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Influence of local potential plant biodiversity as green manure on soil total N, N uptake, and chlorophyll content of rice plants

SUNTORO SUNTORO[1,♥], GANJAR HERDIANSYAH[1], HERY WIDIJANTO[1], HAIKAL RAFI WARDHANA[1], AZHAR DIMAS TJAHJANTO[1], CHELYNA PUSPITASARI[1], EKO AMIADJI JULIANTO[2], MAROETO[3]

[1]Department of Soil Science, Faculty of Agriculture, Universitas Sebelas Maret. Jl. Ir. Sutami 36A, Surakarta 57126, Central Java, Indonesia. Tel.: +62-271-632477, ♥email: suntoro@staff.uns.ac.id

[2]Faculty of Agriculture, Universitas Pembangunan Nasional Veteran Yogyakarta. Jl. Ring Road Utara No. 104, Sleman 55281, Yogyakarta, Indonesia

[3]Faculty of Agriculture, Universitas Pembangunan Nasional Veteran Jawa Timur. Jl. Rungkut Madya No. 1, Surabaya 60294, East Java, Indonesia

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Abstract. *Suntoro S, Herdiansyah G, Widijanto H, Wardhana HR, Tjahjanto AD, Puspitasari C, Julianto EA, Maroeto. 2024. Influence of local potential plant biodiversity as green manure on soil total N, N uptake, and chlorophyll content of rice plants. Biodiversitas 25: 1655-1662.* Continuous use of chemical fertilizers in the agricultural sector causes several problems. One of several solutions that can be offered is using green manure, which could restore soil fertility and be way more sustainable for the environment. A few criteria for plants to be made into green manure are having a fast decomposition process and a low C/N ratio. This research aims to discover local potential green manure based on nutrient content and response to rice plants' soil total N, N uptake, and chlorophyll content. This research used the experiment method with a single factor Randomized Completely Block Design. There were 10 treatments with 3 replications, namely P0: control; P1: NPK 200 kg/ha; P2: Rice straw 10 tons/ha; P3: *Chromolaena odorata* (siam weed) 10 tons/ha; P4: *Ipomoea* spp. (morning glory) 10 tons/ha; P5: *Eichhornia crassipes* (water hyacinth) 10 tons/ha; P6: NPK 100 kg/ha + Rice straw 5 tons/ha; P7: NPK 100 kg/ha + *C. odorata* 5 tons/ha; P8: NPK 100 kg/ha + *Ipomoea* spp. 5 tons/ha; P9: NPK 100 kg/ha + *E. crassipes* 5 tons/ha. The results showed that *C. odorata*, *Ipomoea* spp., and *E. crassipes* have the potential to be developed as green manure because they have high N nutrient content and low C/N ratio. These three green manures can balance the NPK fertilizer and significantly increase soil total-N content, N uptake, and chlorophyll levels in rice plants. They also provide N nutrients, N uptake, and chlorophyll levels in rice plants.

Keywords: Chlorophyll, green manure fertilizer, N uptake, total N

INTRODUCTION

Continuous use of chemical fertilizers in the agricultural sector causes several problems, such as environmental pollution, high production costs, and effects on human health (Tao et al. 2016). Green manure is a more sustainable approach to increasing plant productivity (Tao et al. 2016). Green manure, including unpolluted organic matter, is less toxic and easily decomposed without leaving hazardous residue in land, water, and air. It is also safe for the environment and does not leave residue on food products. Applying green manure becomes one solution to restore soil fertility, especially in soils with a decreasing organic matter content due to land cultivation and the continuous use of chemical fertilizers (Adekiya et al. 2019). The limited availability of organic matter in research locations becomes a problem in organic farming practices, so it is essential to develop the potential of organic materials. One source of organic material that can be developed is green manure.

