COBISS Code 1.01

DOI: 10.2478/acas-2013-0014
**Agrovoc descriptors:** brassica napus, rapeseed, oilseeds, rapeseed oil, crop yield, nitrogen fertilizers, protein content, lipid content, proximate composition, seed, seeds

**Agris category code**: f04, f60

# Effect of nitrogen rate on seed yield, protein and oil content of two canola (*Brassica napus* L.) cultivars

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Received January 28, 2013; accepted August 30, 2013.
Delo je prispelo 28. januarja 2013, sprejeto 30. avgusta 2013.

**ABSTRACT**

A field experiment was conducted at Rice Research Station, Tonekabon, Iran, to determine the effect of N rate on seed yield, protein and oil content of two canola (*Brassica napus* L.) cultivars. Two canola cultivars ('Hayola-308' and 'RGS-003') and five N rates (0, 50, 100, 150, and 200 kg ha$^{-1}$), organized into a randomized complete block design with a factorial treatment arrangement and three blocks, were applied to plot areas. Results showed that N rate effect was significant ($P$ < 0.01) for seed yield, protein content and yield, and oil yield but not for oil content. On the other hand, cultivar had only significant ($P$ < 0.01) effect on seed protein and oil content. Moreover, the interaction between N rate and cultivar was significant at $P$ < 0.01 for seed, protein and oil yield, illustrating that cultivars showed different responses to N rates for these traits. In general, the quadratic equation provided a good description of the relationship between seed, protein and oil yield and nitrogen rate. For 'Hayola-308', seed, protein and oil yield increased significantly as N application rate increased from 0 to 150 kg ha$^{-1}$, but thereafter remained constant. In contrast, for 'RGS-003', seed, protein and oil yield increased significantly as N application rate increased from 0 to 200 kg ha$^{-1}$. Therefore, at the highest N application rate (200 kg ha$^{-1}$), 'RGS-003' produced greater seed, protein and oil yield than 'Hayola-308'. Averaged across N application rate, the seed protein content of RGS-003 was significantly ($P$ < 0.01) higher than that of 'Hayola-308', while the opposite result was observed for seed oil content. This study demonstrated the differential response of two canola cultivars to N rate in terms of seed, protein and oil yield.

**Key words:** canola, nitrogen rate, oil, protein

**IZVLEČEK**

**VPLIV GNOJENJA Z DUŠIKOM NA PRIDELEK SEMEN, VSEBNOST BELJAKOVIN IN OLJA PRI DVEH SORTAH OLJNE OGRŠČICE (*Brassica napus* L.)**

Za določanje vpliva različnega gnojenja z dušikom na pridelek semen in vsebnost beljakovin in olja v dveh sortah oljne ogrščice (*Brassica napus* L.) je bil izveden poljski poskus na Rice Research Station, Tonekabon, Iran. Dve sorti oljne ogrščice ('Hayola-308' in 'RGS-003') sta bili posejani v petih obravnavanjih z dušikom (0, 50, 100, 150, in 200 kg ha$^{-1}$) v naključnem bločnem poskusu s faktorsko obravnavo v treh blokih. Rezultati so pokazali, da je gnojenje z N statistično značilno ($P$ < 0.01) vplivalo na pridelek semen, vsebnost beljakovin in pridelek olja, ne pa na vsebnost olja. Po drugi strani sta imeli sorti statistično značilen vpliv ($P$ < 0.01) samo na vsebnost beljakovin in olja v semenu. Še več, interakcija med obravnavanji z N in sortami je bila statistično značilna ($P$ < 0.01) za pridelek semen, beljakovin in olja, kar kaže na različen odziv sort v teh znakih na gnojenje z dušikom. V splošnem je kvadratna enačba dobro opisala razmerja med pridelkom semen, beljakovin in olja z gnojenjem z dušikom. Pri sorti 'Hayola-308' je pridelek semen, beljakovin in olja statistično značilno naraščal pri uporabi od 0 do 150 kg N ha$^{-1}$, potem je ostal konstanten. Nasprotno, se je pri sorti 'RGS-003' pridelek semen, beljakovin in olja značilno povečeval od 0 do 200 kg N ha$^{-1}$. Sorta 'RGS-003' je pri obravnavanju z največjo količino dušika (200 kg N ha$^{-1}$) dala večji pridelek semen, beljakovin in olja kot sorta 'Hayola-308'. Povprečno je bila pri vseh obravnavanjih z dušikom vsebnost beljakovin značilno večja pri sorti 'RGS-003'($P$ < 0.01) kot pri sorti 'Hayola-308', obratni so rezultati za vsebnost olja. Raziskava je pokazala različen odziv pridelka semen, beljakovin in olja dveh sort oljne ogrščice na gnojenje z dušikom.

