Insecticidal and fungicidal activity of eucalyptol against pest and fungal diseases of soybean

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ABSTRACT

Soybean (*Glycine max*), a globally significant crop, faces significant economic losses from pests and fungi. Farmers often resort to synthetic pesticides, posing potential risks to human and environmental health. In this context, eucalyptol (1, 8 cineole) stands out in agriculture for its toxicity to various insects and fungi, with minimal environmental and human health impacts. This study aimed to assess in the lab the insecticidal activity of eucalyptol against *Spodoptera frugiperda* and *Epicauta atomaria*. Its fungicidal activity was studied against *Cercospora kikuchii*, *C. sojina*, and *Sclerotium rolfsii*. Preliminary exploration of the potential phytotoxic effect on soybean plants has been initiated. Eucalyptol demonstrated significant fumigant insecticidal activity against *S. frugiperda* 1st instar larvae ($LC_{50} = 9.4 \,\mu L/L \,air$) and *E. atomaria* adults ($LC_{50} = 34.6 \,\mu L/L \,air$), along with significant fungicidal activity against *C. kikuchii* and *C. sojina* (mycelial inhibition halo diameters of 6.0 and 10.0 mm, respectively, at a concentration of 5 $\mu L/disk$). Importantly, eucalyptol exhibited no phytotoxic effects on soybean. Eucalyptol potential as a biopesticide for soybean crops, providing an alternative to synthetic pesticides. Further research is needed to determine its economic viability and large-scale applicability.

Keywords: botanical insecticides, sustainable pest management, monoterpene, plant extract, phytotoxicity, fall armyworm, blister beetle, fungicide effect.

RESUMEN

La soja (Glycine max), un cultivo de importancia global, enfrenta pérdidas económicas significativas debido a plagas e infecciones fúngicas. Ante esto, los agricultores suelen recurrir a plaguicidas sintéticos, lo que puede generar riesgos para la salud humana y el medioambiente. En este escenario, el eucaliptol (1,8-cineol) se destaca en la agricultura por su toxicidad contra diversos insectos y hongos, con un impacto mínimo en el medioambiente y la salud humana. El objetivo de este estudio fue evaluar en laboratorio la actividad insecticida del eucaliptol contra Spodoptera frugiperda y Epicauta atomaria. Asimismo, se investigó su actividad fungicida contra Cercospora kikuchii, C. sojina y Sclerotium rolfsii. Adicionalmente, se comenzó a explorar su potencial efecto fitotóxico en plantas de soja. El eucaliptol demostró una significativa actividad insecticida fumigante contra larvas de primer instar de S. frugiperda ($LC_{50} = 9.4 \,\mu$ L/L de aire) y adultos de E. atomaria ($LC_{50} = 34.6 \,\mu$ L/L de aire). También se observó una notable actividad fungicida contra C. kikuchii y C. sojina, con diámetros de halo de inhibición micelial de 6.0 y 10.0 mm, respectivamente, a una concentración de 5 μ L/disco. Es importante resaltar que el eucaliptol no mostró efectos fitotóxicos en la soja. En conclusión, el eucaliptol presenta un gran potencial como biopesticida para los cultivos de soja, ofreciendo una alternativa a los plaguicidas sintéticos. Sin embargo, se requiere investigación adicional para determinar su viabilidad económica y su aplicabilidad a gran escala.

Palabras clave: insecticidas botánicos, manejo sostenible de plagas, monoterpeno, extracto vegetal, fitotoxicidad, gusano cogollero, escarabajo vesicante, efecto fungicida.

