

Research Article

## *Pluchea indica* metabolites production under chicken manure application in the rainy season

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### Abstract

Indian camphorweed (*Pluchea indica*), an indigenous vegetable, has promising potential as a cosmetic raw material because of its bioactive compounds. Research on cultivation techniques is relatively underexplored in agronomic research, particularly in fertilization practices for *P. indica* in the rainy season, a period that is critical for tropical agriculture. This study aimed to determine the optimal chicken manure dosage for the growth and yield of four-month-old Indian camphorweed during the rainy season. The experiment was conducted at IPB University, Indonesia, from November 2023 to February 2024. A randomized complete block design (RCBD) was applied, consisting of four chicken manure dosages: 0, 2.5, 5.0, and 7.5 kg per plant, with three replications. Results showed that chicken manure application was found to enhance vegetative growth, branching, leaf yield, and leaf N and K status, indicating its suitability as an organic nutrient source under high-rainfall conditions. Regression analysis indicated the optimal dosage of chicken manure was 7.00 kg per plant for fresh weight yield at 27 weeks after planting (WAP). Chicken manure application changes the phenolic content and antioxidant capacity, that suggest the trade-offs between yield improvement and certain aspects of functional quality.

**Keywords:** Indian camphorweed; optimum dosage; organic fertilizer

### Introduction

Indian camphorweed (*Pluchea indica* (L.) Less.) is a perennial shrub (Cho et al., 2012) from the family Asteraceae, commonly cultivated as a hedgerow plant in Indonesia and thriving in dry or rocky soils (Syafira et al., 2019). Indian camphorweed is typically found in swamps, saline areas, tidal zones, lowland riparian zones, and wetlands, and can grow in full sun to partial shade. Native to Asia, it grows in India, Thailand, Indochina, southern China, Japan, the Philippines, and Malaysia, and has spread to Australia and the Hawaiian Islands as a non-native species (Peng et al.,

1998). Today, Indian camphorweed is estimated to be distributed across tropical regions worldwide, including Indonesia, as a locally adapted plant and with high potential due to genetic–environment interactions (Rukmana & Yudirachman, 2016).

Indian camphorweed contains protein, fat, fiber, carbohydrates, calcium,  $\beta$ -carotene, and vitamin C (Sudjaroen, 2012). It also shows potential for medicinal and cosmetic applications due to its phytochemical content, which includes flavonoids, tannins, phenolic hydroquinone, sterols (Safitri et al., 2018), alkaloids, and saponins (Lestari et al., 2020). Traditionally, Indian camphorweed has been used to reduce body odor, relieve muscle and rheumatic pain, and treat kidney stones (Ibrahim et al., 2022). Pharmacological studies have reported appetite-stimulating, antipyretic, digestive, antidiarrheal, antitussive, emollient, and antibacterial activities (Andarwulan et al., 2012), as well as against *Escherichia coli* and *Bacillus subtilis*, which cause diarrhoea (Lestari et al., 2020), and against *Streptococcus mutans*, which causes dental caries (Nurrohman et al., 2021). Given these pharmacological properties, attention has increasingly shifted toward cultivation practices that can enhance growth, yield, and metabolite production of *P. indica* under field conditions.

The previous research used chicken manure applied at planting on *P. indica* in the dry season, which significantly improved plant height, leaf number, tertiary branch number, fresh and dry leaf weight at 8 and 14 WAP, and total flavonoid content of the third leaf at 14 WAP (Indriani et al., 2024). Soil nutrients may decline over time due to harvest removal, volatilization, or leaching (Hidayanto, 2019), necessitating repeated fertilization to maintain plant nutrient supply. For perennial crops, fertilization is typically conducted several times a year, often before and after the rainy season (Budiargo et al., 2015).

Conducting fertilization experiments during the rainy season is essential for perennial leafy crops such as Indian camphorweed that are cultivated under tropical conditions because rainfall strongly regulates soil nutrient dynamics, fertilizer efficiency, and plant metabolic responses. High rainfall intensity and frequency increase nutrient solubility and downward transport, enhancing the risk of nitrogen (N) and potassium (K) leaching and causing large seasonal variation in crop response to fertilization, which cannot be adequately captured by dry-season studies (Guo & Chen, 2022; Russo et al., 2017). Therefore, executing this experiment during the rainy season enables accurate assessment of chicken manure dosage effects on growth, yield, nutrient status, and metabolite production under conditions that realistically represent farmers' practices and support the objective of developing season-specific fertilization recommendations for *P. indica*.