**Ključne besede:** oljna ogrščica, gnojenje z dušikom, pridelek, semena, olje, beljakovine

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## 1 INTRODUCTION

Canola (*Brassica napus* L.) is a member of the mustard family that is grown for the production of animal feed and vegetable oil for human consumption. Canola oil has the lowest levels of saturated fat compared to some other vegetable oils. Although canola is a summer crop in the temperate and cool areas of the world, it is mainly grown in the northern Iran as a winter crop in rotation with rice. In this area, canola should be planted in mid October to early November and harvested in mid-late May to achieve the highest yields.

Nitrogen (N) is an essential nutrient for plant growth and is a key limiting factor in agro-ecosystems. Nitrogen is a constituent of amino acids, which are required to synthesize proteins and other related compounds. It plays a role in almost all plant metabolic processes. Nitrogen is a part of chlorophyll, the green pigment of the plant that is responsible for photosynthesis. Plant growth and developmental aspects such as seed germination, leaf development (Walch-Liu et al., 2000), flower and fruit development (Stitt et al., 2000), root architecture (Zhang and Forde, 1998) can be affected by the amount of N supplied to plants. N fertilizer mainly increases canola leaf area index, leaf duration (Wright et al. 1988), growth rates, number of flowering branches, plant height, number of flowers, number and weight of siliquae and seed yield (Grant and Bailey, 1993).

It has been frequently reported that N fertilizer increased seed yield of canola and winter oilseed rape (Taylor et al. 1991; Asare and Scarisbrick, 1995; Hocking et al., 1997; Brennan et al., 2000; Jackson, 2000; Cheema et al., 2001; Hocking and Stapper, 2001). Rathke et al. (2005) reported that application of N fertilizer increased the seed yield of winter oilseed rape. Nevertheless, some researchers documented a stagnation or reduction in seed yield at high N- rates. At the same time, N fertilization generally increases the protein content of canola seed and meal. In contrast, N fertilization usually has little effect on canola seed oil content (Brennan et al. 2000) or may significantly decrease it, especially at higher rates (Cheema et al., 2001). Moreover, it has been reported that canola cultivars had different response to N fertilizer (Svecnjak and Rengel, 2006). Therefore, this study was conducted to study seed yield, protein and oil content response of two canola cultivars to N fertilizer application.

## 2 MATERIALS AND METHODS

A field experiment was conducted at Rice Research Station, Tonekabon (36$^o$ 51' N, 50$^o$ 46' E; -20 m above sea level), northern Iran, from early November 2011 through late May 2012. Soil properties were 2.71% organic matter content, 33% clay, 42% silt, 25% sand and 6.9 pH.

Two canola (*Brassica napus* L.) cultivars (‘Hayola-308’ and ‘RGS-003’) and five N rates (0, 50, 100, 150, and 200 kg ha$^{-1}$), organized into a randomized complete block design with a factorial treatment arrangement and three blocks, were applied to plot areas. Plot size was 3 m × 3 m, with a row spacing of 30 cm and a plant spacing of 5 cm between plants. Planting date for both canola cultivars was 21 October 2011.

After the harvest of rice, the soil was disked (cross-disking) in the autumn to a depth of 15–20 cm consistent with local practices in north of Iran. Half of nitrogen fertilizer (applied as urea) was incorporated into the top 5 cm of soil two weeks before sowing time and remaining half nitrogen was top dressed in two split doses at stem elongation and flowering stages. Moreover, triple super phosphate and potassium sulfate were applied to provide 50 kg $P_2O_5$ ha$^{-1}$ and 75 kg $K_2O$ ha$^{-1}$ at each plot, respectively, and incorporated before sowing. Weeds were controlled by trifluralin (2.5 l ha$^{-1}$) application before seed sowing and after this by manual removal if necessary.

