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ORIGINAL ARTICLE

Biocontrol potential of selected botanicals and *Beauveria bassiana* (Bals.) spore suspension against *Bemisia tabaci* (Genn.)

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Abstract

This study aimed to evaluate the effects of plant extracts of *Capsicum annuum*, *Datura stramonium* and *Allium sativum* and the fungus *Beauveria bassiana* on different developmental stages of *Bemisia tabaci*. The *D. stramonium* extract achieved the highest adult mortality rate of 92.4% at 4% w/v concentration after 96 hours of exposure, which was higher than *A. sativum* (85.7%) and *C. annuum* (76.3%). The isolated *D. stramonium* extracts demonstrated maximum lethal activity against the early developmental stages of *B. tabaci*, with second instar showing the highest vulnerability and mortality rate of up to 97.8%. *B. bassiana* exhibited maximum pathogenicity against *B. tabaci* life stages, specifically with the second instar nymphs showing the highest susceptibility at 93.6% mortality when treated with 1×10^8 conidia/mL. The combination of plant extracts with *B. bassiana* resulted in elevated mortality rates with all combinations demonstrating synergistic effects with co-toxicity factor (CTF) values > 20. Lethal potency emerged from the combination treatment of *D. stramonium* (1% w/v) and *B. bassiana* (1×10^6 conidia · ml^{-1}), which reached a maximum adult mortality of 98.0% ±2.1. Greenhouse trials showed that this combined treatment generated population reductions exceeding 90% across all life stages, including adult insects, nymphs, and eggs within 14 days. These findings confirm that both botanical extracts and *B. bassiana* are suitable components for developing sustainable Integrated Pest Management (IPM) programs due to their efficient and environmentally friendly insect pest control properties.

Keywords: *Allium sativum, Bemisia tabaci*, *Beauveria bassiana*, *Capsicum annuum*, *Datura stramonium*

Introduction

The whitefly species *Bemisia tabaci* Genn. (Hemiptera: Aleyrodidae) is a globally destructive agricultural pest that infests approximately 600 tropical and subtropical plant species (Gilbertson *et al.* 2015). Commercial damage from this polyphagous pest derives from direct feeding effects in addition to the production of honeydew and the transmission of more than 200 plant viruses (Fiallo-Olivé *et al.* 2020). *B. tabaci* and its associated viral agents have caused significant financial damage that was projected to reach 132.3 million USD in 2016 and 161.2 million USD in 2017 (Li *et al.* 2021). The management of *B. tabaci* has been mainly dependent on synthetic chemical pesticides. However, this pest has consequently developed resistance to several chemical pesticides, and other recent studies have reported that its populations are increasingly resistant to multiple pesticides, including new compounds (Guo *et al.* 2019; Wakil *et al.* 2023). Increasing resistance issues and growing concerns about environmental pollution (non-target species), potential risks to human health, highlight the need for research initiatives to develop alternative control methods (Khalifa and Bedair 2023; Sabry *et al.* 2023; Wang *et al.* 2024).

Due to their diverse modes of action, low environmental persistence, and low toxicity to mammals (70%

of insecticides being derived from plants), botanical insecticides represent a promising solution to this problem (Moustafa *et al.* 2024; Rohimatun *et al.* 2024). Numerous plant species have produced secondary metabolites that demonstrated insecticidal, repellent, antifeedant, and growth-regulating properties against various insect pests (Jafarbeigi *et al.* 2012; Campolo *et al.* 2020). Therefore, *C. annuum*, *D. stramonium*, and *A. sativum* are particularly promising for pest management due to their bioactive constituents (Sutanto *et al.* 2025). The alkaloids, capsaicinoids, and flavonoids in *C. annuum* demonstrate important insecticidal and repellent properties. Additionally, extracts obtained from *C. annuum* have been found to be effective against several agricultural pests notably aphids, thrips, and larvae of butterflies (Ristaino *et al.* 2021; Sánchez-Quezada *et al.* 2024). Numerous studies have shown that the *D. stramonium* plant extract is rich in tropane alkaloids that can disrupt insect nervous system function. This extract has also shown promising results against various hemipteran pests (Kumari *et al.* 2017; Sabzevari and Hofman 2022). Mnayer *et al.* (2014) demonstrated that the *A. sativum* plant extract exhibits insecticidal, repellent, and antibacterial properties due to the organosulfur compounds allicin and diallyl disulphide. Studies have indicated that the *A. sativum* extract effective against sucking-piercing insects such as whiteflies and aphids (Kayahan 2023).

