaporation and resuspension in acetonitrile. For nectar samples, after centrifugation, a portion was firstly subjected to clean-up followed by partial evaporation and resuspension in acetonitrile. Finally, for determination of neonicotinoid residues in coffee bushes, 2.5 g of each leaf sample per farm were added to a centrifuge tube with water and acetonitrile and then vortex. Following centrifugation, a portion of extract was transferred to a vial containing mixture of salts for clean-up and activated charcoal, vortexed, centrifuged and filtered. For all three matrices, the final solution was analysed by liquid chromatography tandem mass spectrometry system [LC (Agilent)-MS/MS (SCIEX 5500)], using 0.05% acetic acid + ammonium formate in water as mobile phase A, and 0.05% acetic acid in methanol as mobile phase B, with a C18 column of 150 mm × 2mm × 5µm.

The limit of quantification (LOQ) for thiamethoxam in pollen, nectar, and leaf samples was 0.001 mg/kg, and 100×LOQ corresponded to 0.1 mg/kg. No internal standard (e.g., isotopically labelled neonicotinoid) was used in the analysis.

2.6 Statistical analyses

All statistical analyses and figure generation were performed using R version 4.3.2.

2.6.1 Contributions of stingless bees to coffee yields

Coffee yield (kg branch^{-1} bush^{-1}) was analysed using linear mixed-effects models (LMMs), accounting for the spatial non-independence of sampling sites (average bee foraging range: 0.87 km; Campbell et al., 2019) via the inclusion of random effects ('sampling units' as intercepts and 'plots within farms' as slopes, nested within 'farm ID'). The fixed effect was pollination treatment (near vs. far from bee colonies). The response variable was standardised (mean = 0, SD = 1) and log-transformed to meet assumptions of normality (P = 0.48) and homogeneity of variance (P = 0.73; checked using the performance package; Lüdecke et al., 2021). Model fitting used the *lme4* package (Bates et al., 2015), employing restricted maximum likelihood (REML) and the Bound Optimization BY Quadratic Approximation (BOBYQA) algorithm for parameter estimation. Overdispersion was assessed using the *DHARMa* package (Hartig, 2022).

2.6.2 Generalised linear mixed models: effects of thiamethoxam-based products on bee colony health

We applied generalised linear mixed models (GLMMs) to assess the effects of thiamethoxam-based products on brood production, brood mortality (%), and foraging activity of *S. depilis* colonies from conventional and organic farms. Predictor variables included time period (pre-blooming, post-blooming, and 45, 75, and 105 days post-exposure), farming system (conventional vs. organic), and their interaction. Farm and colony ID were included as crossed random effects.

For each response variable, three candidate models were fitted using different error distributions (Poisson, quasi-Poisson, and negative binomial) but identical fixed and random effects. Model selection was based on Akaike's Information Criterion (AIC) and Akaike weights, using the AICtab function from the *bbmle* R package (Bolker and R Core Team, 2020). The model with the lowest AIC and highest weight was retained for inference.

To account for potential temporal autocorrelation, each selected model was compared with an analogous version incorporating an AR(1) correlation structure. The best-fitting model was again identified using AIC-based criteria. Parameter significance was assessed with the Anova function (*car* package; Fox and Weisberg, 2019). Estimated marginal means (EMMs ± SE) and pairwise contrasts were obtained using the emmeans package (Lenth et al., 2018), with *P*-values adjusted for multiple comparisons via the false discovery rate (FDR) method.

Complete parameter estimates, confidence intervals, and model selection results are presented in Supplementary Tables S2–S4, along with the corresponding R script.

3 Results and discussion

Our field study showed that where managed colonies of *S. depilis* had been introduced, pollination provided by stingless bees significantly increased arabica coffee yield at shrub levels (Figure 1). The deployment of stingless bees resulted in a significant increase of 67% in fruit yield (close vs. far from bee colonies: 0.50 ± 0.002 kg branch^{-1} *vs.* 0.30 ± 0.008 kg branch^{-1}).

