



Research Article

Humic acid and chicken manure improve growth and productivity of organic edamame

Regata Ringga Hanessa Putry ¹, Maya Melati ^{2,*}, and Munif Ghulamahdi ²

¹ Agronomy and Horticulture Study Program, Graduate School, IPB University (Bogor Agricultural University), Jl. Meranti, Kampus IPB Dramaga, Bogor 16680, INDONESIA

² Department of Agronomy and Horticulture, Faculty of Agriculture, IPB University (Bogor Agricultural University), Jl. Meranti, Kampus IPB Dramaga, Bogor 16680, INDONESIA

* Corresponding author (✉ maya_melati@apps.ipb.ac.id)

ABSTRACT

Edamame (Glycine max L.), a soybean harvested for young pods, has high nutritional content and is promising as an export commodity. Here, organic edamame was produced by incorporating chicken manure and humic acid. This study aimed to evaluate the effects of chicken manure, humic acid, and their interaction on the growth and productivity of edamame. The study used a factorial randomized complete block design with two factors and three replications, and it was carried out at the IPB Experimental Station, Cikarawang A, Dramaga, Bogor, from May to September 2024. The first factor was chicken manure: 0, 8, 16, and 24 tons ha⁻¹, and the second factor was humic acid: 0, 30, 60, and 90 kg ha⁻¹. Vegetative variables were observed at 5 weeks after planting (WAP). The relative growth rate (RGR) and net assimilation rate (NAR) were determined from the period between 3 and 5 WAP. Yield components included the number, length, and weight of pods and pod yield. The results showed that the interaction of chicken manure and humic acid significantly increased root length, RGR, number of flowers, number of productive nodes, plant fresh weight, plant dry weight, pod weight per plant, and pod yield. Chicken manure at 9.5 tons ha⁻¹ and humic acid at 90 kg ha⁻¹ were the optimum doses to significantly increase pod yield. This study highlights the potential use of humic acid to reduce the amount of chicken manure demand in organic farming systems.

Keywords: leaf greenness; net assimilation rate; nutrient levels; organic fertilizer; relative growth rate

INTRODUCTION

Edamame (*Glycine max* (L.) Merr.) is a kind of soybean that is harvested for the young pod. It is utilized extensively in the food and manufacturing sectors due to rich in protein, fiber, and antioxidants, including vitamin C and isoflavones (Kumar et al., 2011; Kumar et al., 2014). Edamame has high demand in the international market, especially in Japan, Taiwan, the United States, Malaysia, Singapore, and Europe (Radarjember, 2023). Especially, organic edamame is a promising export commodity from Indonesia.

However, the capacity of Indonesian farmers to fulfil such demand is still low. Indonesia in 2015-2020 produced 674,843 tons annually or 8.3% of the Dutch market and 6.67% of the Japan market demand of edamame (Ministry of Agriculture, 2024). This underscores the urgent need to increase edamame productivity. One focus to improve production is through improving soil health through organic matter and fertilization application (Tao et al., 2022).

Edited by:

Siti Marwiyah
IPB University

Received:

26 April 2025

Accepted:

16 July 2025

Published online:

29 August 2025

Citation:

Putry, R. R. H., Melati, M., & Ghulamahdi, M. (2025). Humic acid and chicken manure improve growth and productivity of organic edamame. *Jurnal Agronomi Indonesia (Indonesian Journal of Agronomy)*, 53(2), 150-160. DOI: <https://dx.doi.org/10.24831/jai.v53i2.63915>

Farmer usually uses inorganic fertilizers in edamame farming, and excess inorganic application might detrimental effect on soil and environment (Kakar et al., 2020). Therefore, organic fertilizers such as chicken manure can be recommended. Chicken manure contains N, P, K, Ca, Mg, and S (McCall, 1980). Nitrogen and phosphorus in chicken manure are considered high compared to other organic fertilizers (Sari & Arifandi, 2019). A previous study reported that the application of 4 tons ha⁻¹ chicken manure increases the growth and productivity of soybean (Ahmadi & Arien, 2022). Chicken manure with a dose of 16 tons ha⁻¹ increases the yield of edamame (Melati et al., 2025).