Entomopathogenic fungi, particularly *B. bassiana* – a leading biopesticide for field applications – have attracted research interest as a more sustainable control method to manage insect infestations (Jasman and Slomy 2021). This fungus is particularly effective against sucking-piercing pests such as whiteflies because it enters directly through the insect's cuticle (Dara 2019). *B. bassiana* has shown high efficacy for controlling *B. tabaci* at various developmental stages (Cuthbertson and Audsley 2016). Furthermore, the development of new formulations based on *B. bassiana* has improved the longevity and efficacy of such biopesticides, thereby enhancing their economic potential (Jaronski and Mascarin 2017). The studies about molecular analysis of intestinal bacterial communities in insects combined with bioassays of botanical insecticides and entomopathogenic fungi have revealed synergistic interactions between botanical insecticides and entomopathogenic fungi against different insect pests (Togbé *et al.* 2015). This synergistic combination may mitigate the disadvantages of individual treatments, including the relatively slow action of entomopathogenic fungi and the short residual activity of botanical insecticides, and could potentially lower the risk of resistance development due to their distinct modes of action (Bamisile *et al.* 2021; Norris and Bloomquist 2021). However, comprehensive research on their relative effectiveness, modes of action and compatibility with native enemies is still lacking. Therefore, this study aimed to: (1) evaluate the insecticidal properties of the *C. annuum*, *D. stramonium*, and *A. sativum* extracts against *B. tabaci*; (2) assess the pathogenic potential of *B. bassiana* against various developmental stages of *B. tabaci*; (3) explore possible synergistic relationships between plant extracts and *B. bassiana*; and (4) validate the effectiveness of promising treatments in greenhouse settings. The findings of this study contribute to the development of sustainable management strategies for this economically important pest.

Materials and Methods

Insect rearing

A colony of *B. tabaci* was established from field-collected individuals from tomato crops and maintained on tomato plants (*Solanum lycopersicum*) in mesh cages (60 × 60 × 60 cm) at 25 ± 2°C, 65 ± 5% relative humidity (RH), and a 16 : 8 h (light:dark) photoperiod. The insects were identified using taxonomic keys by entomologists.

Plant material and extract preparation

Fresh plant materials of hot pepper fruits (*C. annuum* L.; Solanales: Solanaceae), Jimsonweed leaves (*D. stramonium* L.; Solanales: Solanaceae), and Garlic bulbs (*A. sativum* L.; Asparagales: Amaryllidaceae) were collected from the Agricultural College, AL-Qasim Green University, Iraq, in Spring of 2024. The plant species were identified and confirmed by a taxonomist using standard taxonomic keys and morphological descriptions provided in the Flora of Iraq (Townsend and Guest 1985). Plant materials were washed with distilled water and then dried at room temperature (25 ± 2°C) under shade conditions for 7–10 days before grinding into powder using an electric grinder. The preparation of extracts involved the maceration process described by Sutanto *et al.* (2025), with minor adjustments. Each plant powder was dissolved in a solution of 100 g in 1 L of distilled water for 24 h with intermittent shaking. The solution was filtered via Whatman No. 1 filter paper and the resulting filtrate was concentrated using a rotary evaporator. The prepared crude extracts were stored in dark glass bottles at 4°C until analysis. For bioassays, stock solutions containing 10% w/v concentration were prepared by dissolving plant extracts in distilled water combined with 0.1% Tween-80 as an emulsifier. Solutions with different concentrations (0.5, 1.0, 2.0, and 4.0% w/v) were prepared

by diluting the stock solutions with distilled water.

Phytochemical analysis

The major bioactive compounds present in the *D. stramonium*, *A. sativum*, and *C. annuum* plant extracts were identified and quantified using high-performance liquid chromatography (HPLC) following the methods described by Mnayer *et al.* (2014).

Fungal culture and preparation

The fungal isolate *B. bassiana* strain (BB-BAG0; spore concentration 4.4×10^{10} conidia $\cdot$ g^{-1}) was obtained from the Department of Plant Protection, University of Baghdad, and was morphologically confirmed using standard taxonomic keys based on colony characteristics and conidial morphology (Rehner *et al.* 2011).

The commercial product was combined with sterile distilled water and 0.05% Tween-80 to prepare conidial suspension. The suspensions were vortexed for two minutes before filtering the solution using a sterile muslin pad, and the concentrations were adjusted to 1×10^6, 1×10^7, and 1×10^8 conidia $\cdot$ ml^{-1} using a hemocytometer.