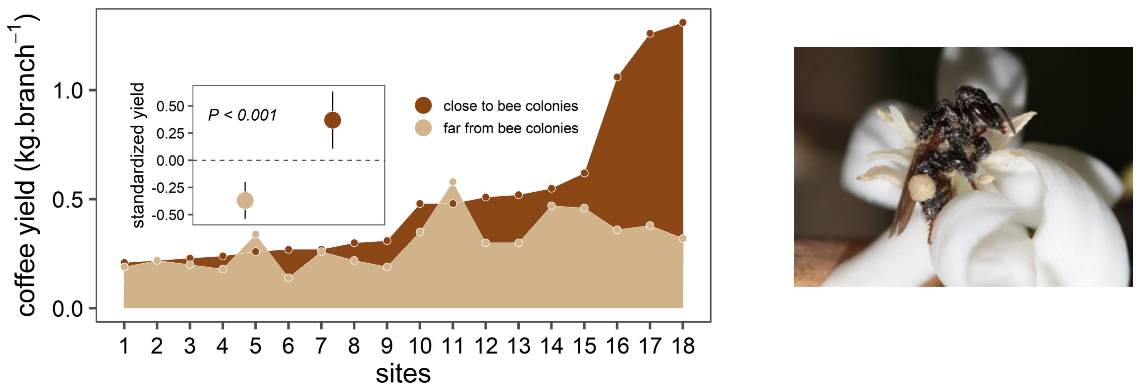

FIGURE 1
Coffee yield (kg branch^{-1}) was higher at sites located close to stingless bee colonies (dark brown) compared to sites located far from colonies (light brown). The inset graph presents the effect of the pollination treatment (close vs. far) on log-transformed coffee yield. Log-transformed yield data were standardised to a mean of 0 ± standard error (SE) to allow for a direct comparison of effect sizes.

Our results support the hypothesis that the native stingless bee *S. depilis*, with typical shorter homing range (~0.9 km) (Campbell et al., 2019) and narrow diet breath, is a highly effective managed pollinator for coffee production in the Neotropical Region (Almeida-Dias et al., 2025). The high pollination efficiency of *S. depilis* is likely explained by its morphological and behavioural traits, including small body size (~5.5 mm), short trip duration (~4.2 min), constant foraging activity from morning to early afternoon, mass-recruitment foraging strategy using scent trails to quickly mobilise nestmates to food sources, and large colony size (~10,000 workers) (Bueno et al., 2023). In addition, although *S. depilis* foragers are similar to honeybees in moistening the collected coffee pollen with nectar and compacting it into their metatibial pollen baskets, apparently, they carry more free pollen grains incidentally attached to their bodies that are more likely to be available for pollination. Even though stingless bees are well suited as managed tropical pollinators, as they pose less of a hazard to farm workers due to the lack of a functional sting, and have relatively well-developed colony management protocols, commercial colony production is still incipient (Jaffé et al., 2015) compared with large-scale honeybee keeping operations, which provides hundreds of honeybee colonies to support crop pollination. Even so, the shortage in pollinator availability is a common problem in many agroecosystems, rising pollination deficits and reducing crop yield (Sáez et al., 2022). At the same time, there is a growing concern over reliance on *Apis mellifera* as a single key species for agricultural pollination, mainly in regions outside its native range, which has increased interest in native bees as alternative managed pollinators to optimise crop pollination (Isaacs et al., 2017).

To optimise pollination services, one of the combined tactics for a successful integrated pollinator and pest management approach is enhancing the farm environment through pesticide stewardship to mitigate pesticide risks for non-target beneficial insects (Isaacs et al., 2017). Honeybees are used as model species for environmental monitoring and risk assessment, but non-*Apis* bee species, such as stingless bees, have been relatively neglected (Tosi et al., 2022; Raine and Rundlöf, 2024). Given that, the diversity of pollinators has consistently been shown to be more relevant than the abundance of a single bee species for crop pollination (Potts et al., 2016; Garibaldi et al., 2017), understanding the underlying factors driving exposure in different bee species is essential for developing strategies to mitigate pesticide risks (Cham et al., 2019).

