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Research articles

Application of perennial legume green manures to improve growth and yield of organic lowland rice

M. Winarni[1,*], P. Yudono[2], D. Indradewa[2], B.H. Sunarminto[2]

[1] Postgraduate Program, Faculty of Agriculture, GadjahMada University, Indonesia.
[2] Faculty of Agriculture, GadjahMada University, Indonesia.

*corresponding author: martiwinarni@yahoo.co.id

Abstract: A pot experiment in green house was done to study the effect of the dosage and speciesof perennial legume green manures to the physiological traits, growth and yield of organic lowland rice (*Oryza sativa*L.), and to obtain the optimal dosage as well. The research was arranged in a factorial randomized block design consistedof two factors with three replications.The first factor was the species of perennial legume thatconsisted of threespecies: Turi (*Sesbaniagrandiflora*), Glirisidia (*Gliricidiasepium*), and Lamtoro (*Leucaenaleucocephala*) and cow manure as control treatment. The second factor was the dosage of green manure thatconsisted of four levels: 5, 10, 20 and 40 t/ha. The results showed that application ofperennial legumesinto the soil significantly improved the growth and yield of rice. The application of 20 t Glirisidia leaves/haproduced the highest grain yield, followed by 20 t Lamtoro leaves/ha and 20 t Turi leaves/ha. The optimal dosages *of S. grandiflora, G. sepium* and L. *leucochepala* leaves that could yield 58.03 g/hill (equivalent to14.51 t/ha), 53.67 g/hill (equivalent to 13.42 t/ha), and 49.67 g/hill (equivalent to 12.42 t/ha) were 28.05, 25.46 and 26.41 t/ha, respectively.

Keywords: *organic lowland rice, perennial legume green manures, species*

Introduction

Rice productivity have increased since the green revolution (Hasanuzzaman et al., 2010) and applied a system that uses large amounts of inorganic fertilizers, pesticides, and herbicides (Khan et al., 2007). Constantly and exaggerated inorganic fertilizer application in every planting season have reduced soil productivity within a certain span of time (Ikemura and Shukla, 2009; Hasanuzzaman et al., 2010; Sanati et al., 2011). Growing awareness about the negative impact of the use of inorganic fertilizers, pesticides and other agricultural inputs to the environment, health, food and human health, have prompted farmers to develop organic farming (Ikemura and Shukla, 2009).

Organic farming is a holistic production management system that improves and develops the agro-ecosystem health, including biodiversity, biological cycles, and soil biological activity (USDA, 2007; Sirikul et al., 2009; BSN, 2013). Organic farming works depending on the soil health and the nutrients cycle through natural processes (Ram et al., 2011). Rice needs a lot of nitrogen (N) nutrients for its growth that macro N often becomes the production limitation. Nitrogen is most needed nutrient for plants. It is a component of amino acids, proteins, nucleic acids, chlorophyll, enzymes, and various other metabolic essential. Moreover, it is the main element to build the blocks of protein, protoplasm, chloroplasts, and enzymes. Its role is associated with photosynthetic activity, indicating its direct and indirect effect in the metabolism and respiration (Rao, 2006).

Organic farmers typically use manure as its source of nutrients, including N. However, the amount of manure to be applied is limited, besides its nutrient content is low and varied. One of the alternative organic fertilizer sources is the green manure. Therefore, green manure which is more ready and available, is an important source of N and organic materials (Kaushal et al., 2010). Green manure is biomass of plant, usually legume that is incorporated into the soil in fresh green form to enrich organic matter and nutrients, including N in the soil. Organic fertilizer derived from green materials will decompose and release N rapidly as well (Purwanto et al., 2014). Green manure is practice of adding organic matter to the soil by ploughing and adding into the soil undecomposed green plant tissues for improving physical structure and fertility of the soil. The purpose of applying the green manure is to improve the content of organic matter and nutrients in the soil, resulting in improving the physical, chemical and biological soil, and ultimately improving the productivity of soil and soil resistance to erosion (Rachman et al., 2006; Ram et al., 2011).

