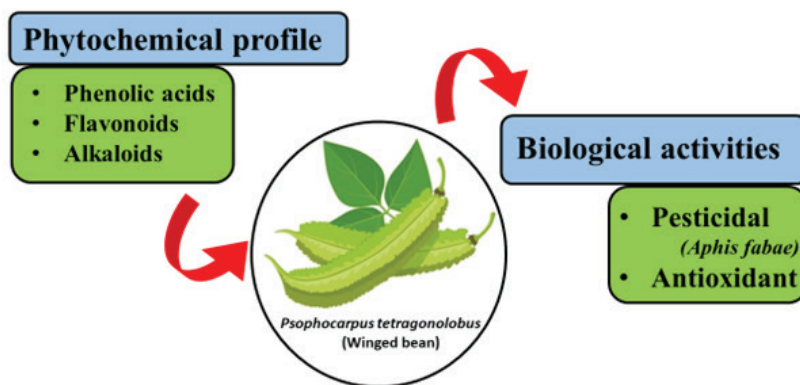


RESEARCH ARTICLE

Pesticidal and Antioxidant Activity of Solvent Extracts of Pods and Leaves of *Psophocarpus tetragonolobus* (Winged Bean)

M.W.M. P. Nawarathne, J.M.S. Jayasinghe*, W.A. Priyanka P. De Silva

**Highlights**

- *Psophocarpus tetragonolobus* pod/leaf extracts tested for pesticidal and antioxidant activity
- Water: acetone extracts of pods and leaves showed the highest pesticidal and antioxidant activities
- Highest pesticidal and antioxidant activities due to the higher amounts of phenolic compounds
- *P. tetragonolobus* extracts can be used as an eco-friendly natural pesticide and antioxidant

RESEARCH ARTICLE

Pesticidal and Antioxidant Activity of Solvent Extracts of Pods and Leaves of *Psophocarpus tetragonolobus* (Winged Bean)

M.W.M. P. Nawarathne^{1,2}, J.M.S. Jayasinghe^{2*}, W.A. Priyanka P. De Silva³

¹Postgraduate Institute of Science, University of Peradeniya, Peradeniya 20400, Sri Lanka

²Department of Chemistry, University of Peradeniya, Peradeniya 20400, Sri Lanka

³Department of Zoology, University of Peradeniya, Peradeniya 20400, Sri Lanka

Received: 20.02.2025; Accepted: 25.08.2025

Abstract: The demand for natural pesticides and antioxidants is rising due to the adverse effects of synthetic chemicals on human health and the environment. *Psophocarpus tetragonolobus* (winged bean), a tropical legume, is recognized as a rich source of nutrients and bioactive compounds. This study evaluated the pesticidal and antioxidant potential of hot water, cold water, and water: acetone (1:1) crude extracts of pods and leaves of *P. tetragonolobus*. The pesticidal activity was tested against *Aphis fabae* (black aphids), while antioxidant activity was assessed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay, total polyphenolic content (TPC), total flavonoid content (TFC), and total antioxidant capacity (TAC). Phytochemical screening was also conducted to identify the compounds present. Among the extracts, water: acetone extracts exhibited the highest pesticidal activity, causing 100% mortality of *A. fabae* at 10,000 mg L⁻¹ for both pods and leaves. Toxicity was concentration- and time-dependent, with LD₅₀ values of 202.50 mg L⁻¹ at 12 hours and 162.26 mg L⁻¹ at 24 hours for leaves. Water: acetone extracts also showed the strongest antioxidant activity, with IC₅₀ values of 356.76 mg L⁻¹ and 395.90 mg L⁻¹, TPC of 241.50 mg(GAE)/g and 159.83 mg(GAE)/g, TFC of 125.60 mg(QUE)/g and 100.60 mg(QUE)/g, and TAC of 245.63 mg(AAE)/g and 212.74 mg(AAE)/g for leaves and pods, respectively. These findings highlight the potent pesticidal and antioxidant properties of water: acetone extracts of *P. tetragonolobus*, suggesting their potential as eco-friendly alternatives for pest management and natural antioxidant sources.

Keywords: Antioxidant activity; *Aphis fabae*; DPPH assay; Pesticidal activity; *Psophocarpus tetragonolobus*

INTRODUCTION

Crops are essential sources of stable food and play a critical role in providing adequate nutrition worldwide, especially in developing countries. Protecting crops from biotic stresses, including weeds, pathogens, and insect pests, is a key aspect of sustainable agriculture (Dedryver et al., 2010). Among these, insect pests pose a major challenge due to their high adaptability and close synchronization of their life cycles with host plant growth, which enhances their survival and reproductive success. These pests employ various strategies to withstand diverse environmental conditions, allowing them to thrive across different habitats (Valenzuela & Hoffmann, 2015).

Aphids are among the most abundant and destructive insect pests affecting vegetables, ornamentals, and greenhouse crops. They cause significant damage by feeding on phloem sap and act as vectors for several plant-debilitating viruses, including necrotic yellow virus, mosaic yellow virus, and leaf roll virus (Tewary et al., 2005). In addition to direct feeding and virus transmission, aphid infestation can reduce photosynthetic activity through the phytotoxic effects of their saliva and the growth of sooty molds on honeydew excreted by the insects (Dedryver et al., 2010). Considering the economic importance of crops and the multifaceted damage caused by aphids, effective management strategies are essential to safeguard crop health and maintain productivity in agriculture.

Chemical pesticides, including carbamates, pyrethroids, organochlorines, and organophosphates, are still widely used worldwide for effective pest control (Damalas & Eleftherohorinos, 2011; Zikankuba et al., 2019). However, the extensive use of synthetic chemicals has caused numerous adverse effects, such as toxicity to non-target organisms, environmental pollution from pesticide residues, bioaccumulation, acute or chronic toxicity to humans, and the development of pest resistance (Ebadollahi et al., 2020). As a result, plant-based insecticides have gained considerable attention as safer alternatives. Rich in bioactive compounds such as phytochemicals and essential oils, these natural products can effectively target pests and pathogens while minimizing harm to non-target organisms and the environment (Pichersky & Gershenzon, 2002; Bakkali et al., 2008; Ebadollahi et al., 2020; Basaid et al., 2020).