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INTRODUCTION

Soybean, Glycine max (L.) Merr (Fabales: Fabaceae), is a globally significant crop, playing a pivotal role in oil and protein production (Graham and Vance 2003; Hamza et al., 2024). Argentina is the world's third-largest producer of soybeans, with an annual output of 51 million tonnes, accounting for 13% of global production (USDA, 2022). Nevertheless, various pests and fungal diseases can cause considerable damage to these crops in Argentina, resulting in significant annual economic losses (Jerez et al., 2023; Murúa et al., 2018; Ploper, 2004). The fall armyworm (FAW), Spodoptera frugiperda JE Smith (Lepidoptera: Noctuidae), and the blister beetle (BB), Epicauta atomaria Germar (Coleoptera: Meloidae), are phytophagous insects that affect various agricultural crops, including soybeans (De Freitas Bueno et al., 2011; Campos-Soldini et al., 2021; Overton et al., 2021). FAW can cause significant yield losses in soybean, especially at postbloom stages, where intervention thresholds have been reported at 25% defoliation, compared to 50% at pre-bloom (Overton et al., 2021). In contrast, no established thresholds have been reported for BB. Additionally, Cercospora kikuchii (Tak. Matsumoto and Tomoy.) MW Gardner, Cercospora sojina K Hara (Mycosphaerellales: Mycosphaerellaceae), and Sclerotium rolfsii Sacc. (Amylocorticiales: Atheliaceae) are phytopathogenic fungi responsible for various diseases in soybean (Barro et al., 2023; Billah et al., 2017; Bhamra and Borah, 2022; Hartman et al., 1999; Sautua et al., 2019). Although soybean yield losses caused by these pathogens are difficult to estimate, as they, along with other pathogens, are part of the "late-season soybean diseases" (LSD). Collectively, these diseases account for an average of 10% of annual losses, but can reach up to 30%, depending on environmental conditions (Carmona et al., 2016). In Argentina, it was estimated that damage caused by C. kikuchii reduced soybean crop yields by 11% in 2018 and 2019 (Lavilla and Ivancovich, 2021). Consequently, farmers often turn to synthetic pesticides to mitigate these issues. These synthetic pesticides can be harmful to both human and environmental health, especially when used excessively or inappropriately (Alaoui et al., 2024; Aparicio and De Gerónimo, 2024; Rani et al., 2021). Therefore, it is essential to seek and promote new tools that facilitate the more sustainable control of these pests and diseases.

Eucalyptol (1, 8-cineole) is a monoterpene oxide comprising up to 85% of the total essential oils extracted from eucalyptus species (Campos and Berteina-Raboin, 2022). It is also present in essential oils from rosemary (*Salvia rosmarinus* Spenn.), lavender (*Lavandula* sp), and laurel (*Laurus nobilis* L.), albeit in smaller proportions (Borges et al., 2019; Cavanagh and Wilkinson, 2002; Chahal et al., 2017). Currently, there is increasing interest in eucalyptol, not only in the pharmaceutical and cosmetic industries (Cai et al., 2021; Hoch et al., 2023) but also in agriculture. This interest in eucalyptol stems from its notable toxicity against various pest insects and phytopathogenic fungi (Jiang et al., 2020; Tahiri et al., 2022; Tripathi and Mishra, 2016), coupled with its facile biodegradability and minimal impact on the environmental and human health (Batish et al., 2008).

So far, the potential of eucalyptol as a prospective active compound in biopesticide formulations for controlling the mentioned insects and phytopathogenic fungi is still understudied. However, some studies have revealed that essential oils rich in eucalyptol exhibit pronounced toxicity against these insects and fungi (Sekhar et al., 2020; Usseglio et al., 2022; Wagner and Campos-Soldini, 2022; Wagner et al., 2021). Based on these considerations, we aim to pursue the following research objectives: i) evaluate the

insecticidal and fungicidal activity of eucalyptol against the mentioned species of insect and fungi, and ii) investigate the possible phytotoxic effect of eucalyptol on soybean plants.

MATERIALS AND METHODS

Chemical compound

Eucalyptol (1, 8-cineole), characterized by its analytical-grade quality (99% purity), was obtained from Merck-Sigma-Aldrich®, Argentina, and is commercially available at http://www.sigmaaldrich.com/. Chlorpyrifos (10.5% w/v) (Huagro Hormix, Huagro SA, Argentina) and difenoconazole (25% w/v) (Janfry®, Gleba SA, Argentina) and 2, 4-D (66.9% w/v) (Enlist®,Corteva Agriscience™, Argentina).