Tomar et al. (2013) reported that application of *Erythrina indica*, tree bean (*Parkiarox burghii*), Acacia (*Acacia auriculiformis*), cassia (*Cassia siamea*) each of 10 t/ha obtained higher grain yields than the grain yield (5.13 t/ha) level that wasobtained with recommended N–P_2O_5–K_2O (80:60:40 kg/ha) in third year. Significantly higher grain yield of rice was recorded with incorporation of Erythrina leaves (5.67 t/ha) that remained at par with all other leaves except Alder (4.67 t/ha). Alagappan and Venkitaswamy (2015) reported that among organic treatments, 100 % recommended dosage of N (RDN) through Dhaincha green manure recorded higher SPAD value and rice grain yield (5.08 and 5.14 t/ha in 2012 and 2013, respectively).

Result of the research conducted by Manjappa (2014) indicated that the rice grain yield were recorded with application of Eupatorium at 10 t/ha (6.6, 6.9 and 6.7 t/ha, respectively), 15 t/ha (6.7, 7.2 and 6.7 t/ha) and 20 t/ha (6.5 ;7.4 and 6.9 t/ha) were found to be on par with each other during first year, second year and in pooled data. Neelima (2008) reported that the treatment of total incorporation of Sunnhemp (35 t/ha consisting of shoot 29.3t/ha and root 6.7 t/ha) has significantly out yielded all other treatments (green manuring shoot 29.3 t/ha, root 6.7 t /ha, and no green manure) in respect on the grain yield of rice.

The information about dosage of various species of green manure suitable for pure organic lowland rice cultivation is still limited. Therefore, the objectives of this research were to study the effect of the dosage of some species of perennial legume as green manures to the physiological traits, growth and yield of organic lowland rice, and to determine the optimal dosage as well.

Materials and Methods

A pot experiment was conducted in a greenhouse of Faculty of Agriculture, Gadjah Mada University, Yogyakarta from February to July 2013. The species of the perennial legume used as green leaf manures were Turi (*Sesbania grandiflora* (L.) Poir), and Lamtoro (*Leucaena leucocephala* (Lam.) de Wit.), Glirisidia (*Gliricidia sepium* (Jacq.) Walp. Cow manure was used as a control treatment. Organic lowland rice seed used for this study was Mentik Wangi variety.

This research was arranged in a factorial randomized block design with two factors and three replications. The first factor was the species of perennial legume that consisted of three species: Turi (*Sesbania grandiflora*), Glirisidia (*Gliricidia sepium*, and Lamtoro (*Leucaena leucocephala*) and cow manure (organic fertilizer used by farmers) as control treatment. The secondfactor was the dosage of green manure that consisted of four levels: 5 t/ha (30 g/pot), 10 t/ha (60 g/pot), 20 t/ha (120 g/pot), and 40 t/ha (240 g/pot) fresh weight based. Combination of the treatments consisted of 16 pieces and 528 experimental units (4 species of green manure x 4 levels of dosage x 3 replications x 11 pot rice hill of each of the treatments).

Soil of organic rice lowland was an Inceptisol with C-organic content, N-total content, C/N ratio, and pH of 2.50% (moderate), 0.16% (low), and 16 (high), and 6.1, respectively (Eviati and Sulaiman, 2009). Soil of organic lowland rice was taken from a depth of 0-30 cm in composites, dried, crushed, and sieved through a 2 mm sieve. Each ecerimental pot was filled with 12 kg of soil and irrigated as high as 5 cm upper soil surface. Two days after flooding, the green manures (Turi, Gliricidia and Lamtoro leaves) and cow manure were incorporated into the soil to a depth of 10 cm from the surface of soil as the basal fertilizer two weeks before transplanting the rice seedlings. The rice seedlings aged three weeks after sowing were transplanted into the pots, two rice seedlings on each per pot.

