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The Effects of Amino Acids Application Techniques on Tobacco (*Nicotiana tabacum* L.) Plant Growth

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Abstract: This study aims to determine the effect of amino acids application techniques on the growth of tobacco plants. These research was carried out at the TEFA (Teaching Factory) of the Nursery, Department of Agricultural Production, Politeknik Negeri Jember. The study used a Randomized Block Design (RBD) with 1 treatment factor, namely: 1) without amino acids (control); 2) amino acids 500 ml/polybag by pouring; 3) amino acids 500 ml/polybag by spraying. Observed parameters of tobacco plant growth were as follows: 1) plant height, 2) leaves number; 3) stem diameter; 4) plant wet weight, and 5) plant dry weight. Observational data were analyzed using the F 5% test, and the LSD 5% test. The result was the amino acids application technique significantly affected the parameters of plant height, number of leaves, and stem diameter of tobacco plants. No significant differences were found in the parameters of plant base weight and dry weight. The amino acid application technique by spraying gave better results on stem diameter, wet weight, and dry weight of plants, while the application by pouring gave better results on plant height and number of leaves of tobacco plant.

Keywords: Amino Acids, Application Techniques, Growth, Tobacco

1. Introduction

Tobacco (*Nicotiana tabacum* L.) is one of the most important plantation crops due to its substantial contribution to farmers' income and national revenue through excise and export activities. Tobacco productivity and quality are mainly determined by leaf yield and chemical composition, which are influenced by the interaction of genetic factors, environmental conditions, and cultivation practices (Huan et al., 2024). Among agronomic practices, fertilization plays a crucial role because tobacco is highly responsive to nutrient availability, particularly nitrogen (N), phosphorus (P), and potassium (K), which regulate vegetative growth, leaf expansion, biomass accumulation, and leaf quality attributes (Paramesh *et al.*, 2023).

Optimal tobacco growth requires a balanced supply of essential nutrients in the soil. Nitrogen is essential for chlorophyll formation and vegetative growth, phosphorus is involved in cell division and root development, while potassium plays an important role in stomatal regulation, translocation of photosynthates, and improvement of tobacco leaf quality, including color and elasticity (Fathi, 2022). Improper fertilization practices, either in terms of dosage or application technique, may result in suboptimal growth, reduced leaf quality, and increased susceptibility of tobacco plants to biotic and abiotic stresses.

In addition to fertilizer type and rate, fertilizer application techniques are a critical aspect in improving nutrient uptake efficiency in tobacco cultivation. Fertilizer application techniques include soil application, foliar application, split application, and localized placement near the root zone. Appropriate application techniques can increase

nutrient availability in the rhizosphere, reduce nutrient losses through leaching or volatilization, and enhance fertilizer use efficiency (Prajapati et al., 2025).

In tobacco cultivation, soil fertilization is commonly applied as basal and top dressing, especially for macronutrients, whereas foliar fertilization is often used to supply micronutrients or as a complementary practice during periods of intensive growth. Previous studies have demonstrated that the appropriate combination of fertilizer application techniques can improve vegetative growth, increase leaves number and leaves area, and enhance both yield and cured leaves quality of tobacco (Xing et al., 2025). Therefore, a comprehensive understanding of effective and efficient fertilizer application techniques is essential to support increased productivity and sustainability of tobacco production.

2. Materials and Methods

These research was carried out at the TEFA (Teaching Factory) of the Nursery, Department of Agricultural Production, Politeknik Negeri Jember. While the research time is June to November 2025. The research materials included tobacco seeds ready to planting, amino acids from lemuru fish, planting media, polybags, and inorganic fertilizers. The tools used are cultivation, measuring, and identification tools.

The study used a Randomized Block Design (RBD) with 1 treatment factor, namely: 1) without amino acids (inorganic fertilizer (SOP dose) = 6.9 grams N + 5.4 grams P + 6.9 grams K per plant) ; 2) amino acids 500 ml/polybag by pouring (inorganic fertilizer (30% of SOP dose) = 2.3 grams N + 1.8 grams P + 2.3 grams K per plant); and 3) amino acids 500 ml/polybag by spraying (inorganic fertilizer (1/3 SOP dose) = 2.3 grams N + 1.8 grams P + 2.3 grams K per plant). Each treatment was repeated four times to obtain 25 experimental units (10 plants each).

Observed parameters of tobacco plant growth were as follows: 1) plant height, 2) leaves number; 3) stem diameter; 4) plant wet weight, and 5) plant dry weight. Observational data were analyzed using the F5% test, and the results were significantly different, then they will be tested further with the LSD 5% test.

