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Research Article

Effects of Nitrogen Level on Nitrogen Partitioning and Harvest Index in *Brassica napus L.*

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ABSTRACT

To investigate the impact of nitrogen (N) mineral on reproductive potential of *Brassica napus* L, plants were treated with different levels of N treatment (N_0; N_{100}; N_{500}). The half of N content for each treatment were applied at the beginning of the early vegetative stage and the rest was applied at the late vegetative stage. Nitrogen content in plant tissues such as root, stem and branch, leaf, pod and seed was analyzed and harvest index (HI) was calculated as percentage of seed yield to total plant weight. Biomass and nitrogen content were significantly affected by different levels of N supply. Biomass was significantly decreased by 59.2% in nitrogen deficiency (N_0) but significantly increased by 50.3% in N excess (N_{500}), compared to control (N_{100}). Nitrogen content in all organs was remarkably increased with nitrogen levels. N distribution to stem and branches, and dead leaves was higher in N-deficient (N_0) and N excessive plants (N_{500}) than in control (N_{100}). However, nitrogen allocated to seed was higher in control (N_{100}) than in other treatments (N_0 or N_{500}), accompanied by higher HI. These results indicate that the optimum level of N supply (N_{100}) improve HI and N distribution to seed and excessive N input is unnecessary.

(**Key words** : *Brassica napus*, Harvest Index, Nitrogen, Nitrogen partitioning)

Ⅰ. INTRODUCTION

Nitrogen (N) is an integral nutrient for plant growth and development, which is a component of amino acids required to synthesize proteins and other related compounds such as providing green color to the plants due to increase of chlorophyll. In addition, N is essential in plants processes such as photosynthesis. For example, high N in plants induces high photosynthesis, consequently leads to high plant growth and development. These indicate that N is a key factor influencing crop yield. Therefore, N deficiency affects the obtained crop yield and quality, which depends on the growth stage and duration (Zhao et al., 2012).

Nitrogen is taken up by roots, transported to leaves and then assimilated to amino acids and proteins at rosette stage. Stored metabolites in leaves are remobilized to sustain growth of reproductive tissues such as pods and seeds. Thus, amount of stored nitrogen in leaves and availability of N transport via the phloem would be important in seed filling and seed yield (Noquet et al., 2004; Malagoli et al., 2005). Habekotté (1993) suggested enhancement of the utilization of assimilates before flowering by increasing accumulation of reserves and enhancement of the efficiency of translocation during seed filling may result in higher final seed weight. In addition, Masclaux-Daubresse et al. (2010) suggested that nitrogen remobilization is important for seed yield. Therefore, it would be interesting to study regulation of balance between stored N metabolites in source leaves and translocation of N into sink organs, pods and seeds.

Oilseed rape (*Brassica napus* L.) has become a major agro-economic crop for human and animal food. Their production has increased by 5-fold in the world for the last three decades (Food and Agriculture Organization). Oilseed rape is high N-demanding plants contrary to cereals (Rathke et al., 2005; Hocking et al., 1997). However, low N utilization

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has been well reported in oilseed rape. Rossato et al. (2001) showed that only 48% of total N was recovered in the mature pods, as previously shown in seeds by Schjoerring et al. (1995). Consequently, this occur ecological problem regarding N loss in the field, leads soil and water pollution. In addition, loss of N fertilizer in farmer resulted in economic loss because of high N cost. Therefore, economic optimum level of N fertilizer should be considered to produce high seed yield and quality (Firbank, 2005) which is a goal of agronomic studies. This study aims to investigate effects of different levels of N supply on N uptake and partitioning, and seed yield.

Ⅱ. MATERIALS AND METHODS

1. Plant growth and treatment

In the greenhouse, seeds of *Brassica napus* L. (cv. Tammi) were sown in seedling trays and covered with moist papers to maintain the humidity for germination. After germination, the moist papers were removed and the seedlings were carefully maintained with normal irrigations for about one month and then transplanted into the fields. For plant cultivation, several plots of $1m^2$ size were prepared and four rows in each plot were made. Five plants were transplanted in each row and thus 20 plants were transplanted in each plot. The gaps between the plants were 20 cm and between the rows were 25 cm in each plot. Between the two plots, 1 m gap was kept for the maintenance of the plants. After the transplantation of the seedlings, sufficient irrigations were applied regularly for the normal growth and development of the plants. For the treatments, urea ($CO(NH_2)_2$) was used as the sources of nitrogen (N). Three different levels of N mineral were applied in the field, such as N_0 (N deficiency); N_{100} (Control, 150 kg ha^{-1}); N_{500} (N excess, 750 kg ha^{-1}). To maximize effect of N excess to soil buffering capacity in field condition, five times higher N was supplied. The half of N content for each treatment were applied at the beginning of the early vegetative stage and the rest was applied at the late vegetative stage. Plants were harvested at the mature seed stage and then divided into root, stem and branch, leaves, pod, and seed. Dry weight (DW) of each part was determined by drying samples in an oven at 60 °C for seven days to remove water.