The experiment was observed by the method of destruction, included the parameters of rice N uptake, leaf area/hill, chlorophyll content, transpiration rate, CO_2-cell content, photosynthesis rate, plant height, plant dry weight, number of panicles/hill, panicle length, number of grain/panicle, the percentage of grain filled, weight of thousand grains and grain yield/hill. The N content of the rice plants was measured from the roots, stems, leaves and grains by applying Kjeldahl method. The N uptake was calculated by multiplying the plant dry weight x N content of the plants. Leaf area/hill was measured by using a measurement system area At Delta T Devices LTD. The chlorophyll content was observed from the rice leaf extract given with 95 % acetone and measured using the spectronic 21D instrument. Transpiration rate, CO_2-cell content, and photosynthesis rate were observed by using the plant canopy analyzer LAI-2000. Plant height was measured from the ground to the tip of the highest leaf. Plant dry weight was measured by weighing all parts of the plant after being put in the oven within a temperature of 80^{o}C for 48 hours by using the Electronic Precision Balance ACIS AD. 300 H. Grain yield/hill weight was measured after being dried in the sun with a moisture content of 14% as determined by the digital grain multi tester instrument. Analysis of Variance was applied to analyze the data at 5% significance level, followed by Duncan's Multiple Range Test (DMRT) at 5% significance level. Data were analyzed using the software program SAS version 9.1.3.

Results and Discussion

Data presented in Table 1 show that the cow manure contained higher C/N ratio compared to the perennial legumes. However, the cow manure fulfilled the requirements of C-organic content (>15%), and C/N ratio (15-25) as soil amelioratiant (Permentan, 2011). The leaves of Turi, Lamtoro and Gliricidia can be used as green manures due to the high content of N-total (≥ 2,5%). Palm et al. (2001) suggested that based on quality, plant materials can be grouped into high quality if it contains at least 2.5% N, lignin content < 15 % and polyphenols < 4 %. The plant material containing N < 2.5%, relatively high lignin and polyphenols is considered as poor quality. Organic manures that have C/N ratio close to C/N ratio of soil (10-12) can be used to the crops (Setyorini et al., 2006). Both the leaves of green manure and cow manure were organic manures as a source of nutrients, including N and C-organic to maintain soil fertility.

Physiological traits and growth organic lowland rice

The data on rice N uptake, leaf area, and chlorophyll content were presented in Table 2, while those of transpiration rate, CO_2-cell of leaf contents, photosynthesis rate, plant height, plant dry weight were in Table 3. There were significant interactions among the dosage and species of perennial legume green manure treatments to the physiological traits and growth of rice.

Application of different dosages and species of perennial legume green manures had significant different effect on the physiological trait and growth of rice (Table 2 and 3).The dosage 20 to 40 t Glirisidia/ha gave the highest physiological trait and growth of rice, viz. rice N uptake, leaf area/hill, chlorophyll content, transpiration rate, CO_2-cell of leaf contents, photosynthesis rate, plant height, and plant dry weight. It was the same as to the chlorophyll content and transpiration rate for rice, applied with Lamtoro manure. Both were higher than those in Turi manure and much higher compared to cow manure.

Those results indicated that the dosage of 20 t/ha was the best treatment of Gliricidia leaf, followed by 20 t/ha of Lamtoro and Turi, which could improve the traits of physiological and growth of the rice plant. The increase of N uptake in rice was due to the release of N from the perennial legume green manure. Gliricidia showed the highest of N-total content (4,14%). The release of N from Glirisidia leaves to the soil was higher than that released by Lamtoro and Turi leaves due to the lower content of total-N, 3.56% and 3,47%, respectively.

Earlier research conducted by Tomar (2013) showed different results with 10 t Erytrina/ha that produced the highest N uptake after three year application of Erytrina. The results of similar research showed that 100% fertilizer dosage recommendation (RDF) through Daincha green manure gave the highest SPAD value (Alagappan and Venkitaswamy, 2015).

Table 1. Chemical analysis of perennial legumes and cow manure

Species of Perennial Legumes	Dry weight (%)	Total-N (%)	Organic-C (%)	C/ N
Turi (*S. grandiflora*)	26.30	3.47	39.02	11.24
Lamtoro (*L. leucocephala*)	27.29	3.56	44.98	12.63
Glirisidia (*G. sepium*)	28.18	4.14	47.96	11.58
Cow Manure	30.00	1.49	21.90	14.70

Table 2. N uptake, leaf area, and chlorophyll content of rice as affected byapplication of dosage and species of perennial legume green manures