On the other hand, humic acid application might enhance manure efficiency. The application of chicken manure in combination with humic acid improves chicory (*Cichorium intybus* L.) (Gholami et al., 2018). Application of fulvic acid, a kind of humic acid, increases plant growth and flavonoid content in moringa (Kailola et al., 2023). Humic acid is an amphiphilic polyelectrolyte macromolecule with an acidic pH; it increases the cation exchange capacity in the soil (Azahin et al., 2014; Rahmandhias & Rachmawati, 2020). The utilization of humic acid enhances nutrient availability for plants and improves the quality of the growth media, thereby promoting plant growth (Akladios & Mohamed, 2018; Al-Taey et al., 2019). A previous study reported that application of humic acid at a rate of 50 kg ha⁻¹ combined with 200 kg ha⁻¹ SP36 increased soybean production by 43% due to humic acid-assisted release of P bound by soil metals so that they become available to plants (Setyawan & Setyawan, 2019). Application of 10 kg ha⁻¹ humic acid combined with inorganic fertilizers can also increase available P, available K, and dry weight of soybean seeds. The presence of carboxyl and phenolic groups in humic acid facilitates the binding of ions like Al, Fe, and Ca, therefore enhancing phosphorus availability. Moreover, humic acid contributes to enhancing the Cation Exchange Capacity (CEC), hence facilitating improved nutritional absorption (Rahayu et al., 2021).

However, research on the combination of chicken manure and humic acid on the edamame is still lacking. This study aimed to examine the impact of different doses of chicken manure and humic acid, as well as their interaction, on the growth and productivity of edamame, and to identify the ideal dosage of both to enhance edamame yield. This research may support sustainable organic fertilization techniques in edamame.

MATERIALS AND METHODS

The experiment was carried out at the IPB Experimental Station in Cikarawang A, Dramaga (6°33'30" S, 106°43'43" E; 160 m above sea level), Bogor, West Java, Indonesia, from May to September 2024. The experiment was conducted using a factorial randomized complete block design with two factors and three replications. The first factor was chicken manure, which consisted of 0, 8, 16, and 24 tons ha⁻¹, and the second factor was humic acid: 0, 30, 60, and 90 kg ha⁻¹. There were 16 fertilizer combinations with three replications each, so there were 48 experimental units.

Soil was plowed, and chicken manure and dolomite were applied. Subsequently, raised beds were formed with dimensions of 2 m long, 2 m wide, and 0.3 m high. Each bed was made with 80 planting holes at a spacing of 0.25 m x 0.2 m. Edamame Biomax 1 seeds were planted in planting holes 2-4 cm deep in the experimental plot with two seeds per hole. Chicken manure was applied two weeks before planting. The method for applying humic acid through seed coating is as follows: soybean seeds are moistened with a small amount of water, then inoculated with *Rhizobium* sp. (5 g per kg of seed) and humic acid according to the treatment dosage. Any remaining humic acid that does not stick to the seeds is poured into the seed hole.

Chemical content of the chicken manure was evaluated (Table 1). Soil analysis was conducted twice, i.e., before planting and after fertilizer application at three weeks after planting (WAP). Soil sampling was conducted by taking composite soil samples at a depth of 20 cm from five different points diagonally. The analysis consisted of pH (H₂O), C-organic content (Walkley and Black method), CEC (N NH₄OAC), N total (Kjeldahl method), P potential (Bray method), P available (HNO₃:HClO₄), K total (HNO₃:HClO₄), and K available (HNO₃:HClO₄).

Table 1. Chemical properties of chicken manure.

Manure properties	Value	Standard*	Criteria
Moisture content % (w/b)	19.02	8-20	Appropriate
C-organic (%)	20.36	Minimum 15	Appropriate
pH	8.68	4-9	Appropriate
N-total (%)	2.23	≥2	Appropriate
P ₂ O ₅ -total (%)	2.95	≥2	Appropriate
K ₂ O-total (%)	2.27	≥2	Appropriate

Note: *Criteria based on the Ministry of Agriculture (2019).

