Changes in biochemical characteristics in shade soybean plants

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Abstract. Shade conditions will change the morpho-physiological and biochemical of plants. This study examined the biochemical characteristic changes including four shade soybeans varieties, total leaf sugar, seed protein, and seed fat content. A split-plot factorial design was used in this study with three replications. The treatments consisted of shading level as the main plot with 4 treatment levels, i.e. no shade, 30%, 50%, and 70% shade; soybean varieties as the subplot consisted of four varieties, i.e. Anjasmoro, Pangrango, Tanggamus, and Nanti. Analysis of total leaf sugar was using the total sugar measurement by seed protein and seed fat content measurement was conducted using Proximate Composition Analysis. The results showed an increase on total leaf sugar in shade condition. The Anjasmoro and Tanggamus varieties produced a lower seed protein and seed fat content compared to the other varieties at all shade levels. In contrast, the Nanti variety showed a high seed protein and seed fat content at all shade level conditions.

1. Introduction
One of the problems with soybean production was the cropland narrowing process. One of the existing solutions is by using the lands under the plantation crops, such as oil palm and rubber. However, this condition will cover the light and resulting in a low light intensity which affects the growth and yield of soybeans [1]. Shade plants will receive limited sunlight, even though the light has an important role for the plant growth and the development process. The pressure of low light intensity also results in agronomic, anatomical, physiological, molecular and biochemical characteristics changes which are related to photosynthetic efficiency that affect the plant yields [2].

2. Materials and Methods
This research was conducted in April to October 2009 using polybags in the arrangement shading house at the Experimental Farm, Faculty of Agriculture, Agriculture University Putra Malaysia (UPM) located at 25 m above the sea level.

2.1. Materials
The materials used were soybean seeds including a variety of Anjasmoro, Pangrango, Tanggamus, and Nanti (Table 1). According to the classification of [3], the size of soybeans could be classified into three
groups, namely small seed (<10 g/100 seeds), medium seed (10-12 g/100 seeds), and large seed (13-18 g/100 seeds). The Anjasmoro variety was in large seed group, the Pangrango and Tanggamus varieties were in medium seed groups and the Nanti variety was in small seed group. In this research, all varieties became the determinant of development type.

These varieties were screened from the earlier research, which showed that the Anjasmoro and Pangrango varieties were the consistently tolerant to shade condition, on the other hand, the Tanggamus and Nanti varieties were not. Based on grain yield per plant (g), it was found that Nanti variety had a small tolerance index while the Pangrango variety was able to tolerate 70% of shade condition and the Anjasmoro variety was able to tolerate 30% and 50% of shade condition [4].

Table 1. Description of soybean varieties [5]

| Source                  | Anjasmoro                      | Pangrango                      | Tanggamus                      | Nanti                      |
|-------------------------|--------------------------------|--------------------------------|--------------------------------|---------------------------|
| Source                  | Mass selection from pure strain Mansuria | Crossing local variety of Lampung x Davros in 1983 | Derivate selection of Kerinci x No. 3911 | Single cross of Dempo variety and No. 3623 |
| Potential Production(ton/ha) | 2.03-2.25                      | 1.4-2.0                        | 1.22                           | 1.24                       |
| Development type        | Determinite                    | Determinite                    | Determinite                    | Determinite               |
| The weight of 100 seeds | 14.8-15.3 g ± 10 g             | 11 g                           | 44 g                           | 10 g, oval shape          |
| Seed protein (%)        | 41.8-42.1                      | 39                              | 44.5                           | 42.8                      |

2.2. Methods.
A split-plot factorial design was used in this study with three replications. The treatments consisted of I. shading level (S) as the main plot with 4 treatment levels, i.e. no shade (S0), 30% shade (S1), 50% shade (S2) and 70% shade (S3); II. Soybean varieties (V) as the subplot consisted of four varieties, i.e. Anjasmoro (V1), Pangrango (V2), Tanggamus (V3), and Nanti (V4). Analysis of total leaf sugar was using the total sugar measurement by [6], seed protein and seed fat content measurement were conducted using Proximate Composition Analysis [7]. Then, the data were analysed by ANOVA (F test) at a test level of 5%, followed by the DMRT test at a test level of 5% [8]. The data were processed by SAS and Microsoft Excel program.

