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Evaluation of plant densities on analysis of growth indices in two canola forage (*Brassica napus* L.)

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ABSTRACT

In order to investigate the effect of the amount plant density on yield, yield components and growth characteristics of spring type of forage canola in summer cultivation, this experiment was conducted in Ghazvin(Esmail Abad) at 2011- 2012. This experiment was done in split plot form and with a randomized complete block design with three replications. The main factor included five levels of plant density :(100, 125, 150, 175 and 200 plant per m^2). The sub factor included two varieties of spring type of Canola including RGS003 and SARIGOL. The impact of plant density ($p \leq 0.01$) on final dry forage in stem elongation was significant. The highest dry forage yield in stem elongation was obtained from applying 175 plants per m^2 and RGS003 variety with the average of 665.4($kg.h^{-1}$). Increasing or decreasing in plant density resulted to decreasing in LAI. The effect of plant density and cultivars were significant on crop growth rate (CGR), relative growth rate (RGR), net assimilation rate (NAR), specific leaf weight (SLA), leaf area ration (LAR) and leaf area index (LAI).

Key words: Canola (*Brassica napus* L.), Plant density (PD), forage fat yield, protein yield.

INTRODUCTION

Oilseed rape is cultivated and processed for many different purposes. The importance of rape has thus increased in recent years and today it is one of the most important oil seed crops in the world (Bybordi et al., 2009)[3]. Canola (*Brassica napus* L.) belonged to *Crucifer* a family has received remarkable attentions for forage production potential as well as oil and meal source, to the best of our knowledge, there are rare researches in literatures on forage canola in Iran, however, in recent years, it has been central focused research area. Canola forage has been widely cultivated and used since 600 years ago for feeding livestock (Fitzerald and Black, 1991), although its water demand is exorbitant as summer forage [9]. Average Canola forage yield in three harvesting dates ranged from 4350 to 5690 ($kg.h^{-1}$). Harvesting at September gave 5540 ($kg.h^{-1}$) forage yield, while at end of October, it was amounted to 7900 ($kg.h^{-1}$) (Morison, 1990)[22]. Canola is first choice to supplying needed vegetable oil to country. According studies Canola planting is more considerable than other oily seeds due to its compatibility with most the country region and it's higher qualitative oil. In this experiment studied effect of planting density on growth traits of canola varieties. Canola contains 40-48% oil, 38-45% protein in the meal with 5% grain moisture. Linoleic to linolenic acids ratio in canola oil is known to be 2:1 which is normal for human diets purposes. Canola meals contain 13% fiber. Much fiber concentration present in meal serves as a limiting factor for feeding livestock, because it loses potential to release energy in ration. Analysis of quantitative aspects of growth of whole plant can be effectively conducted using the functional growth analysis techniques which use regression procedure. Yield is a complex trait resulting from the interaction of morphological, physiological and environmental parameters on the growth of plants. Identification of the variations of morphological and physiological traits influencing the yield of a plant in a certain