The functions of nitrogen for plant growth and development include increasing plant vegetative growth, increasing leaf production in plants such as vegetables or livestock grasses, increasing protein levels in plants, and functioning as a source of amino acids and proteins in plants (Patti et al., 2013). The phosphorus element is an important component in providing energy (ATP, ADP, NADPH), which in turn plays a role in plant metabolic processes and responses to abiotic stresses (Taiz & Zeiger, 2002). Potassium plays a role in the translocation of photosynthetic products from leaves to reproductive organs and storage. Potassium is also an element that forms proteins and carbohydrates, functions to increase resistance to pests and plant diseases, and helps open and close stomata (Purnomo et al., 2017). Chicken manure also contains microelements, including calcium, magnesium, sulphur, sodium, iron, copper, and molybdenum (Hidayati et al., 2021). Chicken manure has the potential to improve the physical, chemical, and biological structure of soil, namely in improving

soil aeration, increasing buffer capacity against pH changes, increasing cation exchange capacity, and becoming a source of energy for soil microorganisms (Sari et al., 2020). This study aims to determine the optimal fertilizer dose using chicken manure for the growth and production of 18-week-old Indian camphorweed in the rainy season.

## Materials and methods

### **Research site and procedure**

The research was conducted at IPB University, Bogor, Indonesia, from November 2023 to February 2024. Fertilizer and NPK leaf concentration analysis was carried out at the Testing Laboratory, Department of Agronomy and Horticulture. Fertilizer analysis – for water content with gravimetry (SNI 7763:2018 section 6.3); C-organic (%): gravimetry (SNI 7763:2018 section 6.5); pH: pH meter (SNI 7763:2018 section 6.4); total N (%): Titrimetry (SNI 7763:2018 section 6.6.1); total P<sub>2</sub>O<sub>5</sub> (%): Spectrophotometry (SNI 7763:2018 section 6.7); total K<sub>2</sub>O (%): AAS (SNI 7763:2018 section 6.7). Leaf pigment analysis, fresh and dry weight measurements, and phenolic and flavonoid contents were carried out in the Postharvest and Quality Testing Laboratory, Department of Agronomy and Horticulture. Phytochemical and antioxidant capacity tests were performed at the Tropical Biopharmaca Research Center, IPB University.

Plants used were 18 weeks after planting (WAP) Indian camphorweed with 1 m × 1 m spacing. They had previously been treated with chicken manure at varying dosages (0, 2.5, 5, and 7.5 kg per plant) at planting and harvested at 8 and 14 WAP during the dry season (Indriani et al., 2024). At 18 WAP, the fertilizer was applied by soil incorporation around the plants, plants were pruned to 40 cm above ground level and 30 cm horizontally from the main stem, then re-fertilized with chicken manure at the same dosages at 19 WAP.

### **Experimental design and trait measurements**

This research used a randomized complete block design (RCBD), consisting of four chicken manure dosages: 0.0 (control), 2.5, 5.0, and 7.5 kg per plant. Each treatment was replicated three times (Yan, 2021), with 20 plants per experimental unit, for a total of 240 plants, and using ten (10) sample plants per plot.

Growth parameters measured every 2 weeks included plant height (cm) and the number of primary, secondary, tertiary, quaternary, and *de novo* branches. Seven leaves from ten sample plants were composited and then analyzed for leaf pigments (Yudiansyah et al., 2024) measured included chlorophyll a (mg g<sup>-1</sup>), chlorophyll b (mg g<sup>-1</sup>), total chlorophyll (mg g<sup>-1</sup>), anthocyanins (μmol), and carotenoids (mg g<sup>-1</sup>) at 23 WAP. Leaf NPK content was determined from the seventh leaf from the leaf buds at 23 WAP. Phytochemical composition in total phenolic and flavonoid contents, and antioxidant capacity (ascorbic acid equivalent, ppm) were measured from dried leaves at 27 WAP.

Two harvests were conducted after fertilizer application, at 23 and 27 WAP. At 23 WAP, all branches were pruned to 45 cm above ground level, with lateral branches cut 30 cm from the main stem (measured horizontally). At 27 WAP, all plants were pruned to 50 cm above ground level and 30 cm from the main stem. Dry leaf weight was obtained by air-drying the leaves for one day, followed by oven-drying at 70 °C for 48 hours.