Insecticidal activity of plant extracts

Plant extract evaluation for insecticidal properties against adult *B. tabaci* was performed through leaf-dip bioassays following Moustafa *et al.* (2024) with minor modification. Tomato leaf discs (3.5 cm diameter) were immersed in each plant extract solution (0.5, 1.0, 2.0, and 4.0% w/v) for ten seconds with gentle agitation, then dried for 30 min. The control solution contained distilled water and 0.1% Tween-80. Each treated leaf disc was placed individually in ventilated Petri dishes (9 cm diameter) containing a 1.5% agar layer to maintain proper leaf turgidity. The experimental procedure included placing 20 adult whiteflies of mixed sex into each Petri dish using an aspirator shortly after their emergence (1–2 days old). Each ventilated Petri dish (9 cm diameter) was filled with 1.5% agar to maintain leaf turgidity under controlled conditions at 25 ± 2°C with 65 ± 5% RH and a 16 : 8 h (light : dark) photoperiod. The survival was assessed at four time points, from 24 to 96 hours after insecticide application. Whiteflies were considered dead if they showed no movement when probed using a fine brush. Each treatment was replicated five times. The 4% w/v concentration, which resulted in the highest adult mortality rate, was chosen for testing its insecticidal activity against immature stages. Using a similar leaf-dip method, tomato leaf discs were placed on agar in Petri dishes, and 50 adult whiteflies were released for oviposition. After 24 h, the adults were removed, and the number of eggs on each leaf disc was counted under a stereomicroscope. The leaf discs with eggs were dipped in different concentrations of the plant extracts, as described above. The development and mortality of immature stages (eggs, the first to the fourth instar nymphs) were monitored daily until adult emergence. Each treatment was replicated fivetimes.

Pathogenicity of *Beauveria bassiana*

The pathogenicity of *B. bassiana* against different life stages of *B. tabaci* was evaluated using the methods described by Cuthbertson and Audsley (2016) with modifications. For adults, tomato leaf discs were sprayed with conidial suspensions (1×10^6, 1×10^7, and 1×10^8 conidia $\cdot$ ml^{-1}) using a hand-held sprayer until runoff and allowed to air dry. Twenty adult whiteflies were released onto each treated leaf disc as described in the previous section. The control leaf discs were sprayed with a 0.05% Tween-80 solution. The records of deaths were collected from all insects over 7 days. Surface sterilization of dead insects occurred through exposure to a solution of 1% sodium hypochlorite for 30 s, while sterile distilled water was used for two rinses. The solidified fungi were placed on water agar plates to verify the infection by observing mycelial growth. Since the 2nd instar is the most affected in plant extract tests, the effects of *B. bassiana* on adults, the 2nd instar, and eggs were tested. For the immature stages, the procedure was similar to that described for plant extracts, with leaf discs containing eggs or specific nymphal instars sprayed with conidial suspensions. The development and mortality of each stage were monitored daily. Each treatment was replicated five times.

Combined effect of plant extracts and *Beauveria bassiana*

The effects of the plant extract and *B. bassiana* were evaluated based on the methodology described by Togbé *et al.* (2015). Leaf discs were first exposed to 1% w/v sublethal concentrations of plant extracts before drying. Next the leaf discs were sprayed with sublethal *B. bassiana* concentrations of 1×10^6 conidia $\cdot$ ml^{-1}. Three separate control conditions consisted of unaltered leaf discs and leaf discs treated with neither *B. bassiana* nor the plant extracts. The 20 adult whiteflies were placed on each leaf disc and the mortality was recorded at four time points from 24 h to 96 h after

treatment. The experiment included five repetitions for each group of test conditions. The evaluation of plant extracts and *B. bassiana* interaction types required the use of the co-toxicity factor (CTF) described by Mansour *et al.* (1966), using the following formula:

$$\text{CTF} = \frac{\text{Observed mortality} - \text{Expectet mortality}}{\text{Expectet mortality}} \times 100,$$

where the expected mortality was calculated using the formula:

$$\text{Expected mortality} = O_1 + O_2(100 - O_1)/100,$$

where: O_1 and O_2 are the observed mortalities for individual treatments. A CTF value > 20 indicates synergism, –20 to 20 indicates an additive effect, and < –20 indicates antagonism.

Greenhouse experiments

The greenhouse experiments were performed to confirm the effectiveness of the chosen treatments in semi-field environments. We used plastic pots (20 cm in diameter) to grow tomatoes at the 4–5 leaf stage using a mixture of soil sand and compost at a ratio of 2 : 1 : 1. The treatments were arranged in randomized complete block design with five replicates. Each plant was infested with 50 adult whiteflies (1–2 days old) by releasing them onto the third leaf from the top and covering the leaf with a clip cage for 24 h. The treatments included: (1) untreated control (water + 0.1% Tween-80), (2) the *C. annuum* extract (2% w/v), (3) the *A. sativum* extract (2% w/v), (4) the *D. stramonium* extract (2% w/v), (5) *B. bassiana* (1×10^8 conidia · ml^{-1}), and (6) combined treatment of the *D. stramonium* extract (1% w/v) + *B. bassiana* (1×10^6 conidia · ml^{-1}). The treatments were applied using a hand-held sprayer until runoff. The plants were maintained under a natural photoperiod in a greenhouse.