Plant-based antioxidants are important bioactive compounds that support human health and help manage reactive oxygen species, unlike some synthetic antioxidants which may pose health risks (Yea et al., 2014; Cadet & Davies, 2017). In addition to their health benefits, these natural antioxidants can protect plants from pests through multiple mechanisms. Compounds such as polyphenols, flavonoids, and tannins can be toxic to pests, disrupt their biological processes, act as natural repellents, inhibit pest growth and reproduction, and provide antifungal and

*Corresponding Author's Email: susanthij@sci.pdn.ac.lk



antibacterial effects. Plants like neem, tobacco, and garlic are widely used for their pesticidal properties, and ongoing research continues to explore how plant antioxidants and other bioactive compounds can be harnessed for sustainable pest management.

Psophocarpus tetragonolobus, commonly known as the winged bean, is a tropical legume in the family Fabaceae with edible leaves, flowers, pods, seeds, and tubers. It is a perennial herb rich in proteins, oils, vitamins, and carbohydrates (Calvindi et al., 2020). The plant also contains bioactive phytochemicals, including phenolics, flavonoids, and tannins, which contribute to its antioxidant, anti-inflammatory, antinociceptive, antibacterial, antifungal, antiproliferative, and cytotoxic activities (Bassal et al., 2020; Ruma, 2016). Interestingly, winged beans experience fewer pest attacks, such as from aphids, compared to other legumes. Previous studies have reported its insecticidal (Gupta et al., 2008; Kaur & Sohal, 2018) and antioxidant properties (Bassal et al., 2021; Hlosrichok & Aunpad, 2023).

The effects of bioactive compounds differ among aphid populations, influenced by their exposure to various bioactive substances. Notably, there is a scarcity of reports regarding the insecticidal efficacy of winged bean against black aphids, as well as the antioxidant properties of winged

bean extracts using water-acetone solvent systems. This study investigated the pesticidal and antioxidant activities of three distinct solvent extracts (hot water, cold water, and water: acetone) derived from winged bean pods and leaves in Sri Lanka.

MATERIALS AND METHODS

Preparation of plant extracts

Healthy and fresh winged bean leaves and pods were collected from Kandy District, Sri Lanka. Plant materials were washed thoroughly with running tap water, followed by distilled water. An equal amount (100 g) of plant material was weighed, cut into small pieces, and ground into a fine paste using a blender. Three solvent systems: hot water, cold water, and water: acetone (1:1) were used for the extraction of active compounds. Hot water extracts were prepared using refluxing for 3 hours. Cold water and water: acetone (1:1) extracts were prepared by maceration using a bottle shaker (24 hours x 3). Subsequently, extracts were filtered through no 1 Whatman filter papers. Following the filtration, samples were concentrated using a rotary vacuum evaporator to obtain crude plant extracts. Trace amounts of water were removed by freeze drying and were stored below 0°C until further use.

Phytochemical screening tests

Table 1: Preliminary qualitative phytochemical screening tests, methodologies, and expected observations

Phytochemical and the test	Methodology	Observations (based on published literature)
Alkaloids (Iodine test)	A few drops of diluted iodine solution were added to the 3 mL test solution.	Blue color appeared, disappeared when boiling, and reappeared when cooling (Nain et al., 2013)
Carbohydrates (Benedict's test)	2 mL of plant extract was mixed with a few drops of Benedict's reagent and boiled in a water bath	Formation of reddish-brown precipitate (Bhandary et al., 2012)
Coumarins	3 mL of 10% NaOH was added to 2 mL of the plant extract	Formation of yellow color (Savithramma et al., 2011)
Fatty acids	2 mL of extract was mixed with 5 mL of ether. This extract allowed for evaporation on filter paper, and then dried the filter paper	The appearance of transparency on filter paper (Savithramma et al., 2011)
Flavonoids (Alkaline test)	3 mL of each plant extract was treated with ammonia solution	Increase in the intensity of yellow color, which was colorless on the addition of a few drops of H ₂ SO ₄ (Nain et al., 2013)
Flavonoids (Lead acetate test)	3 mL of each plant extract was treated with a few drops of 10% lead acetate	Formation of yellow precipitate (Nain et al., 2013)
Glycosides (Keller-Killani test)	5 mL of each plant extract was treated with 2 mL of glacial acetic acid. One drop of 10% FeCl ₃ and 1 mL of concentrated H ₂ SO ₄ was added. Formation of two layers was observed	Lower reddish-brown layer and upper acetic acid layer, which turns bluish green (Doss, 2009)
Phenolic acids	1 mL of 10% FeCl ₃ solution was added to 2 mL of plant extract	Formation of green to blackish deposits (Nain et al., 2013)
Proteins (Biuret test)	2 mL of plant extract was treated with 10% sodium hydroxide solution and two drops of 0.1% Copper Sulphate solution	Formation of violet/pink color (Bhandary et al., 2012)

Phytochemical and the test	Methodology	Observations (based on published literature)
Saponin (Foam test)	5 mL of each plant extract was mixed with water and shaken	Formation of foams, which is stable for 10-15 minutes (Bhandary et al., 2012)
Steroids	1 mL of the extract was dissolved in 5 ml of chloroform and concentrated H ₂ SO ₄	The upper layer turns red, and the H ₂ SO ₄ layer shows yellow with green fluorescence (Savithramma et al., 2011)
Tannins	3 mL of extract was added to a few drops of 1% lead acetate	Formation of a yellowish precipitate (Savithramma et al., 2011)
Terpenoids (Salkowski test)	3 mL of each plant extract was mixed with 2 mL of chloroform, and concentrated H ₂ SO ₄ was added slowly to form a layer	Reddish brown coloration of the inner surface (Doss, 2009)

Pesticidal bioassays

The 10,000 mg L⁻¹ stock solution for each plant extract was prepared with distilled water, and a dilution series of 5000, 2000, 1000, 500, 250, and 100 mg L⁻¹ was prepared. This step was followed for plant crudes obtained from hot water, cold water, and acetone: water. Bioassay experiments were conducted using mature black aphids (*Aphis fabae*) collected from agricultural fields in Kurunegala District. A cabbage leaf with 25 mature black aphids was transferred into the treatment chamber (50 mL beaker). Then, 0.5 mL of each test solution (100, 250, 500, 1000, 2000, 5000, and 10,000 mg L⁻¹) was sprayed on aphids using a bottle spray. The aphids were exposed to the treatment for a duration of one hour. Treated aphids were transferred to a new beaker, and mortality was observed after 3 hours, 6 hours, 12 hours, and 24 hours. Three replicates per concentration and per solvent extract were carried out. Control experiments were done using distilled water alone. Observations were noted, and percentage mortalities were calculated using the formula given below. LD₅₀ values were calculated for each solvent extract using Origin software.

