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The effect of nitrogen-fixing bacteria in the plant *Rhizosphere* on the quality of forage nutrient

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Abstract

The presence of soil microbes around plant roots (*Rhizosphere*) plays a role in providing nutrients needed by plants. Research to determine the effect of nitrogen-fixing bacteria population in the plant *Rhizosphere* on the quality of forage nutrient. Sampling in the plant *Rhizosphere*: NR = Non *Rhizosphere*, RG = *Rhizosphere Gliricidia sepium*, RL= *Rhizosphere Leucaena leucocephala*, and RI = *Rhizosphere Indigofera zollingeriana*, and each treatment was repeated 7 times. Observed variables include: soil nutrient content, population microbes, nitrogen-fixing bacteria, and quality of forage nutrient. The results showed that the C-organic content of the soil in the *Rhizosphere Indigofera zollingeriana* (RI) treatment was 4.16% (very high) and total Nitrogen was 0.34% (moderate). The highest total microbes and nitrogen-fixing bacteria populations of 6.0 x 10^6 and 2.5 x 10^5 Cfu/g soil were found in the *Rhizosphere Gliricidia sepium* (RG) treatment, and the lowest results were 2.6 x 10^5 and 2.0 x 10^5 Cfu/g soil in the Non *Rhizosphere* (NR). The quality of forage nutrient in the *Rhizosphere Gliricidia sepium* (RG) treatment, produced the highest crude protein (28.56%) and the lowest crude fiber (14.41%). It can be concluded that the population nitrogen-fixing bacteria in the *Rhizosphere Gliricidia sepium* (RG) is able to improve the quality of forage nutrient produced.

Keywords: Legumes; Nitrogen-Fixing Bacteria; Forage Nutrient Quality; Rhizosphere; Soil Nutrients

1. Introduction

Forage is the main feed for ruminant livestock so that the availability of forage throughout the year needs to be considered to increase the productivity of ruminant livestock. Almost 90% of ruminant livestock feed comes from forage with fresh consumption/day of 10 - 15% of body weight, while the rest is concentrate and additional feed [7]. Inhibiting factors in providing forage such as the influence of seasonal factors and dry land conditions that can affect soil fertility. The influence of the season can reduce production especially during the dry season, while in the rainy season forage production can increase [6]. Dry land conditions can reduce soil fertility because they have a low organic matter content of less than 1%, and when compared to fertile land conditions can contain 3-5% organic matter. Therefore, dry land conditions must be managed properly, and one effort that can be made is to increase the population of nitrogen-fixing bacteria.

The population of soil microbes in the plant root area (*Rhizosphere*) can help provide nutrients needed by plants to increase the productivity of forage. The types and populations of *Rhizosphere* bacteria are dynamic, they differ in each location depending on the type of soil, plant species, different seasons, and local climate conditions [2]. The *Rhizosphere* of gamal plants contains a population of nitrogen-fixing bacteria (non-symbiotic) from the genus *Azotobacter* of 83 × 10^5 cfu/g soil [5]. Soil microbial activity can provide nitrogen nutrients, and microbial activity is highly dependent on land conditions and plant species. The presence of nitrogen in the form of N_2 in the air is very abundant at around 78%, but cannot be utilized directly by plants. Soil microbes can help provide nutrients for plants by fixing N_2 in the air. Nitrogen can only be absorbed by plants in the form of ammonium ions (NH_4^+) or nitrate ions (NO_3^-) [3]. Optimizing

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biological nitrogen-fixation in the air by microbes is the right alternative to reduce the use of nitrogen fertilizers. Nitrogen-fixing activity by microbes provides benefits for plants, so it is widely used for agricultural practices and is an alternative to anorganic fertilizers [8]. Microbes that are useful for restoring soil fertility include *Plant Growth Promoting Rhizobacteria* (PGPR) bacteria such as: nitrogen-fixing bacteria from the genus *Rhizobium, Azotobacter*, and *Azospirillum*.