Adults, eggs, and nymphs of *B. tabaci* were recorded from three randomly selected leaves per plant before treatment and at 3, 7, and 14 days after treatment. The percentage reduction in population was calculated using the following formula:

Reduction (%) = [(Pre-treatment count – Post-treatment count) / Pre-treatment count] × 100.

Statistical analysis

Data analysis was performed using Statistical Package for the SPSS version 18.0. A correction method based on Abbott's formula was used to treat the control mortality figures in the data sets. The research utilized the Analysis of Variance (ANOVA) statistical method ($p < 0.05$) for group mean comparisons between treatments. Prior to data analysis, the percentage of mortality and population reduction values underwent arcsine transformation to normalize their variance. The results contained non-transformational mean values.

Results

Phytochemical analysis

Phytochemical screening revealed the presence of various bioactive compounds in plant extracts (Table 1). The *D. stramonium* extract contained alkaloids, flavonoids, phenols, glycosides, and steroids. The *A. sativum* extract was rich in organosulfur compounds, flavonoids, and saponins, while the *C. annuum* extract contained capsaicinoids, flavonoids, phenols, and terpenoids.

Insecticidal activity of plant extracts

All three plant extracts exhibited significant insecticidal activity against *B. tabaci* adults, with mortality increasing with the concentration and exposure time (Fig. 1). At the highest concentration tested (4% w/v), the *D. stramonium* extract achieved the highest mortality (92.4 ± 3.1%) at 96 h post-treatment, followed by *A. sativum* and *C. annuum*. The control treatment showed minimal mortality (5.2 ± 1.1%). The differences in the mortality rates among the three plant extracts were statistically significant. These results indicated that the *D. stramonium* extract had the highest insecticidal activity against adult *B. tabaci* among the tested plant extracts.

Figure 2 shows that the insecticidal activity of plant extracts against immature stages of *B. tabaci* varied according to developmental stage. All three plant extracts were more effective against the first and the second instar nymphs compared to eggs, third instar, and fourth instar nymphs. At 4% concentration, the mortality rates of the second instar nymphs were 97.8 ± 2.3%, 89.7 ± 3.1%, 82.5 ± 3.5% and 30.2 ±2.3% for the *D. stramonium*, *A. sativum*, *C. annuum* extracts, and the control treatments respectively, at 96 h post-treatment.

Pathogenicity of *Beauveria bassiana*

B. bassiana was highly pathogenic to all *B. tabaci* stages with mortality rate increasing with conidial concentration and exposure time (Fig. 3). The highest mortality rate was recorded in the second instar nymphs (93.6%) at 1×10^8 conidia · ml^{-1}, while eggs showed the lowest

Table 1. Phytochemical composition of *Capsicum annuum, Datura stramonium, and Allium sativum* extracts

Phytochemical group	Specific compounds	*C. annuum*	*D. stramonium*	*A. sativum*
Alkaloids	capsaicin	+++	–	–
	dihydrocapsaicin	++	–	–
	scopolamine	–	+++	–
	hyoscyamine	–	+++	–
	atropine	–	++	–
Organosulfur compounds	allicin	–	–	+++
	diallyl disulfide	–	–	+++
	diallyl trisulfide	–	–	++
Flavonoids	quercetin	++	+	+
	luteolin	++	+	–
	kaempferol	+	++	–
Phenolic compounds	chlorogenic acid	++	+	–
	caffeic acid	+	+	–
	ferulic acid	+	+	–
Terpenoids	limonene	++	–	–
	caryophyllene	+	–	–
Saponins		–	+	++
Tannins		+	+	–
Glycosides		–	++	+
Steroids		–	+	–

Note: +++ – high concentration; ++ – moderate concentration; + – low concentration; (–) – not detected. Concentrations were determined based on HPLC analysis compared to standard compounds

susceptibility. Mortality in the control treatment group remained very low for all developmental stages.

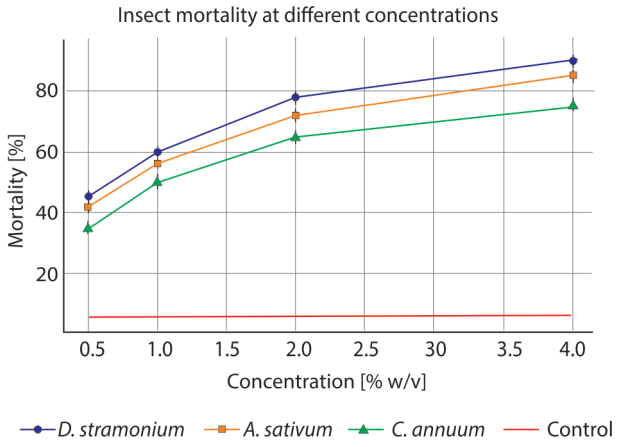

Fig. 1. Adult mortality of *Bemisia tabaci* following exposure to botanical extracts of *Datura stramonium*, *Allium sativum*, and *Capsicum annuum* at various concentrations (1%, 2%, and 4%) after 96 hours. Bars represent mean ± SD of three replicates. Different letters above bars indicate significant differences ($p < 0.05$) according to LSD test

Combined effect of plant extracts and *Beauveria bassiana*

The combination of plant extracts (1% w/v) and *B. bassiana* (1×10^6 conidia · ml^{-1}) significantly increased adult *B. tabaci* mortality compared to the individual treatments (Table 2). The most effective treatment was the combination of *D. stramonium* and *B. bassiana*, achieving 98.0% ± 2.1 mortality of insects observed at 96 h after treatment and indicating a strong synergistic effect.