2. Determination of Nitrogen

To determine the N content in plant tissues, dried samples were ground into fine powder. Approximately 200 mg were analyzed using a C/N/S analyzer (EA3000, Euro Vector, Milan, Italy). nitrogen partitioning was calculated the N proportion of each plant organ in whole plant N content.

3. Determination of harvest index (HI)

In general, harvest index is grain weight as a percentage of total above-ground dry weight at maturity, and it was calculated as followed:

Harvest index (HI) = seed weight (g DW per plant)/ shoot biomass (g DW per plant)

4. Statistical Analysis

Duncan multiple range test were used to compare the means of three replicates in three treatments. Different letters in each series of treatments indicate that the mean values are significantly different at $p < 0.05$ according to Duncan’s multiple range test by using SAS 9.4 software.

Ⅲ. RESULTS & DISCUSSION

1. Biomass

It has been well reported that nitrogen (N) is important factor for plant growth. As expected, in the present study, plant biomass was significantly affected by different levels of nitrogen (N) supply (Table 1). Total biomass was significantly increased (+50.4%) and decreased (-60%) by N-excessive (N_{500}) and -deficient (N_0) supply, respectively, compared to control (N_{100}). A significant increase of root biomass (2.6-fold) was observed in N-excessive plants but not in N deficient plants, compared to control. Shoot biomass was significantly affected by different levels of N supply, showing increase (+46.9%) in N excess and decrease (-62.6%) in N deficiency treatment compared to control. Biomass of the stem and branch, as well as dead leaves, was increased accordance with increasing levels of nitrogen supply. Similar results were observed in pod. Seed yield was significantly decreased by N deficiency treatment, whereas it was not changed in N excess treatment compared to control. Similar results were observed by Kulsum (2007), who reported the

Table 1. Biomass of *Brassica napus* under different levels of nitrogen treatment at mature seed stage

Treatment	Biomass (g DW)						
	Root	Stem and branch	Dead leaves	Pods	Seeds	Shoot biomass	Total biomass
N_0	1.41±0.34^b	4.84±0.22^c	1.70±0.09^c	4.77±0.76^c	3.32±0.63^b	14.63±1.7^c	16.04±2.04^c
N_{100}	1.15±0.63^b	9.16±0.99^b	3.14±0.28^b	15.79±2.42^b	10.97±2.25^a	39.06±5.38^b	40.21±6.01^b
N_{500}	3.06±0.32^a	14.79±2.12^a	5.72±0.21^a	22.92±0.76^a	13.97±0.55^a	57.39±2.12^a	60.45±1.81^a

Values are means ± *SD* for n = 3. Values in a vertical column followed by different letters are significantly different at $p < 0.05$ according to Duncan's multiple range test.

Table 2. Nitrogen content of root, stem and branches, dead leaves, pods, and seeds of Brassica napus under different levels of nitrogen treatment at mature seed stage

Treatment	Nitrogen content (mg N/plant DW)					
	Root	Stem and branch	Dead leaves	Pods	Seeds	Total
N_0	14.03±4.66^a	38.18±3.26^c	19.03±2.33^c	63.45±9.62^c	112.09±20.05^b	246.79±39.92^c
N_{100}	17.71±2.35^a	74.90±8.85^b	35.26±7.05^b	199.81±26.23^b	360.32±94.64^b	688.00±125.01^b
N_{500}	19.46±0.86^a	137.65±25.58^a	71.22±3.35^a	312.28±10.25^a	460.54±45.69^a	1001.16±12.35^a

Values are means ± SD for n = 3. Values in a vertical column followed by different letters are significantly different at $p < 0.05$ according to Duncan's multiple range test.

highest seed yield in moderate level of nitrogen. These results were conducted with previous results observed by He et al. (2017). Therefore, these results indicate that optimum level of the N supply is crucial for the seed production.

2. Nitrogen content in plant tissues

The N content in plant tissues was strongly reflective of increasing N levels (Table 2). As expected, the total N amount was the lowest in the N deficient plants, while it was the highest in the N excessive plants. N contents in stem and branch and dead leaves were more than 1.83-fold higher in N excessive plants than control, whereas they were less than 49% lower in N deficient plants. Similarly, Dreccer et al. (2000) reported that the highest N content was observed in dead leaves of N excess treatment. Thus, the high N residues in dead leaves would be resulted in low seed yield, as suggested by Avice and Etienne (2014). In the present study, N excessive plants increased N content of pods and seeds by 56.3% and 27.8%, respectively, compared to control. However, N deficient plants decreased more than 68%. No significant difference observed in roots. These results indicate that a large part of the N stored in source organ such as source leaves, branch and stem is not fully remobilized into sink organ such as pods and seeds, especially N excessive plants.