Nitrogen is the main nutrient for the vegetative growth process of plants such as increasing plant height, increasing the number (leaves, stems, and roots). The nitrogen element will be used by plants to increase the chlorophyll content of leaves, so that the photosynthesis process runs optimally and the results of photosynthesis as components of protein and nucleic acids, as well as components of protoplasm as a whole [1].

Based on the information above, this study needs to be conducted to determine the effect of the population of nitrogen-fixing bacteria in the plants *Rhizosphere* on the quality of forage nutrient in dry land.

2. Material and methods

2.1. Research Flowchart

Soil samples by determining the plant *Rhizosphere* area, from each plant *Rhizosphere* treatment were taken using a soil drill to a depth of 20 cm. The of each plant by taking soil samples for analysis. Analysis of soil samples to determine the population of nitrogen-fixing bacteria and soil nutrient content. Sampling of plant parts (leaves and stems) in each treatment that were analyzed proximately to determine the quality of forage nutrient. Leguminous plants are plants that grow naturally on designated land.

2.2. Treatment

Soil sampling in the plant *Rhizosphere* and plant parts for each treatment was done 7 times. The sampling for each plant *Rhizosphere* was: Non *Rhizosphere* (NR), *Rhizosphere Gliricidia sepium* (RG), *Rhizosphere Leucaena leucocephala* (RL), and *Rhizosphere Indigofera zollingeriana* (RI).

2.3. Observed Variables

The observed variables were: (1) nitrogen-fixing bacterial population; (2) total plate count; (3) soil pH; (4) soil texture; (5) soil nutrient content; and (6) quality of forage nutrient.

3. Results and discussion

The results of the study (Table 1) showed that the soil nutrient content in the *Rhizosphere Indigofera zollingeriana* (RI) gave the best results with C-organic content of 4.16% and nitrogen content of 0.34% (moderate). The nutrient content in other treatments was much lower than *Rhizosphere Indigofera zollingeriana* (RI) treatment and was classified as very low. The nutrient content in the soil can affect the soil microbial population. High nutrient content is able to provide nutrients for plants to carry out the growth process, but on the other hand it can reduce the activity of soil microbes. Soil microbes will not work optimally in providing nutrients, because the soil already has sufficient nutrients for plant needs to carry out growth, so that the presence of nitrogen-fixing bacteria decreases. The results of this study indicate that the *Rhizosphere Indigofera zollingeriana* (RI) treatment which contains the highest nitrogen nutrients among other treatments produces the lowest nitrogen-fixing bacterial population (Table 2). This opinion is supported by [5], that the presence of nitrogen-fixing bacteria around the *Rhizosphere* of leguminous plants will increase if the nutrients in the soil are low, so that the bacteria will continue to work to bind nitrogen in the atmosphere to be given to the plants. Furthermore [5], found that the *Rhizosphere* of gamal plants has a population nitrogen-fixing bacteria (non-symbiotic) of the genus *Azotobacter sp*. with a colony count of 83×10^5 cfu/g soil. Factors that affect the number of soil microbes depend on the type of soil, plant species, different seasons, and are influenced by nutrient soil content [2].

Table 1 Soil Nutrient Content in Plants *Rhizosphere*

Soil content [2)]	**Treatment**[1)]			
	NR	RI	RG	RL
pH	7.12 (N)[3)]	7.14 (N)	6.95 (N)	7.05 (N)
C-Organik (%)	1.26 (R)	4.16 (TS)	2.51 (S)	2.92 (S)
N Total (%)	0.06 (SR)	0.34 (S)	0.15 (R)	0.17 (R)
P available (ppm)	228.06 (ST)	34.90 (T)	39.63 (ST)	24.73 (S)
K available (ppm)	336.19 (T)	174.67 (S)	173.69 (S)	164.82 (S)
KU (%)	7.58	6.64	7.15	6.89
KL (%)	27.35	33.73	34.41	34.46
Teksture				
Sand (%)	17.71	29.05	19.59	34.93
Dust (%)	65.45	33.08	34.10	22.56
Clay (%) Information	16.84 Dusty clay	37.88 Clay loam	46.31 clay	42.51 clay