$$\% \text{ mortality} = \frac{\text{number of dead aphids}}{\text{total number of aphid exposed}} \times 100$$

Determination of antioxidant activity

DPPH radical scavenging assay

The antioxidant activity was determined using the DPPH radical scavenging assay (2,2-diphenyl-1-picrylhydrazyl), as described by Garcia et al. (2012) with slight modifications. A concentration series of 3 ml was prepared from the extract, and initial absorbance values were measured at 517 nm using a UV-Vis spectrophotometer. A 1.5×10^{-3} mol dm⁻³ DPPH methanolic stock solution was prepared by dissolving (0.0059±0.0001) g DPPH in 50 mL of reagent-grade methanol. For the negative control, 1.20 mL of DPPH stock solution was mixed with 3 mL of methanol, and the absorbance was measured at 517 nm. Next, 1.2 mL of DPPH stock solution was added to each test solution, and the mixture was allowed to react in the dark at room temperature for 30 minutes. The absorbance of each test sample was then measured at 517 nm. The percentage inhibition was calculated using the formula provided below, and the IC₅₀ values were determined (Garcia et al., 2012). This procedure was repeated for all crude extracts.

% inhibition of DPPH

$$= \frac{(\text{A negative control} - \text{A sample} - \text{A blank})}{\text{A negative control}} \times 100$$

Total Antioxidant Capacity (TAC)

The total antioxidant capacity was determined using the phosphomolybdate method with some modifications (Prieto et al., 1999). The 0.4 mL of plant extract was treated with 4 mL of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate, and 4 mM ammonium molybdate). Test tubes were capped and incubated in a water bath at 95°C for 90 minutes and cooled to room temperature. The absorbance of the solutions was measured at 695 nm against the blank. The blank solution contained an equal amount of reagent solution and distilled water and was incubated under the same conditions as the rest of the samples. The TAC was calculated from the ascorbic acid calibration curve and expressed as ascorbic acid equivalent.

Total Phenolic Content (TPC)

The total phenolic content of pods and leaves of *P. tetragonolobus* was determined by the Folin-Ciocalteu phenol reagent method with some modifications (Kim et al., 2003). 1 mL of plant extract was mixed with 1 mL of 10% Folin-Ciocalteu phenol reagent. After 5 minutes, 5 mL of 7% Na₂CO₃ solution was added to the mixture, and 5 mL of distilled water was added. After mixing the solutions thoroughly, the solutions were kept in the dark conditions for 90 minutes at room temperature. The absorbance was read at 750 nm. Total phenolic content of pods and leaves of *P. tetragonolobus* was expressed as mg gallic acid equivalents. This procedure was repeated for all crude extracts with three replicates.

Total Flavonoids Content (TFC)

The total flavonoid content of pods and leaves of *P. tetragonolobus* was determined using the method described by Park et al. (2008). 1 mL of plant extract was treated with 3.4 mL of 30% methanol, 0.15 mL of 0.5M NaNO₂, and 0.15 mL of 0.3M AlCl₃. After 5 minutes, 1 mL of 1M NaOH was added, and absorbance was read at 506 nm. Total flavonoid was determined using the quercetin calibration curve and expressed as quercetin equivalents. The procedure was repeated three times for each plant extract (Park, et al., 2008).

RESULTS

Preliminary phytochemical screening of *Psophocarpus tetragonolobus* pods and leaves revealed the presence of several secondary metabolites. The extracts contained terpenoids, glycosides, alkaloids, flavonoids, phenols, saponins, coumarins, tannins, carbohydrates, and proteins (Table 2). Steroids and fatty acids were absent in all extracts. Glycosides were detected only in cold water and water: acetone extracts, while they were absent in hot water extracts. Conversely, alkaloids were present only in the hot water extract and absent in the cold water and water: acetone extracts.

The study's findings highlighted the pesticidal properties of the pods and leaves of *P. tetragonolobus* against *Aphis fabae*. Percent mortalities of exposed aphids were recorded at six and twelve-hour intervals, continuing until the 24-hour mark. Figure 1 presents the mortality rates of aphids after 3, 6, 12, and 24 hours of exposure. After three-hour exposure, the most significant mortality rates were observed with water-acetone extracts, achieving 70% for *P. tetragonolobus* pods and 75% for leaves at the highest concentration of 10,000 mg L⁻¹. In contrast, both cold and hot water extracts did not demonstrate significant mortality after three hours of exposure.

Results showed that 6 hours post-exposure, water: acetone extracts of *P. tetragonolobus* exhibited the highest pesticidal activity, causing 80% and 90% mortality in pods and leaves, respectively. Cold water extracts induced 45% and 55% mortality, while hot water extracts caused 45% and 50% mortality at 10,000 mg L⁻¹. Overall, leaf extracts demonstrated higher pesticidal activity than pods. Mortality rates increased with longer exposure times, and complete aphid mortality was observed with water: acetone extracts of both pods and leaves after 12 hours. At the same point, cold water extracts caused 70% mortality in pods and 80% in leaves, and similar rates observed for hot water extracts. The study found that aphid mortality was highest at 10,000 mg L⁻¹, with mortality increasing over time post-exposure. Among the extracts tested, water: acetone

showed the greatest efficacy, followed by cold and hot water extracts. After 24 hours, cold and hot water extracts of winged bean pods caused 80% aphid mortality, while leaf extracts achieved 90% mortality. The control treatment with distilled water showed no mortality.

Table 3 shows the LD₅₀ values obtained from bioassay experiments. The water: acetone solvent extracts exhibited the lowest LD₅₀ values for both the pods and leaves of *P. tetragonolobus*, in comparison to cold and hot water extracts. Specifically, the water: acetone extract of winged bean pods recorded LD₅₀ values of 484.39 mg L⁻¹ and 202.82 mg L⁻¹, while the leaves showed LD₅₀ of 220.50 mg L⁻¹ and 162.26 mg L⁻¹ after 12 and 24 hours, respectively. Notably, the leaf extracts reported the lowest LD₅₀ values across all three solvent systems, indicating a relatively higher pesticidal activity compared to pod extracts.