Under the conditions of our study, with thiamethoxam applications occurring 8 months before coffee flowering as per label recommendations, the resulting low residue levels did not cause persistent negative effects on stingless bee colony strength parameters. The residues in leaves collected from the crop and pollen and nectar collected from the stores within the colonies are shown in the Table 2.

Stingless bee colonies kept in conventional and organic farms exhibited similar brood production (χ^2 = 2.61, df = 1, P = 0.10; Figure 2a) and brood mortality (χ^2 = 0.02, df = 1, P = 0.87; Figure 2b), with no significant interaction between farming system and sampling period (P > 0.1 for both variables). In contrast, brood production and mortality varied significantly across sampling periods within each system (P < 0.001; Supplementary Tables 2, 3).

Foraging activity, however, differed between systems (χ^2 = 7.2, df = 1, P = 0.006; Figure 2c), with colonies in conventional farms showing lower baseline activity, particularly before coffee flowering, suggesting pre-existing differences in floral resource availability. Despite this, both systems exhibited similar proportional increases in foraging activity from pre- to post-bloom periods (χ^2 = 29.3, df = 4, P < 0.001; Supplementary Table 4).

Notably, this lower baseline activity in conventional farms was observed before coffee flowering began, suggesting pre-existing differences in available forage resources between the two farming systems. When examining the pattern of increase from pre-bloom to post-bloom periods, both treatments showed similar proportional increases in foraging activity.

While we observed a temporary difference in foraging activity between conventional and organic farms before flowering, this difference equalised in subsequent assessments, and no significant differences were observed in brood production or mortality. These findings suggest that when applied according to label instructions with sufficient time before flowering, thiamethoxam-based products may be compatible with stingless bee pollination services in coffee agroecosystems. However, we acknowledge that distinct application timings or rates, could potentially yield different results, and further research is warranted to explore different label recommendations. Variations in average brood mortality rates in different periods may result from natural fluctuations in colony population, as these stingless colonies were recovering from the winter season and were transported to the farms and back to the forest patch, changing the ecological conditions in each transportation event, which can be a significant source of stress. Abrupt changes in the environmental and/or weather conditions, confinement and disturbance during transportation can increase brood mortality, impair brood production, foraging activity, and even the overall colony strength. Furthermore, the rate of brood mortality in stingless colonies placed on conventionally managed and organic coffee farms remained within the typical range for stingless bees (Bueno et al., 2023). In contrast, foraging activity showed significant differences between the conventional and control groups. In both groups, foraging activity was minimal before coffee blooming, but it was notably lower at conventionally managed farms. At this period, foragers had not yet been exposed to the nectar and pollen of coffee flowers. Therefore, other factors likely contributed to the difference on foraging activity, such as the difference in the composition of forage within the surrounding landscape between the conventional and organic farms; difficulty for foragers locating their nests in the crop field setting or other characteristic in the farming systems. However, in the first assessment after coffee bloom, foraging activity in both groups increased similarly, and colonies with low initial foraging rates recovered in the following weeks, equalising between the two groups in subsequent assessments. For these reasons, the exposure to thiamethoxam should not be considered as the primary cause of the difference in foraging activity.

Despite the temporary difference in foraging activity before flowering, we observed no significant differences in brood production or brood mortality between conventional and organic farms. These parameters are crucial indicators of colony reproductive capacity and overall strength. Extensive literature on bee colonies corroborates that temporary effects on foraging activity do not represent a significant threat to colony viability if they are recovered and do not translate into negative impacts on brood production and mortality (Thompson and Maus, 2007; Blacquière et al., 2012; Pilling et al., 2013; Cutler and Scott-Dupree, 2014; Thompson et al., 2016; Pamminger et al., 2025). Our findings align with this established understanding, as the transient differences in foraging activity did not result in detectable effects on these critical reproductive parameters.