Plant variables at the vegetative phase, namely root length (cm), leaf area (cm²), and leaf greenness index, were observed on 2 sample plants per experimental unit at 5 WAP. Measurement of the leaf greenness index was carried out using the SPAD-502 Chlorophyll Meter on three mature leaves (Freidenreich et al., 2019). Relative growth rate (RGR) and net assimilation rate (NAR) were examined throughout the interval of 3 to 5 WAP (Simatupang et al., 2023) and calculated using the formula:

$$\bullet \text{ RGR (g g week}^{-1}\text{)} = \frac{\ln(W_2) - \ln(W_1)}{t_2 - t_1} \quad \bullet \text{ NAR (g cm}^{-2} \text{ week}^{-1}\text{)} = \frac{W_2 - W_1}{A_2 - A_1} \times \frac{\ln(W_2) - \ln(W_1)}{t_2 - t_1}$$

Description:

ln = Natural logarithm

W₁ and W₂ = Plant dry weight at the 1st and 2nd observations, respectively

t₁ and t₂ = Plant age at the 1st and 2nd observations

A₁ and A₂ = Leaf area of plants in the 1st and 2nd observations

Ten plants were collected to assess the number of flowers, number of productive nodes, fresh weight of the plant (g), dry weight of the plant (g), number of pods per plant, pod length per plant (cm), percentage of empty pods per plant (%), pod weight per plant (g), and pod yield (tons ha⁻¹) during harvest 70-72 days post planting (stage R6). Pod yield of edamame's pod was calculated using the formula:

$$\bullet \text{ Pod yield (tons ha}^{-1}\text{)} = \frac{\text{Net pod weight per plot} \times 10,000 \text{ m}^2}{2 \text{ m} \times 2 \text{ m}}$$

The optimal dose of chicken manure in increasing edamame pod yield was determined through quadratic regression analysis. Based on the regression equation $y = -0.0283x^2 + 0.5355x + 8.944$, the optimal dose of chicken manure is indicated by the peak point of the parabolic curve and is calculated using the formula:

$$\bullet x = \frac{-b}{2a} = \frac{-0.5355}{2(-0.0283)}$$

The data were analyzed using an F-test at a 5% significance level, followed by Duncan's multiple range test (DMRT). Statistical analysis was conducted utilizing SPSS 27.0 (IBM® SPSS®, Chicago) and Microsoft® Office Excel 2021.

RESULTS AND DISCUSSION

Soil properties

The usage of chicken manure and humic acid can elevate the pH and nutritional levels in the soil. This can be seen from the results of soil analysis before treatment (Table 2) and post-application of chicken manure and humic acid with various doses (Figure 1). The combination of treatments can increase soil pH from slightly acidic to neutral. The content of organic C, total N, and available P increased with the addition of chicken manure and humic acid doses. Potassium potential increased from low to medium due to the application of chicken manure and humic acid doses.

Soil pH influences the solubility, mobility, and availability of nutrients, as well as their transfer inside plants. The dissociation of acidic functional groups caused by increased pH can result in an enhancement of soil organic matter content, facilitating the appropriate mineralization of nutrients. Moreover, an increase in pH to an optimal level can enhance microbial activity and soil respiration (Neina, 2019). The regulation of pH with the

addition of chicken manure and humic acid facilitates an environment conducive to the enhanced absorption of key nutrients, including carbon (C), nitrogen (N), phosphorus (P), potassium (K), and numerous micronutrients by plants. Chicken manure can be a source of organic matter and essential elements that significantly enhance soil nutrient content (Dikinya & Mufwanzala, 2010).

Table 2. Soil properties before treatment.

Soil properties	Method	Value	Status*
pH	H ₂ O	6.13	Slightly acidic
C-organic (%)	Walkley and Black	2.11	Medium
N-total (%)	Kjeldahl	0.23	Medium
P-available (ppm P ₂ O ₅)	Bray	78.78	High
P-potential (mg P ₂ O ₅ 100g ⁻¹)	HNO ₃ :HClO ₄	123.09	High
K-available (ppm K ₂ O ₅)	HNO ₃ :HClO ₄	337.95	High
K-potential (mg K ₂ O 100g ⁻¹)	HNO ₃ :HClO ₄	19.95	Low
CEC (cmol kg ⁻¹)	NH ₄ OAc	17.81	Medium

Note: *Criteria based on the Indonesian Ministry of Agriculture's criteria of soil properties

Without humic acid application (0 kg ha⁻¹), the P-available value was relatively low at all levels of manure application (Figure 1). This indicates the importance of humic acid as a supplement for chicken manure. The application of humic acid will help release P bound by soil metals so that they become available to plants (Setyawan & Setyawan, 2019). Humic acid can form chelates with iron (Fe), aluminum (Al), and calcium (Ca) in the soil solution, as well as to solubilize insoluble phosphate rock minerals such as FePO₄ (Rosolem et al., 2024). The nutrients supplied by this organic fertilizer are kept in the soil for an extended duration and utilized effectively during the plant growth phase (Rostaei et al., 2024). The combined impact of chicken manure and humic acid enhances the chemical properties of soil and increases nutrient availability.