Figure 2 shows the IC₅₀ values obtained from the DPPH assay. Among the six extracts of *P. tetragonolobus* tested, the water: acetone extract exhibited the strongest antioxidant activity, with IC₅₀ values of 356.76 mg L⁻¹ for leaves and 395.90 mg L⁻¹ for pods. Cold water extracts showed IC₅₀ values of 423.91 mg L⁻¹ (leaves) and 441.54 mg L⁻¹ (pods), while hot water extracts had IC₅₀ values of 474.70 mg L⁻¹ (leaves) and 496.43 mg L⁻¹ (pods). Overall, the leaves demonstrated higher antioxidant activity compared to the pods.

Figure 3 shows the total flavonoid content (TFC), total phenolic content (TPC), and total antioxidant capacity (TAC) of *P. tetragonolobus* pods and leaves. The highest TFC was observed in water: acetone extracts, with 100.6 mg (QUE)/g for pods and 125.6 mg (QUE)/g for leaves. Cold water extracts contained 61.6 mg (QUE)/g (pods) and 69.6 mg (QUE)/g (leaves), while hot water extracts showed slightly higher values of 73.6 mg (QUE)/g (pods) and 79.6 mg (QUE)/g (leaves). The highest TPC was also recorded in water: acetone extracts, at 159.83 mg (GAE)/g for pods and 241.50 mg (GAE)/g for leaves, followed by cold water (74.28 and 155.94 mg (GAE)/g) and hot water extracts (66.50 and 150.39 mg (GAE)/g). Phenolic

Table 2: Qualitative analysis of phytochemicals of pods and leaves of *Psophocarpus tetragonolobus*

Test	Hot water		Cold water		Water: acetone (1:1)	
	Pods	Leaves	Pods	Leaves	Pods	Leaves
Alkaloids	+	+	-	-	-	-
Carbohydrates	+	+	+	+	+	+
Coumarins	+	+	+	+	+	+
Fatty acids	-	-	-	-	-	-
Flavonoids	+	+	+	+	+	+
Glycosides	-	-	+	+	+	+
Phenolic acid	+	+	+	+	+	+
Proteins	+	+	+	+	+	+
Saponins	+	+	+	+	+	+
Steroids	-	-	-	-	-	-
Tannin	+	+	+	+	+	+
Terpenoids	+	+	+	+	+	+

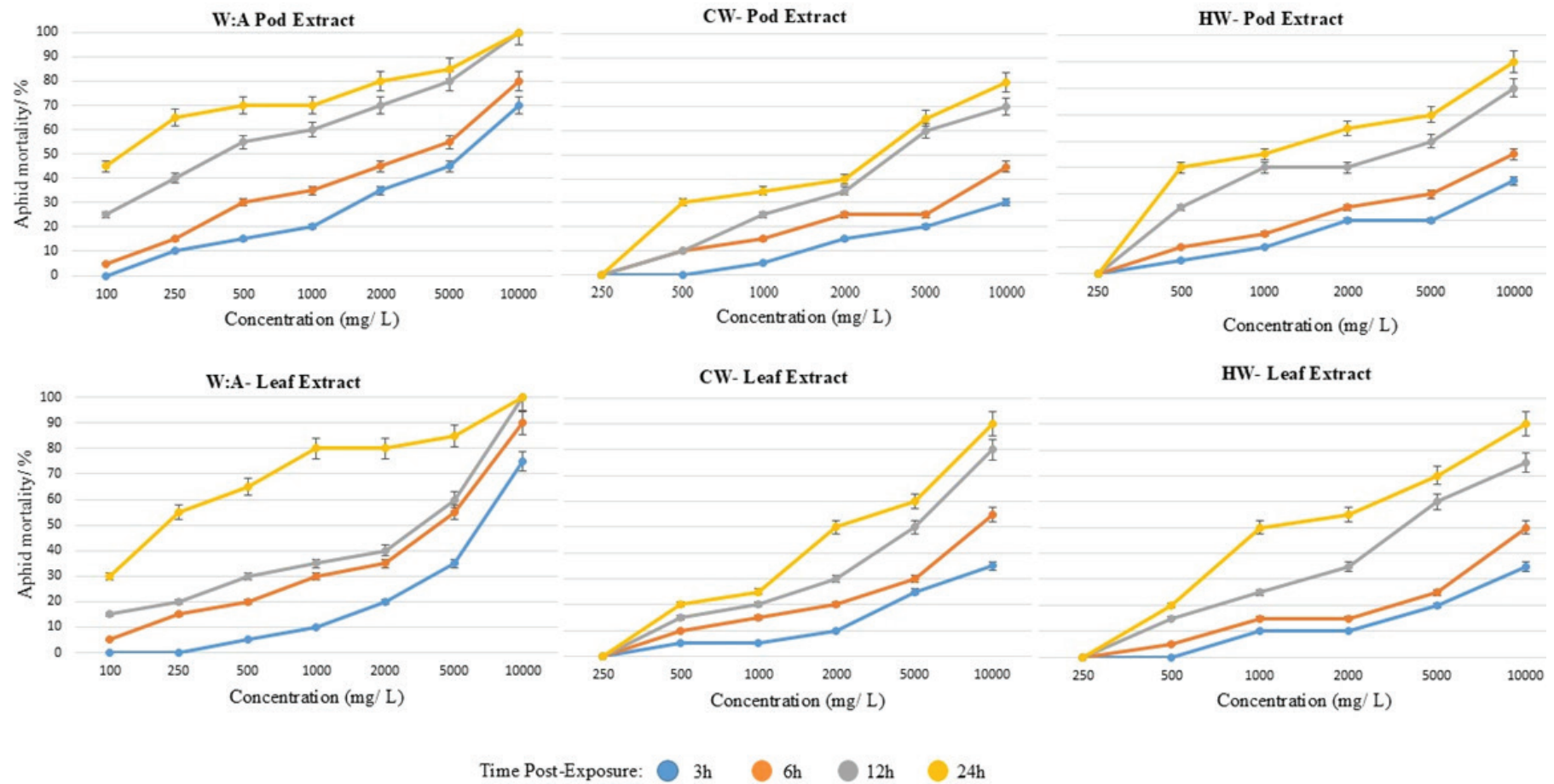
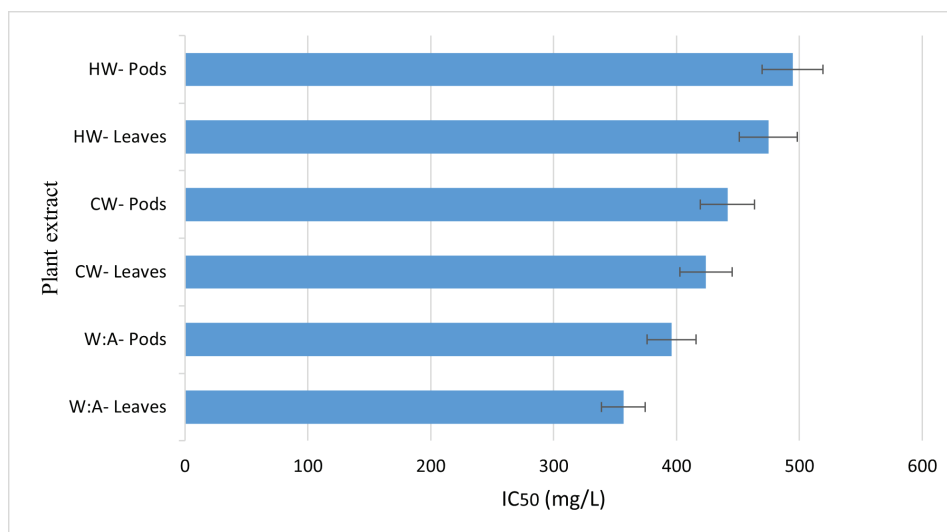
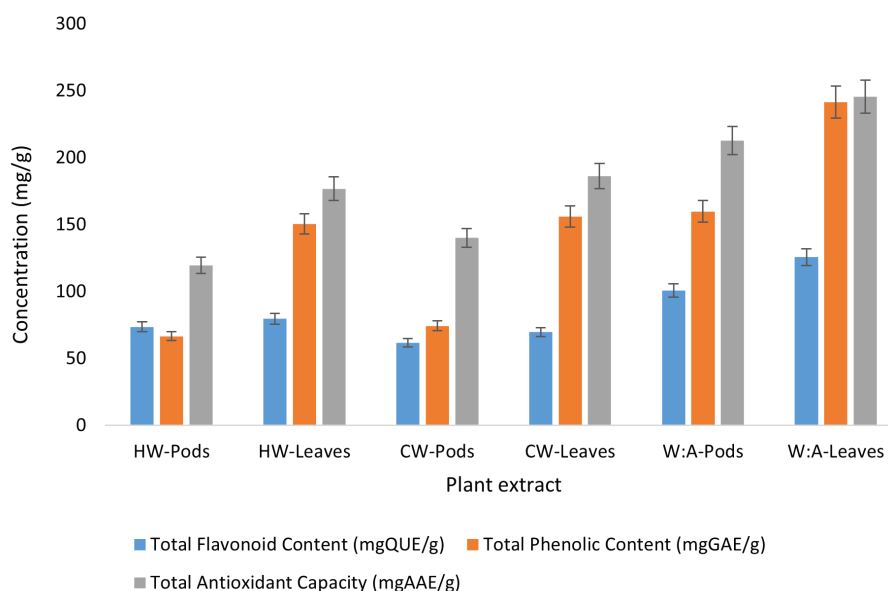