Insects and fungi

BB adults were collected manually from their host plants (Salpichroa origanifolia (Lam.) Baill. and Amaranthus hybridus L.) located in the vicinity of soybean fields near Diamante, Argentina, during the spring period (October and November) of 2022. FAW larvae were acquired from the moth colony in the CICYTTP insectarium, Diamante, established in 2020. Both species were maintained in the laboratory under controlled conditions at $27 \pm 1^{\circ}\text{C}$, $70 \pm 5\%$ relative humidity, and a light-dark photoperiod of 16/8 h. BB adults were feed with fresh chard leaves, while FAW larvae were nourished using an artificial diet based on chickpea flour and wheat germ (Murúa et al., 2003).

The fungi *C. kikuchii* (strain NRBC 6713), *C. sojina* (strain NRBC 6715), and *S. rolfsii* (strain CCC 143-2018), were sourced from microbial collections affiliated with the Facultad de Bioquímica y Ciencias Biológicas at the Universidad Nacional del Litoral, Argentina, and the Centro de Referencia de Micología (CEREMIC) at the Universidad Nacional de Rosario, Argentina.

Insecticidal activity

The fumigant insecticidal activity of eucalyptol was evaluated following the protocol outlined by Baghouz et al. (2024), with slight adaptations. Briefly, groups of five insects (BB adults or FAW 1st instar larvae) were placed in 127 mL glass vials fitted with rubber lids. A 1 cm² Whatman filter paper disk was affixed to the underside of each rubber lid. The filter paper disks were impregnated with varying aliquots of pure eucalyptol, using an automatic pipette, to achieve concentrations of 19.7, 27.6, 35.4. 47.2. and 78.7 µL/L air (for experiments with BB) and 3.1. 6.3, 9.4, 12.6, and 19.7 μ L/L air (for experiments with FAW). To support the filter paper on the lid and prevent direct contact of the insects with eucalyptol, a porous fabric mesh was used between the lids and the vials. To prevent vapour escape, the vials were hermetically sealed with Parafilm. Negative controls only contained a filter paper disk free of chemical substances, while positive controls incorporated a filter paper disk with chlorpyrifos at equivalent concentrations to those used with eucalyptol in both species. Each treatment was replicated five times under controlled conditions at a temperature of 27 ± 1°C and a lightdark photoperiod of 16/8 h. Each replication was conducted on different days. Mortality was recorded after 6 h of exposure, for BB and 24 h, for FAW. Insects were deemed dead if they remained motionless in response to stimuli provided by entomological brushes and forceps.



Fungicidal activity

The fungicidal activity of eucalyptol against *C. kikuchii*, *C. sojina*, or *S. rolfsii* was evaluated using the disc diffusion method, as described by Sequín et al. (2023), with some adaptations. Fungal hyphal suspensions were obtained by carefully collecting mycelium from 7-day-old colonies of *C. kikuchii*, *C. sojina*, or *S. rolfsii* grown on PDA (Potato Dextrose Agar). The hyphal fragments in each suspension underwent microbiological surface quantification and were subsequently adjusted to a concentration ranging from 4.0×10^2 to 4.3×10^3 CFU/mL. Following this, $100~\mu\text{L}$ of each suspension was inoculated at the centre of a 9 cm diameter Petri dish containing 10 mL of YMDA (4 g/L yeast extract, 4 g/L malt extract, 10 g/L dextrose, 15 g/L agar), ensuring even distribution across the medium surface.

Sterile 5 mm diameter Whatman N° 4 filter paper discs were impregnated with 5, 2.5, and 1 μ L of eucalyptol per disc, plus 5 μ L of dimethyl sulfoxide (DMSO). Negative controls consisted solely of discs saturated with 5 μ L of DMSO per disc, while positive controls contained 5 μ L of difenoconazole (concentration: 0.2 μ g of a.i for μ L) from a commercial source, which was used as a positive control per disc. Subsequently, the impregnated discs were placed on the surface of the PDA-inoculated Petri dishes. The Petri dishes were flipped and incubated in an oven at 25 ± 1°C, maintaining a 16/8 h light-dark cycle for a duration of 7 days. Each treatment was replicated four times on different days. Mycelial inhibition halo diameter was measured at the experiment's conclusion using a digital caliper, values expressed in millimetres (mm).