Species of Perennial Legumes	Dosage (t/ha)	N Uptake/ Hill (g)	Leaf Area/Hill (dm^2)	Chlorophyll Leaf Contents (mg/g)
Turi (*S. grandiflora*)	5	1.143 d	23.62 c	0.997 c
	10	1.357 c	25.62 c	1.015 c
	20	1.787 b	31.23 b	1.048 b
	40	1.777 b	31.55 b	1.049 b
Glirisidia (*G. sepium*)	5	1.170 d	24.58 c	1.014 c
	10	1.447 c	26.06 c	1.018 c
	20	2.077 a	37.32 a	1.107 a
	40	2.083 a	37.51 a	1.108 a
Lamtoro (*L. leucocephala*)	5	1.157 d	23.73 c	1.012 c
	10	1.393 c	25.82 c	1.106 c
	20	1.863 b	31.88 b	1.072 a
	40	1.880 b	32.09 b	1.075 a
Cow Manure	5	0.863 e	16.76 d	0.886 d
	10	0.890 e	16.93 d	0.896 d
	20	0.907 e	17.47 d	0.898 d
	40	0.920 e	19.21 d	0.903 d
Interaction		(+)	(+)	(+)
CV (%)		5.57	9.33	1.74

Remaks: The numbers in the same column followed by the same letter are not significant at 5% DMRT. (+): The interaction among the dosage and species of perennial legume green manure is significant.

Table 3. The transpiration rate, CO_2-cells of leaf content, photosynthesis rate, plant height and plant dry weight of rice as affected byapplication of dosage and species of perennial legume green manures

Species of Perennial Legumes	Dosage (t/ha)	Transpiration Rate (µmol H_2O m^2/s)	CO_2-cell of Leaf Contents (µmol CO_2/molair)	Photosynthesis Rate (µmol CO_2 m^2/s)	Plant height (cm)	Plant Dry Weight (g)
Turi	5	6,03 cd	403,93 d	42,17 c	120,43 c	89,26 d
(*S. grandiflora*)	10	6.33 bcd	411.17 cd	43.50 c	120.97 c	84.27 c
	20	7.07 ab	448.11 bc	46.00 c	128.47 b	123.32 b
	40	6.06 cd	404.28 d	46.33 c	128.78 b	122.83 b
Glirisidia	5	5.77 d	407.74 d	42.27 c	120.70 c	90.15 d
(*G. sepium*)	10	6.95 abc	414.71 cd	46.93 c	121.17 c	108.81 c
	20	7.71 a	489.51 a	62.00 a	136.88 a	136.99 a
	40	7.67 a	483.49 ab	63.00 a	137.97 a	135.75 a
Lamtoro	5	6.07 cd	405.77 d	41.67 c	120.67 c	89.90 d
(*L. leucocephala*)	10	6.21 bcd	413.03 cd	45.17 c	121.03 c	106.09 c
	20	7.61 a	482.87 ab	54.33 b	129.28 b	124.59 b
	40	7.67 a	480.90 ab	53.67 b	129.50 b	123.37 b
Cow manure	5	4.04 e	332.36 e	31.17 d	111.63 d	73.31 e
	10	4.39 e	333.09 e	34.50 d	112.13 d	74.70e
	20	4.48 e	335.43 e	35.50 d	112.33 d	76.56e
	40	4.63 e	337.76 e	36.00 d	113.55 d	77.27e
Interaction		(+)	(+)	(+)	(+)	(+)
CV (%)		8.44	5.15	6.86	2.73	5.92

Remarks: The numbers in the same column followed by the same letter are not significant at 5% DMRT. (+): The interaction among the dosage and species of perennial legume green manure is significant.

Manjappa (2014) reported that maximum result of the straw recorded by the Eupatorium 20 t/ha, but a par with 10 t/ha and 15 t/ha. The condition of N availability in the soil determines N content to be absorbed by plants (Purwanto, 2014). Plants with sufficient N supply will develop leaves and high chlorophyll content, so that the plant is able to produce sufficient amount of assimilation to sustain the vegetative growth (Wijaya, 2008). Nitrogen is a component of amino acids, proteins, nucleic acids, chlorophyll and many other metabolic essential for survival of the plant (Rao et al., 2006)

Component yield and grain yield

The data ofnumber of panicles, panicle length, number of grains, percentage grains filled, grains thousand weight and grain yield of rice werepresented in Table 4. The application of different dosage and species treatments of perennial legume green manures had significantly different effect on their interaction in the yield component and grain yield of rice. Application of 20 t Glirisidia/ha gave the highest yield components, i.e. number of panicles, and panicle length, number ofgrains, percentage of grain filled, and grains thousand weight. It was the same to the number of panicles and panicle length at 40 t Girisidia manure/ha. Both were higher than those of Lamtoro and Turi each at 20 to 40 t/ha and were much higher compared to cow manure. Different dosages of various spesies of perennial legume led to different dry grain yield at 14 % moisture content.