Figure 1: Percentage mortalities of black aphids obtained from bioassay experiments conducted for water: acetone (W: A), cold water (CW), and hot water (HW) extracts of pods and leaves of *Psophocarpus tetragonolobus* after 3-, 6-, 12-, and 24-hour post-exposure period

Table 3: LD₅₀ values, correlation coefficient (r), and p values for water: acetone, cold water, and hot water extracts of *Psophocarpus tetragonolobus* pods and leaves (LD₅₀ - Lethal Dosage required to kill 50% of the population).

Solvent system	Plant part	LD ₅₀ / mg L ⁻¹				Correlation coefficient	P value (p<0.05)
		3hrs post-exposure	6hrs post-exposure	12hrs post-exposure	24hrs post-exposure		
Water: acetone (1:1)	Pods	5652.66	2344.22	484.39	202.82	0.9391	0.0179
	Leaves	4495.57	2329.81	220.50	162.26	0.9253	0.0242
Cold water	Pods	>10000	>10000	3563.53	2186.86	0.9811	0.0031
	Leaves	>10000	>10000	3390.78	1868.72	0.9828	0.0027
Hot water	Pods	>10000	>10000	3466.26	1563.15	0.9414	0.0169
	leaves	>10000	>10000	3229.97	1348.96	0.9911	0.0010

**Figure 2:** IC₅₀ values (Inhibitory Concentration) for water: acetone, cold water, and hot water extracts of *Psophocarpus tetragonolobus* pods and leaves**Figure 3:** Total Flavonoid Content (TFC), Total Phenolic Content (TPC), and Total Antioxidant Capacity (TAC) of water: acetone (W:A), cold water (CW), and hot water (HW) extracts of *Psophocarpus tetragonolobus* pods and leaves

content in leaves was approximately double that of pods across all solvent systems. Similarly, TAC was highest in water: acetone extracts, with values of 212.74 mg (AAE)/g (pods) and 245.63 mg (AAE)/g (leaves), followed

by cold water (140.10 and 186.16 mg (AAE)/g) and hot water extracts (119.80 and 176.68 mg (AAE)/g). Overall, leaves consistently exhibited higher TFC, TPC, and TAC compared to pods.

DISCUSSION

The results of this study revealed significant pesticidal activity in water: acetone extracts of *Psophocarpus tetragonolobus* pods and leaves. This high efficacy may be attributed to the extraction of both water-soluble polar compounds and non-polar organic compounds, in contrast to the extracts obtained using cold and hot water solvents. Both hot and cold water extracts also demonstrated considerable aphid mortality, suggesting that the active compounds responsible for aphid control are stable and not degraded at high temperatures. Using all three extraction systems allowed a comprehensive evaluation of the plant's bioactivity, as each solvent targets different classes of compounds.