environment is an essential tool for selecting and breeding of yield (Abayomiand and Adedoyin, 2004)[1]. The growth of the plants in certain environmental conditions can be measured by classic growth analysis. One of the main goals in agriculture is determining best plant density to yielding desired yield. Desired density obtain when canopy have maximum leaf area to up taking sunlight at the beginning of reproductive stage (larry et al., 2002)[18]. Goals such as improving absorbed sunlight by changing plant density and also changing row spacing perused in agricultural plants planting (Maddonni et al., 2001)[19]. Increasing light penetrating into lower parts of canopy by changing its structure is a management way with cause to improving yield (Reta-Sanches and Fowler, 2002)[28]. Heikkinen, and Auld, (1991) recommended densities more than to plants.m^{-2} to canola [13]. Considering canola density status has a great deal of importance to achieve high yield and quantity forage yield. The main objective for the present research is to shed light on the best plant density treatment and subsequently to determine suitable cultivar for cultivation. Al-Barzinjy et al. (1999) investigated the effects of different plant densities ranging from 20 to 130 plants.m^{-2} in rape seed [2]. They concluded that dry matter per plant decreased as plant density increased. Previous studies have shown that plant density is an important factor affecting rapeseed yield. Plant density in rapeseed governs the components of yield, and thus the yield of individual plants. A uniform distribution of plants per unit area is a prerequisite for yield stability (Diepenbrock 2000)[6]. In oilseed rape, row spacing or plant density vary considerably worldwide, depending on the environment, production system and cultivar. The growth is analyzed by measuring two factors, namely leaf area and dry weight of the organs and other quantities are calculated based on these two factors. When necessary, these quantities may be calculated either for whole plants or for different parts of the plants like root crown and leaves (Karimi, 2005) [14]. Crop growth rate (CGR) is slow at early growth stages because the plant cover is incomplete and the plants absorb just a part of the solar radiation. As the plants develop, their growth rate is quickly increased because of the expansion of leaf area and the penetration of less radiation through plant cover to the soil surface. Maximum CGR (the steepest slope in total biomass variations graph) is realized when the plants are tall and dense enough to be able to maximally utilize all environmental parameters (Radford, 1967) [25]. Zajac *et al*. (2005) found a positive relation between dry matter yield and growth indices like CGR and LAD [35]. Also, Mahdavi *et al*. (2006) and Katsura *et al*. (2007) reported that rice grain yield can be increased by selection on the basis of physiological growth indices like LAD, CGR, relative growth rate (RGR) and net assimilation rate[15, 20]. NAR is determined primarily by the ratio of carbon gained through photosynthesis and carbon lost through respiration. LAR reflects the amount of leaf area a plant develops per unit total plant mass and, therefore, depends on the proportion of biomass allocated to leaves relative to total plant mass (leaf mass ratio, LMR) and how much leaf area a plant develops per unit leaf biomass (specific leaf area, SLA), where LAR = LMR x SLA. (NAR) and leaf area ratio (LAR) are good measures of solar radiation capture during growth with NAR and LAR for an individual plant and LAI for population helping to explain differences in RGR. Sanches (1997) stated that investigation of forage fat and protein percent in eight canola varieties in Brazil showed that oil and protein percent are 41.3, 36.8, 24.7 and 20.9 respectively and varieties difference significantly in terms of forage fat and protein percent yield. The studies on lentil showed that such traits as biological yield, harvest index as well as leaf area index (LAI) and CGR can be used as indices for improving seed yield of lentil (Haghnazari *et al*., 2005)[11]. Siahpoosh *et al*. (2003) indicated that out of the studied physiological indices, net assimilation rate (NAR) and leaf area duration (LAD) were effective indices in increasing yield [32]. In a three-year study on linseed cultivars, Zajac *et al*. (2005) found a positive relation between dry matter yield and growth indices like CGR and LAD [35]. Also, Mahdavi *et al*. (2006) and Katsura *et al*. (2007) reported that rice grain yield can be increased by selection on the basis of physiological growth indices like LAD, CGR, relative growth rate (RGR) and net assimilation rate[15, 20].

MATERIALS AND METHODS

The experiment was carried out at Esmael Abad agricultural research station (Lat49° 54´ E, long 36° 15´ N), Iran in 2011- 2012. In order to evaluate effect of different plant density applications on quantity and quality forage of two spring canola cultivars in summer cultivation, an experiment was conducted in Ghazvin province in agronomical year of 2011-2012. Study area is located at 1285 m above sea level with annual average rainfall 310-320(mm), annual average temperature 13.9(C), minimum and maximum absolute annual temperatures of 17.4 and 37.8(C) respectively. Soil texture in study area is loam and silt loamy with pH 7.9-8 and its electrical conductivity found to be 1.1-1.29(ds.m^{-1}) (table1). This experiment was arranged as split plot in completely randomized block designs in the 3 replication. Plant density was considered as the main factor involving five levels of 100, 125, 150, 175 and 200 (plant.m^{-2}). Two spring canola cultivars RGS003 and SARIGOL were used in the present research. Seeds provided from department of oil seed researches, research center of seed and seedling breeding and preparation in Karaj (RGS003: German and spring type, SARIGOL: Iranian and spring type).