At maturity stage, to determine seed yield, seeds were collected from 2 m$^2$ in each plot and subsequently was adjusted to 9% moisture content. N content in seed was determined by the Kjeldahl method and then protein content in seed was calculated by multiplying the N content in seed by 6.25 (Williams et al., 1998). Protein yield was calculated by multiplying seed yield by protein content. Oil content was determined by nuclear-magnetic resonance as described by Robertson and Morrison (1974). Oil yield was calculated by multiplying seed yield by oil content.

Statistical analysis of the data was done by standard analysis of variance (ANOVA) using the SAS software package version 9.1.3 (SAS Institute, 2004). For cultivar factor, where the F-ratios were found to be significant, treatment means were compared by fisher's protected LSD at the 5% level. For N rate factor, where the F-ratios were found to be significant, quadratic regressions with standard error of the mean were used to describe the relationship between N application rate and dependent variables such as grain yield, protein content and yield, and oil yield.

## 3 RESULTS AND DISCUSSION

### 3.1 Seed yield

The main effect of N rate was significant ($P$ < 0.01) for canola seed yield, but the main effect of cultivar was not significant. Nevertheless, the interaction between cultivar and N rate was significant at $P$ < 0.01 level (Table 1), illustrating that cultivars showed different responses to N rates for seed yield. For both cultivars, a positive quadratic equation expressed the relationship between N application rate and canola seed yield (Figure 1). However, for ‘Hayola-308’, seed yield increased rapidly as N application rate increased from 0 to 150 kg ha$^{-1}$, but did not significantly increase at higher N rate. In contrast, for ‘RGS-003’, seed yield increased significantly as N application rate increased from 0 to 200 kg ha$^{-1}$. Therefore, at the highest N application rate, ‘RGS-003’ produced greater seed yield than ‘Hayola-308’ (Figure 1). This result is consistent with data of Qayyum et al. (1998) who reported that canola grain yield increased significantly when N rate was increased from 0 to 180 kg ha$^{-1}$. Moreover, Cheema et al. (2001) reported that the seed yield of canola increased as N application rate increased from 0 to 90 kg ha$^{-1}$, while at the highest N application rate (120 kg ha$^{-1}$), canola seed yield was significantly reduced.

**Table 1**: Mean squares of ANOVA for seed yield (Y), seed protein content, protein yield, seed oil content, and oil yield as affected by cultivar, and N rate.

| Source | df | Seed yield | Seed protein content | Protein yield | Seed oil content | Oil yield |
|---|---|---|---|---|---|---|
| R | 2 | 71158 $^{ns}$ | 2.58$^{ns}$ | 9251$^{ns}$ | 1.56 $^{ns}$ | 5443 $^{ns}$ |
| Nitrogen (N) | 4 | 6356476$^{**}$ | 3.70 $^{**}$ | 444005 $^{**}$ | 3.75 $^{ns}$ | 1023596$^{**}$ |
| Cultivar (C) | 1 | 17376 $^{ns}$ | 13.12$^{**}$ | 11029 $^{ns}$ | 20.68$^{**}$ | 1887 $^{ns}$ |
| N×C | 4 | 200138 $^{**}$ | 0.30 $^{ns}$ | 12625 $^{**}$ | 0.001$^{ns}$ | 29298 $^{**}$ |
| Error | 18 | 53076 | 0. 18 | 2840 | 1.53 | 6502 |

*, ** represent significance at 0.05 and 0.01probability level, respectively.
ns represents no significant difference