The study further indicated that leaves of *P. tetragonolobus* exhibit higher pesticidal efficacy than pods, as reflected by lower LD₅₀ values across water: acetone, cold water, and hot water extracts. Limited data exists on the pesticidal activity of *P. tetragonolobus* against agricultural pests. For example, isolated proteins from *P. tetragonolobus* seeds have been reported to hinder the growth and development of *Helicoverpa armigera* (cotton bollworm) and *Spodoptera litura* (tobacco cutworm) (Gupta et al., 2008). Similarly, seed proteins demonstrated a concentration-dependent mortality effect on the melon fruit fly (*Bactrocera cucurbitae*) and prolonged larval and pupal development (Kaur & Sohal, 2018). While this study used unpurified extracts without isolating specific bioactive compounds, it is noteworthy that no prior studies have investigated leaf extracts. Here, the water: acetone leaf extract exhibited superior pesticidal activity compared to other solvent extracts and pod extracts. Qualitative phytochemical analysis confirmed the presence of terpenoids, glycosides, alkaloids, flavonoids, phenols, saponins, coumarins, tannins, carbohydrates, and proteins, while quantitative analysis showed high concentrations of polyphenols and other bioactive compounds in leaves, potentially explaining the observed higher pesticidal activity.

Several studies have reported pesticidal effects of plant extracts against aphids and other insect pests (Rahman et al., 2016; Temba et al., 2019; Ebadollahi et al., 2020; Gospodarek et al., 2022). For instance, the water extract of aerial parts of *Artemisia dracunculoides* caused 78–84% mortality of *Aphis fabae* after 108 hours at 30% fresh plant extract (Gospodarek et al., 2022), which is considerably lower than the mortality achieved in this study, where water extracts of *P. tetragonolobus* reached similar levels within 12 hours. Six plant extracts, *Ballota undulata*, *Teucrium polium*, *Phlomis aurea*, *Pulicaria incisa*, *Seriphidium herba-alba*, and *Euphorbia saint catherine*, achieved 74.3%, 73.2%, 72.7%, 65.9%, 62.5%, and 56.8% mortality of *A. fabae* at 50,000 mg L⁻¹ after 48 hours, respectively, which is significantly lower than the 100% mortality observed at 10,000 mg L⁻¹ after 24 hours in this study. Mortality of natural predators, *Chrysoperla carnea* and *Coccinella undecimpunctata*, was below 10%, demonstrating the selectivity and high efficacy of *P. tetragonolobus* extracts (Rahman et al., 2019). Comparatively, the commercial product Naturalis-L showed 62.39%, 78.35%, and 87.66%

mortality of *A. fabae* at 3000 mg L⁻¹ over 24, 48, and 72 hours, respectively, whereas water: acetone extracts in this study caused over 80% mortality at the same concentration within 24 hours, indicating higher effectiveness (Abbas et al., 2023). Chemical insecticides such as pyrethroids (fenvalerate) and neonicotinoids (thiacloprid) produced 81.14 ± 4.2% and 89.21 ± 3.6% mortality against *A. fabae* at 144 × 10³ mg L⁻¹ after 24 hours, further highlighting the potency of *P. tetragonolobus* extracts at much lower concentrations (Purhematy et al., 2014).

This study also evaluated the antioxidant potential of *P. tetragonolobus* extracts (water: acetone, cold water, and hot water) using the DPPH assay, total flavonoid content (TFC), total antioxidant capacity (TAC), and total phenolic content (TPC). The water: acetone leaf extract demonstrated the highest antioxidant activity, TFC, TPC, and TAC among all extracts, likely due to its higher concentration of polyphenolic compounds, which are known to reduce oxidative stress. Similarly, water: acetone extracts exhibited the highest pesticidal activity, suggesting that the elevated phenolic content may contribute to both antioxidant and aphid mortality effects. Pod extracts in water: acetone also showed significantly higher activity compared to cold and hot water extracts. The higher performance of water: acetone extracts can be attributed to their ability to solubilize a broader range of bioactive compounds, including both polar and non-polar components, particularly polyphenols, which act as reducing agents, hydrogen donors, and singlet oxygen quenchers (Khalili et al., 2013; Bassal et al., 2020).

DPPH radical scavenging activity can be compared with standard antioxidants such as ascorbic acid (49.08 ± 6.08 mg AAE/g), gallic acid (14.43 ± 3.53 mg GAE/g), Trolox (67.47 ± 7.35 mg TE/g), and catechin (30.32 ± 8.82 mg CE/g) (Hwang & Lee, 2023). These standards exhibit higher antioxidant activity than the crude extracts in this study, likely because the extracts are derived from unpurified plant material. Previous studies on *P. tetragonolobus* also report variability in antioxidant activity. For example, Bassal et al. (2021) reported an IC₅₀ of 178.40 mg L⁻¹ for 70% methanol leaf extract, whereas Hlosrichok and Aunpad (2023) observed IC₅₀ values of 10.30 mg L⁻¹ (DPPH) and 3.10 mg L⁻¹ (ABTS) for protein extracts from seeds. The higher activity in these studies may reflect the extraction of proteins and other bioactive compounds that are absent in the water or acetone extracts used here. Olaiya et al. (2018) evaluated six-winged bean accessions and reported TPC (0.853–0.941 mg GAE/g), TFC (0.678–1.169 mg QUE/g), and TAC (1.278–1.806 mg AAE/g), which are lower than the values observed in this study. This suggests that water:acetone, cold water, and hot water extracts of pods and leaves are effective radical scavengers with diverse antioxidant mechanisms.

Variations in antioxidant activity may result from differences in extraction solvent, method, plant geographical origin, and assay conditions (Singh et al., 2019; Hlosrichok & Aunpad, 2023). Natural antioxidants such as polyphenols often accumulate in response to environmental stressors, including extreme temperatures, drought, heavy metals, and nutrient deficiencies—enabling plants to adapt and

survive under challenging conditions (Singh et al., 2019). Consequently, IC₅₀, TFC, TPC, and TAC values reported here cannot be directly compared with literature data. Notably, there are no prior reports on variations of these parameters for water:acetone, cold water, and hot water extracts of pods and leaves of *P. tetragonolobus*. The high antioxidant content observed may also explain the lower pest attacks on leaves and pods compared to other legumes, supporting the hypothesis that polyphenols contribute to the observed pesticidal activity. Future studies should focus on identifying the specific antioxidants responsible for these bioactivities.