Leaf nitrogen (N) content was analyzed using the Kjeldahl method (Nelson & Sommers, 1980), phosphorus (P) content by UV-VIS spectrophotometry (Adelowo & Oladeji, 2016), and potassium (K) content by Atomic Absorption Spectrophotometry (AAS) (Hasanuzzaman et al., 2018). Samples for NPK analysis consisted of dried powder from the seventh leaf collected at 23 WAP (Indriani et al., 2024).

Total phenolic content was determined following (Vongsak et al., 2013) using the seven dried leaf samples from ten sample plants at 27 WAP harvest. Samples were oven-dried (Santo drying oven MOV-112) at 70 °C for 48 hours. Each 200 µL aliquot of sample extract (1000 g mL<sup>-1</sup>) was mixed with 500 µL of Folin-Ciocalteu reagent (diluted 1:10 with deionized water) and 800 µL of sodium bicarbonate solution (7.5%, w/v). The mixture was allowed to stand at room temperature for 30 minutes with occasional mixing, and absorbance was measured at 765 nm using a UV-VIS spectrophotometer, Shimadzu type UV-1280. Results were expressed as milligrams of gallic acid equivalents (GAE) per 100 g dry weight.

Total flavonoid content was determined following (Vongsak et al., 2013) using dried leaf samples from the 27 WAP harvest. Samples were oven-dried at 60 °C for 24 hours. Each 500 µL aliquot of sample extract (1000 g mL<sup>-1</sup>) was mixed with 500 µL of 2% aluminum chloride solution. The mixture was kept at room temperature for 10 minutes with occasional mixing, and absorbance was measured at 415 nm using a UV-VIS spectrophotometer, with a blank (without aluminum chloride) as the control. Results were expressed as milligrams of quercetin equivalents (QUE) per 100 g dry weight.

Antioxidant capacity (Chaves et al., 2020) was evaluated using dried leaf powder from the 27 WAP harvest. Samples were extracted with 96% ethanol, and antioxidant activity was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging method at 50 ppm. Ten milligrams of plant extract or vitamin C (positive control) were dissolved in 1 mL DMSO, sonicated until fully dissolved, and vortexed. A total of 40 µL of sample or vitamin C was transferred into a microplate (in triplicate, with a negative control). DPPH solution was added to the samples, while ethanol was added to the negative control. The mixture was incubated in the dark at room temperature for 30 minutes before absorbance was measured at 517 nm using an ELISA reader, BioTek Epoch 2.

### **Data analysis**

Statistical analysis was based on individual plants and was analyzed using analysis of variance (ANOVA) at the 5% significance level with SPSS IBM SPSS Statistics 26 and Duncan's Multiple Range Test (DMRT). Regression analysis and graphical presentation were performed using Microsoft Excel 2019. Statistical significance was assessed at  $p \leq 0.05$ .

## **Results and discussion**

### **General conditions**

During the experimental period, the average temperature was 26.75 °C, monthly rainfall averaged 691.43 mm, relative humidity was 83.35%, and average daily sunshine duration was 5.44 hours. Rainfall was highest in November (1068.00 mm) and decreased progressively in December (596.75 mm), January (409.44 mm), and February (232.80 mm) (Meteorology, Climatology, and Geophysical Agency, 2024).

### Vegetative growth

Chicken manure application significantly affected plant height at 23–27 WAP. Plants receiving 2.5 kg per plant showed a 13.48% increase in height compared to the control at 27 WAP; no significant difference was found between the 2.5 to 7.5 kg treatments (Table 1).

Table 1. Average plant height of Indian camphorweed with different doses of chicken manure treatment

Dose of chicken manure (kg per plant)	Plant height (cm)					
	21 WAP	22 WAP	23 WAP	24 WAP	26 WAP	27 WAP
0.0	55.89	79.42	90.05b	57.20b	69.33c	76.78b
2.5	60.78	87.71	100.07a	63.22a	79.81b	89.92a
5.0	59.11	85.57	100.42a	60.03a	82.06ab	90.92a
7.5	62.50	86.94	101.22a	59.92ab	88.92a	95.97a

Note: Numbers followed by different letters in the same column indicate significant differences with DMRT at 5% level; WAP = week after planting.