Green manure can act as a soil conditioner and a source of nutrition for plants. Green manure can also reduce negative environmental impacts (Cherr et al. 2006). The criteria for plants that can be used as green manure are that they have a low C/N ratio, lignin, and polyphenols, have a fast decomposition process, and are a source of nutrients available to plants (Aboyeji 2019). Green manure releases Nitrogen (N), which can meet the needs of plants after undergoing the decomposition process (Gao et al. 2020). Applying high levels of inorganic fertilizers can cause a significant loss of soil nutrients, affecting groundwater quality and soil quality. Green manure contains large amounts of N, but the nutrients are released more slowly, resulting in a more stable N source for plants (Talgre et al. 2012). Decomposition of green manure also increases the amount of nutrients and benefits the plant's rhizosphere (Zhang et al. 2017). Applying green manure has an advantage over other organic manures because it can be incorporated during land preparation. Applying green manure can increase the water-holding capacity, cation exchange capacity, and soil pH buffer. Green manure can reduce Nitrate (NO_3^-) loss through leaching, and the dose of inorganic N fertilizer is sufficient to meet plant needs (Latt et al. 2009). Green manure is a good agricultural management practice because it can stimulate plant growth and soil microbial activity and increase soil fertility (Jadhao et al. 2019).

Many river basins in Java experience land fertility degradation (Maroeto et al. 2017). Rice soil fertility is degraded due to low levels of soil organic matter (Suntoro et al. 2020). The soil type at the research location is Vertisols. Vertisols have high nutrient reserves but are absorbed by clay, so their availability for plants is low

(Sudadi et al. 2020). The growth of rice plants is greatly influenced by the availability of soil nutrients, especially Nitrogen, which is needed in large quantities. Nitrogen is essential nutrient, and it is the main component in the structure of chlorophyll. The N content in plants has a very close relationship with the chlorophyll content of plants (Fathi 2022). Nitrogen application increases leaf chlorophyll, dry weight, and seed yield (Muhammad et al. 2022). Nitrogen nutrients have a mobile property in the natural environment and are easily lost through various causes such as denitrification, volatilization, chemical and microbial fixation, and leaching (Kumar et al. 2014). Nitrogen deficiency in plants can inhibit plant growth and development, reduce photosynthesis and leaf area, and finally reduce the productivity of the plant (Mu and Chen 2021). An issue in Vertisols soil requires fertilization activities to increase nutrients. One attempt that can be made is organic fertilization using green manure. The rice variety used was Inpari 32. Inpari 32 is an improvement of Ciherang for its resistance to Bacterial Blight (BLB) disease and is more resistant to rice blast disease. The productivity of the Inpari 32 reached 7.2 tons/ha (Agustian et al. 2022).

The type of green manure used can be adapted from local potential, making it easier for farmers to obtain. There are four sources of organic material from potential plants that are abundant in the research area, namely rice straw (*Oryza sativa*), *Chromolaena odorata* (siam weed), *Ipomoea* spp. (morning glory), and *Eichhornia crassipes* (water hyacinth). *C. odorata* is a weed that is easily found on abandoned land and on roadsides. The most environmentally friendly way to control this weed is to utilize it for productive purposes in agriculture (Manjappa 2014). *C. odorata* has a total N content of 3.4% and a C/N ratio of 11:1 (Hamdani et al. 2017). *C. odorata* is a plant that grows rapidly, has high biomass, and contains 2.56% N (Okalia et al. 2022). *Ipomoea* spp. is a wild plant that usually grows in agricultural wetlands, including marginal wetlands. This plant is green manure and can act as a biopesticide (Sutarman et al. 2022). *Ipomoea* spp. (morning glory) has a total N content of 0.74% based on fresh weight (Rathore et al. 1993). Fresh *E. crassipes* has a total N content of 0.04% and organic matter of 3.5%, while in dry conditions, it has a total N content of 1.5% and organic matter of 75.8% (Jafari 2010). It is hoped that local potential green manure can be a solution to overcome nutrient deficiencies, especially N, in Vertisols soil. Green manure can be applied to various soil types, with different results in providing nutrient availability. This research aims to discover local potential green manure based on nutrient content and response to rice plants' soil total N, N uptake, and chlorophyll content.

MATERIALS AND METHODS

Study area

This research was conducted in Tegalsari Village, Weru District, Sukoharjo Regency, Central Java. The type of soil in this area is Vertisols. Soil and plant analysis were conducted at the Soil Chemistry and Fertility Laboratory, Faculty of Agriculture, Sebelas Maret University, Surakarta.