CONCLUSION

The study revealed that pods and leaves of *Psophocarpus tetragonolobus* exhibit significant pesticidal activities and moderate antioxidant properties. The water: acetone (1:1) extracts showed the highest mortality rates in aphids, which were influenced by both concentration and duration of exposure. Additionally, these extracts showed better antioxidant activity, characterized by the lowest IC₅₀ values, the highest TPC, TFC, and TAC when compared to other solvent extracts, and could be the reason for the observed pesticidal activity. The overall findings suggest that *Psophocarpus tetragonolobus* can be effectively utilized for the development of eco-friendly natural pesticides and antioxidants, potentially benefiting pest management.

DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest

FUNDING INFORMATION

This research was financially supported by a University Research Grant (Grant no: URG/2023/29/S), University of Peradeniya, Peradeniya, Sri Lanka.

AUTHOR CONTRIBUTION

Conceptualization: **J.M.S.J.**; Manuscript editing and reviewing, project supervision: **J.M.S.J.**, **W.A.P.P.D.S.**; Data collection, methodology, statistical analysis, writing the original manuscript: **M.W.M.P.N.**

REFERENCES

- Abbas, M. A., Husani, A. H. A., & Mohammadali, M. T. (2023). Evaluation of the efficacy of some alcoholic plant extracts and the biocide Naturalis-L against *Aphis fabae* Scopoli (Homoptera: Aphididae) under laboratory conditions. *Pakistan journal of science*, **66**, 215-219. <https://journal.utripoli.edu.ly/index.php/Alqalam>.
- Bakkali, F., Averbeck, S., Averbeck, D., & Idaomar, M. (2008). Biological effects of essential oils—A review. *Food Chemistry and Toxicology*, **46**, 446-475. <https://doi.org/10.1016/j.fct.2007.09.106>.
- Basaid, K., Chebli, B., Mayad, E., Furze, J., Bouharrou, R., Krier, F., & Paulitz, T. (2020). Biological activities of essential oils and lipopeptides applied to control plant pests and diseases: A review. *International Journal of Pest Management*, **2**, 1-24. <https://doi.org/10.1080/09670874.2019.1707327>.
- Bassal, H., Hijazi, A., Farhan, H., Trabolsi, C., Ahmad, B. S., & Omar, F. (2021). Study of the antioxidant and anti-inflammatory properties of the biological extracts of *Psophocarpus tetragonolobus* using two extraction methods. *Molecules*, **26**, 4435-4447. <https://doi.org/10.3390/molecules26154435>.
- Bassal, H., Merah, O., Ali, A. M., Hijazi, A., & Omar, F. E. (2020). *Psophocarpus tetragonolobus*: An underused species with multiple potential uses. *Plants*, **9**(12), 1730-1738. <https://doi.org/10.3390/plants9121730>.
- Bhandary, S. K., Kumari, S., Bhat, V. S., Shamila, K. P., & Bekal, M. P. (2012). Preliminary phytochemical screening of various extracts of *Punica granatum* peel, whole fruit and seeds. *Nitte University Journal of Health Science*, **2**(4), 34-38. <https://doi.org/10.1055/s-0040-1703609>.
- Cadet, J., & Davies, K. J. (2017). Oxidative DNA damage & repair: An introduction. *Free Radical Biology and Medicine*, **107**, 2-12. <https://doi.org/10.1016/j.freeradbiomed.2017.03.03>.
- Calvindi, J., Syukur, M., & Nurcholis, W. (2020). Investigation of biochemical characters and antioxidant properties of different winged bean (*Psophocarpus tetragonolobus*) genotypes grown in Indonesia. *Biodiversitas*, **21**, 2420-2424. <https://doi.org/10.13057/biodiversity/210612>.
- Damalas, C., & Eleftherohorinos, I. (2011). Pesticide exposure, safety issues, and risk assessment indicators. *International Journal of Environmental Research & Public Health*, **8**, 1402-1419. <https://doi.org/10.3390/ijerph8051402>.
- Dedryver, C. A., Ralec, A. C., & Fabre, F. (2010). The conflicting relationships between aphids and men: A review of aphid damage and control strategies. *Comptes Rendus Biologies*, **33**, 539-553. <https://doi.org/10.1016/j.crv.2010.03.009>.
- Doss, A. (2009). Preliminary phytochemical screening of some Indian Medicinal Plants. *Ancient Science of Life*, **29**(2), 12-16. PMID: 22557345; PMCID: PMC3336308.
- Ebadollahi, A., Ziaee, M., & Palla, F. (2020). Essential oils extracted from different species of the *Lamiaceae* plant family as prospective bioagents against several detrimental pests. *Molecules*, **25**, 1556-1571. <https://doi.org/10.3390/molecules25071556>.
- Garcia, E. J., Oldoni, T. L., Alencar, S. M., Reis, A., Loguercio, A. D., & Grande, R. H. (2012). Antioxidant activity by DPPH assay of potential solutions to be applied on bleached teeth. *Braz Dent Journal*, **33**(1), 22-27. <https://doi.org/10.1590/s0103-64402012000100004>.
- Gospodarek, I., Endalamew, A., Worsdale, M., & Pasmionka, I. B. (2022). Effects of *Artemisia dracuncululus* L. water extracts on selected pests and aphid predator *Coccinella septempunctata* L. *Agronomy*, **12**, 788-801. <https://doi.org/10.3390/agronomy12040788>.
- Gupta, G. P., Birah, A., & Rani, S. (2008). Growth-inhibitory effects of winged bean (*Psophocarpus tetragonolobus*) proteinase inhibitors on two problematic Lepidopteran