Phytotoxic activity

The potential phytotoxic effect of eucalyptol on soybean was evaluated, following a protocol similar to that of the International Seed Testing Association (1976), but with slight modifications. Briefly, single soybean seeds were planted inside plastic boxes containing a 2 cm-thick layer of sterilized sand (weight: 30 g). Each sand layer was treated with a 3 mL solution of eucalyptol (concentration: 1,150 μ g/mL) for the experimental samples, or distilled water for negative controls, or 2, 4-D (1.25 μ g/mL) for positive controls. In all cases, distilled water with 1% v/v Tween 20 as diluent was used.

Each box with its soybean seed was considered one replicate, totalling 12 replicates per treatment. The boxes were hermetically sealed with a lid and placed in a germination chamber at a constant temperature of $25 \pm 1^{\circ}$ C, with a photoperiod of

16/8 h light-dark cycle for six days. At the end of this period, the hypocotyl and radicle of each seedling were measured using a digital caliper. The dry weight of each seedling was also determined after dehydrating them in an oven at 50°C for 3 days until a constant weight was achieved.

Statistical analysis

For BB and FAW experiments, lethal concentration doses producing 50% (LC $_{50}$) and 90% (LC $_{90}$) mortality were determined using Probit analysis (Finney, 1971) with POLO-PLUS Software (LeOra Software, 2002–2014). Significant differences between LC $_{50}$ and LC $_{90}$ values were considered when the 95% confidence limits did not overlap. Insecticidal, fungicidal and phytotoxic activity were assessed using the Kruskal-Wallis test, followed by a Conover test for post hoc comparisons (Conover, 1999), utilizing InfoStat version 2018 statistical software, and with α = 0.05.

RESULTS

Insecticidal activity

Figure 1 illustrates the mortality percentages caused by the fumigant action of different eucalyptol concentrations against FAW and BB. Table 1 summarises the calculated LC $_{50}$ and LC $_{90}$ values for eucalyptol and chlorpyrifos (positive control) against both species. Eucalyptol exhibited significant fumigant insecticidal activity in a concentration-dependent manner against FAW 1st instar larvae (H = 24.61; df = 5; p < 0.05) and BB adults (H = 22.33; df = 5; p < 0.05). Median mortality reached 100% at concentrations equal to or higher than 9.4 μ L/L air for FAW and 47.2 μ L/L air for BB, respectively. The calculated LC $_{50}$ values were 6.1 μ L/L air for FAW and 34.6 μ L/L air for BB. Chlorpyrifos (positive control) showed potent fumigant activity with LC $_{50}$ values below 3.1 and 19.7 μ L/L of air against FAW and BB, respectively (table 1). Negative controls showed no mortality in either species.

Fungicidal activity

Table 2 presents the median values of the mycelial inhibition halo diameters caused by the fungicidal action of eucalyptol at different concentrations against *C. kikuchii*, *C. sojina*, and *S. rolfsii*. Eucalyptol demonstrated significant fungicidal activity against *C. kikuchii* and *C. sojina*. Specifically, only mycotoxicity was observed at the highest evaluated concentration (5 μL/

Insects	Compound	LC ₅₀ (µL/L air)	95% CL (µL/L air)	LC ₉₀ (µL/L air)	95% CL (µL/L air)	Slope ± SE	(x ²) ^a
S. frugiperda	Eucalyptol	6.1	5.2-7.0	10.5	9.0-13.3	5.5 ± 0.8	1.4
	Chlorpyrifos	< 3.1		< 3.1			
E. atomaria -	Eucalyptol	34.6	31.9-37.4	46.6	42.1-55.6	9.9 ± 1.7	0.8
	Chlorpyrifos	< 19.7		< 19.7			

LC: lethal concentration. CL: confidence limits. Chlorpyrifos: positive control.