Table 4. Number of panicles/hill, panicle length, number of grains/panicle, percentage grains filled, grains thousand weight and grain yield as affected by application of dosage and species of perennial legume green manures

Species of Perennial Legumes	Dosage (t/ha)	Number of Panicles/ Hill	Panicle Length (cm)	Number of Grains/ Panicle	Percentag e of grain filled (%)	Grains Thousand Weight (g)	Grain Yield/Hill (g)
Turi	5	12.33 d	24.48 c	119.83 e	88.41 c	25.52 c	33.56 e
(*S. grandiflora*)	10	15.17 c	24.56 c	132.06 d	89.03 c	25.65 c	39.06 d
	20	18.33 b	26.72 b	143.39 c	91.06 b	27.08 b	47.94 c
	40	18.50 b	26.25 b	143.00 c	91.26 b	27.07 b	45.22 c
Glirisidia	5	13.50 d	24.51 c	121.72 e	88.90 c	25.55 c	35.12 e
(*G.sepium*)	10	16.67 c	24.69 c	135.45 d	89.31 c	25.65 c	41.18 d
	20	21.67 a	29.20 a	158.16 a	93.70 a	28.62 a	57.70 a
	40	21.00 a	28.72 a	151.33 b	91.90 b	27.10 b	45.45 c
Lamtoro	5	12.67 d	24.41 c	121.17 e	88.77 c	25.53 c	34.37 e
(*L. leucocephala*)	10	15.67 c	24.62 c	134.11 d	88.93 c	25.75 c	39.64 d
	20	19.17 b	27.03 b	150.28 b	91.84 b	27.18 b	52.84 b
	40	18.83 b	26.72 b	148.83 bc	91.58 b	27.18 b	45.30 c
Cow Manure	5	11.83 d	23.36 c	101.95 f	85.42 d	24.03 d	26.03 f
	10	11.83 d	24.22 c	102.39 f	85.61 d	24.07 d	27.37 f
	20	12.00 d	24.25 c	104.72 f	85.62 d	24.18 d	28.65 f
	40	12.17 d	24.31 c	106.39 f	86.20 d	24.18 d	29.62 f
Interaction		(+)	(+)	(+)	(+)	(+)	(+)
CV (%)		5,86	3.36	2.81	11.13	2.84	5.86

Remarks: The numbers in the same column followed by the same letter are not significant at 5% DMRT. (+): The interaction among the dosage and species of perennial legume green manure is significant.

The highest rice grain yield (57.70 g/hill equivalent to 14.43 t/ha) was achieved by applying the dosage of 20 t Gliricidia/ha, followed by 20 t Lamtoro/ha, 20 to 40 t Turi/ha, 40 t/haof Gliricidia and Lamtoro, 10 t/ha of Gliricidia, Lamtoro, and Turi, and 5 t/ha of Gliricidia, Lamtoro and Turi. The lowest grain yield (26.03 g/hill equivalent to 6.51 t/ha) was obtained by 5 t/ha of cow manure and insignificantly difference that those obtained 10 to 40 t/hacow manure (Table 4). There was a positive correlation among variable number of panicles, panicle length, number of grains/panicle, the percentage of filled grain, weight of thousand grains and grain yield.