- pests. *Indian Journal of Agricultural Sciences*, **78**(2), 159-162. <https://epubs.icar.org.in/index.php/IJAgS/article/view/2664>.
- Hlosrichok, A., & Aunpad, R. (2023). Antioxidant potential of gamma ray irradiated winged bean (*Psophocarpus tetragonolobus*) seed protein hydrolysate. *Functional Foods in Health and Disease*, **13**(4), 208-229. <https://doi.org/10.31989/ffhd.v13i4.1090>.
- Hwang, S-J., & Lee, J-H. (2023). Comparison of antioxidant activities expressed as equivalents of standard antioxidants. *Food Science Technology*, **43**, 1-5. <https://doi.org/10.1590/fst.121522>.
- Kaur, A. P., & Sohal, S. K. (2018). Purified winged bean protease inhibitor affects the growth of *Bactrocera cucurbitae*. *Bulletin of Entomological Research*, **1**, 1-9. <https://doi.org/10.1017/S0007485318000913>.
- Khalili, R. M., Shafekh, S. E., Norhayat, A. H., Fatahudin, I. M., Rahiman, R., Norkamalia, H. N., & Azimah, A. N. (2013). Total phenolic content and in vitro antioxidant activity of winged bean (*Psophocarpus tetragonolobus*). *Pakistan Journal of Nutrition*, **12**(5), 416-422. <https://doi.org/10.3923/pjn.2013.416.422>.
- Kim, D.-o., Jeong, S. W., & Lee, C. Y. (2003). Antioxidant capacity of phenolic phytochemicals from various cultivars of plums. *Food Chemistry*, **81**, 321-326. [https://doi.org/10.1016/S0308-8146\(02\)00423-5](https://doi.org/10.1016/S0308-8146(02)00423-5).
- Nain, J., Bhatt, S., Dhyani, S., & Joshi, N. (2013). Phytochemical screening of secondary metabolites of *Datura stramonium* L. *International Journal of Current Pharmaceutical Research*, **5**(2), 151-153. <https://www.researchgate.net/publication/281376906>.
- Olaiya, C. O., Soetan, K. O., & Karigidi, K. O. (2018). Evaluation of in vitro antioxidant capacities of six accessions of winged Beans (*Psophocarpus tetragonolobus*). *EC Nutrition*, **13**(8), 589-595. <https://doi.org/10.3390/plants9121730>.
- Park, Y.-S., Jung, S. T., Kang, S.-G., Heo, B. G., Avila, P. A., & Toledo, F. (2008). Antioxidants and proteins in ethylene-treated kiwifruits. *Food Chemistry*, **107**(2), 640-648. <https://doi.org/10.1016/j.foodchem.2007.08.070>.
- Pichersky, E., & Gershenzon, J. (2002). The formation and function of plant volatiles: Perfumes for pollinator attraction and defense. *Current Opinion of Plant Biology*, **5**, 237-243. [https://doi.org/10.1016/s1369-5266\(02\)00251-0](https://doi.org/10.1016/s1369-5266(02)00251-0).
- Prieto, P., Pineda, M., & Aguilar, M. (1999). Spectrophotometric quantitation of antioxidant capacity through the formation of a phosphomolybdenum complex: Specific application to the determination of vitamin E1. *Analytical Biochemistry*, **269**, 337-341. <https://doi.org/10.1006/abio.1999.4019>.
- Purhematy, A., Ahmadi, K. & Moshrefi, M. (2014). Toxicity of thiacloprid and fenvalerate on the black bean aphid, *Aphis* toxicity of thiacloprid and fenvalerate on the black bean aphid, *Aphis*. *Toxicity and biosafety of insecticides*, **6**(2), 207-210. <https://www.researchgate.net/publication/266478383>.
- Rabasse, J.-M., & Van steemis, M. J. (1999). Biological control of aphids. *Integrated Pest and Disease Management in Greenhouse Crops*, **14**, 235-243. https://doi.org/10.1007/0-306-47585-5_16.
- Rahman, S., Biswas, S. K., Barman, N. C., & Ferdous, T. F. (2016). Plant extract as selective pesticide for integrated pest management. *Journal of Biotechnology Research*, **2**(1), 6-10. <http://br.biomedpress.org/index.php/br/article/view/706>.
- Rahman, R. S. K., Ismail, I. A. E., Mohamed, T. A., Hegazy, M. E. F. & Abdeisuafeek, K. A., Biswas, S. K., Barman, N. C., & Ferdous, T. F. (2019). Laboratory and field evaluation of certain wild plant extracts against *Aphis fabae* Scop. (Homoptera: Aphididae) and its predators. *Bulletin of the National Research Centre*, **43**, 1-5. <https://doi.org/10.1186/s42269-019-0084-Z>.
- Ruma, O. C. (2016). Phytochemical screening of selected indigenous edible plants from the towns of Isabela, Philippines. *Asian Journal of Natural & Applied Sciences*, **5**(1), 36-45. <https://www.ajsc.leena-luna.co.jp/ajscvol5nl.php>.
- Savithramma, N., Rao, M., & Subralatha, D. (2011). Screening of medicinal plants for secondary metabolites. *Middle-East Journal of Scientific Research*, **8**(3), 579-584. <https://www.researchgate.net/publication/267690135>.
- Singh, M., Dubey, R. K., Koley, T. K., Maurya, A., Singh, P. M., & Singh, B. (2019). Valorization of winged bean (*Psophocarpus tetragonolobus* (L.) DC) by evaluation of its antioxidant activity through chemometric analysis. *South African Journal of Botany*, **121**, 114-120. <https://doi.org/10.1016/j.sajb.2018.10.026>.
- Temba, Y., Mkindi, A. G., Mekenda, P. A., Mpumi, N., Mwanauta, R., Stevenson, R., & Belmain, S. R. (2019). Pesticidal plant extracts improve yield and reduce insect pests on legume crops without harming beneficial arthropods. *Frontiers in Plant Science*, **9**, 1425-1435. <https://doi.org/10.3389/fpls.2018.01425>.
- Tewary, D. K., Anu, B., & Shanker, A. (2005). Pesticidal activities in five medicinal plants collected from mid hills of western Himalayas. *Industrial Crops and Products*, **22**, 241-247. <https://doi.org/10.1016/j.indcrop.2005.01.004>.
- Valenzuela, I., & Hoffmann, A. A. (2015). Effects of aphid feeding and associated virus injury on grain crops in Australia. *Austral Entomology*, **54**, 292-305. <https://doi.org/10.1111/aen.12122>.
- Yea, C. S., Ebrahimpour, A., Hamid, A. A., Bakar, J., Muhammad, K., & Saari, N. (2014). Winged bean [*Psophocarpus tetragonolobus* (L.) DC] seeds as an underutilised plant source of bifunctional proteolysate and biopeptides. *Food and Function*, **5**, 1007-1017. <https://doi.org/10.1039/c3fo60667h>.
- Zikankuba, V., Mwanyika, G., Ntwenya, J., & James, A. (2019). Pesticide regulations and their malpractice implications on food and environment safety. *Cognet Food Agriculture*, **5**, 1601544-160166. <https://doi.org/10.1080/23311932.2019>.