Physiological characters of soybean cultivars with application of nitrogen sources under dry land conditions

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Abstract. The objective of this study was to evaluate the influence of nutrient N management on physiological characteristics of three different soybean cultivars under dry land conditions. The study was conducted under dry lands of Desa Sambirejo (Langkat Regency) in the dry season. The study was conducted with a Randomize Block Design with two factors and three replication. The research was used a randomized block design with 2 factors and 3 replications. The first factor was soybean cultivars (Anjasmoro, Wilis, Sinabung). The second factor was N source, with Urea (50 kg/ha), Bradyrhizobium sp., farmyard manure (10 ton/ha), a combination of Bradyrhizobium sp. + farmyard manure (5 ton/ha) and a control with no N. The parameter observed in this study was the content of root N, shoot Nitrogen, shoot Phosphor, shoot Potassium and total of chlorophyll content. The results suggest that Anjasmoro and Sinabung cultivars had higher physiological characteristics (root N, shoot P and shoot K) compared to Wilis. Nitrogen source of Urea gave a higher physiological characteristics (content of root N, shoot Phosphor and shoot Potassium) compared to different treatment of N source in this study. The interaction between Anjasmoro cultivar and Urea gave the highest of content of shoot Phosphor and shoot Potassium, otherwise the interaction between Sinabung cultivar and Bradyrhizobium sp. gave the highest of content of shoot Nitrogen.

1. Introduction

Soybean [Glycine max (L.) Merril], as an important source of vegetable and oil for human and animal consumption [1]. As one of the most important legumes species, soybean contains about 40% highly nutritious protein with all essential amino acids, rich in magnesium, phospholipids, vitamins and minerals [2-3] and also contains secondary metabolites such as isoflavones [4], saponins, phyticacid, oligosaccharides [4] and phytoestrogens [5] which are beneficial to health. In addition to its nutritional values, soybean is also used as important nitrogen (N2)-fixing crop throughout the world for the restoration and maintenance of soil fertility in a sustainable way and consequently the improvement of crop yields [6-7].

Demand for soybean in Indonesia continues to increase, along with increasing public knowledge of the benefits of soy as a functional food. Production of soybean is not keeping up with demand, so that efforts are necessary to improve national soybean production [8]. This is done through the approach of increasing...
productivity, increasing the intensity of soybean cultivation and expansion of planting soybean area to marginal land (sub optimal) such as dry land. Dry land has many problems such as drought stress during dry season, soil pH less suitable for soybean cultivation (pH 5.0), low macro nutrient N, P, K content. Based on these problems, dry land management for increasing soybean production can be done with two main approaches namely choosing adaptive soybean varieties of dry land and improving soil fertility through the management of nitrogen [9]

Nitrogen is one of the essential nutrients for plants, the basic components of protein formation, nucleic acids, amino acids and protoplasm, required in the synthesis of chlorophyll, the formation and growth of vegetative parts of plants (leaves, stems, and roots). Application of N nutrient under dry land conditions will influence on physiological characteristics of soybean varieties. Therefore, the objective of this research was to evaluate the influence of nutrient N management on physiological characteristics of three different soybean cultivars under dry land conditions.

2. Materials and methods
Research was conducted in Sambirejo Village, Binjei District, Langkat, Sumatra Utara (Indonesia), a dry land area, June to September 2012. Location determination refers to soybean production center in North Sumatera and according to Agroecological Zone map, FSZ North Sumatra. The soil texture of the experimental site was a sandy clay loam which had 11% coarse sand, 38% fine sand, 29% silt and 22% clay. Nitrogen content was low (0.14%), organic matter was 1.02%, with a pH of 5.0 [8]

2.1. Experimental design and crop management
The research conducted by using a Factorial Randomized Block Design with 2 factors and 3 replications. The first factor is soybean cultivars are Anjasmoro (V1), Wilis (V2) and Sinabung (V3). As the second factor is the source of N consisted of without N application (N0) ; Urea 50 kg/ha (N1) ; Bradyrhizobium sp. (N2) ; manure of 10 tons/ha (N3) and combination Bradyrhizobium sp. + manure 5 tons/ha (N4).

The land used consists of 45 plot with the size of plot 2 m x 2 m. Among the plots are limited by drainage channels. Liming with dolomite 500 kg/ha was done during the second tillage by spread evenly 2 weeks before planting. Application of P and K fertilizer when planting is done at each time planting with the same dose (recommended dose of P and K fertilizer for soybean crop is 150 kg TSP/ha and 75 kg KCl/ha, respectively).

Three seeds per planting hole were planted at the study. Thinning is done 1 week after planting by cutting plants that grow poorly and leaving 1 plant per planting hole. Isolate Bradyrhizobium sp. is inoculated according to treatment. Isolate is mixed with soybean seeds, done in the morning just before planting in the shade. Soybean seeds that have been mixed isolates Bradyrhizobium sp.

Application of Urea and farmyard manure according to treatment done at planting time. Urea fertilizer is given half the dose at the time of planting and half the remaining dose at 30 HST. Application of cow manure is done by mixing cow manure with planting media evenly.