Vegetative phase variables

Table 3 shows that without chicken manure, the provision of humic acid increased root length by 68.47% and RGR by 111.54%, and in combination with chicken manure, it increased by 82.49% and 288.46%, respectively. Chicken manure at a rate of 8 tons ha⁻¹ plus the highest dose of humic acid produced the longest root and highest RGR. Application of chicken manure increased the nitrogen and phosphorus in the soil (Figure 1). It is likely that increasing root length and RGR could be related to increasing availability of soil P and N. The provision of chicken manure can enhance the root length of soybean plants by up to 2 cm compared to those not given fertilizer (Bandyopadhyay et al., 2010). The application of chicken manure can directly provide nitrogen and phosphorus elements and increase the activity of soil microbes that contribute to the mineralization of nitrogen and phosphorus in the soil (Kacprzak et al., 2023). Humic acid decreases the absorption and precipitation of phosphorus in the soil. The complexation of humic acid with iron and aluminum can enhance phosphorus availability for plants (Rosolem et al., 2024; Salihi et al., 2024). An extensive root system marked by enhanced root length can significantly enhance the growth efficiency of edamame plants, particularly during the vegetative stage.

The relative growth rate (RGR) indicates the efficiency of plants in adding new dry biomass per unit of biomass that has been accumulated in a particular time unit. This parameter provides an overview of the efficiency of plants in using resources to produce new biomass (Hlushach & Avksentieva, 2024). The better RGR value is likely due to a better plant root system. Plants treated with the addition of 8 tons ha⁻¹ chicken manure and 90 kg ha⁻¹ humic acid had the highest root length, which was 25.95 cm (Table 3). Good nutrient absorption affects photosynthesis activity and biomolecule synthesis, which contribute to increasing plant biomass (Rostaei et al., 2024).