Thiamethoxam and its metabolite clothianidin were detected in all coffee leaves sampled at conventional coffee-producing farms (Table 2). Also, thiamethoxam residues were found in nectar and pollen stored in the stingless bee nests kept in all conventionally managed farms during coffee blooming. At organic farms, residues of thiamethoxam and its metabolite clothianidin in coffee leaves were below the limit of quantification. At the colony level for *A. mellifera*, effects on colony development were observed after 6-week continuous feeding with 100 ppb thiamethoxam, with no adverse effects at 50 ppb and no effects observed (NOEC) at 37.5 ppb thiamethoxam (Thompson et al., 2019). This NOEC for honeybees is 1.8 to 2-fold higher than the residues detected in the pollen and nectar collected from food stores of the stingless bee nests in this study. Although laboratory studies are crucial for indicating the potential risks of pesticide use for non-target organisms, studies under realistic field settings are more reflective of the exposure and effects of pesticides but significantly more challenging due to the inherent difficulty of conducting controlled experiments in the field (Carreck and Ratnieks, 2014). For social pollinators, colony-level studies, which integrate all sublethal effects on individuals, are important to validate regulatory decision-making in evaluating the potential risk of bee-toxic pesticide use (Sgolastra et al., 2020); effects on colony growth and survival cannot be fully assessed by testing individuals in the laboratory. In such studies, attention should be given to the peculiarities of the farming system and characteristics of the studied agricultural crop. In this sense, for systemic insecticides, the results found for one specific crop should not be directly extrapolated for another crop without clear criteria, such as crop attractiveness and resulting residues in pollen and nectar (Cham et al., 2019).

The extended monitoring period of 105 days post-exposure to coffee flowers — spanning approximately three complete

TABLE 2 Residue levels (mean ± SD) of thiamethoxam and clothianidin detected in leaves, nectar, and pollen stored within stingless bee nests sampled in September 2022 at conventionally (CS) and organically (OS) managed farms.

Farm	Thiamethoxam (mg kg^{-1})			Clothianidin (mg kg^{-1})		
	Leaves	Nectar	Pollen	Leaves	Nectar	Pollen
CS1	0.1071	0.0052 ± 0.0010	0.0070 ± 0.0017	0.0509	< 0.0010	0.0020 ± 0.0010
CS2	0.0353	0.0034 ± 0.0001	0.0034 ± 0.0013	0.0349	< 0.0010	0.0015 ± 0.0007
CS3	0.0093	0.0026 ± 0.0001	0.0016 ± 0.0006	0.0102	< 0.0010	< 0.0010
CS4	0.0317	0.0025 ± 0.0015	0.0037 ± 0.0014	0.0340	< 0.0010	< 0.0010
CS5	0.0418	0.0214 ± 0.0014	0.0174 ± 0.0030	0.0327	< 0.0010	0.0010*
CS6	0.0110	0.0032 ± 0.0004	0.0045 ± 0.0006	0.0320	< 0.0010	< 0.0010
OS1	< LOQ	–	–	< LOQ	–	–
OS2	< LOQ	–	–	< LOQ	–	–

*Average value estimated for two pollen samples (<0.001 and 0.0020).