3. Results and Discussion

3.1. Total leaf sugar
The shade level treatment had a significant effect to total leaf sugar parameter, in contrast, the variety, and the two factors interaction had no significant effect. Table 2 showed the lowest total leaf sugar in soybean plants with no shade treatment which was significantly different from all shade levels. The highest total leaf sugar was found at 50% shade but not significantly different with 30% and 70% shade condition.
Table 2. Total leaf sugar in soybean at several shade levels and varieties

| Varieties | 0%   | 30%  | 50%  | 70%  | Average |
|-----------|------|------|------|------|---------|
| Anjasmoro | 1.02 | 1.23 | 1.80 | 1.20 | 1.31    |
| Pangrango | 0.96 | 1.15 | 1.25 | 1.17 | 1.13    |
| Tanggamus | 1.16 | 1.14 | 1.15 | 1.14 | 1.15    |
| Nanti     | 1.12 | 1.12 | 1.13 | 1.23 | 1.15    |
| Average   | 1.06 b| 1.16 ab| 1.33 a| 1.18 ab^a|

^a the numbers followed by an unqualified notation on the line indicated as significantly different at 5% level based on DMRT test

Figure 1 showed that a higher shade level or a lower of light received by the plant showed a higher percentage of total leaf sugar. Soverda [9] reported that a low light pressure intensity decreased the PGA kinase activity, a smaller decrease was found in the genotype of upland rice which was tolerant to shades compared to the sensitive genotypes. A low light intensity showed the carbohydrate translocation disorders, and the total sugar (in the part of non-reduction sugar and starch amount) actually was decreased in all parts of the plant. Meanwhile, Benkeblia [10] reported that total sugar varied in green leaves of onions, and increased progressively from week 10th to 14th, then sharply increased by two-fold after week 16th which mean that the total leaf sugar was depended on the age of leaves. In this research, the soybean leaves were picked up on the V5 phase, where the full leaves growth on the fifth internode [11]. This was presumably the best performance of vegetative growth [12].

3.2. Seed protein and seed fat content

The treatments of shade level, soybean varieties and the interaction of two factors were significantly influenced the seed protein and the seed fat content. Comparison of seed protein and seed fat content in percentage (%) between each variety at various shade levels could be seen in Figure 2. The highest soybean seed protein content was produced by Nanti variety in 70% and 50% shade level, while the lowest seed protein content was found in Anjasmoro variety with no shade (0%) which was not significantly different from Tanggamus variety.
The highest seed fat content was found in Nanti variety (70% shade) which was not significantly different from Pangrango at the same shade level. While the lowest seed fat content was found in Anjasmoro variety with no shade which was not significantly different to the 30%, 50% and 70% shade levels.

Figure 2. Comparison of (a) seed protein content and (b) seed fat content between each variety at various shade levels

In this study, we found a contradictory that Anjasmoro and Tanggamus varieties produced a lower seed protein and seed fat content than the other varieties at all shade levels. On the other hand, Nanti variety produced the highest seed protein and seed fat content at all shade levels. Seed protein and seed fat content of Anjasmoro variety actually were 41.8-42.1 and 17.2-18.6 respectively (Table 1). Meanwhile, Nanti variety produced a higher seed protein and seed fat content than its description, which was understandable that the condition was caused by genetic responses on environmental conditions [13]. Furthermore, Figure 2 showed that the seed protein and seed fat content was related to each other, increased and decreased in the same condition.

4. Conclusions
Total leaf sugar was increased in shade condition which was caused by the leaves that were picked up on the primary vegetative growth (V5). Anjasmoro and Tanggamus varieties produced a lower seed protein and seed fat content than the other varieties at all shade levels. On the other hand, Nanti variety produced the highest seed protein and seed fat content at all shade levels condition.
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