3. Results and Discussion

The analysis of variance showed that amino acids application techniques presented in Table 1.

Table 1. Analysis of variance of tobacco growth parameters

Parameters	Plant Age (dap)	F-test	F 0.05	F 0.01	Notation	CV (%)
Plant height	45	4.55	3.63	6.23	*	17.29
	60	8.10			**	10.34
	75	6.47			**	14.56
Leaves number	45	8.17			**	5.91
	60	2.30			ns	9.92
	75	6.84			**	6.19
Stem diameter	45	7.65			**	8.13
	60	6.27			**	10.64
	75	5.39			*	10.58
Plant wet weight	80	0.71			ns	12.99
Plant dry weight	80	1.17			ns	18.90

Description: ns: not significant; *: significant at 0.05; **: significant at 0.01; dap: days after planting.

Table 1 showed that amino acids application techniques had a very significant effect on the plant height 60 and 75 days after planting (dap), on the leaves number 45 an 75

dap, and on the stem diameter 45 and 60 dap. Amino acids application techniques had a significant effect on the plant height 45 dap, and the stem diameter 75 dap, and had no significant effect on leaves number 60 dap and the wet weight and plant dry weight of plants.

3.1. Plant Height of Tobacco Plant

Based on Table 1, the analysis results for plant height parameters show a very significant difference. Further testing with 5% LSD is presented in Table 2.

Table 2. The Effect of Amino Acids Application Techniques on Tobacco Plant Height.

Treatment	Plant Height (cm)		
	45 dap	**60 dap**	**75 dap**
Amino acids by pouring	5.22 a	19.26 A	71.67 a
Amino acids by spraying	4.91 a	18.81 A	67.33 a
Without amino acids (control)	4.09 b	16.00 B	56.11 b
LSD 5%	0.82	1.86	9.47

dap = days after planting. Unequal letters in the same column indicate significant differences within each variable, according to LSD test ($p<0.05$).

Based on Table 2 it can be seen that administering amino acids by pouring and spraying is significantly different, and produces better plant height compared to not administering amino acids. This is proven by the increase in the height of tobacco plants with amino acids poured (71.61 cm) and sprayed (67.33 cm), compared to the control (56.11 cm) at plant age 75 dap.

The amino acids provided come lemuru fish, which contains free amino acids and short-chain peptides, with glutamic acid, aspartic acid, alanine, glycine, and proline commonly dominating the composition (Mahrus et al., 2023). These compounds are readily absorbed by plants and function as sources of organic nitrogen, plant biostimulants, and natural chelating agents for micronutrients. Therefore, lemuru fish protein hydrolysates have strong potential as raw materials for the production of amino acid–based fertilizers, particularly within sustainable and environmentally friendly agricultural systems (Sun et al., 2024).

Plant height is an important indicator of vegetative growth, reflecting the processes of cell division and cell elongation, particularly in stem meristem tissues. Fertilizer application techniques significantly influence plant height because they determine nutrient availability, distribution, and uptake efficiency by plant roots and tissues. Soil application of fertilizers, especially when nutrients are placed near the root zone, enhances the availability of macronutrients such as nitrogen (N), phosphorus (P), and potassium (K), which play direct roles in stem growth. Nitrogen is a key component of amino acids and proteins that support cell division and internode elongation, leading to increased plant height. Studies have shown that proper soil application of nitrogen fertilizers results in significantly taller plants compared to surface broadcasting or poorly targeted applications (Barłóg et al., 2022; Ali et al., 2025). Whereas foliar fertilizer application serves as a supplemental nutrient supply, particularly for micronutrients and low concentrations of nitrogen that can be rapidly absorbed by leaf tissues. Foliar fertilization during active growth stages has been shown to enhance photosynthetic activity and metabolic processes, which support stem elongation and contribute to increased plant height, especially under soil nutrient limitations (Chandra et al., 2025).

3.2. Leaves Number of Tobacco Plant

Leaves number is an important indicator of vegetative growth because it is directly related to photosynthetic capacity and biomass production potential. The application of amino acids, either through soil or foliar application, has been reported to increase leaves

number in various plant species through several physiological and biochemical mechanisms. Specifically in tobacco plants, the number of leaves determines the amount of production, because tobacco leaves are the part of the plant that is harvested and has high economic value. The parameter data for the number of leaves and the results of the LSD 5% further tests can be seen in Table 3.