Table 1. Fumigant toxicity of eucalyptol against Spodoptera frugiperda first-instar larvae and Epicauta atomaria adults.

^a Chi-square values, significant at p < 0.05 level



disk). At this concentration, mycelial inhibition halo diameters of 6.0 \pm 0.0 mm and 10.0 \pm 2.5 mm (median \pm interquartile range) were observed for *C. kikuchii* and *C. sojina*, respectively. These values differed significantly from the negative controls, where no mycelial inhibition was evident (table 2). In contrast, the positive control (difenoconazole: 5 μ l/disk), applied to both *C. kikuchii* and *C. sojina*, exhibited mycelial inhibition halo diameters significantly similar to the maximum eucalyptol concentration. However, neither eucalyptol nor the negative control nor difenoconazole inhibited the growth of *S. rolfsii* mycelium.

Phytotoxic activity

Figure 2 shows the phytotoxic effect caused by eucalyptol on the radicle growth, hypocotyl growth, and dry weight of soybean seedlings. Eucalyptol at 1,150 μ g/mL (\approx 1,249 μ L/L) exhibited no phytotoxic activity on soybeans despite even at a concentration 1 \times 10³ times higher 2, 4-D (positive control: 1.25 μ g/mL). In contrast, 2, 4-D demonstrated significant phytotoxicity, as evidenced by a significant reduction in both radicle length growth (H = 15.17; df = 2; p < 0.05) and seedling dry weight (H = 7.09; df = 2; p < 0.05). However, hypocotyl growth remained unaffected by both eucalyptol and 2, 4-D.

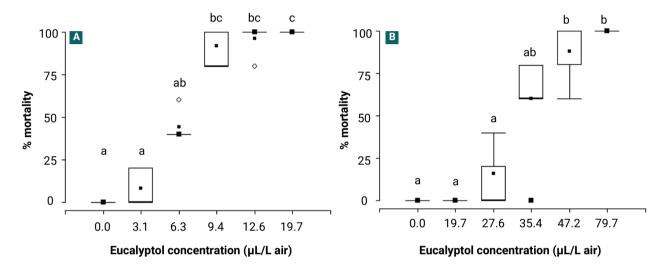


Figure 1. Box and whisker plots showing the percentage mortality caused by the fumigant effect of different eucalyptol concentrations on (A) *Spodoptera frugiperda* first-instar larvae and (B) *Epicauta atomaria* adults. Each box represents the median (horizontal line), the mean (central point), and the interquartile range (percentiles 25 and 75). Whiskers extend to percentiles 10 and 90. Data analysis was conducted using a Kruskal-Wallis test followed by a Conover multiple comparison test (p < 0.05; n = 12). Different letters above the boxes indicate significant differences among treatments.

Fucelyntal (ul. /dica)	Mycelial inhibition halo diameter median ± interquartile range (mm)				
Eucalyptol (μL/disc)	C. kikuchii	C. sojina	S. rolfsii		
5	6.0 ± 0.0 bc	10.0 ± 2.5 b	8.0 ± 10.0 a		
2.5	5.0 ± 1.0 abc	8.5 ± 3.0 ab	0.0 ± 7.0 a		
1	4.0 ± 4.0 ab	6.50 ± 0.5 ab	0.0 ± 0.0 a		
Control (-)	0.0 ± 0.0 a	0.0 ± 0.0 a	0.0 ± 0.0 a		
Control (+)	9.0 ± 2.0 c	7.0 ± 2.0 b	0.0 ± 0.0 a		
Н	12.75	10.31	3.28		
df	4	4	4		
р	0.0099	0.0326	0.1520		

Medians with different letters represent significant differences between treatment groups (Kruskal–Wallis test followed by Conover post–hoc comparisons, significant at p < 0.05 level)