The CTF test revealed that *B. bassiana* formed synergistic interactions with the plant extracts (Table 2). Results indicated that the CTF values were 25.64, 26.25, and 33.0 for the combinations of *D. stramonium* with *B. bassiana*, *A. sativum* with *B. bassiana* and *C. annuum* with *B. bassiana* respectively. The CTF values of 33.0 obtained from testing *C. annuum* + *B. bassiana* indicated synergism (CTF > 20) for all three combinations, with the strongest synergistic effect being observed for the *C. annuum* + *B. bassiana* combination. Multiple factors may explain the synergistic effects of plant extracts and *B. bassiana* in entomopathogenic bioassays.

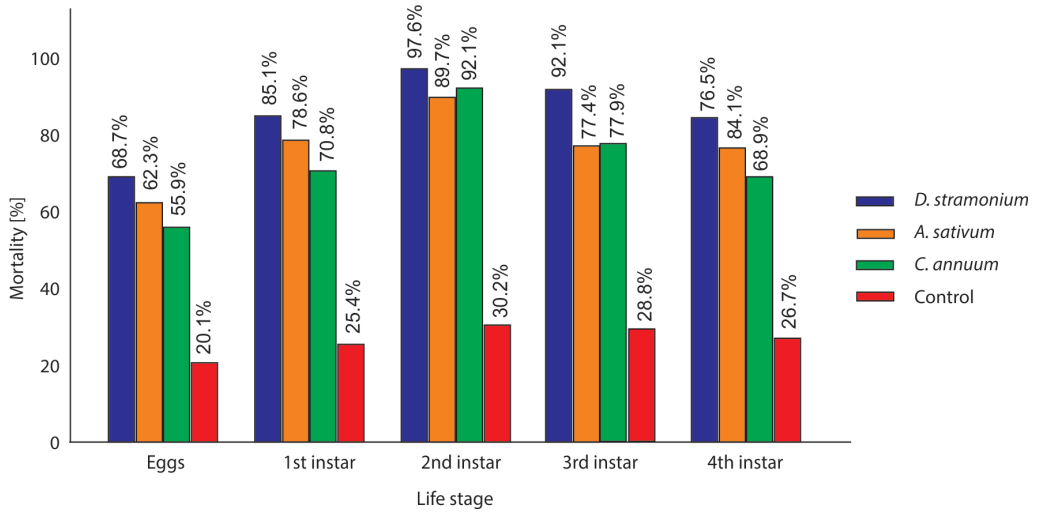

Fig. 2. Mortality of different *Bemisia tabaci* life stages treated with three botanical extracts (*Datura stramonium*, *Allium sativum*, and *Capsicum annuum*) at 4%concentrations. Observations were recorded at 96 hours post-treatment. Error bars indicate standard deviation ($n = 3$)

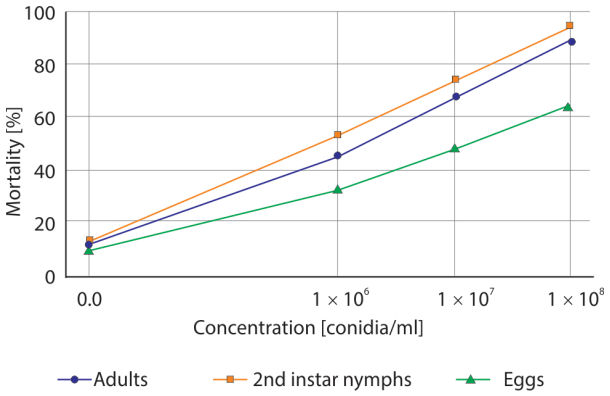

Fig. 3. Pathogenicity of *Beauveria bassiana* against different life stages of *Bemisia tabaci* (eggs, 2nd instar nymphs, and adults) at conidial concentrations of 1×10^6, 1×10^7, and 1×10^8 conidia · ml^{-1}. Mortality rates were recorded after 7 days post-treatment. Data represent mean ± SD. Different letters represent statistically significant differences ($p < 0.05$)

Greenhouse experiments

Greenhouse experiments showed that the strategies were effective under semi-field conditions. Mortality rates were recorded at 3, 7 and 14 days after treatment. In all the tests, the highest mortality for all life stages of *B. tabaci* was observed when the *D. stramonium* extract (1% w/v) was used with *B. bassiana* (1×10^6 conidia · ml^{-1}). The mortality rates of adults, nymphs and eggs after 14 days of treatment were 88.0 ± 4.2%, 91.0 ± 4.5%, and 95.0 ± 4.7%, respectively. These mortality rates were higher than those achieved by the two treatments applied individually (Figs. 4, 5, and 6).