Procedures

Research method

The research used an experimental method with a Randomized Completely Block Design (RCBD), which included 10 treatments and 3 replications to obtain 30 test plots (Figure 1). The plots measured 3 × 2.5 m. Rice plants were planted with a plant spacing of 25 × 25 cm. The variety of rice used was Inpari 32. The different treatments used are shown in Table 1.

Land tillage is done by plowing the land until it becomes muddy and leveling. Green manure is chopped into 3-5 cm pieces and applied after tillage, 7 days before planting. NPK fertilization was carried out twice at 7 and 21 Days After Planting (DAP). Plant maintenance includes irrigation, weeding, replanting, and controlling pests and diseases. Soil and plant samples were taken at the maximum vegetative phase (60 days old). Soil samples were taken from each plot (30 samples) and plants, including the wet and dry weight of the top of the plants and roots.

Soil and plant analysis

Soil analysis includes pH (electrometric method) (Balai Pengujian Standar Instrumen Tanah dan Pupuk 2023), Cation Exchange Capacity (CEC) (ammonium acetate extraction method) (Balai Pengujian Standar Instrumen Tanah dan Pupuk 2023), Organic C (Walkley and Black Method) (Balai Pengujian Standar Instrumen Tanah dan Pupuk 2023), and Soil Total N (Kjeldahl Method) (Balai Pengujian Standar Instrumen Tanah dan Pupuk 2023). Plant analysis includes plant tissue N (Kjeldahl method) (Balai Pengujian Standar Instrumen Tanah dan Pupuk 2023). Chlorophyll Levels (Arnon Method) (Arnon 1949). Nutrient uptake of the upper plant (shoot) is obtained by multiplying the upper plant's dry weight by the upper part's nutrient content.

Table 1. Treatments used in the study

Code	Treatments	Dose			
		Green manure		NPK	
		(kg/ha)	(kg/plot)	(kg/ha)	(kg/plot)
P0	Control	0	0	0	0
P1	NPK	0	0	200	0.15
P2	Rice straw	10,000	7.5	0	0
P3	*C. odorata*	10,000	7.5	0	0
P4	*Ipomoea* spp.	10,000	7.5	0	0
P5	*E. crassipes*	10,000	7.5	0	0
P6	NPK + Rice straw	5,000	3.75	100	0.075
P7	NPK + *C. odorata*	5,000	3.75	100	0.075
P8	NPK + *Ipomoea* spp.	5,000	3.75	100	0.075
P9	NPK + *E. crassipes*	5,000	3.75	100	0.075

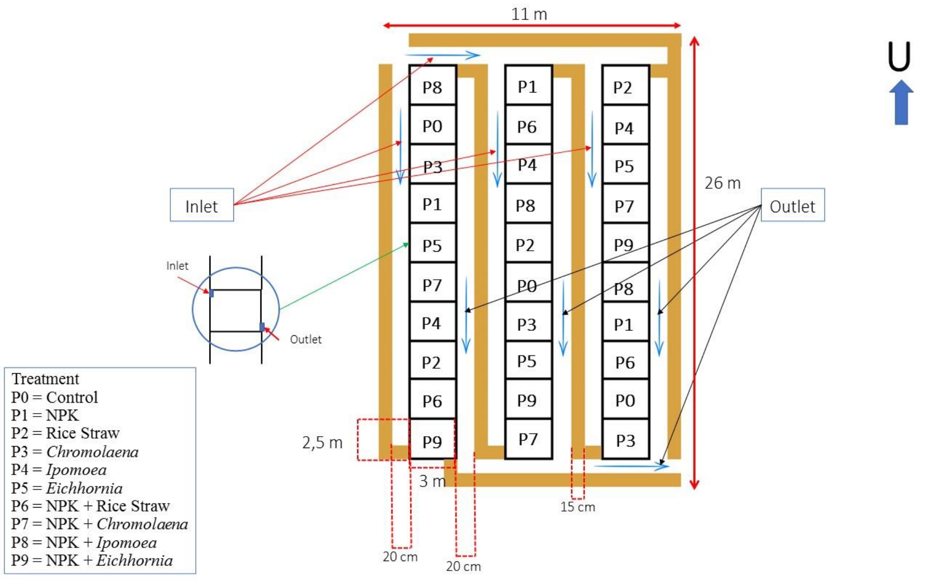

Figure 1. Experimental layout

Table 2. Soil characteristics

Analysis	Value	Classification
pH H_2O	7.70	Slightly alkaline
Organic C (%)	1.10	Low
KTK (me/100g soil)	31.11	High
Total N (%)	0.10	Low