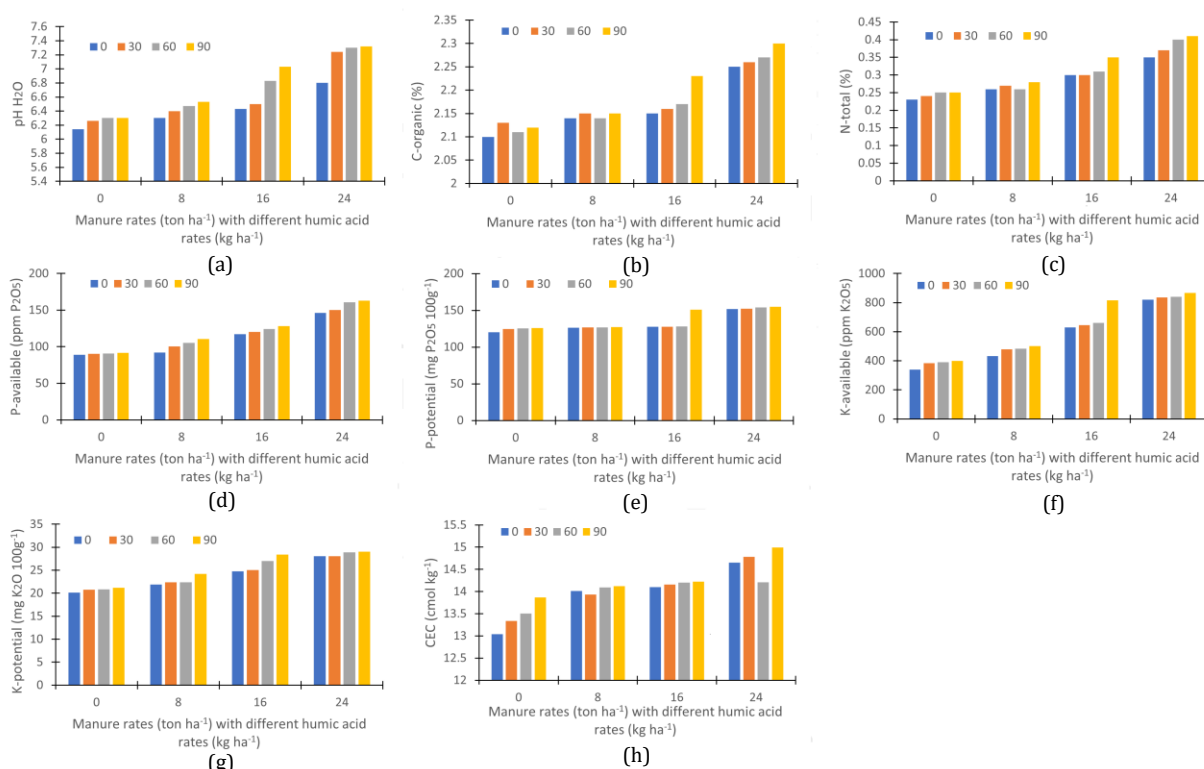


Figure 1. Effect of different chicken manure rates and humic acid rates on soil properties: (a) soil pH H₂O; (b) organic carbon (%); (c) total nitrogen (%); (d) available phosphorus (ppm P₂O₅); (e) potential phosphorus (mg P₂O₅ 100g⁻¹); (f) available potassium (ppm K₂O); (g) potential potassium (mg K₂O 100g⁻¹); (h) cation exchange capacity (cmol kg⁻¹).

Table 3. Root length and relative growth rate of the edamame from different chicken manure and humic acid treatments.

Humic acid (kg ha ⁻¹)	Chicken manure (tons ha ⁻¹)			
	0	8	16	24
Root length (cm)				
0	14.22±1.61h	23.48±0.18bcd	24.31±1.01b	20.13±0.28e
30	22.36±1.86cd	17.95±0.10fg	18.25±0.20fg	18.91±1.8efg
60	23.03±0.53bcd	19.20±0.28efg	21.93±0.07d	18.35±0.69fg
90	23.96±0.50bc	25.95±0.20a	17.83±0.53g	19.55±1.0ef
Relative growth rate (g g ⁻¹ week ⁻¹)				
0	0.26±0.05c	0.41±0.05bc	0.41±0.23bc	0.52±0.32bc
30	0.26±0.09c	0.75±0.54ab	0.25±0.17c	0.25±0.10c
60	0.32±0.08c	0.57±0.15bc	0.43±0.05bc	0.33±0.04c
90	0.55±0.21bc	1.01±0.22a	0.82±0.29ab	0.24±0.16c

Note: Values followed by the same letter were not significantly different according to DMRT at $\alpha = 5\%$; mean \pm standard error.

Plants given the highest doses of chicken manure and humic acid had the lowest RGR, i.e., 0.24 g g⁻¹ week⁻¹ (Table 3). The low RGR at the highest dose, which supplies approximately 535.2 kg N ha⁻¹, is likely due to excessive nitrogen causing nutrient imbalance (Table 1; Figure 1). In a single season, soybean uptake nitrogen 12.6-41.2 kg and 93.2-300.0 kg ha⁻¹ for biomass and seed, respectively, dependent on variety (Fatkhunnisa et al., 2025). Nitrogen is essential for plant growth, namely in promoting root length, diameter, branching, and biomass formation in the roots. However, excessive amounts of it will cause a physiological imbalance in plants so that growth is inhibited (Farhan et al., 2024).

The interaction of chicken manure and humic acid did not significantly influence leaf area, net assimilation rate, and leaf greenness index. The application of 8 tons ha⁻¹ of chicken manure produced the highest values for leaf area at 3 and 5 weeks after planting

(WAP), as well as for net assimilation rate and leaf greenness index. Meanwhile, the 90 kg ha⁻¹ dose of humic acid led to the maximum leaf greenness index (Table 4).

Table 4. Leaf area, net assimilation rate, and leaf green index of the edamame from different chicken manure and humic acid treatments.