Table 2. Co-toxicity factors for combined treatments of plant extracts and *Beauveria bassiana* against adults *Bemisia tabaci* at 96 hours post-treatment

Combined treatment	Observed mortality [%]	Expected mortality [%]	Co- toxicity factor	Interaction type
Datura stramonium(1%)+B. bassiana [1 × 10⁶ conidia · ml⁻¹]	98.0 ± 2.1	78.0	25.64	synergistic
Allium sativum(1%)+B. bassiana [1 × 10⁶ conidia · ml⁻¹]	95.0 ± 2.7	75.25	26.25	synergistic
Capsicum annuum(1%)+B. bassiana [1 × 10⁶ conidia · ml⁻¹]	93.5 ± 3.1	70.3	33.0	synergistic
Datura stramonium [1%]	60.8 ± 2.3	–	–	–
Allium sativum [1%]	55.3 ± 2.9	–	–	–
Capsicum annuum [1%]	46.5 ± 3.4	–	–	–
Beauveria bassiana [1 × 10⁶ conidia · ml⁻¹]	45.0	–	–	–
Control	12.0	–	–	–

Note: Values for observed mortality represent mean ± standard error ($n = 5$). Expected mortality was calculated using the formula: $E = O_1 + O_2(100 - O_1)/100$, where: O_1 and O_2 are the observed mortalities for individual treatments. Co-toxicity factor = [(Observed mortality – Expected mortality) / Expected mortality] × 100. Co-toxicity factor > 20 indicates synergism, –20 to 20 indicates additive effect, and < –20, intigates antagonism

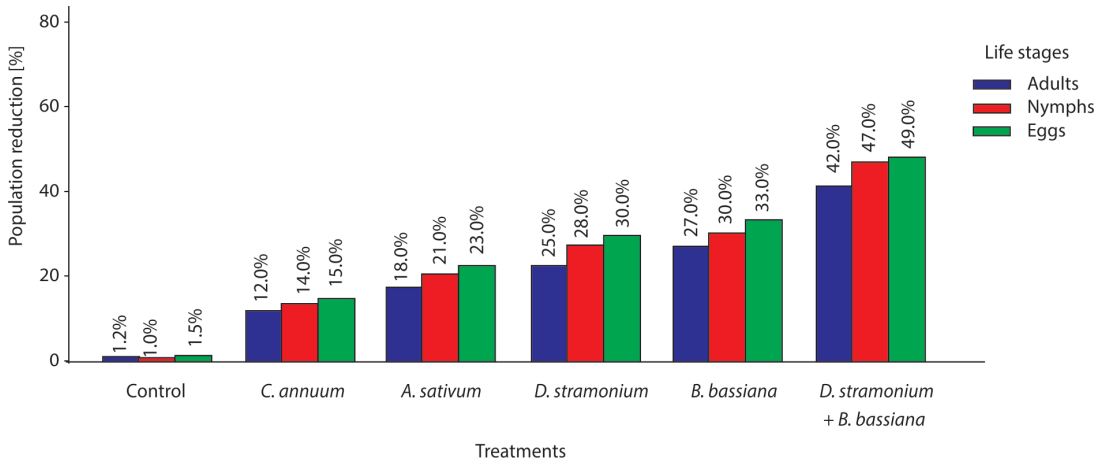

Fig. 4. Percentage reduction in egg, nymph, and adult populations of *Bemisia tabaci* in greenhouse trials at 3 days after treatment. Treatments included *Datura stramonium*, *Allium sativum*, *Capsicum annuum* extracts (4%), *Beauveria bassiana* (1×10^8 conidia · ml^{-1}), and their combinations. Means with different letters indicate significant differences ($p < 0.05$) using LSD test