Application also significantly affected the number of primary branches at 26 WAP, tertiary branches at 24–27 WAP, and quaternary branches at 24 and 26 WAP, but not secondary or *de novo* branches. Untreated plants generally had fewer primary, tertiary, and quaternary branches compared to treated plants (Table 2).

Plant height and branch number decreased at 24 WAP due to the 23 WAP harvest, after which growth resumed. Tertiary branches were the most abundant, likely contributing substantially to leaf yield. The chicken manure organic fertilizer used has a total N content of 2.14%, P<sub>2</sub>O<sub>5</sub> 3.84%, K<sub>2</sub>O 3.14%, with a moisture content of 18.76% (w/b), C-organic 23.14%, and pH 8.09. The fertilizers used have met the quality criteria for solid organic fertilizer according to SNI 7763: 2018. Nitrogen plays a very important role in growth and development, as well as plant production (Luo et al., 2020). Research by Yaldız et al. (2019) states that the application of chicken manure increases plant height and the number of branches of basil (*Ocimum basilicum* L.) plants compared to plants without being given chicken manure. This is in line with the research of Indriani et al. (2024) in the drought season, which stated that the application of chicken manure to 0 WAP Indian camphorweed plants had a significantly higher effect on plant height and the Indian camphorweed branch number.

### Leaf yield

Fresh leaf weight at 18 WAP (before re-fertilization) differed significantly among treatments, reflecting residual effects from previous fertilization at planting in the dry season (Indriani et al., 2024). At 18 WAP in the rainy season, control plants (without manure application) were significantly lower than plants that had residual fertilizer treatment of 2.5 and 5 kg per plant, but not significantly different from plants that had residual fertilizer treatment of 7.5 kg per plant. The plant with the 2.5 kg treatment increased leaf fresh weight by 84.26% compared to the control (Table 3). According to Indriani et al. (2024), Indian camphorweed in the drought season produced an average fresh leaf weight of 338.54 g per plant at 14 WAP, while control plants without manure yielded 188.85 g per plant. These values suggest that the fresh leaf weight observed at 18 WAP in the present study during the rainy season was relatively lower by 27.8–34.3% than in the drought season.

Table 2. Branch number per plant of Indian camphorweed with different doses of chicken manure treatment

Dose of chicken manure (kg per plant)	Week after planting (WAP)					
	21	22	23	24	26	27
	Primary branch number					
0.0	9	9b	11	15	13 b	14
2.5	11	11ab	14	14	14 ab	15
5.0	11	11a	13	13	15 a	15
7.5	10	11a	13	13	13 b	14
	Secondary branch number					
0.0	42	35	52	46	45	55
2.5	45	44	59	50	52	54
5.0	44	43	53	50	51	49
7.5	42	44	47	52	49	46
	Tertiary branch Number					
0.0	66	51	75	55 b	61 b	68 b
2.5	59	61	83	70 b	79 a	89 a
5.0	75	75	76	71 b	80 a	90 a
7.5	71	73	75	93 a	85 a	79 ab
	Quaternary branch number					
0.0	17	13 b	31	11 c	21 c	31
2.5	19	19 ab	32	24 bc	34 b	45
5.0	18	32 a	30	29 ab	40 ab	51
7.5	32	26 ab	27	43 a	46 a	50
	<i>De novo</i> branch number					
0.0	2	2	4	2	2	1
2.5	1	4	5	3	2	3
5.0	1	4	5	2	3	2
7.5	1	4	4	2	2	2

Note: Numbers followed by different letters in the same column and trait indicate significant differences with DMRT at 5% level.

Table 3. Fresh weight of Indian camphorweed at 18 WAP

Dose of chicken manure (kg per plant)	Fresh weight (g per plant)
0.0	124.15b
2.5	228.76a
5.0	244.31a
7.5	157.64b
F-test	**
CV (%)	10.85

Note: \*\*: highly significant effect at 1% level; Numbers followed by different letters in the same column indicate significantly different with DMRT at 5% level.

At the first harvest (23 WAP), leaf fresh weight was increased by 36.04, 62.02, and 54.02% for the 2.5, 5, and 7.5 kg treatments, respectively, compared to the control. At the second harvest (27 WAP), increases were 80.11, 88.82, and 114.62%, respectively (Figure 1). Dry leaf weight at 23 WAP increased significantly by 73.96% (5 kg) and 54.89% (7.5 kg) compared to the control. At 27 WAP, increases were 56.65, 60.66, and 79.68% for the 2.5, 5, and 7.5 kg treatments, respectively (Figure 2).