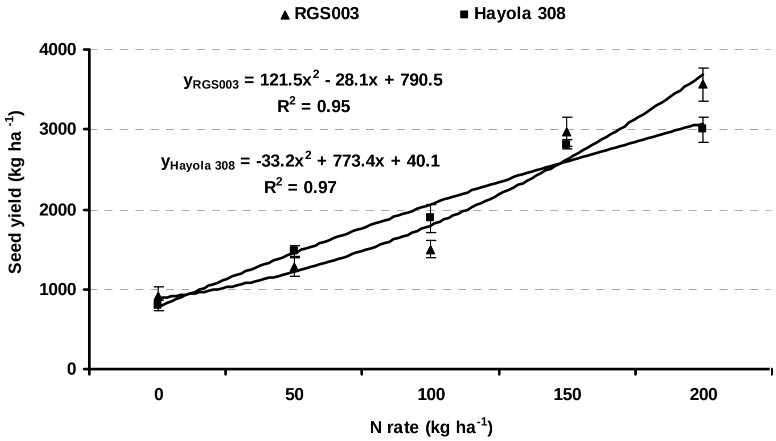


**Figure 1:** Effect of N rate on seed yield of two canola cultivars (‘RGS-003’ and ‘Hayola-308’)

### 3.2 Protein content

Analysis of variance showed that protein content varied significantly ($P$ < 0.01) because of cultivar and N rate, but these factors did not interact significantly (Table 1). Averaged across N application rate (Table 2), the seed protein content of ‘RGS-003’ (25.5%) was significantly ($P$ < 0.01) higher than that of ‘Hayola-308’ (24.1%). Regardless of canola cultivar, seed protein content followed a positive quadratic relationship as N rate increased from 0 to 200 kg ha$^{-1}$. The highest protein content of 25.8% was recorded when 200 kg N ha$^{-1}$ was applied, this value being significantly higher than the values recorded in other N rates except 150 and 100 kg N ha$^{-1}$, where the difference was statistically non-significant (Figure 2). Between the N rates of 0 to 200 kg N ha$^{-1}$, seed protein content increased by 2.1% when averaged across canola cultivar. These results are confirmed by those reported by Kutcher et al. (2005), Asghar et al. (2003) and Saleem et al. (2001) who concluded that increasing nitrogen fertilizer rate had a significant positive effect on the protein content of canola seed. As nitrogen is the major constituent of protein, increases in N fertilizer application frequently lead to an increase in protein content (Brennan and Bolland, 2007 a, b; Malhi and Gill, 2007).

**Table 2:** Seed yield, protein content, protein yield, oil content, and oil yield of two canola cultivars when averaged across N rate.

| Trait / Cultivar | Seed yield (kg ha$^{-1}$) | Protein content (%) | Protein yield (kg ha$^{-1}$) | Oil content (%) | Oil yield (kg ha$^{-1}$) |
|---|---|---|---|---|---|
| ‘RGS-003’ | 2043 | 25.5 | 526 | 41.02 | 828 |
| ‘Hayola-308’ | 1995 | 24.1 | 488 | 42.68 | 845 |
| LSD (0.05) | 221 | 0.3 | 52 | 0.95 | 77 |