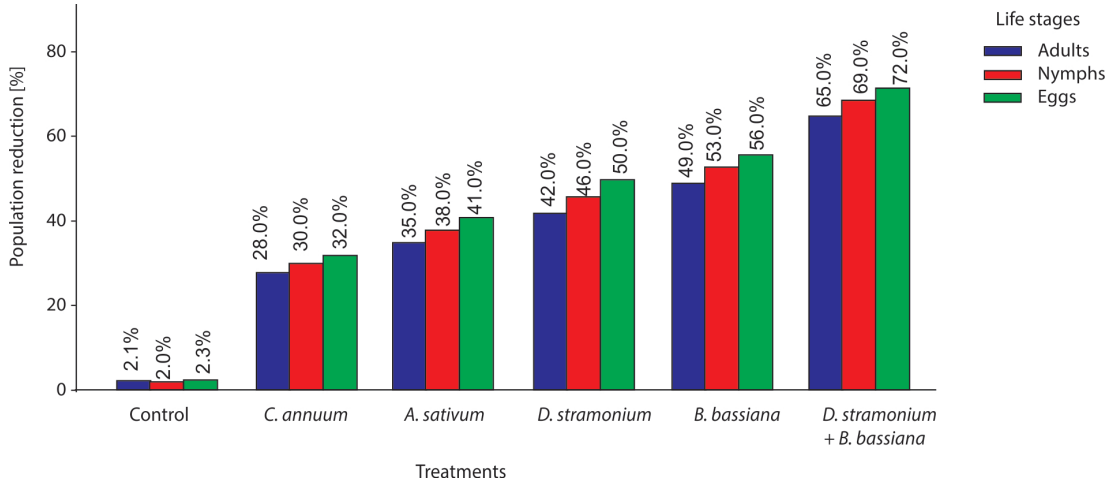

Fig. 5. Percentage reduction in egg, nymph, and adult populations of *Bemisia tabaci* in greenhouse trials after 7 days after treatment. Treatments included *Datura stramonium*, *Allium sativum*, *Capsicum annuum* extracts (4%), *Beauveria bassiana* (1×10^8 conidia · ml^{-1}), and their combinations. Means with different letters indicate significant differences ($p < 0.05$) using LSD test

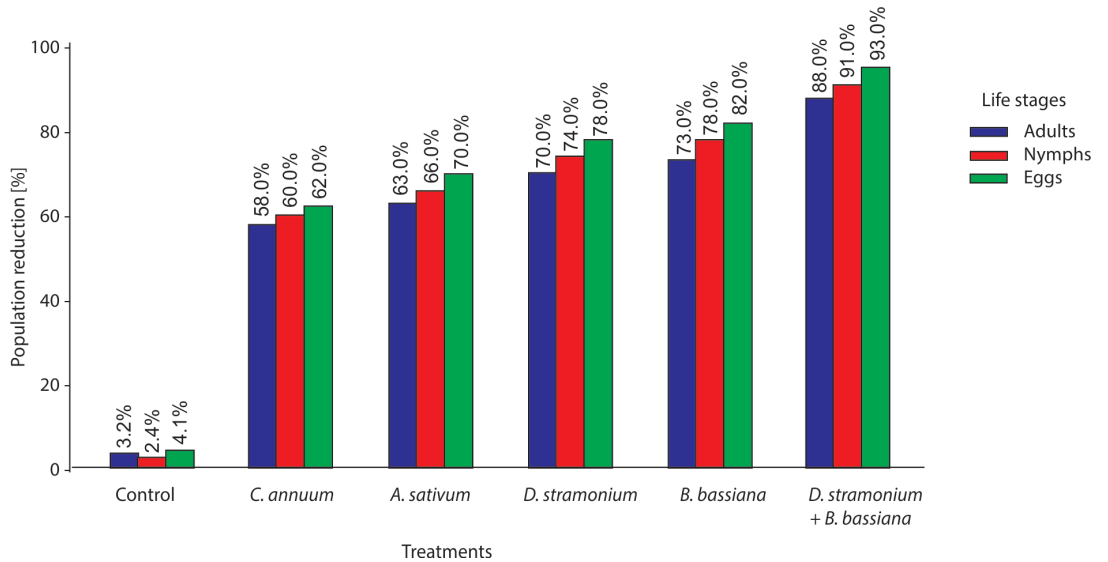

Fig. 6. Percentage reduction in egg, nymph, and adult populations of *Bemisia tabaci* in greenhouse trials at 14 days after treatment. Treatments included *Datura stramonium*, *Allium sativum*, *Capsicum annuum* extracts (4%), *Beauveria bassiana* (1×10^8 conidia · ml^{-1}), and their combinations. Means with different letters indicate significant differences ($p < 0.05$) using LSD test

Discussion

The phytochemical screening revealed variation in active compounds of the plant extracts such as alkaloids, flavonoids, and organosulfur compounds, and these indicate their effectiveness in pest control. These results agree with earlier observations of the phytochemical constituents of these plants (Mnayer *et al.* 2014; Kumari *et al.* 2017**;** Sánchez-Quezada *et al.* 2024).