Treatment	Leaf area (cm ²)		Net assimilation rate (g cm ⁻² day ⁻¹)	Leaf greenness index
	3 WAP	5 WAP		
Chicken manure (tons ha ⁻¹)				
0	18.78±3.17c	25.16±3.78b	0.162±0.78b	37.9±2.9c
8	24.05±3.74a	30.10±3.27a	1.152±0.771a	40.3±3.4ab
16	23.45±4.22a	30.57±3.39a	0.509±0.444b	39.5±2.3b
24	21.08±2.48b	25.03±3.93b	0.299±0.041b	41.6±2.5a
Pr>F	**	**	**	**
Humic acid (kg ha ⁻¹)				
0	21.54±4.53	26.90±4.30	0.473±0.291	37.8±3.4b
30	21.67±4.34	27.60±4.00	0.491±0.066	40.0±2.7a
60	21.97±3.52	27.78±4.04	0.273±0.088	40.4±2.1a
90	22.18±3.88	28.59±5.43	0.884±0.400	41.3±3.0a
Pr>F	ns	ns	ns	**

Note: Values followed by the same letter were not significantly different according to DMRT at $\alpha = 5\%$; ns-non-significant different, ** significant different at $\alpha = 1\%$; mean \pm standard error; WAP-weeks after planting

The positive effect of chicken manure application might relate to increasing availability of N, P, and K (Figure 1), which play a role in plant photosynthesis. Chicken manure contained water of 19.02%, C-organic of 20.36%, pH of 8.68, N-total of 2.23%, P₂O₅ total of 2.95%, and K₂O total of 2.27% (Table 1). Meanwhile, humic acid enhances nitrogen availability and uptake by improving health and promoting root growth. Nitrogen is an important component of chlorophyll, proteins, and enzymes that play an essential role in photosynthesis (Farhan et al., 2024). Magnesium is a core element in chlorophyll, which determines photosynthesis performance (Xie et al., 2024).

Generative phase variables

The combination of chicken manure and humic acid significantly influences the quantity of flowers and productive nodes. Without humic acid, the application of chicken manure at a dose of 8 tons ha⁻¹ increased the number of flowers by 33.5% and the number of productive nodes by 30.77%, while when combined with humic acid at a dose of 90 kg ha⁻¹, they increased 106.8% and 110.26%, respectively (Table 5). The enhancement in flower production may result from improved nutrient availability, particularly phosphorus, which is crucial for floral growth (Zhao et al., 2021). Chicken manure provides nutrients like phosphorus faster than other manures (Kamaluddin et al., 2022).

The enhancement in the number of flowers and productive nodes is due to humic acid significantly increasing the availability of phosphorus in the soil compared to chicken manure alone. Figure 1 shows that at all levels of chicken manure doses, the P-available value increased along with the increase in humic acid doses. Humic acid chelates metal ions, releasing the phosphorus; the humic acid is able to chelate metal ions due to having aromatic and lipid rings, which are connected to carboxyl, hydroxyl, and carbonyl groups (Xiong et al., 2023). Boron, alongside phosphorus, is a vital element for the processes of blooming and fruit development (Azizah & Rosantika, 2023). The utilization of humic acid in organic soil has demonstrated efficacy in enhancing boron absorption via the production of tetrahedral complexes with boric acid (Goli et al., 2019).

Table 5. Number of flowers and number of productive nodes of the edamame from different chicken manure and humic acid treatments.

Humic acid (kg ha ⁻¹)	Chicken manure (tons ha ⁻¹)			
	0	8	16	24
Number of flowers				
0	20.6±2.4b	27.5±6.6b	27.8±6.5b	34.7±2.5b
30	31.0±8.4b	23.2±4.0b	38.5±2.6b	25.7±5.4b
60	35.2±2.9b	42.2±4.8a	38.2±2.5b	26.2±5.6b
90	27.0±6.1b	42.6±5.2a	42.4±4.9a	31.9±7.5b
Number of productive nodes				
0	3.9±0.7e	5.1±0.9cd	7.3±0.7ab	6.6±0.7bc
30	4.1±0.8de	5.5±0.7ab	7.4±0.8ab	4.8±1.1de
60	4.6±0.9de	8.0±0.8a	7.3±0.8ab	4.5±0.9de
90	4.9±0.7de	8.2±0.8a	8.1±0.8a	5.9±0.8cde

Note: Values followed by the same letter were not significantly different according to DMRT at $\alpha = 5\%$; mean \pm standard error.

Table 6 shows that without chicken manure, increasing the dose of humic acid from 0 to 24 kg ha⁻¹ resulted in an increase in fresh weight from 66.46 g to 94.87 g, and dry weight from 15.21 g to 32.81 g. With a dosage of 90 tons ha⁻¹ chicken manure and 8 kg ha⁻¹ humic acid, the fresh weight of the plant reached 140.62 g, the highest value among all treatments. Likewise, the highest dry weight was recorded at 46.02 g in the same combination. The RGR, as shown in Table 3, is closely related to the efficiency of photosynthesis. Plants that have a higher relative growth rate have a better ability to carry out photosynthesis (Simatupang et al., 2023).