Table 3. The Effect of Amino Acids Application Techniques on Tobacco Leaves Number

Treatment	Leaves Number (sheets)		
	45 dap	60 dap	75 dap
Amino acids by pouring	6.78 a	9.70	19.30 A
Amino acids by spraying	6.59 a	9.52	19.04 A
Without amino acids (control)	6.07 b	8.81	17.44 B
LSD 5%	0.38		1.15

dap = days after planting. Unequal letters in the same column indicate significant differences within each variable, according to LSD test ($p<0.05$).

Table 3 shows that the administration of amino acids, whether by dripping or spraying, was significantly different and gave better results on the number of leavess compared to the control. This is proven by the increase in the leaves number of tobacco plants with amino acids poured (19.30 sheets) and sprayed (19.04 sheets), compared to the control (17.44 sheets) at plant age 75 dap.

Amino acids act as a readily available source of organic nitrogen for plants. Nitrogen is a key element in the synthesis of structural proteins and enzymes that support cell division in the shoot apical meristem. Enhanced cell division activity at the growing point promotes faster leaf initiation, resulting in an increased number of leaves per plant (Fathi, 2022; Bryant, 2025). Certain amino acids such as glycine, glutamic acid, and aspartic acid play important roles in chlorophyll synthesis and improve photosynthetic efficiency. Enhanced photosynthesis increases the availability of energy and assimilates required for leaf initiation and expansion, thereby stimulating vegetative growth and increasing leaves number (Sowmya et al., 2023; Rahman et al., 2024). Besides that amino acid–based fertilizers often contain peptides and other biostimulant compounds that may stimulate the activity of endogenous plant hormones such as auxins and cytokinins. Cytokinins play a crucial role in promoting cell division and leaf differentiation, thereby contributing to an increased number of leaves (Heidarzadeh, 2025).

3.3. Stem Diameter of Tobacco Plant

The analysis results showed that the amino acid application technique significantly affected the stem diameter parameter (Table 1). Furthermore, the results of the 5% LSD test are presented in Table 4.

Table 4. The Effect of Amino Acids Application Techniques on Tobacco Stem Diameter

Treatment	Stem Diamater (cm)		
	45 dap	60 dap	75 dap
Amino acids by pouring	0.99 a	1.41 a	2.03 A
Amino acids by spraying	0.99 a	1.32 ab	1.92 Ab
Without amino acids (control)	0.87 b	1.18 b	1.72 B
LSD 5%	0.08	0.14	0.20

dap = days after planting. Unequal letters in the same column indicate significant differences within each variable, according to LSD test ($p<0.05$).

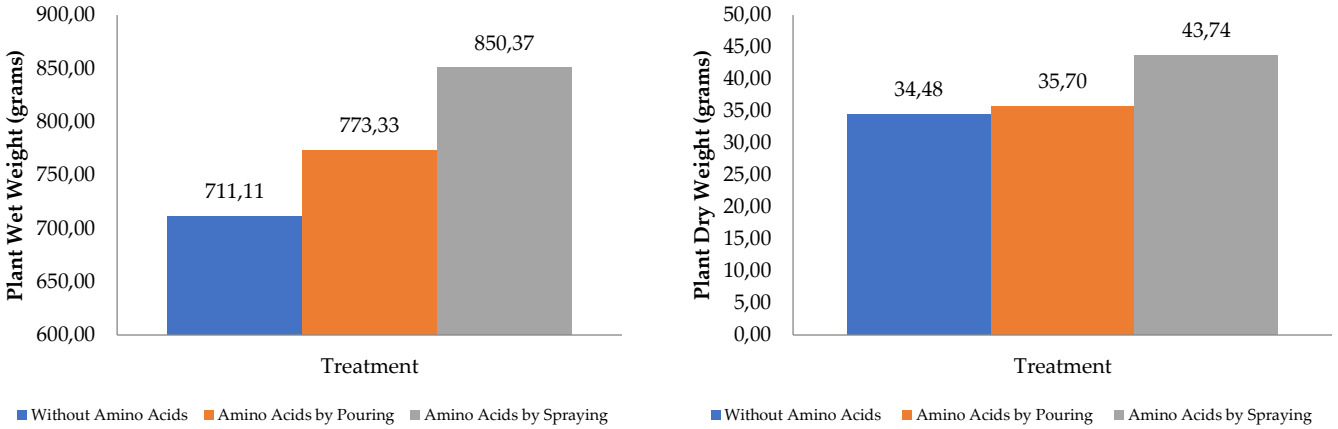
The result of the LSD 5% test presented in Table 4 shows that the amino acid application technique by spraying tends to have a better effect on the diameter of tobacco plant stems, followed by the application technique by pouring and without amino acids,

at plant age 45, 60, and 75 dap. Fertilizer application techniques play a crucial role in influencing plant stem diameter growth by regulating nutrient availability, uptake efficiency, and spatial distribution within the root and shoot systems. Proper fertilizer placement and application methods enhance the absorption of essential macronutrients which are directly involved in cambial activity, cell division, and cell enlargement in stem tissues.