The differences in mortality rates among the three plant extracts were statistically significant. These results indicate that the *D. stramonium* extract had the highest insecticidal activity against adult *B. tabaci* among the tested plant extracts. The higher susceptibility of immature instar nymphs could be attributed to their thinner cuticle and less developed detoxification systems compared to later instars (Wakil *et al.* 2023). The high insecticidal activity of the *D. stramonium* extract can be attributed to its richness of secondary metabolites which are known for their effects on insects (Kumari *et al.* 2017). Similarly, the insecticidal property of the *A. sativum* extract is probably attributed to its organosulfur compounds, which may interfere with insect cellular mechanisms in a number of ways, such as enzyme inhibition and membrane disruption (Mnayer *et al.* 2014). The insecticidal effects of the *C. annuum* extract are considered moderate because it disrupts sensory receptors and metabolic activities in insects (Sánchez-Quezada *et al.* 2024).

Previous studies have verified the insecticidal properties of plant extracts on various insect species. The *D. stramonium* leaf extract compromised the survival of *Aphis gossypii* at a concentration of 3–4% resulting in 85–90% mortality rate (Sabzevari and Hofman 2022). The mortality rates observed by Kayahan (2023), in the control of green peach aphid (*Myzus persicae*) with *A. sativum* plant extract varied between 1.5–2.2% according to LC_{50} testing. Another study reported that *C. annuum* plant extract caused 65–75% mortality in *Frankliniella occidentalis* at 3–5% concentration (Ristaino *et al.* 2021).

Microscopic examination of dead whiteflies confirmed *B. bassiana* infection, with white mycelial growth emerging from cadavers within 2–3 days after death. The fungus typically emerges from intersegmental regions, particularly between the head and thorax, and between the abdominal segments. This pattern of emergence is consistent with previous reports on *B. bassiana* infection in various insect hosts (Jaronski and Mascarin 2017).

The high efficiency of *B. bassiana* against *B. tabaci* observed in this study agrees with the results of earlier studies. According to Cuthbertson and Audsley (2016), *B. bassiana* led to 80–90% mortality of *B. tabaci* adults at the concentration of 1×10^8 conidia $\cdot$ ml^{-1}. Wang *et al.* (2024) demonstrated that different *B. bassiana* isolates result in substantial mortality rates among *B. tabaci* nymphs. Also Dara (2019) reported that the second instar nymphs were more sensitive to *B. bassiana* since the cuticles of these nymphs are thinner than those of older nymphs. The second instar nymphs have reduced mobility due to their lack of protective wax cover, and they remain less mobile than adult stages thus increasing their susceptibility to fungal infection (Jaronski and Mascarin, 2017). The protective nature of the chorion around the eggs creates a fungal penetration barrier, resulting in reduced susceptibility of the eggs (Wang *et al.* 2024).

Multiple factors can explain the synergistic effects of the plant extracts and *B. bassiana* in entomopathogenic bioassays. Plant extracts ensure that insects are less capable of protecting themselves against fungi by damaging their immune systems (Togbé *et al.* 2015). Plant extract compounds have been found to boost the fungal germination rate and enhance both the growth and penetration ability of fungi into the insect cuticles (Norris and Bloomquist 2021). The alkaloids present in *D. stramonium* influence the activity of the nervous system of insects, resulting in their failure to groom their bodies; hence fungal conidia cannot be removed (Kumari *et al.* 2017). Organosulfur compounds present in *A. sativum* affect insect detoxification enzymes, making fungal metabolites more toxic to the target organism (Mnayer *et al.* 2014). Previous studies have documented the combined beneficial effects of botanical insecticides created using entomopathogenic fungi. Bamisile *et al.* (2021) established broader synergistic effects between neem oil and *B. bassiana* to kill *B. tabaci*, based on recorded CTF values between 25 and 35. Greater mortality of *A. gossypii* occurred when Togbé *et al.* (2015) mixed plant essential oils with *B. bassiana* which yielded synergistic effects demonstrated by CTF values.

Many researchers support the conclusion that combining these treatments worked better than using them independently, as demonstrated through greenhouse assessments (Togbé *et al.* 2015). Field conditions may benefit from these treatments because their multiple methods of action include direct killing effects while also acting as repellents, deterring oviposition, and strengthening plant defense systems (Norris and Bloomquist 2021).

These results provide evidence for new sustainable management methods for *B. tabaci*. Combining plant extracts and *B. bassiana* has shown to

be very effective for controlling this main pest (Bamisile *et al*. 2021). Also, since there are several approaches these treatments take, they often lower the risk of developing resistance which is a big problem for routine chemical insecticides (Wakil *et al*. 2023).

Conclusions

Laboratory and greenhouse tests indicated that the highest rate of *B. tabaci* death and the greatest decrease in all life stages occurred when *D. stramonium* 1% (w/v) and *B. bassiana* (1×10^6 conidia · ml^{-1}) were combined. The study found that mixing plant extracts and biocontrol fungi provides the same benefits as using synthetic pesticides. Further study is required to support sustainable management of whiteflies in diverse agricultural systems by integrating IPM methods.

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