Table 1. Analysis results of soil experiment

Depth	0-30(cm)	30-60(cm)
EC(ds.m^{-1})	1.1	1.29
PH	8	7.9
SAR	3.80	4.2
T.N.V%	7.5	7.8
O.C%	0.64	0.57
Total N %	0.09	0.06
Texture	Silt Loam	Loam

In this experiment was fertilized before sowing by to the following fertilization rates: 60 kg N/ha as ammonium sulphate and 60 kg P2O5/ha as triple superphosphate. Additional 60 kg N/ha was applied in the study. In order to analyze and calculate the growth indices, the plots were sampled four times; each time 0.5 m of each row was harvested. In laboratory, the organs of the plants were dissected and then, their fresh weights were measured. Afterwards, the leaf blade area of the samples was measured. Next, the samples were transferred to in bags to lose their moisture. After one week, they were completely oven-dried at 105°C. Then, their dry weight was measured by a 0.001g digital scale. After collecting the data of leaf area and shoot dry and fresh weights, the growth indices were calculated as follows (Sarmadnia and Koucheki, 1989) [30]: Leaf area index (LAI): To measure LAI, one m^2 was sampled from each plot. Then, the leaves of the plants were parted and their area was measured by leaf-area meter.

$$LAI = \frac{\text{Leaf area}}{\text{Land area}}$$

Crop growth rate (CGR): It was calculated in terms of g.m^{-2}.day^{-1} by the following equation (Rahnama, 2006) [27]:

$$CGR = \frac{W_2 - W_1}{GA(T_2 - T_1)}$$

Net assimilation rate (NAR): It was calculated in terms of g.m^{-2} leaf area.day^{-1} by the following equation (Rahnama, 2006) [27]:

$$NAR = \frac{W_2 - W_1}{T_2 - T_1} \times \frac{\ln LA_2 - \ln LA_1}{LA_2 - LA_1}$$

Relative growth rate (RGR): It was calculated in terms of g.g^{-1}.day^{-1} by the following equation (Rahnama, 2006) [27]:

$$RGR = \frac{W_2 - W_1}{W_1(T_2 - T_1)}$$

Leaf area ratio (LAR): It was calculated in terms of cm^2.g^{-1} by the following equation (Rahnama, 2006)[27]:

$$LAR = \frac{\frac{LA_1}{W_1} + \frac{LA_2}{W_2}}{2}$$

Leaf weight ratio (LWR): This dimensionless index was calculated by the following equation:

$$LWR = \frac{\frac{LW_1}{W_1} + \frac{LW_2}{W_2}}{2}$$

Specific leaf area (SLA): It was calculated in terms of cm^2.g^{-1} by the following equation:

$$SLA = \frac{\frac{LA_1}{LW_1} + \frac{LA_2}{LW_2}}{2}$$

The symbols used in foregoing equations were as follows:

W1: total biomass measured at the first sampling	LA1: leaf area measured at the first sampling
W2: total biomass measured at the second sampling	LA2: leaf area measured at the second sampling
T1: first sampling time	LW1: leaf biomass measured at the first sampling
T2: second sampling time	LW2: leaf biomass measured at the second sampling

Oil content was determined by extracting the oil with diethyl ether in a Soxleth extraction apparatus, while content protein was determined using DUMAS, s procedure.

The data were subjected to analysis of variance using the SAS software. When the *F*-test indicated statistical significance at the $P = 0.05$or 0.01 levels, Duncan's multiple- range test was used to determine the significance between means.