The application of chicken manure can increase the yield of Indian camphorweed due to the nutrient content and physicochemical properties of the fertilizer. Chicken manure, derived from composted poultry waste, generally contains higher nitrogen and potassium levels compared to other livestock manures (Pagliarini et al., 2024). Azmi et al. (2019) conveyed that the application of chicken manure

significantly increased soil pH, total organic C, C/N ratio up to 10.39, macronutrient content such as N-total, P, K, Ca, and exchangeable Mg, and micronutrient content such as Fe, Cu, Zn, exchangeable Mn. Chicken manure can also increase soil organic matter and is slow-released (Thepsilvisut et al., 2022). So that improving soil structure and adding nutrients can support plant growth and production.

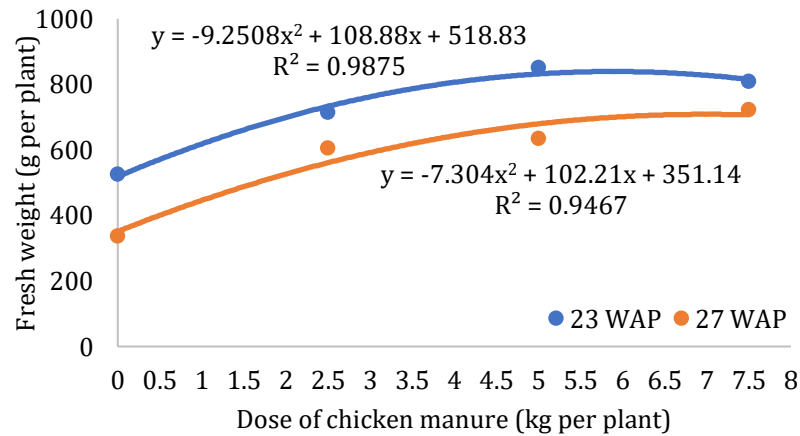


Figure 1. Regression of chicken manure on leaf fresh weight at 23 and 27 WAP

The average leaf fresh weight of Indian camphorweed at 27 WAP decreased by 11-36%, and the dry weight of leaves decreased by 42% compared to the harvest at 23 WAP. Several factors may occur, including the reduction of available nutrients over time, so that at 27 WAP, the plants may not receive the optimal amount of nutrients as at 23 WAP. A decrease in nutrients in the soil after harvesting indicates the utilization of nutrients by plants for growth and production (Dewanto et al., 2013). Manure has a lower nutrient content than inorganic fertilizers and decomposes relatively quickly within a few growing seasons, so the application of manure must be repeated frequently to maintain soil productivity (Lima et al., 2021). Other factors, such as climate and pathological disorders, can also contribute to reduced yields. Some powdery mildew symptoms were observed during the experiment, which is commonly associated with high humidity and reduced air circulation during the rainy season. This study was conducted during the rainy season, and high rainfall can also be a factor in nutrient leaching (Rashmi et al., 2017).