Harvesting is done when soybean have shown the criteria of harvest that the pod has brownish color and the stems and leaves have dried up.

2.2. Analysis of shoot N, P, K content and total chlorophyll
Analysis of shoot N, P, K content was conducted at Asian Agri Laboratory, while the total chlorophyll was analyzed at Integrated Laboratory, Faculty of Agriculture, Universitas Sumatera Utara. Analysis of shoot N content were determined using method from Kjeldahl, shoot P content were determined using Spechtrhophotometry method, shoot K content were determined using AAS method and total chlorophyll were determined using method from Arnon.
2.3. Statistical analysis
Data were subjected to analysis of variance (ANOVA) for comparison of means. Means were separated using Duncan’s Multiple Range Test at the 0.05 probability level.

3. Results and Discussion
3.1. N shoot content
Based on Table 1 it can be seen that the cultivars of Anjasmoro and Sinabung have significantly higher N content of shoot than Wilis. Source N Bradyrhizobium sp. has significantly higher N content of shoot (3.78%) than with other treatments, whereas the treatment of without N and combinations of Bradyrhizobium sp. + farmyard manure (5 tons / ha) has the lowest N shoot content. In the three varieties, the treatment of Bradyrhizobium sp. increased the N shoot content of Anjasmoro and also Sinabung varieties compared with other treatments.

3.2. N root content
The cultivars of Anjasmoro and Sinabung have N content of roots significantly higher than Wilis. Sources of N Urea has significantly higher N root content (1.29%) than Bradyrhizobium sp. + farmyard manure (5 tons/ha) and without N application, but not significantly different from Bradyrhizobium sp. and farmyard manure. The interaction between the varieties of Sinabung and Urea has a significantly higher N root content than other treatments, whereas the interaction of Wilis treatment and without N administration and the interaction of Wilis and farmyard manure treatment has the lowest N root content (Table 1)

3.3. P shoot content
Anjasmoro and Sinabung cultivars have significantly higher P content than Wilis. Sources of N Urea have significantly higher P shoot content than other treatments. The interaction between Anjasmoro variety and Urea has the highest P content (0.39%), while the interaction between Anjasmoro variety and farmyard manure (10 tons/ha) and interaction between Wilis and combination of Bradyrhizobium sp. + farmyard manure (5 tons/ha) have the lowest P shoot content (Table 1).

3.4. K shoot content
The cultivars of Anjasmoro and Sinabung have significantly higher K shoot content than Wilis. Sources of N Urea and Bradyrhizobium sp. have significantly higher K shoot content than other N treatments. The interaction between Anjasmoro variety and Urea has the highest K shoot content, whereas Bradyrhizobium sp. + farmyard manure (5 ton / ha) or manure on Wilis varieties give the lowest K content (Table 1)

3.5. N uptake of shoot
N uptake of shoot in Anjasmoro cultivar tend to be higher than Wilis and Sinabung. Source N Bradyrhizobium sp. has significantly higher N uptake of shoot compared with other N source treatments, whereas treatment without N application has the lowest of N uptake of shoot. The interaction between the Anjasmoro variety and Bradyrhizobium sp. tend to be the highest N uptake of shoot (Table 1)

3.6. Total of Chlorophyll
Anjasmoro cultivar tend to have the lower total chlorophyll content than Wilis and Sinabung. Treatment without N application tend to produce the lowest total chlorophyll compared with other N source treatments. Treatment of Bradyrhizobium sp. tend to produce the highest total chlorophyll. Urea treatment on Sinabung variety tend to increase the total leaf chlorophyll (5.68 mg/g wet weight of leaf).
Table 1. Physiological characters of three soybean cultivars with application of Nitrogen sources under dry Land conditions on June to September growing season.