Note: Classification according to Balai Pengujian Standar Instrumen Tanah dan Pupuk (2023)

Table 3. Green manure characteristics

Analysis	Rice straw	*C. odorata*	*Ipomoea* spp.	*E. crassipes*
Organic C (%)	37.14	36.66	36.28	38.42
N (%)	1.31	3.38	3.54	2.92
C/N	28.35	10.84	10.24	12.89

Soil characteristics

The soil at the research location is classified as the Vertisols order, with the main characteristics being the presence of vertic characteristics (wrinkle-swelling characteristics) and a blackish-gray color (presence of a reduction reaction). This soil has a pH of 7.70 (slightly alkaline), Organic C 1.10% (low), CEC 30.41 me/100g soil (high), and Total N 0.10% (low) (Table 2). Sudadi et al. (2020) state that Vertisols have high CEC and low organic C Land use in this research area is paddy rice cultivation using a paddy rice-paddy rice pattern. Suntoro et al. (2023) stated that Vertisols are very suitable for cultivating rice because it requires flooding, especially in the vegetative phase.

Green manure characteristics

Green manure, an organic material, releases complete nutrients such as N, P, and K in uncertain, relatively small amounts and slow release. Das et al. (2020) state that green manure is considered a complete nutritional fertilizer because it contains the essential elements for plants and is released slowly during decomposition. Continuous use of green manure can increase nutrients for the soil. Based on the organic material source survey that has been carried out, there are 4 abundant sources of organic material at the research location, namely rice straw, *C. odorata*, *Ipomoea* spp. (morning glory), and *E. crassipes*. It is hoped that these four abundant sources of organic material can be used as fertilizer.

These three green manures meet the requirements as green manure has relatively high N content, is succulent, and has low C/N, so it provides plant nutrients quickly. The C/N ratio parameter determines the speed of mineralization of organic fertilizer in releasing soil nutrients. Judging from the N and C/N nutrient content, it can be seen that *C. odorata*, *Ipomoea* spp. (Morning Glory), and *E. crassipes* have relatively large potential for use as fertilizer (Table 3). *Ipomoea* spp. (morning glory) has a relatively high potential compared to other green manures. This can be seen from the higher N content and lower C/N than other green manure types.

Data analysis

The results obtained were analyzed using Analysis of Variance (ANOVA), followed by Duncan Multiple Range Test (DMRT) 95% to see the best (significant) effect, and Pearson Correlation Analysis (PCA) to determine the closeness of the relationship between the observed variables.

RESULTS AND DISCUSSION

Effect of green manure on soil chemical properties

Soil pH is described as a "master soil variable" influencing soil's biological, chemical, and physical properties, influencing plant growth and crop yields (Neina 2019). Soil pH greatly affects plant nutrient availability (Cai et al.

2019). Green manure can increase soil pH with the highest results in the 10 tons/ha (*C. odorata* treatment), increasing the pH to 7.85 (Table 4). According to Gondal et al. (2021), plant growth problems often occur in soil that is too alkaline. The pH acidity value of the soil solution greatly influences nutrient availability, plant growth, pesticide effectiveness, soil productivity, and microbial activity. Providing green manure can also reduce soil pH. The decrease in soil pH occurs due to the production of CO_2 and organic acids during the green manure decomposition process (Islam et al. 2019). Providing inorganic fertilizer and a combination of green manure with inorganic fertilizer tends to reduce the soil pH value compared to the control. According to Bhatt et al. (2019), excessive application of inorganic fertilizer can cause an imbalance of nutrients, which causes soil acidity, thereby reducing crop yields.