Increasing biomass weight (Table 6) from the treatment of chicken manure + humic acid indicates enhanced photosynthetic efficiency. This aligns with the findings of Chen et al. (2024) that chicken manure improves soil fertility and soil microbial activity. Permatasari & Nurhidayati (2014) stated that soil microorganisms indirectly increase photosynthetic efficiency by supplying nutrients and producing phytohormones. Ampong et al. (2022) highlighted the significant function of humic acid in enhancing soil physical and biochemical activity through the improvement of structure, texture, water retention, and microbial population.

Table 6. Total fresh weight and total dry weight of the edamame plant at harvest from different chicken manure and humic acid treatments.

Humic acid (kg ha ⁻¹)	Chicken manure (tons ha ⁻¹)			
	0	8	16	24
Plant fresh weight (g)				
0	66.46±8.31h	83.01±0.98fgh	104.20±5.58cde	94.87±5.51def
30	75.95±1.81gh	115.97±3.48bc	105.27±13.78cde	114.71±15.31bc
60	83.51±2.63fgh	130.59±4.50ab	114.42±4.04bc	103.69±15.68cde
90	88.93±2.55efg	140.62±11.02a	107.74±20.10cd	101.98±6.21cde
Plant dry weight (g)				
0	15.21±3.03e	34.14±3.86bc	38.26±18.05ab	32.81±7.83bc
30	19.20±0.63de	41.19±0.63ab	32.71±6.97bc	31.03±2.07bc
60	24.66±1.98cde	33.69±6.67bc	30.16±2.63bc	33.72±3.63bc
90	29.78±1.24bcd	46.02±1.65a	31.50±5.98bc	30.45±3.93bc

Note: Values followed by the same letter were not significantly different according to DMRT at $\alpha = 5\%$; mean \pm standard error.

Edamame productivity

The combination of chicken manure and humic acid did not substantially influence the number of pods, pod length, and percentage of empty pods. Chicken manure level significantly affected empty pods. Application of the highest manure level resulted in the highest percentage of empty pods (Table 7). An excess nitrogen supply in chicken manure might cause this increase in empty pods. The application of chicken manure with a dose of 24 tons ha⁻¹ has excess nitrogen content (Figure 1).

Table 7. Number of pods, pod length, and percentage of empty pods of the edamame from different chicken manure and humic acid treatments.

Treatment	Number of total pods	Pod length (cm)	Empty pods (%)
Chicken manure (tons ha ⁻¹)			
0	28.0±7.5	5.96±0.19	15.26±6.39ab
8	31.2±9.4	5.98±0.17	12.24±5.52b
16	28.8±8.6	5.92±0.11	15.87±5.46ab
24	27.9±8.0	5.97±0.21	22.13±8.50a
Pr>F	ns	ns	*
Humic acid (kg ha ⁻¹)			
0	24.6±4.5	5.94±0.11	13.83±7.16
30	31.4±10.3	5.95±0.19	13.65±10.16
60	29.9±5.7	6.01±0.16	18.61±2.83
90	30.6±10.0	5.94±0.22	19.40±10.95
Pr>F	ns	ns	ns

Note: Values from the same column followed by the same letter were not significantly different according to DMRT at $\alpha = 5\%$; ns-non-significant difference, * significant difference at $\alpha = 5\%$; mean \pm standard error.

Excess nitrogen levels usually stimulate excessive vegetative growth in plants, resulting in an increase in the number of leaves and stems, while simultaneously inhibiting seed formation and development within pods (Farhan et al., 2024). Consequently, the plant failed to fulfill all pods and leading to a high incidence of empty pods. In addition, excess organic matter from high doses of manure might cause nutrient imbalances. This can interfere with absorbing important nutrients such as P and K, which are essential for seed formation and development (Yusdian et al. 2025).

The application of 8 tons ha⁻¹ of chicken manure and 90 kg ha⁻¹ of humic acid resulted in the highest pod weight per plant (121.41 g). Without humic acid, the provision of chicken manure increased pod weight per plant by 74.96%, while when combined with humic acid, it was 293.87% (Table 8).