Soil-applied fertilizers, when positioned within the effective root zone, promote continuous nutrient uptake that supports steady stem thickening (Wen-xuan, et.al., 2023). Meanwhile, foliar fertilizer application provides rapid nutrient absorption through leaf surfaces, accelerating physiological processes such as photosynthesis and assimilate translocation to the stem, particularly during active vegetative growth stages (Bai, et.al., 2024). The use of liquid fertilizers enriched with amino acids has also been reported to enhance nitrogen assimilation, enzymatic activity, and protein synthesis, thereby stimulating cell proliferation and stem tissue strengthening. Collectively, these fertilizer application techniques optimize nutrient utilization and physiological performance, resulting in increased stem diameter and improved structural stability of plants (Wang, et.al., 2025).

3.4. Plant Wet Weight and Dry Weight of Tobacco Plant

Data on the effect of application techniques on the wet weight and dry weight parameters of tobacco plants can be seen in Figure 1.



(a) (b)

Figure 1. Plant wet weight (a) and plant dry weight (b) of tobacco plant

Figure 1 shows that the amino acid application technique by spraying tends to be better than the application by pouring and without amino acids, both in terms of wet weight and dry weight parameters of tobacco plant, that is 850.37 grams (wet weight plant) and 43.74 grams (dry weight plant). Amino acid application techniques significantly influence plant fresh weight and dry weight by affecting the rate of absorption, metabolic efficiency, and biomass accumulation. Foliar application of amino acids enables rapid uptake through leaf surfaces, enhancing chlorophyll synthesis, photosynthetic activity, and cellular osmotic regulation, which in turn increases water absorption and cell expansion, resulting in higher fresh weight (Henderson, et.al., 2025). In contrast, soil application supports sustained amino acid availability in the rhizosphere, improving nitrogen assimilation and root-mediated nutrient uptake, thereby contributing to long-term biomass formation (Heneron, et.al., 2020). Amino acids act not only as organic nitrogen sources but also as metabolic signals that stimulate enzyme activity, protein synthesis, and carbon–nitrogen metabolism, leading to increased accumulation of structural carbohydrates and proteins (Heidarzadeh, 2025).

The spraying application of amino acids gives better results, presumably because amino acids can be absorbed quickly through the stomata and leaf cuticles, thus directly involved in the physiological processes of the plant. Amino acids by spraying enhance chlorophyll synthesis and photosynthetic rate, leading to increased cell expansion and tissue water content, which results in a higher fresh weight of plant (Mirtaleb, 2021). Amino acids also play an important role in regulating cellular osmotic pressure and improving water use efficiency, which is particularly critical for tobacco during active vegetative growth stages. The enhancement of photosynthetic activity and nitrogen metabolism following foliar amino acid application also promotes the accumulation of photosynthates in the form of carbohydrates and structural proteins. This process directly contributes to an increase in dry weight, reflecting greater biomass accumulation of plants (Ouyang, et.al., 2025).

This study used the inorganic fertilizer dosage according to the SOP for fertilizing tobacco plants in the control (without amino acids), namely 6.9 grams N + 5.4 grams P + 6.9 grams K per plant. Meanwhile, in the treatment of giving amino acids either by pouring or spraying, the inorganic fertilizer dosage given was 30% of the dosage according to the SOP, namely 2.3 grams N + 1.8 grams P + 2.3 grams K per plant. Tobacco plant growth data analysis, including plant height, number of leaves, stem diameter, wet weight and dry weight of the plant, shows that amino acids treatment with a reduction in the use of inorganic fertilizer by 70% of the dose according to SOP, proves that amino acids, whether given by pouring or spraying, are able to replace the role of inorganic fertilizer by 70% of the SOP dose.

4. Conclusions

Based on the results and discussion, it can be concluded that the amino acid application technique significantly affected the parameters of plant height, number of leaves, and stem diameter of tobacco plants. No significant differences were found in the parameters of plant base weight and dry weight. The amino acid application technique by spraying gave better results on stem diameter, wet weight, and dry weight of plants, while the application by pouring gave better results on plant height and number of leaves.

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