RESULTS AND DISCUSSION

Total dry weight in stem elongation: Results from variance analysis indicate that cultivar, plant density and cultivar*plant density interactions were significant in probability levels of 1%. Mean comparison of cultivar showed that cultivar RGS003 showed highest dry weight with average 501.256(kg.h^{-1}) followed by SARIGOL with average 380.019(kg.h^{-1}). Plant density were classified in various statistical classes so that the highest dry weight was obtained by plant density (175 plant.m^{-2}) on average 560.5(kg.h^{-1}) and the least was attributed to 100(plant.m^{-2}) treatment with average 218.7(kg.h^{-1}). Mean comparison of plant density*cultivar interaction showed the highest dry weight in cultivar RGS003 and 175(plant.m^{-2}) with average 665.4(kg.h^{-1}) and least dry weight in cultivar SARIGOL and 100(PLANT.M^{-2}) with average 195.4(KG.H^{-1}). (Tables 2, 3, 4). Accumulation of dry matter in above ground organs and transporting it to grain have been reported in some crops such as rice , soybean, wheat and canola [16, 17]. As a whole, firstly, accumulation of dry matter in above ground is slow, but it increases rapidly with increase canopy and subsequently slowing down as leaves senescent while grain refilling. Dry matter at following is maximum rate while flowering as well (Wisoki et al. 2005; Yasari et al. 2008) [33, 34]. The highest total dry matter per plant was produced from the lowest plant density. This high total dry matter production per plant can be attributed to the fact that the plants from low densities were more vigorous, thicker in stems with more branches per plant. This can be a result of lesser interplant competition among plants and a better radiation distribution through open canopy. The negative effect of increasing plant population on total dry matter production is also reported by other workers (McGreegor, 1987; Morrison et al., 1990) [21, 23].

Forage fat yield: Results of variance analysis revealed that fat yield in forage was affected by plant density and cultivar individually in probability levels of 1% and but it was not significantly for nitrogen*cultivar interaction although. Analysis of mean comparison on cultivar effect showed that SARIGOL had the less fat (81.895 kg.h^{-1}) in Comparison to RGS003 (144.884 kg.h^{-1}). Mean comparison of nitrogen*cultivar interaction revealed the highest fat yield (203.4 kg.h^{-1}) in RGS003 when plant density (100 plant.m^{-2}) was applied. The lowest fat yield was achieved in SARIGOL with plant density (200 plant.m^{-2}) was applied with average 49.19 kg.h^{-1} (Table 2, 3, 4).

Forage Protein yield: forages raw protein serves as one of the most important criteria widely used to evaluate forage quality. Variance analysis showed that cultivar was significant at protein yield in probability levels of 1% but there were not significantly for plant density and nitrogen*cultivar interaction although. Mean comparison on cultivar effect showed that RGS003 had much protein (829.625 kg.h^{-1}) than SARIGOL (624.331 kg.h^{-1}). Different plant density levels were classified in two statistical classes. The lowest protein was related to 200 (plant.m^{-2}) treatments with average (613.6 kg.h^{-1}). Applying 100 and 125 plant.m^{-2}, resulted in 860.3 and 749.7 kg.h^{-1} proteins yield respectively. Mean comparison of nitrogen*cultivar interaction revealed the least protein yield(497.6 kg.h^{-1}) in SARIGOL when 200(plant.m^{-2}) treatment was applied. The highest protein was achieved in RGS003 once 100(plant.m^{-2}) was applied with average 968.5(kg.h^{-1})(Table 2, 3, 4).

glucosinolate contents(mg.g^{-1}): The simple effects of plant density and cultivar on the glucosinolate content were significant at the one percent level and the interaction effects of these factors on the glucosinolate content was not significant (Table 3). Comparison of means test relate to the interactive effect between plant density and cultivar on the glucosinolate content showed the treatment of RGS003 and plant density (200 plant.m^{-2}) was found to have the least glucosinolate content with an average of 13.30 mg.g^{-1} weight and the treatment of SARIGOL planted on 100 plant.m^{-2} was found to have the most glucosinolate content with an average of 29.50 mg.g^{-1} weight (Table 4). It can be seen that decrease plant density causes an increase in glucosinolate content either cultivar(Table 2, 3, 4).