Information: NR = Non Rhizosfer, RI = Rhizosfer *Indigofera zollingeriana,* RG = Rhizosfer *Gliricidia sepium,* dan RL = Rhizosfer *Leucaena leucocephala;* KU = Air dry, KL = Field capacity, C = Carbon, N = Nitrogen, P = Phosfor, K = Kalium; N = Neutral, SR = Very low, R = Low, ST = Very high , T = High, S = Moderate

The population of nitrogen-fixing bacteria (Table 2) in the RG, RL, and RI treatments was higher compared to the NR treatment. This is because in the rhizosphere of leguminous plants there are root exudates which are one of the supporting factors for the growth of microorganisms. These root exudates are known to be a mixture of complex sugar compounds such as glucose, amino acids, organic acids, fatty acids, and others [4]. Root exudates in the *Rhizosphere* can affect the soil microbial community, the availability of macro and micro nutrients, especially nitrogen and phosphorus. The results of this study are in accordance with the results [9], hat the microbial population in the *Rhizosphere* is higher than in the Non *Rhizosphere* both in quantity and quality.

Table 2 Population of Nitrogen-Fixing Bacteria in Plants *Rhizosphere*

Treatment	**Total Plate Count (Cfu/g soil)**	**Total Nitrogen-Fixing Bacteria (Cfu/g soil)**
NR	2.6×10^5	2.0×10^5
RI	2.0×10^5	2.0×10^5
RG	6.0×10^6	2.5×10^5
RL	4.6×10^6	2.1×10^5

Information: NR = Non Rhizosfer, RI = Rhizosfer *Indigofera zollingeriana,* RG = Rhizosfer *Gliricidia sepium,* dan RL = Rhizosfer *Leucaena leucocephala*

The relationship between the number of nitrogen-fixing bacteria and the quality of forage nutrients (Table 3), the higher the population of nitrogen-fixing bacteria causes the nutrient content, especially crude protein to increase and the crude fiber of the forage to decrease. In the treatment of *Rhizosphere Gliricidia sepium* (RG) with the highest number of nitrogen-fixing bacteria of 2.5×10^5 Cfu/g of soil, it produced the highest crude protein of 28.56% and the lowest crude fiber content of 14.41 %, then the RL treatment with crude protein (28.24%) and crude fiber (15.91 %), RI treatment with crude protein (23.49%) and crude fiber (16.12%), but it was very different from the NR treatment with crude protein (10.58%) and crude fiber 26.01% (very high). This is because nitrogen-fixing bacteria are able to bind N_2 in the air maximally so that the nutrients provided to plants can be utilized for the plant growth process properly and produce high quality forage nutrients. Nitrogen is the main nutrient for plant growth, because nitrogen is a component of protein and nucleic acid and is a component of protoplasm as a whole. The plant growth process runs well with the availability

of nutrients for the growth process and allows the plant to increase the photosynthesis process. The photosynthesis process runs higher, so that the results of photosynthesis produced in the form of maximum carbohydrates and proteins.

Table 3 Nutrient Content of Forage

Nutrient Content	**Treatment**			
	NR	RI	RG	RL
Dry weight (%)	75.29	80.00	84.21	75.32
ash (%)	13.26	13.37	31.05	9.65
Organic Materials (%)	86.73	86.62	68.94	90.34
Crude protein (%)	10.58	23.49	28.56	28.24
Crude fiber (%)	26.01	16.12	14.41	15.91
Crude fat (%)	3.19	5.71	8.85	5.83
TDN (%)	23.38	48.18	55.71	48.78

Impormation: NR = Non Rhizosfer, RI = Rhizosfer *Indigofera zollingeriana,* RG = Rhizosfer *Gliricidia sepium,* dan RL = Rhizosfer *Leucaena leucocephala*

4. Conclusion

It can be concluded that the population of nitrogen-fixing bacteria in the *Rhizosphere Gliricidia sepium* (RG) is able to improve the quality nutrients forage produced.

Compliance with ethical standards

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Disclosure of conflict of interest

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