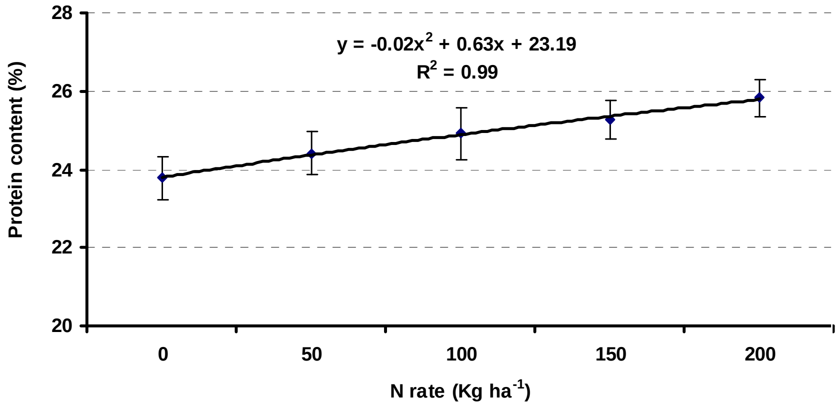


**Figure 2:** Seed protein content of canola as influenced by N application rate, averaged over cultivars.

### 3.3 Protein yield

The rate of N fertilizer application significantly ($P < 0.01$) affected protein yield (Table 1). Despite observed differences in seed protein contents between 'RGS-003' and 'Hayola-308', the cultivars had similar protein yield (Table 2) as variations in protein content were offset by differences in seed yield. Moreover, effect of N rate × cultivar interaction was significant at $P < 0.01$ level (Table 1), indicating different response of canola cultivars in protein yield to N application rate. For 'RGS-003' and 'Hayola-308', quadratic equations (Y= 31.4 $X^2$ +1.5 X + 184.9, $R^2$= 0.96 and Y= -3.3 $X^2$ +196.5 X + 16.0, $R^2$= 0.97, respectively) provided a good description of the relationship between protein yield and nitrogen rate. As shown in figure 3, at lower N rate (0, 50, and 100 kg ha$^{-1}$), there was no significant difference between 'RGS-003' and 'Hayola-308' for protein yield, but at higher N rate, 'RGS-003' produced higher protein yield than 'Hayola-308' (Figure 3).

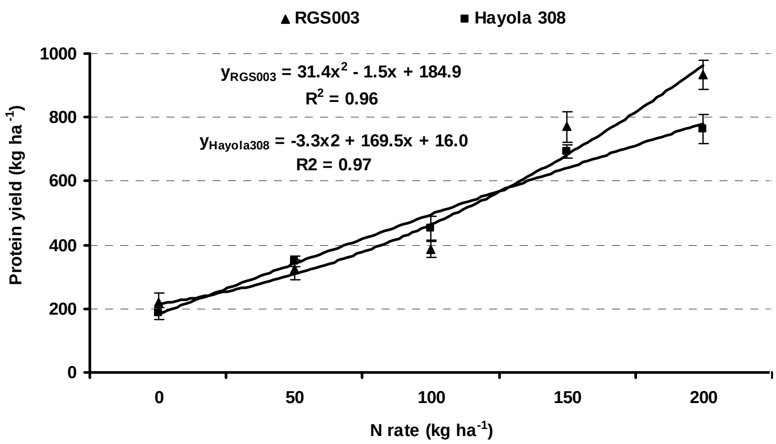


**Figure 3:** Effect of N rate on protein yield of two canola cultivars ('RGS-003' and 'Hayola-308').

### 3.4 Oil content

N rate had no significant effect on oil content, but the effect of cultivar was significant ($P < 0.01$). The interaction between N rate and cultivar was not significant at $P < 0.01$ level, illustrating that cultivars showed similar responses to N rates for oil content (Table 1). Seed oil content ranged from 42.8% in the unfertilized plot to 40.9% in plot which received the highest N rate, although these differences were not statistically significant (data not shown). Similar result was reported by Dreccer et al. (2000) for winter oilseed rape. In contrast, it has been reported that seed oil content in canola (Jan et al., 2002; Saleem et al., 2001; Cheema et al., 2001; Hocking et al., 1997; Taylor et al., 1991) and winter oilseed rape (Rathke et al., 2005) reduced significantly as N application rate increased. Moreover, Cheema et al. (2001) reported that the highest oil content was recorded in unfertilized winter oilseed rape while the lowest one appeared at high N- supply. Seed oil content of 'Hayola-308' was significantly ($P$ < 0.01) higher than that of 'RGS-003' (Table 2).

### 3.5 Oil yield

Although N rate had no significant effect on oil content, oil yield was significantly ($P$ < 0.01) affected by N rate (Table 1). This was due to higher seed production at higher N application rate. Moreover, oil yield did not significantly affect by cultivar (Tables 1 & 2). On the other hand, the interaction between N rate and cultivar was significant for oil yield, indicating varietal differences of oil yield response to N application rate. The relationship between N rate and oil yield was well fitted by a quadratic curve for both cultivars. For 'Hayola-308', oil yield increased significantly as N application rate increased from 0 to 150 g ha$^{-1}$, whereas there was only a small rise in oil yield from 150 to 200 kg N ha$^{-1}$ (Figure 4). In contrast, for 'RGS-003', oil yield increased significantly as N application rate increased from 0 to 200 kg ha$^{-1}$. At the N rate of 50 and 100 kg ha$^{-1}$, 'Hayola-308' produced greater oil yield compared to 'RGS-003', but at the highest N rate, oil yield of 'RGS-003' was significantly ($P$ < 0.01) higher than that of 'Hayola-308'. Rathke and Schuster (2001) reported that seed oil yield of canola remained constant when N application rate increased from 160 to 240 kg ha$^{-1}$. Moreover, Cheema et al. (2001) declared that increasing the rate of N fertilizer application up to 90 kg ha$^{-1}$ significantly increased oil yield, but thereafter oil yield was significantly reduced.