Table 2. Variance analysis of dry weight, Forage fat and Forage protein

SOV	df	Final dry weight in stem elongation ($kg.h^{-1}$)	Forage fat ($kg.h^{-1}$)	Forage protein ($kg.h^{-1}$)	Glucosinolate content($mg.g^{-1}$)
Replication	2	405.752^{ns}	1419.172^{ns}	80913.195^{ns}	0.819^{ns}
density(D)	4	128727.896^{**}	8409.088**	52063.454^{ns}	158.605**
error	8	357.488	464.381	23598.786	2.654
Cultivars (V)	1	110238.682^{**}	29757.036**	316094.448**	56.307**
N* D	4	5923.482^{**}	480.117^{ns}	605.335^{ns}	2.269^{ns}
error	10	66.591	314.026	11356.295	0.789
Total	29				
CV%		1.85	15.63	14.66	3.96

**, ** and ^{ns}: significantat5%, 1% probability levels, and Non-significant.*

Table 3. Mean comparison of effects plant density and cultivars

Plant density	Final dry weight in stem elongation($kg.h^{-1}$)	Forage fat ($kg.h^{-1}$)	Forage protein ($kg.h^{-1}$)	Glucosinolate content($mg.g^{-1}$)
100	218.7d	163.7a	860.3a	28.55a
125	363.6c	135.2a	749.7ab	25.85b
150	519.9b	104b	669.2ab	22.70c
175	560.5	98.68b	742.2ab	19.40d
200	540.5ab	65.44c	613.6ab	15.55e
RGS003	501.256a	144.884a	829.625a	21.04b
SARIGOL	380.019b	81.895bb	624.331b	23.78a

Means in each column having similar letter (s), are not significantly at the 5% level.

Table 4. Mean comparison of effect plant density * cultivar interaction

Plant density	Cultivar	Final Dry weight in stem elongation ($kg.h^{-1}$)	Forage fat ($kg.h^{-1}$)	Forage protein ($kg.h^{-1}$)	Glucosinolate content($mg.g^{-1}$)
100	**RGS003**	241.9g	203.4a	968.5a	27.60b
100	**SARIGOL**	195.4g	123.9cd	752bc	29.50a
125	**RGS003**	427.2e	170.2b	851.1ab	25.30cd
125	**SARIGOL**	300.1f	100.1de	648.2bcd	26.40bc
150	**RGS003**	559.2c	136.5c	769.3abc	21.20e
150	**SARIGOL**	480.5d	71.50ef	571.1cd	24.20d
175	**RGS003**	665.4a	132.6cd	831.7ab	17.60f
175	**SARIGOL**	455.6de	64.71f	652.7bcd	21.20e
200	**RGS003**	612.5b	81.69ef	729.5bc	13.30g
200	**SARIGOL**	468.5de	49.19f	497.6d	17.60f

Means in each column having similar letter (s), are not significantly at the 5% level.