3. Partitioning of nitrogen in plant

Nitrogen distribution in plant tissues was shown in Fig. 1. Most of the nitrogen distributed to sink organs such as pod and seed (more than 71%). N distributed to seeds was the highest in the control as a 52%. However, N distributed to pods was the highest in N excess treatments, accordance with previously reported results (Malagoli et al., 2005; Koeslin-Findeklee and Horst, 2016). On contrast, N distributed to dead leaves, and branch and stem was the lowest in the control as 5% and 11%, respectively, providing high N remobilization. Their relatively high proportion was observed in N excess and deficiency treatments, as shown by Schjoerring et al. (1995) who suggested that large amounts of nitrogen loss occurred in source organ of oilseed rape because of low nitrogen use efficiency. In addition, N proportion of root was decreased as increasing levels of nitrogen supply. These results indicate that N excess or N deficiency restricts N remobilization from source organ to sink organ, resulting high N loss.

4. Harvest Index (HI)

For determining crop yield, harvest index (HI) is an important index because of a positive correlation between HI and grain yield to measure retranslocation efficiency of absorbed N from vegetative plants parts to grain (Sinclair, 1998; Li et al, 2003).

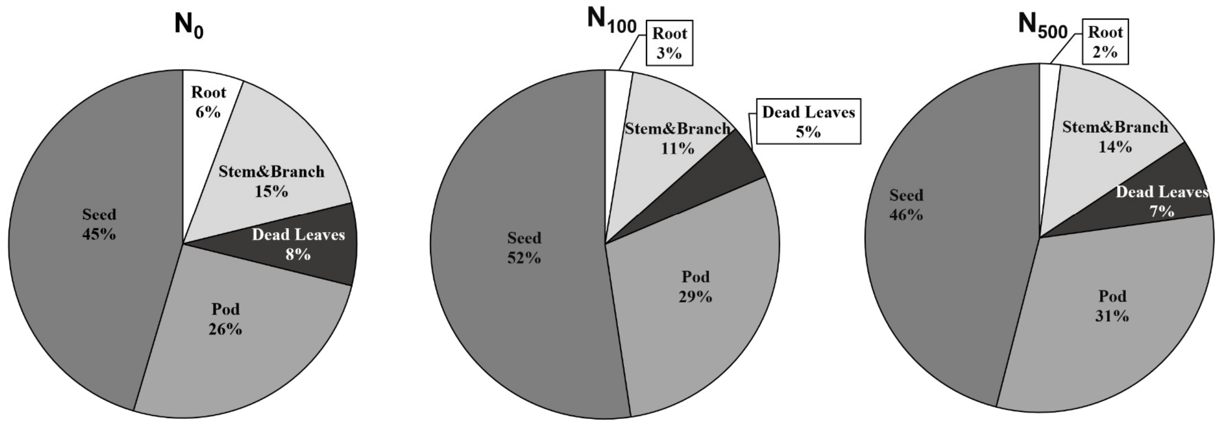

Figure 1. Nitrogen partitioning in the different tissues of *Brassica napus* under under different levels of nitrogen treatment at mature seed stage.

HI is defined to the ratio of grain yield to total biomass. HI was shown 0.225, 0.278, and 0.244 in N deficiency, control, and N excess treatment, respectively (Fig. 2). Compared to control, N deficient treatment showed (−19%) significantly lower HI. On the other hand, N excess treatment was 13.9% lower than control but not significant whatever 5 times higher nitrogen supply. It has been reported that HI increased depends on increasing levels of nitrogen, and it was constant over specific levels of N (Shivay et al., 2016; Farooq et al., 2017) or decreased (Keivanrad et al., 2012). Therefore, these results indicate that optimum level of N is very important for the remobilization of N from source to sink as well as productivity.

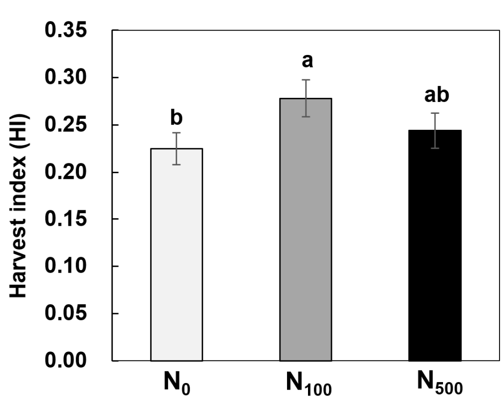

Figure 2. Nitrogen index (HI) in Brassica napus under different levels of nitrogen treatment at mature seed stage. Data presented as mean ± *SD* for n = 3. Bar labelled with the different letters are significantly different at $p < 0.05$ according to Duncan’s multiple range test.

Ⅳ. CONCLUSION

In conclusion, N nutrition effects on seed yield and HI was significant. Excessive N-supplied plants produced higher biomass and N uptake. However, high N content was observed in source organ such as leaves, stem and branch and HI was lower than control, providing low N remobilization form source to sink organ. Similarly, N-deficient plants showed low HI with high N residues in source organs. Therefore, our results suggest that the optimum level of N supply is economic method to enhance HI due to increase of N distribution to seed but excessive N input is unnecessary for increase seed production.

Ⅴ. ACKNOWLEDGEMENT

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