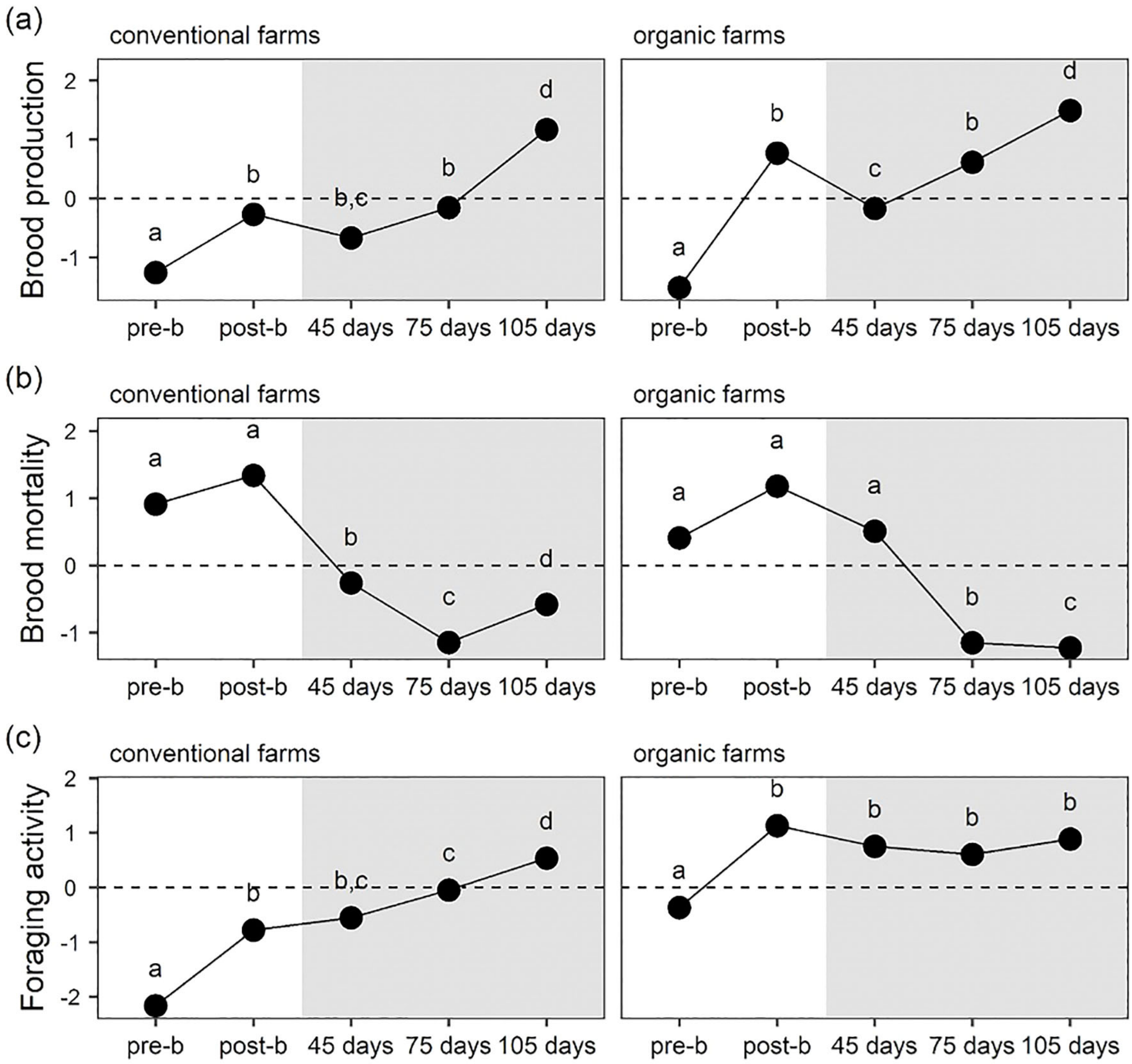

FIGURE 2
Effects of thiamethoxam-based products on stingless bee colony strength in coffee farms. Brood production **(a)**, brood mortality **(b)** and foraging activity **(c)** were assessed on both conventionally managed (left) and organic (right) farms in five periods: pre- and post-blooming and the subsequent 45, 75, and 105 days post-exposure. Evaluations were carried out at coffee farms (in white) and at a forest patch located over 90 km south of coffee farms (in grey). Data are mean values ± SE, and the black dotted line represents the overall mean values. To facilitate multiple comparisons of effect sizes the raw data were standardised with mean 0 ± SE).

generations of stingless bees (Simões and Bego, 1991) — provides robust evidence regarding potential long-term effects. Throughout this timeframe, we observed no significant negative effects on brood mortality, brood production, and foraging activity. The multi-generational nature of our monitoring allows for detection of potential delayed or cumulative effects that might not be apparent in shorter-term studies (Thompson et al., 2019).