Leaf Area Index in stem elongation(LAI): Variance analysis Showed that simple effects (plant density and cultivar) and interaction effect of plant density*cultivar were significant at probability level of 1%. Mean comparison of cultivar effect indicated highest leaf area index in cultivar RGS003 with average 8.168 followed by SARIGOL with average 7.271. Different plant density levels were categorized in statistical classes. The highest leaf area index was observed during applying 175($plant.m^{-2}$) with average 10.26. In contrast, the least value was attributed to 100 ($plant.m^{-2}$) application treatments on average 4.552. Mean comparison of plant density*cultivar interaction indicated that different plant density*cultivar levels fall into various statistical classes. The highest and the least LAI were observed in RGS003 and SARIGOL (with averages 10.70 and 4.402 respectively), when 175 and 100($plant.m^{-2}$) were applied respectively (Table 5, 6, 7). Yesari et al., (2008) pointed out that low leaf area index at start and end of growth season is common, presumably attributes to leaves senescent and scattering, specifically those old ones located at lower canopy layers [34]. Canola leaves serve as the main photosynthesis source from emerging until middle of flowering period. Although they may not have direct contribution in development process, they, however, are vital in developing sink capacity. Not only maximum leaf area, but also leaf area durability (consistency) is important to quantify leaf development [34]. Salehian *et al*. (2002) showed that the highest plant density (i.e. 110 plants m^{-2}) produced the highest LAI. LAI plays a key role in determining CGR, both because it acts directly and substantially, and because of its indirect negative effect on NAR. LAR plays an important, albeit negative, role both directly and indirectly through NAR. The negative effects on NAR both of LAI and LAR may be attributed to reciprocal shading of the leaves when leaf area becomes excessive, which means that the crop requires the right sowing density while in crop management it is necessary to control practices that lead both to a deficit and an excess of leaf development. This explains the great interest shown in LAI as regards its interception of light energy and production of plant dry matter(Sarkar and pal, 2005)[29].

Leaf area ratio (LAR) at stem elongation: Results of variance analysis showed that plant density, cultivar and plant density * cultivar was significant influence on leaf area ratio at probability level of 1%. Results obtained by mean

comparison analysis in cultivars that genotype SARIGOL dedicated itself higher specific leaf area by 0.019 $m^2.g^{-1}$ TDW followed by RGS003 with 0.016 ($m^2.g^{-1}$ TDW). Different plant density levels were categorized in the different statistical class. The highest leaf area ratio was observed during applying 100(plant.m^{-2}) with average 0.020($m^2.g^{-1}$ TDW). In contrast, the least value was attributed to 125 (plant.m^{-2}) application treatments on average 0.015($m^2.g^{-1}$ TDW). Mean comparison of plant density*cultivar interaction indicated that the highest leaf area ratios (0.022 $m^2.g^{-1}$ TDW) were recorded in SARIGOL when 100(plant.m^{-2}) were applied (Table 5, 6, 7). Observed that LAR was highest during the early vegetative stage but later decreased rapidly with the advancement of plant age, possibly due to abscission of older leaves. Similar result was reported by Haque (1993) and Rahman (1993) [12].

Specific leaf area (SLA) at stem elongation: Analysis of variance denoted significant effects of plant density, cultivar and plant density*cultivar interaction on specific leaf area on probability levels of 1%. Mean comparison of cultivar effect indicated that genotype RGS003 dedicated itself higher specific leaf area by 0.032 $m^2.g^{-1}$ TDW followed by SARIGOL with 0.030 ($m^2.g^{-1}$ TDW). Different plant density application levels were categorized in the different statistical class and showing significant difference. Result of mean comparison on nitrogen*cultivar interaction indicated that different plant density levels and cultivar were classified in the different statistical class and showing significant difference. The highest specific leaf areas (0.048 $m^2.g^{-1}$ TDW) was recorded in RGS003, when amounts of plant density 100(plant.m^{-2}) were applied (Table 5, 6, 7). The lowest specific leaf areas (0.023 $m^2.g^{-1}$ TDW) was recorded in RGS003 and when amounts of plant density 150(plant.m^{-2}) were applied (Table 5, 6, 7). This central role of SLA in determining seedling potential RGR is thus general across European grasses, herbs and woody perennials (Cornelissen *et al*., 1996)[5]. This refers to the fact that amount of leaf area per unit total plant weight is more important (as related to light attenuation) than allocation of biomass per unit leaf area. The increased LAR enhances the RGR and thus the competitive potential (Peltzer and Kochy, 2001)[24]. Thus the high RGR of grass in competition can be attributed to NAR and LAR.