Canola grain yield was positively correlated with seed protein content, seed protein yield, and seed oil yield at $P$ < 0.01 level, but negatively correlated with seed oil content at $P$ < 0.01 level (Table 3). Moreover, there was a significant ($P$ < 0.01) negative correlation between oil content and protein content. Protein and oil are two main components of canola seed. Increases in protein content in response to N fertilizer application normally result in a corresponding decrease in oil content (Ahmad et al., 1999; Brennan et al., 2000; Prithchard et al., 2000; Brennan and Bolland, 2007 a, b; Malhi and Gill, 2007). Many researchers reported that N fertilizer application enhanced the protein content at the expense of oil content (Andersen et al., 1996; Rathke et al., 2005). In general, high protein content is correlated with low oil content and vice versa (Asare and Scarisbrick, 1995; Andersen et al., 1996). Higher oil content would be beneficial for oil extracting industry, however low protein will decrease the quality of feed to be used for livestock.

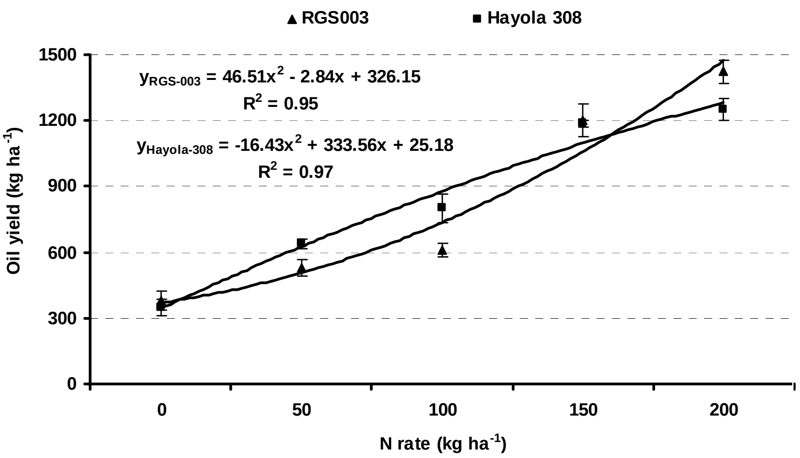


**Figure 4:** Effect of N rate on oil yield of two canola cultivars ('RGS-003' and 'Hayola-308').

**Table 3:** Correlation coefficients for measurements of canola as influenced by N rate and cultivar.

| Parameter | Seed yield | Protein content | Protein yield | Oil content |
|---|---|---|---|---|
| Protein content | 0.55 ** | | | |
| Protein yield | 0.99 ** | 0.60 ** | | |
| Oil content | -0.56 ** | -0.64 ** | -0.59 ** | |
| Oil yield | 0.99 ** | 0.52 ** | 0.99 ** | -0.51 ** |

** Significant at the 0.01 probability levels

## 4 CONCLUSION

This experiment documented that N fertilizer had significant ($P < 0.01$) positive effect on seed, protein and oil yield of canola cultivars, although seed, protein and oil yield responses of two canola cultivars to N rate were different. For ‘Hayola-308’, seed, protein and oil yield increased significantly as N application rate increased from 0 to 150 kg ha$^{-1}$, but thereafter remained constant. In contrast, for ‘RGS-003’, seed, protein and oil yield increased significantly as N application rate increased from 0 to 200 kg ha$^{-1}$. Averaged across N application rate, the seed protein content of ‘RGS-003’ was significantly higher than that of ‘Hayola-308’. This study demonstrated the differential response of canola cultivars to N rate in terms of seed, protein and oil yield.

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