Overall, similar findings have been found in other field studies (Carreck and Ratnieks, 2014). While laboratory-based studies have demonstrated that neonicotinoids can have sublethal effects at individual-level on foraging behaviour, cognitive abilities, and larval development, these detrimental effects have often not been observed in the field (Carreck and Ratnieks, 2014; Balfour et al., 2017; Rundlöf and Lundin, 2019). Here, we explore the primary hypothesis aimed at elucidating these apparent discrepancies. Exposure to sublethal doses can vary in duration, depending on the crop blooming period. Additionally, coffee flowers remain open and available for pollination for only 3 to 4 days, which further limits the exposure period for foraging bees. Typically, coffee flowers open synchronously, with flowering highly dependent on rainfall events, creating brief but intense flowering periods that concentrate bee foraging activity within narrow temporal windows (Silva et al., 2009). Stingless bees visit a wide array of flowering plants to collect food resources (Bueno et al., 2023), which leads to a mixing of pollen and nectar from treated crops with that from other plants, thereby diluting the residue levels. Stored pollen and nectar undergo multiple processing, storage, and exposure routes. Besides that, in general, systemic pesticide residues on flowers resulting from applications well before blooming typically remain very low. In our study areas, applications were conducted an average of 8 months before coffee blooming as indicated on the product label, accounting for the low residue levels we detected. In apple orchards, for instance, no residues of systemic pesticides were detected in the whole flower, pollen and nectar sampled in the spring when applied via foliar spray in the previous fall (Heller et al., 2020). Moreover, both the nutritional value of crops and wildflowers and the recovery mechanisms of social bees may compensate for some negative pesticide effects (Balfour et al., 2017; Knapp et al., 2022), allowing the costs of losing foraging workforce without compromising colony survival (Cham et al., 2019). Further studies could usefully explore the fate of these neonicotinoids within the colony, and the exposure levels of stingless bee brood, adult workers, and the egg-laying queen.

We present an original study that combines manipulating pollinator abundance in coffee farms by introducing managed stingless bee colonies to assess coffee yield, and using stingless bees in a higher-tier assessment to evaluate colony strength under field-realistic thiamethoxam exposure. The results of our study contribute to a better understanding of how combining managed and wild pollinators with responsible pesticide use and habitat management enhances agroecosystem quality (Tosi et al., 2022; Siviter et al., 2021). Introducing stingless bee colonies gives farmers a direct method to increase coffee yield and generate long-term sustainable profit, which can be invested in native forest restoration on their farms (d'Albertas et al., 2024). We underscore the importance of pollination for intensively managed coffee production and the synergistic link between agriculture and environmental conservation in achieving maximum profitability. This profitability is vital in encouraging farmers to practice good land stewardship through nature-positive approaches, ultimately providing diverse benefits to society.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material. Further inquiries can be directed to the corresponding authors.

Ethics statement

The manuscript presents research on animals that do not require ethical approval for their study.

Author contributions

JR: Writing – original draft, Writing – review & editing, Conceptualization, Investigation, Methodology, Validation, Visualization. GS: Writing – review & editing. CF: Data curation, Formal analysis, Writing – review & editing. TS: Writing – review & editing. AC: Project administration, Writing – review & editing. DA: Writing – original draft, Writing – review & editing. FC: Methodology, Writing – review & editing. AC: Conceptualization, Validation, Writing – review & editing. AP: Writing – review & editing. HT: Writing – review & editing. AM: Writing – review & editing. JB: Writing – review & editing. CM: Conceptualization, Investigation, Methodology, Project administration, Writing – original draft, Writing – review & editing.

Funding

The author(s) declared that financial support was received for this work and/or its publication. This work was supported by Embrapa and Syngenta (grant SEG 10.20.00.143.00.00), the National Council for Scientific and Technological (CNPq, grants 164743/2020-0 to D.A.A. and 350679/2022-3 to C.F.S.).

Acknowledgments

We are grateful to farmers and beekeepers for their support to this study, allowing us to access coffee farms, assisting with coffee harvest and bee management. We also thank the Embrapa teamwork for helping with field logistics.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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