Table 2. Mycelial inhibition halo diameters values recorded for eucalyptol against Cercospora kikuchii, Cercospora sojina, and Sclerotium rolfsii



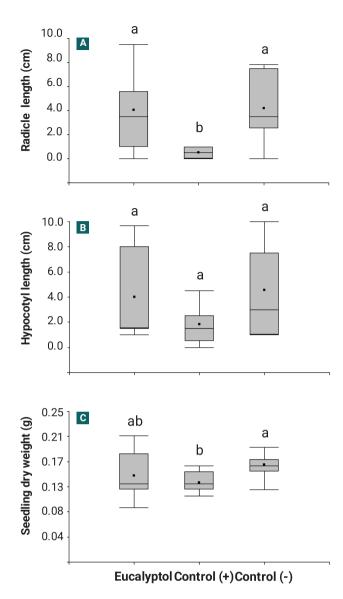


Figure 2. Box and whisker plots represent the effect of eucalyptol at 1.15 μg/mL on A) radicle growth, B) hypocotyl growth, and C) dry weight of soybean seedlings. Positive control: 2, 4-D at 1.25 μg/mL, and negative control: distilled water with 1% v/v Tween 20. Each box represents the median (horizontal line), the mean (central point), and the interquartile range (percentiles 25 and 75). Whiskers extend to percentiles 10 and 90. Data analysis was conducted using a Kruskal-Wallis test followed by a Conover multiple comparison test (p < 0.05; n = 12). Different letters above the boxes indicate significant differences among treatments.

DISCUSSION

In general, eucalyptol exhibited fumigant insecticidal activity against FAW 1st instar larvae and BB adults, as well as fungicidal activity against *C. kikuchii* and *C. sojina*. Importantly, eucalyptol showed no phytotoxic effects on soybean.

Insecticidal activity

To date, the insecticidal activity of eucalyptol through fumigant action against FAW larvae and BB has not been reported. However, studies have shown that, when applied topically, eucalyptol exhibits toxicity (59.6% mortality) against FAW at a dose of $\approx 0.01223~\mu L/larva$ (Bibiano et al., 2022). In contrast, at lower doses ($\approx 0.00326~\mu L$ of eucalyptol/mg of larva), its toxicity is practically negligible (Niculau et al., 2013). Within the mode of application by fumigation, previous studies reveal that essential oils from lavender and rosemary, rich in eucalyptol (34.33 and 18.72%, respectively), demonstrate outstanding activity against BB, with LC $_{50}$ values of 28.9 and 23.3 $\mu l/L$ air, respectively (Wagner and Campos-Soldini, 2022; Wagner et al., 2021). Although this better activity than eucalyptol could be attributed to the addition or synergy with other compounds present in the oils as it was suggested in the case of Tenebrio molitor (L) (Coleoptera: Tenebrionidae) and Spodoptera littoralis Boisd (Lepidoptera: Noctuidae) (Lima et al., 2011; Pavela, 2014).

Eucalyptol also shows fumigant toxicity in other beetle pests, such as stored grain insects *Sitophilus oryzae* L (Coleoptera: Curculionidae), *Tribolium castaneum* Herbst, *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae), and *Rhyzopertha dominica* Fabricius (Coleoptera: Bostrichidae) (Lee et al., 2004; Abdelgaleil et al., 2009; Kheloul et al., 2023).

As observed in other insect species, the toxicity induced by eucalyptol in FAW and BB could be attributed to the inhibition of acetylcholinesterase (AChE) enzyme activity, which disrupts the insect's nervous system, leading to paralysis and eventual death (Abdelgaleil *et al.*, 2009; Picollo *et al.*, 2008).