Neelima (2008) showed that green manure total of Sunnhemp 35 t/ha (29.3 t/ha of shoot and 6.7 t/ha of root portion) give highest rice grain yield (4.21 t/ha) than all other treatment, viz. green manure shoot 29.3 t/ha, root 6.7 t/ha, and no green manure, that these give grain yield 4.18; 3.56, and 3.27 t/ha respectively. Manjappa (2014) reported that the rice grain yield are obtained with apply of Eupatorium at 10 t/ha (6.6 ; 6.9 and 6.7 t/ha, respectively), 15 t/ha (6.7, 7.2 and 6.7 t/ha) and 20 t/ha (6.5, 7.4 and 6.9 t/ha) are found to be on par with each other during first year, second year and in pooled data. Alagappan and Venkitaswamy (2015) report that among organic treatment, 100 % recommended dosage of N (RDN) through Dhaincha green manure record higher rice grain yield (5.08 and 5.14 t/ha in 2012 and 2013, respectively). The results of the research conduct by Tomar et al (2013) indicated that the highest grain yield of rice in third year (5.67 t/ha) is record with incorporation of Erythrina leaves at 10 t/ha that similar result areobtained with all other leaves except Alder (4.67 t/ha).

Figure 1 shows that the relationship between dosage of green manures and grains yield in quadratic polynomial curve. It was shown that, Glirisidia gave the higest grain yield (58.03 g/hill equivalent to 14. 51 t/ha) for organic lowland rice and needed lower optimal dosage (25.46 t/ha), compared to Lamtoro yielding 53.67 g/hill equivalent to 13.42 t/ha, with optimal dosage at 26.41 t/ha. Meanwhile Turi gave yield 49.67 g/hill equivalent to 12.42 t/ha, but it needed the optimal dosage at 28.05 t/ha. Cow manure had the least quality in chemical analysis, gave the lowest yield (29.72 g/hill equivalent to 7.43 t/ha) and for optimal dosage must be applied the manure as much as 36.14 t/ha.

There was a negative relationship between the N content of leaves and dosage of green manure used (R^2 = 0.7946). The higher leaf N content, the lower the required dosage of green manure as a source of N in organic lowland rice cultivation would be. This was due to higher N content signified more N-available released into the soil. To produce to the maximum grain yield, the optimal dosages of Turi (3.47% N), Lamtoro (3.56% N), Gliricidia (4.14% N) and manure (1.49 % N) were 28.05 t/ha, 26.41 t/ha, 25.46 t/ha and 36.14 t/ha, respectively.

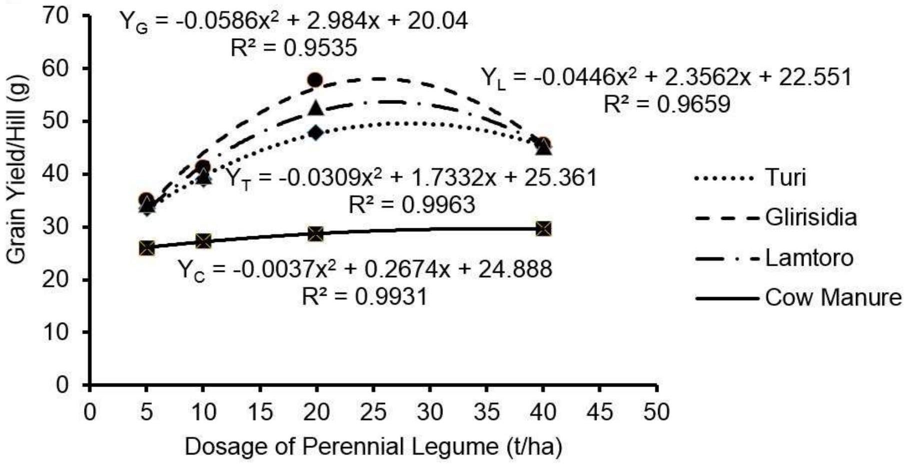

Figure 1. The relationship among the dosage of perennial legume and rice grain yield/hill with in speciesof perennial legume green manures

Conclusion

Application of 20 t Glirisidia/ha gave the highest yield, compared to Lamtoro and Turi each at 20 t/ha. The high yield was due to the higher growth and component yield. There was a negative relationship between the N content of leaves and dosages of green manure. The optimal dosages of Glirisidia, Lamtoro, dan Turi were 25.46 t/ha, 26.41 t/ha, and 28.05 t/ha, respectively, could yield 58.03 g/hill (equivalent to 14.51 t/ha), 53.67 g/hill (13.42 t/ha), and 49.67 g/hill (12.42 t/ha), respectively.

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