Soil organic C content is one component that greatly influences soil fertility levels. Gurmu (2019) stated that soil organic matter contains large amounts of crucial plant nutrients released slowly (especially N), increasing nutrient exchange, retaining moisture, reducing compaction, and increasing water infiltration. Conventional tillage can cause strong disaggregation and aeration of the soil to support the decomposition of organic matter by increasing oxygen availability (Trost et al. 2013). Providing green manure has a significant effect on soil organic C content. Green manure can increase soil organic matter, vital for maintaining soil fertility (Dubey et al. 2015). The highest yield was shown in the 10 tons/ha (*C. odorata* treatment), namely 1.92% or an increase of 0.72%. Das et al. (2020) stated that applying green manure can increase soil organic matter, improving the soil's physical properties. Franzluebbers (2002) states that low soil organic matter content can indicate low soil quality. This can happen because organic matter is vital in nutrient conservation and water infiltration.

Providing green manure has a significant effect on soil CEC. The increase in soil CEC occurs due to humus compounds contained in organic material. Murphy (2015) stated that most of the action of soil organic matter contributes to soil CEC. Providing green manure with a high organic material content can increase the CEC of the soil (Amede et al. 2021). The highest yield was found in the NPK 100 kg/ha + *E. crassipes* 5 tons/ha treatment of 42.47 me/100g of soil. Soil CEC greatly influences nutrient availability for plants. Soil with high CEC requires fertilization with large amounts of certain cations to be available to plants; giving small amounts affects low availability for plants because more cations are bound to the soil. Xu et al. (2012) stated that increasing CEC increases soil fertility through greater nutrient availability because it can retain nutrients from leaching.

Effect of treatment on soil total N and N uptake

Plant growth will occur optimally if the soil has sufficient nutrients to be absorbed by the plant. Nitrogen has an irreplaceable role, especially in organ formation, metabolism, and plant yields. Sufficient N application can increase leaf photosynthesis efficiency and promote flower bud differentiation (Xu et al. 2020). The results of the analysis of variance show that the application of green manure has a significant effect on the availability and uptake of Nitrogen, as shown in Table 5.

Providing green manure has a significant effect on soil total N. The treatment with the highest results was the NPK 100 kg/ha + *C. odorata* 5 tons/ha treatment, namely 0.25% or an increase of 0.14% compared to the control. Combining green manure with inorganic fertilizer can compensate for giving inorganic fertilizer. Das et al. (2020) stated in their research that giving inorganic N fertilizer together with green manure can increase the effectiveness of N fertilizer. Giving green manure at a dose of 10 tons/ha shows total N results of 0.19-0.21%, which offers lower results compared to NPK fertilizer of 200 kg/ha. This follows the statement by Tigka et al. (2021) that providing green manure can increase the availability of N for plants through a decomposition process that releases N slowly and according to plant needs. Apart from that, applying green manure can be a solution to reduce the dose of inorganic fertilizer. The application of green manure can also increase the effectiveness of N fertilization, considering that the N element is very mobile in the soil and has the potential to be lost through various processes. Nitrogen can be lost through volatilization, denitrification, and leaching (Padilla et al. 2018a). The presence of nitrogen always increases plant growth. Nitrogen in sufficient quantities can stimulate the production of new leaves from stem terminal meristems and lateral buds of old leaves (Fathi 2022).

The application of green manure will have a real effect on plant biomass. According to Das et al. (2020), green manure constantly supplies N, which is released slowly according to plant needs to increase plant growth. Providing *C. odorata*, *Ipomoea* spp., and *E. crassipes* green manure has a higher effect compared to straw and can even offset the application of NPK fertilizer. The treatment with the highest yield was the combination of NPK 100 kg/ha + *C. odorata* 5 tons/ha, resulting in 25.45 g/clump. This is in accordance with the statement of Yang et al. (2014) that applying green manure can increase soil organic matter and stimulate N fixation in the soil, resulting in an increase in N in the soil. Apart from that, the use of green manure also significantly affects plant biomass.