Fungicidal activity

While previous studies highlight the significant potential of eucalyptol as a fungicide against various phytopathogenic fungi (Morcia et al., 2012; Shukla et al., 2012; Oxenham et al., 2005; Dammak et al., 2019; Jiang et al., 2020), this study is the first to report its fungicidal activity against C. kikuchii and C. sojina. However, our research group previously determined that, at the same concentration of 5 μ l/disk, the essential oil extracted from lavender, rich in eucalyptol (34.33%), exhibits potent fungicidal activity against C. kikuchii and C. sojina, with mycelial inhibition halos of 34.0 and 29.5 mm, respectively (Wagner et al., 2021). This indicates a fungicidal activity much superior of the natural oil to that observed for pure eucalyptol. This enhanced activity is likely attributed to additive or synergistic effects with other major components present in the essential oil (Hassan et al., 2020; Yan et al., 2021).

On the contrary, in our experiments, eucalyptol does not exert a significant inhibitory effect on the mycelial growth of *S. rolfsii*. These results contradict findings by Kottearachchi *et al.* (2012), who report 100% inhibition of *S. rolfsii* mycelial growth when treated with high concentrations of eucalyptol (0.5 and 1.5%). However, the same authors note that at lower concentrations, the inhibition percentage decreases to less than 30%.

It is well-established that many essential oils, rich in terpenes, possess the ability to induce alterations in the fungal cell wall, plasma membrane, and mitochondria, thereby substantiating their toxicity (Kishore et al., 2007; Pawar and Thaker, 2006). Indeed, toxicity studies conducted against the phytopathogenic fungus Botrytis cinerea confirm the detrimental effects of eucalyptol on the organelles of this fungus's cells (Yu et al., 2015). It is plausible that the observed mycotoxicity of eucalyptol against C. kikuchii and C. sojina may be attributed to some of these reasons.



Phytotoxicity

Previous studies have underscored the strong phytotoxicity of eucalyptol against various plant species as lettuce (Qiu et al., 2010), annual ryegrass, radish (Barton et al., 2014), redroot amaranth and annual bluegrass (Shao et al., 2018; Zhou et al., 2019), our preliminary findings suggest that even at high concentrations, eucalyptol is not phytotoxic to soybean seedlings. Consistent with our study on the herbicidal effects of eucalyptol, Vaughn and Spencer (1993), using a similar methodology, observed that this terpene does not exhibit significant phytotoxicity in soybean seedlings.

It is acknowledged that the phytotoxicity of eucalyptol in other plant species, such as *Arabidopsis*, potato and onion, is attributed to various causes, such as disruptions in microtubule organization and the inhibition of key processes like mitochondrial respiration, mitosis, and phytohormone production, leading to a negative impact on plant development and growth (Baskin *et al.*, 2004; Verdeguer *et al.*, 2020). Nevertheless, it is conceivable that only very high concentrations of eucalyptol may be detrimental to soybean plants at the seedling stage, opening prospects for its future application in protecting this crop.

CONCLUSIONS

The protection of soybean crops against pest insects and phytopathogenic fungi within the framework of Integrated Pest Management (IPM), based on methods like biological control, planting resistant cultivars, and using biopesticides, has sharply declined in recent decades (Bueno et al., 2023; Bueno et al., 2021; Panizzi, 2013). This decline is primarily attributed to the prevalent use of synthetic pesticides as the main control method, despite the well-known adverse impacts on the environmental and human health. Our findings indicate that eucalyptol has notable potential as an active ingredient for future biopesticide formulations, aiming to reduce, at least partially, the use of synthetic pesticides in soybean cultivation. This potential is primarily based on its insecticidal and fungicidal properties found in our studies. However, as with essential oils, its possible high volatility and rapid degradation, induced by exposure to air, sunlight, moisture, and high temperatures, could significantly reduce its effectiveness. Therefore, the application of eucalyptol may face significant limitations, requiring more frequent and higher quantities of application to the crop, thus increasing costs. These stability issues could be addressed through the implementation of formulations based on nanoencapsulation, which protect the compounds from adverse environmental factors, thereby improving their stability and efficacy (Giunti et al., 2021; Campolo et al., 2018; Gupta et al., 2023; Šunika and Mechora, 2022). Therefore, we believe further studies are needed to evaluate the economic feasibility and applicability of eucalyptol in large-scale cultivation areas.

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