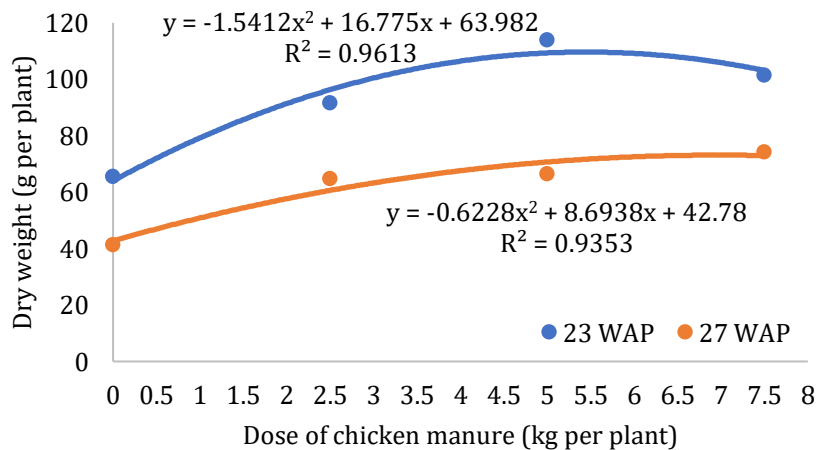


Figure 2. Regression of chicken manure on dry weight at 23 and 27 WAP

Regression analysis showed a quadratic relationship between manure dosage and both fresh and dry leaf weight (Figure 2). Optimum dosages were 5.88 kg per plant for fresh weight and 5.44 kg per plant for dry weight at 23 WAP, and 7.00 kg and 6.98 kg per plant, respectively, at 27 WAP. Based on regression analysis, the optimum chicken manure dosage for maximizing fresh leaf yield at 27 WAP was 7.00 kg per plant. This result may be associated with the continued vegetative growth of *P. indica* at this stage, as the plants were maintained under a repeated harvesting system rather than approaching maturity. Under such conditions, adequate nutrient supply is required to support sustained leaf regrowth following harvest.

### Leaf pigment and NPK content

Manure application had no significant effect on chlorophyll, anthocyanin, or carotenoid content (Table 4), suggesting that pigment formation was more influenced by environmental and genetic factors (Afida et al., 2020).

Table 4. Indian camphorweed leaf pigment analysis at 23 WAP

Dose of chicken manure (kg per plant)	Leaf pigments				
	Chlorophyll a (mg g <sup>-1</sup> )	Chlorophyll b (mg g <sup>-1</sup> )	Anthocyanins (μmol g <sup>-1</sup> )	Carotenoids (mg g <sup>-1</sup> )	Total chlorophyll a (mg g <sup>-1</sup> )
0.0	0.57	0.21	0.58	0.22	0.78
2.5	0.72	0.26	0.54	0.26	0.98
5.0	0.52	0.22	0.68	0.20	0.74
7.5	0.69	0.23	0.45	0.24	0.92
F-test	ns	ns	ns	ns	ns
CV (%)	26.81	23.11	26.77	24.47	25.56

Leaf N and K content increased significantly with increasing manure dosage, whereas P content did not differ significantly among treatments (Table 5). The 2.5 kg treatment increased N and K by 24% and 116%, respectively, compared to the control. In field experiments under high rainfall intensity and frequency, variability is typically amplified due to the heterogeneous nutrient distribution, micro-topography, and differential leaching intensity. This condition masked treatment differences despite increasing fertilizer rates (Russo et al., 2017).

Table 5. Indian camphorweed leaf NPK at 23 WAP

Dose of chicken manure (kg per plant)	N (%)	Status <sup>T</sup>	P (%)	Status <sup>T</sup>	K (%)	Status <sup>T</sup>
	0.0	3.27b	Medium	0.26	Medium	0.44c
2.5	4.07a	Medium	0.29	Medium	0.95b	Deficient
5.0	4.14a	Medium	0.30	Medium	1.19ab	Deficient
7.5	4.20a	Medium	0.35	Medium	1.33a	Deficient
F-test	*		ns		**	
CV (%)	6.32		13.15		15.76	

Note: <sup>T</sup>Jeong et al. (2009). \*: significant effect at 5% level; \*\*: highly significant effect at 1% level; numbers followed by different letters in the same column indicate significantly different on DMRT test at 5% level

### Secondary metabolites

Secondary metabolites are chemical compounds that are naturally produced by plants. Secondary metabolites function to provide color, taste, and aroma to plants

(Ashraf et al., 2018) and play a role in plant interactions with the environment (Pagare et al., 2015). The formation of secondary metabolites is influenced by genetic factors as well as biotic and abiotic factors (Rabeh et al., 2025).

Qualitative phytochemical tests detected flavonoids, tannins, saponins, and steroids in all treatments, indicating these compounds are constitutively produced as part of the plant's adaptive response. Alkaloids, quinones, and triterpenoids were not detected (Table 6).

Table 6. Phytochemical analysis results of Indian camphorweed leaves

Phytochemical parameters	Presence of compound at chicken manure dose			
	0 kg per plant	2.5 kg per plant	0 kg per plant	7.5 kg per plant
Flavonoids	+	+	+	+
Wagner	-	-	-	-
Mayer	-	-	-	-
Dragendorff	-	-	-	-
Tannins	+	+	+	+
Saponins	+	+	+	+
Quinone	-	-	-	-
Steroids	+	+	+	+
Triterpenoids	-	-	-	-

Total phenolic content ranged from 2.1 to 3.8 g GAE per 100 g dry weight, with the control having 52–84% higher phenolic content than fertilized plants (Table 7). This value is still close to the range of phenolic content of Indian camphorweed leaves by (Vongsak et al., 2018), which is 5.1-8.1 g GAE per 100 grams of extract, depending on leaf position and flowering stage, although the plants in this experiment are not flowering. Conversion results using yield data of 35-45% obtained a total phenolic content ranging from 2.04-3.81 g per 100 g dry matter.