Table 4. Effect of green manure on soil chemical properties

Code	Treatments	pH H_2O	Organic C (%)	CEC (me/100g soil)
P0	Control	7.72bc	1.20a	31.11a
P1	NPK	7.64a	1.32b	34.73abc
P2	Rice straw	7.74c	1.81e	33.51ab
P3	*C. odorata*	7.85d	1.92f	35.82bc
P4	*Ipomoea* spp.	7.82d	1.45c	38.34cd
P5	*E. crassipes*	7.72bc	1.47c	36.26bc
P6	NPK + Rice straw	7.68ab	1.34b	35.93bc
P7	NPK + *C. odorata*	7.66ab	1.46c	40.37de
P8	NPK + *Ipomoea* spp.	7.68ab	1.63d	41.01de
P9	NPK + *E. crassipes*	7.71bc	1.39bc	42.47e

Note: Numbers followed by the same letter in the same row are not significantly different at α=5%

Table 5. Effect of treatments on soil total N and N uptake

Code	Treatments	Total N (%)	Plant biomass (g/clump)	Plant tissue N content (%)	Plant N uptake (g/clump)
P0	Control	0.11a	10.19a	1.26a	0.13a
P1	NPK	0.22cde	23.70c	2.73ef	0.65f
P2	Rice straw	0.18b	17.54b	1.97b	0.35b
P3	*C. odorata*	0.21bc	23.59c	2.25c	0.53d
P4	*Ipomoea* spp.	0.20bc	24.93c	2.42de	0.60e
P5	*E. crassipes*	0.21bc	21.47bc	2.40cd	0.51d
P6	NPK + Rice straw	0.20bc	17.44b	2.49d	0.43c
P7	NPK + *C. odorata*	0.25e	25.45c	2.72e	0.69fg
P8	NPK + *Ipomoea* spp.	0.23de	24.82c	2.88f	0.71g
P9	NPK + *E. crassipes*	0.22cde	24.34c	2.66e	0.65f

Note: Numbers followed by the same letter in the same row are not significantly different at α=5%

Table 6. Effect of treatments on chlorophyll content

Code	Treatments	Chlorophyll-a (mg/g leaf)	Chlorophyll-b (mg/g leaf)	Total chlorophyll (mg/g leaf)
P0	Control	1.47a	0.56a	2.02a
P1	NPK	2.49f	2.01g	4.49g
P2	Rice straw	2.37cd	1.04b	3.37b
P3	*C. odorata*	2.52f	2.13h	4.58g
P4	*Ipomoea* spp.	2.23b	1.40c	3.62c
P5	*E. crassipes*	2.41cd	1.67e	4.04e
P6	NPK + Rice straw	2.35c	1.88f	4.33f
P7	NPK + *C. odorata*	2.43de	2.12h	4.52g
P8	NPK + *Ipomoea* spp.	2.40cd	1.52d	3.91d
P9	NPK + *E. crassipes*	2.48ef	1.80f	4.27f

Note: Numbers followed by the same letter in the same row are not significantly different at α=5%

The decomposition and mineralization process will release N in the form of NO_3^- and NH_4^+, which plants can absorb, but plants absorb more N in the form of NO_3^- (Durán-Lara et al. 2020). The maximum absorption of Nitrogen elements occurs in the vegetative phase. It begins to decline when entering the generative phase because the need for Nitrogen in the generative phase is not as great as during the vegetative phase. Continuous application of green manure can increase total soil N and available N that plants can absorb (Gao et al. 2015). The application of green manure significantly affected plant tissue N and N uptake. The highest results were shown by the treatment of NPK 100 kg/ha + *Ipomoea* spp. 5 tons/ha with a plant tissue N content of 2.88% and N uptake of 0.71 g/clump. This aligns with research (Rathore et al. 1993), which shows that applying *Ipomoea* spp. 5 tons/ha showed higher N than the control. Liu et al. (2014) stated in their research that the release of Nitrogen in organic fertilizer is slower than inorganic fertilizer because the Nitrogen in organic fertilizer is not available to plants, so the absorption of nutrients occurs more effectively.

Figure 2 shows the correlation test results for total soil N with plant tissue N levels and N uptake by plants, producing a real and positive relationship. Total-N levels influence plant tissue N levels and N uptake of plants. The higher the soil N-total value, the higher the plant tissue N content and N uptake. This can happen because the amount of plant uptake is determined by the availability of nutrients in the soil.