| Treatment          | Shoot N (%) | Root N (%) | Shoot P (%) | Shoot K (%) | N uptake (g/plant) | Total of chlorophyll (mg/g ww) |
|--------------------|-------------|------------|-------------|-------------|------------------|-------------------------------|
| V1 = Anjasmoro     | 3.40a       | 1.17a      | 0.31b       | 2.06a       | 16.87            | 4.93                          |
| V2 = Wilis         | 3.19b       | 0.98b      | 0.34a       | 1.53b       | 13.14            | 5.03                          |
| V3 = Sinabung      | 3.40a       | 1.28a      | 0.35a       | 1.86a       | 14.05            | 5.06                          |
| N source (N)       |             |            |             |             |                  |                               |
| N0 = Without N application | 3.09c     | 1.05b      | 0.32b       | 1.76b       | 11.40c           | 4.87                          |
| N1 = Urea          | 3.46b       | 1.29a      | 0.38a       | 2.00a       | 13.58b           | 5.07                          |
| N2 = Bradyrhizobium sp. | 3.78a     | 1.14ab     | 0.34ab      | 1.88a       | 19.44a           | 5.26                          |
| N3 = Farmyard manure | 3.33b      | 1.16ab     | 0.31b       | 1.72b       | 14.97b           | 4.98                          |
| N4 = Bradyrhizobium sp. + farmyard manure | 2.98c   | 1.10b      | 0.32b       | 1.73b       | 14.05b           | 4.85                          |
| Interaction V x N  |             |            |             |             |                  |                               |
| V1 N0              | 3.07fg      | 1.00def    | 0.32c       | 2.08bcd     | 11.01            | 5.15                          |
| V1 N1              | 3.66bc      | 1.18bcd    | 0.39a       | 2.49a       | 15.16            | 4.70                          |
| V1 N2              | 3.97a       | 1.18bcd    | 0.33c       | 2.22bc      | 22.66            | 5.58                          |
| V1 N3              | 3.41cde     | 1.35b      | 0.26d       | 2.01cde     | 15.61            | 5.31                          |
| V1 N4              | 2.87gh      | 1.16cd     | 0.38ab      | 1.48ij      | 19.92            | 4.93                          |
| V2 N0              | 3.29ef      | 0.86f      | 0.31c       | 1.58ghij    | 11.23            | 5.02                          |
| V2 N1              | 2.87gh      | 0.94ef     | 0.38ab      | 1.76efg     | 11.44            | 4.84                          |
| V2 N2              | 3.43cde     | 1.10de     | 0.34bc      | 1.51hij     | 20.26            | 5.26                          |
| V2 N3              | 3.00g       | 0.90f      | 0.35abc     | 1.43j       | 10.66            | 4.54                          |
| V2 N4              | 3.35de      | 1.12cd     | 0.25d       | 1.39j       | 12.11            | 5.48                          |
| V3 N0              | 2.91gh      | 1.29bc     | 0.33c       | 1.62ghij    | 11.97            | 4.44                          |
| V3 N1              | 3.85ab      | 1.74a      | 0.36abc     | 1.74fgh     | 14.13            | 5.68                          |
| V3 N2              | 3.94a       | 1.13cd     | 0.35abc     | 1.90def     | 15.40            | 4.94                          |
| V3 N3              | 3.59bcd     | 1.02def    | 0.32c       | 1.71fghi    | 18.65            | 5.09                          |
| V3 N4              | 2.71h       | 1.02def    | 0.34bc      | 2.33ab      | 10.10            | 5.16                          |

Note: Different letters at the same of group treatment and colomn represent significant differences at Duncan’s Multiple Range Test (p < 0.05).

3. Discussion

Inoculation Bradyrhizobium sp. on Anjasmoro cultivar has higher shoot N content and N uptake of shoot compared with other N source treatments (Table 1). Anjasmoro cultivar required higher N nutrients for growth compared to Wilis and Sinabung, probably because Anjasmoro has higher plant growth rate and nett assimilation rate, so that N nutrients are indispensable for plant growth. N nutrient is obtained by mutualistic symbiotic between soybean and Bradyrhizobium sp. through the mechanism of assimilation of N in the form of direct ammonium from the nodule. In addition, the compatibility between Anjasmoro variety and Bradyrhizobium sp. determine the success rate in N absorption as indicated by an increase in shoot N content and N uptake of shoot (Table 1).
The high content of P and K canopy due to the urea treatment on the Anjasmoro cultivar showed that the Anjasmoro cultivar is more responsive to Urea fertilization compared to Wilis and Sinabung. Plant response to nutrient availability is highly dependent on nutrient uptake mechanisms by plants. Absorption of Urea containing 45% N results in increased availability of N so that vegetative growth is well and the ability of plant to absorb P increases. As a result, the ability of plants in the formation of seeds is also increased because P is very instrumental in the formation of primordia flowers, plant reproductive organs and as a key substrate in energy metabolism [10].

The high content of K due to Urea treatment on Anjasmoro cultivar is suspected to be related to the synergy between N and K nutrients. The increase in N uptake due to Urea fertilization results in increased K content, because K functions as an activator of enzymes in the process of plant metabolism and translocation of photosynthetic carbohydrates [11] so that the metabolic processes of plants is well done.

The role of *Bradyrhizobium* sp. in fixing N is seen from the increasing trend of total chlorophyll by the treatment of *Bradyrhizobium* sp. in this study. Total chlorophyll of leaf is important to see the contribution of biological nitrogen fixation to the fulfillment of plant nitrogen needs. As it is known that nitrogen is one of the constituents of the porphyrin ring in chlorophyll (Salisbury and Ross, 1995). The fact that nitrogen is one of the constituents of chlorophyll, while chlorophyll itself is the engine for the process of photosynthesis, making N also a factor affecting the rate of photosynthesis. If nitrogen supply is limited, chlorophyll may not form and will eventually decrease the rate of photosynthesis (Yoshiki *et al*., 2013).

4. Conclusions

Anjasmoro and Sinabung cultivars had a higher physiological characteristics (root N, shoot P and shoot K) compared to Wilis cultivar. Nitrogen source of Urea gave a higher physiological characteristics (content of root N, shoot Phosphor and shoot Potassium) compared to different treatment of N source in this study. The interaction between Anjasmoro cultivar and Urea gave the highest of content of shoot Phosphor and shoot Potassium, otherwise the interaction between Sinabung cultivar and *Bradyrhizobium* sp. gave the highest of content of shoot Nitrogen.

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