Table 7. Total phenolic content and production of Indian camphorweed leaves

Dose of chicken manure (kg per plant)	Total phenolic content (mg GAE per 100 g dry weight)	Total production (g GAE per plant)		
		23 WAP	27 WAP	Total
0.0	3876.0a	2.43	1.61	4.04
2.5	2541.8b	1.85	1.42	3.28
5.0	2239.1b	2.86	1.71	4.56
7.5	2107.7b	2.21	1.64	3.85
F test	**	ns	ns	ns
CV (%)	14.23	20.68	23.28	19.62

Note: ns: not significant effect; \*\*: highly significant effect at 1% level; numbers followed by different letters in the same column indicate significantly different on DMRT test at 5% level

Higher manure dosages tended to reduce phenolic content (Table 8). Phenolic compounds are compounds produced as a form of plant defense against environmental stress such as nutrient deficiencies, pathogen attacks, wounds, temperature stress, UV radiation, and heavy metals (Naikoo et al., 2019). Nitrogen has an impact on the concentration of secondary metabolites in plants (Ibrahim et al., 2011). Total flavonoid content ranged from 364.9 to 978.1 mg QUE per 100 g dry weight, within previously reported ranges for *P. indica*.

Table 8. Total flavonoid content and leaf production

Dose of chicken manure (kg per plant)	Total flavonoids (mg QUE per 100 g dry weight) <sup>t</sup>	Total flavonoid production (mg QUE per plant)		
		23 WAP <sup>t</sup>	27 WAP <sup>t</sup>	Total <sup>t</sup>
0.0	364.9	221.5	146.0	367.5
2.5	676.2	677.0	417.8	1094.8
5.0	956.3	988.6	629.9	1618.5
7.5	978.1	1031.0	738.4	1769.4
F test	ns	ns	ns	ns
CV (%)	30.00	30.00	29.98	30.00

Note: ns: not significant effect; t: transformed data; numbers followed by different letters in the same column indicate significant differences based on DMRT (Duncan's Multiple Range Test) test at 5% level; QUE: quercetin equivalent.

Antioxidant capacity decreased significantly with manure application, with reductions of 39%, 51%, and 55% for the 2.5, 5.0, and 7.5 kg treatments, respectively, compared to the control (Table 9). This may reflect reduced ROS levels under better nutrient supply, reducing the need for antioxidant defense. On *Gynura procumbens*, the same phenomenon was recorded, that increasing chicken manure doses caused a noticeable decline in total phenolic and flavonoid content, and in antioxidant capacity. When passing from lower to higher fertilizer rates, total phenolic content in mature leaves decreased after the application, but inversely in the young leaves (Wan Majid et al., 2024).

Table 9. Antioxidant capacity of Indian camphorweed plants at different doses of chicken manure

Dose of chicken manure (kg per plant)	Capacity to ascorbic acid (ppm)
0.0	18.09a
2.5	11.12b
5.0	8.95bc
7.5	8.06c
F test	**
CV (%)	11.46

Note: \*\*: highly significant effect at 1% level; numbers followed by different letters in the same column indicate significantly different based on DMRT test at 5% level

## Conclusions

The application of chicken manure to four-month-old Indian camphorweed in the rainy season has a significant effect on several variables of growth and production of fresh and dry leaf, total phenolic content, and antioxidant capacity. Application of chicken manure can increase the growth and production of Indian camphorweed leaves, but reduce its levels of phenolic compounds and antioxidant capacity. Chicken manure application is optional for producing flavonoids from four-month-old camphorweed plants in the rainy season.

The results of regression analysis showed that the optimal dose of chicken manure was 5.88 kg per plant for fresh weight production and 5.44 kg per plant for dry weight at 23 WAP, and 7.00 kg per plant for fresh weight production and 6.98 kg per plant for dry weight production at 27 WAP. The plant can be harvested every month. Further research is needed to evaluate the potential of *P. indica* as a cosmetic raw material for anti-aging and skin-protective applications.

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