Effect of treatment on leaf chlorophyll levels

Nitrogen (N) is a constituent component of proteins, enzymes, chlorophyll, and growth regulators. Nitrogen availability is an inhibiting factor in plant growth. Nitrogen deficiency results in stunted plant growth and yellow leaves. Nitrogen strongly influences chlorophyll content because most of the Nitrogen in leaves plays a role in photosynthesis (Padilla et al. 2018b). Plant chlorophylls are chlorophyll a and b (Garousi et al. 2015). The analysis of variance showed that the application of green manure has a significant effect on leaf chlorophyll levels, as shown in Table 6.

Chlorophyll is an essential pigment in photosynthesis, a source of plant growth energy. Photosynthesis reactions are divided into three stages: primary reactions, photosynthetic electron transport, photophosphorylation, and carbon assimilation. Li et al. (2018) state that chlorophyll a and b are important for primary reactions. Chlorophyll a and b absorb sunlight at different wavelengths (chlorophyll a absorbs orange-yellow light, and chlorophyll b absorbs blue-violet light), and total leaf chlorophyll levels (a+b) directly influence the photosynthetic capacity of plants.

The application of green manure with the highest results for chlorophyll-a, b, and the total was in the 10 tons/ha *C. odorata* treatment with respective yields of 2.52, 2.13, and 4.58 mg/g leaf. This can happen because leaf chlorophyll levels are very closely related to Nitrogen, one of the constituent elements of chlorophyll. Mfilinge et al. (2014) stated that chlorosis symptoms characterize nitrogen deficiency in the form of yellow leaves, which can be seen directly.

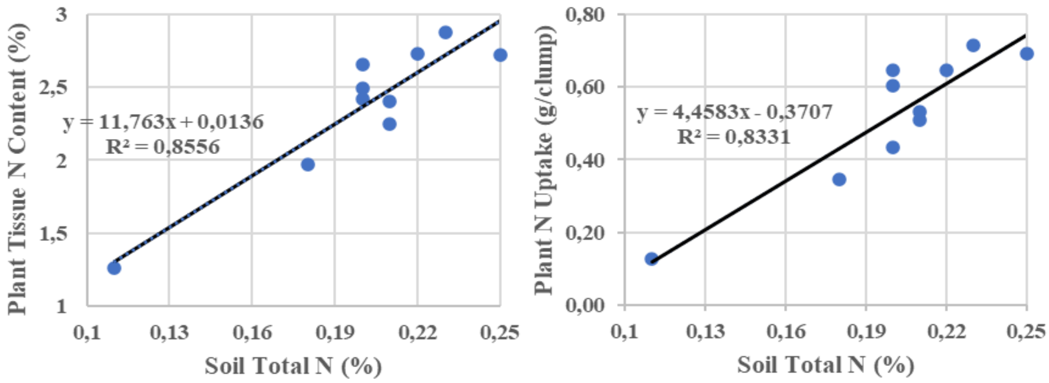

Figure 2. Relationship of soil N-total to plant tissue N content and plant N uptake

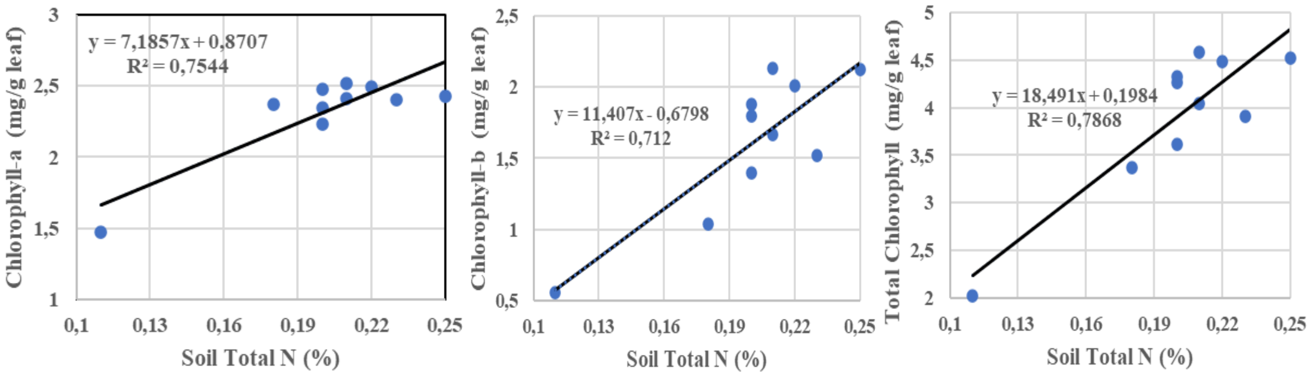

Figure 3. Relationship of soil N-total to plant chlorophyll content (a, b, and total)