Table 8. Pod weight of the edamame from different chicken manure and humic acid treatments.

Humic acid (kg ha ⁻¹)	Chicken manure (tons ha ⁻¹)			
	0	8	16	24
Pod weight per plant (g)				
0	30.82±0.74g	53.93±3.56ef	72.44±5.53cde	63.47±3.21cdef
30	43.59±9.58fg	82.73±10.89bcd	71.47±12.21cde	66.47±9.21cde
60	56.67±2.22ef	95.39±4.94b	83.91±14.26bc	62.68±9.41def
90	60.26±0.47ef	121.41±14.47a	63.41±19.28cdef	69.34±22.53cde

Note: Values followed by the same letter were not significantly different according to DMRT at $\alpha = 5\%$; mean \pm standard error.

Pod yield increased with increasing humic acid (Figure 2). A higher dose of humic acid resulted in a lower optimum dose of chicken manure. The equation in Figure 2 shows that 90 kg ha⁻¹ of humic acid in combination with 9.5 tons ha⁻¹ of chicken manure resulted in optimum dosages for pod yield. It was a consequence of the better vegetative growth of such treatments. Previous reports found that manure application enhances edamame yield by supplying important nutrients such as N, P, and K, while also enhancing soil organic carbon, hence facilitating soil aggregation formation (Melati et al., 2025).

Humic acid had a synergistic effect with manure to increase soil fertility. Humic acid increases soil nutrient availability through the mechanism of activating the H⁺-ATPase pump in the plasma membrane. Moreover, humic acid directly facilitates the co-transport of nutrients across the plasma membrane. Aromatic and aliphatic groups in humic acid have essential physiological roles in stimulating root growth, increasing nitrogen absorption, and encouraging the production of soluble sugars (Ampong et al., 2022). It is important in the future to evaluate the mechanism of how humic acid facilitates nutrient

availability in the presence of manure, like chicken manure, to establish sustainable organic edamame production.

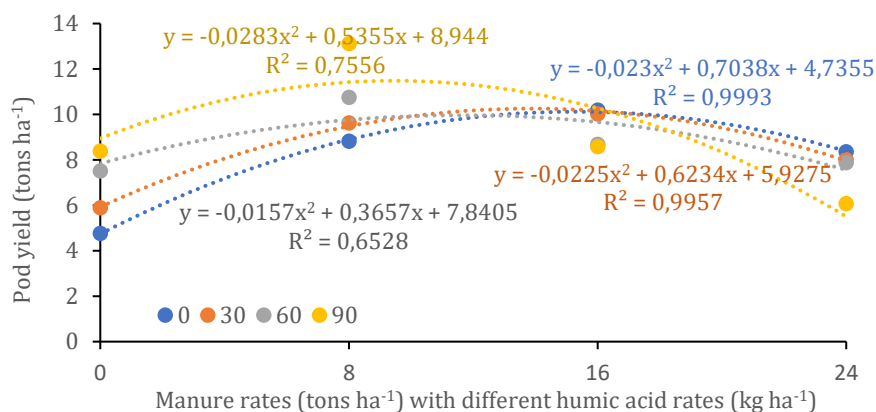


Figure 2. Pod yield from the interaction of chicken manure and humic acid doses

CONCLUSIONS

The dose of 8 tons ha⁻¹ chicken manure and 90 kg ha⁻¹ humic acid increased root length, relative growth rate, number of flowers, number of productive nodes, plant fresh weight, plant dry weight, pod weight per plant, and pod yield. The application of chicken manure at 8 tons ha⁻¹ significantly increased leaf area, net assimilation rate, and leaf greenness index, while decreasing the percentage of empty pods per plant. The application of humic acid 60 kg ha⁻¹ as a single treatment increased the leaf greenness index. Chicken manure at 9.5 tons ha⁻¹ and humic acid at 90 kg ha⁻¹ were the optimum doses to significantly increase pod yield. The utilization of chicken manure and humic acid enhanced nutrient availability for the growth and production of organic edamame.

ACKNOWLEDGEMENTS

The authors acknowledge Lembaga Pengelolaan Dana Pendidikan (LPDP) for financing this research entitled "Plant growth, productivity, and isoflavone content of edamame (*Glycine max* (L.) Merr.) with the application of chicken manure and humic acid".

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