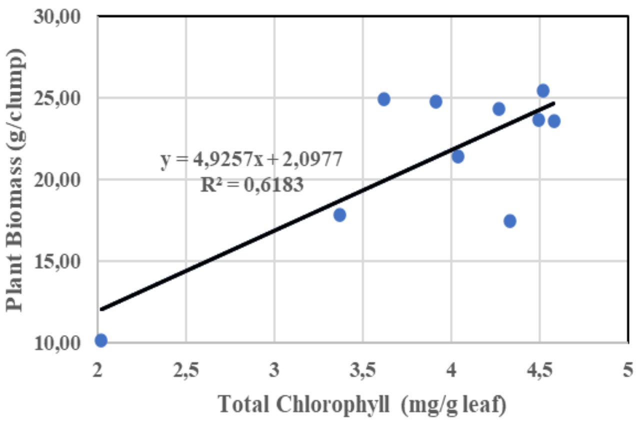

Figure 4. Relationship between total chlorophyll levels and plant biomass

Chlorophyll color can indicate a plant's physiological condition, which is often not considered in agriculture. Lack of water in leaves can also affect chlorophyll synthesis, promote chlorophyll decomposition, and accelerate leaf yellowing. Chlorophyll in rice plants is an important component for photosynthetic activity, which produces carbohydrates to form plant tissue and rice seeds. Low levels of chlorophyll and plant biomass are indicators that the plant is experiencing a nitrogen deficiency. N nutrient levels in plants significantly correlate with chlorophyll levels (Gholizadeh et al. 2017).

The correlation test results of total soil N with chlorophyll-a, b, and total levels produced a real and positive relationship (Figure 3). Total N levels affect plant chlorophyll levels (a, b, and total); the higher the N in the soil, the higher the chlorophyll-a, b, and total levels in the plant. Nitrogen has a very significant role in the formation of chlorophyll. As a structural unit of chlorophyll, N forms porphyrins used in chlorophyll metabolism (Suntoro et al. 2018). Fathi (2022) stated that higher N content in leaves is associated with higher chlorophyll content and will cause increased chloroplast activity, thereby increasing photosynthetic productivity.

Figure 4 shows the correlation test results between chlorophyll levels and plant biomass, which produces a positive relationship. Total chlorophyll levels in plants affect plant biomass. The higher the chlorophyll levels in rice plant leaves, the higher the plant biomass will be. Chlorophyll is the main factor in photosynthesis, which utilizes solar energy, triggering O2 fixation to produce carbohydrates, which are then converted into proteins, fats, nucleic acids, and other organic molecules for plant growth. The chlorophyll content is significant in determining the rate of photosynthesis and dry matter production (Fathi 2022). Finney (2016) stated that the increase in plant biomass was caused by increased N uptake. Nitrogen is the main component of chlorophyll molecules and plays a vital role in photosynthesis (Mfilinge et al. 2014).

In conclusion, the results of this study indicate that the application of green manure significantly affects soil total-N, N uptake, and chlorophyll levels in rice plant leaves. The variation of green manure with inorganic fertilizer showed higher yields but was not significantly different from the green manure treatment. Green manure *C. odorata*, *Ipomoea* spp. (morning glory), and *E. crassipes* have great potential as fertilizer, considering their high N nutrient content and low C/N. There is a positive correlation between the availability of N nutrients in the soil, N levels in rice plant tissue, and rice plant chlorophyll levels, both chlorophyll a, b, and total. Chlorophyll levels will affect increasing plant biomass. This research shows that green manure can compensate for NPK fertilizer by providing N nutrients, N uptake, and chlorophyll levels in rice plant leaves (chlorophyll a, b, and total chlorophyll).

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