Table 5. Variance analysis of SLA, LAR and LAI

SOV	df	LAI in stem elongation	LAR in stem elongation ($m^2.g^{-1}$)	SLA in stem elongation ($m^2.g^{-1}$)
Replication	2	0.090^{ns}	0.017^{ns}	0.003^{ns}
Density(D)	4	37.181**	25.675**	243.343**
error	8	0.086	0.023	0.008
Cultivars (V)	1	6.032**	54.945**	18.252**
N* D	4	0.488^{ns}	6.284**	100.475**
error	10	0.016	0.022	0.008
Total	29			
CV%		1.63	0.83	0.28

**, ** and ^{ns}: significantat5%, 1% probability levels, and Non-significant.*

Table 6. Mean comparison of effects plant density and cultivars on SLA, LAR and LAI

Plant density	LAI in stem elongation	LAR in stem elongation ($m^2.g^{-1}$)	SLA in stem elongation ($m^2.g^{-1}$)
100	4.552e	0.020a	0.041a
125	5.628d	0.015e	0.027d
150	8.746c	0.017d	0.024e
175	10.26a	0.019b	0.034b
200	9.413b	0.017c	0.029c
RGS003	8.168a	0.016b	0.032a
SARIGOL	7.271b	0.019a	0.030b

Means in each column having similar letter (s), are not significantly at the 5% level.

Table 7. Mean comparison of density * cultivars interaction on SLA, LAR and LAI

Plant density	Cultivar	LAI in stem elongation	LAR in stem elongation ($m^2.g^{-1}$)	SLA in stem elongation ($m^2.g^{-1}$)
100	**RGS003**	**4.701f**	**0.019c**	**0.048a**
100	**SARIGOL**	**4.402g**	**0.022a**	**0.033c**
125	**RGS003**	**6.520**	**0.015h**	**0.027g**
125	**SARIGOL**	**4.737f**	**0.015g**	**0.027f**
150	**RGS003**	**9.246c**	**0.016e**	**0.023j**
150	**SARIGOL**	**8.245d**	**0.017d**	**0.025h**
175	**RGS003**	**10.70a**	**0.016f**	**0.035b**
175	**SARIGOL**	**9.816b**	**0.021b**	**0.032d**
200	**RGS003**	**9.669b**	**0.015g**	**0.025i**
200	**SARIGOL**	**9.157c**	**0.019c**	**0.032e**

Means in each column having similar letter (s), are not significantly at the 5% level.

Leaf weight ratio (LWR) at stem elongation: Variance analysis showed there are significant difference of plant density, cultivar and plant density *cultivar interaction in 1% level. Mean comparison cultivar individually denoted that cultivar SARIGOL had higher leaf weight ratio (0.63 $g.g^{-1}$TDW) than RGS003 (0.54 $g.g^{-1}$ TDW). Mean comparison plant density showed that 150(plant.m^{-2}) had higher leaf weight ratio (0.68 $g.g^{-1}$ TDW) than

200(plant.m^{-2}) (0.61 g.g^{-1} TDW). Mean comparison of nitrogen*cultivar interaction showed that the highest leaf weight ratio was observed in RGS003 and plant density (150 plant.m^{-2}) (0.70 g.g^{-1} TDW) and least value (0.40g.g^{-1} TDW) was attributed to cultivar RGS003 and plant density (100 plant.m^{-2}). (Table 8, 9, 10). LAR is determined by both LAR and SLA (Causton and Venus, 1981)[4]. This increase in LAR is largely determined by due to changes in LWR and often due to the changes in SLA.

Net assimilation rate (NAR) at stem elongation: Results of variance analysis showed that plant density, cultivar and plant density *cultivar interactions in probability level of 1% were significant. Mean comparison of cultivar revealed that cultivar RGS003 had higher net assimilation rate (3.489 g.day^{-1}.m^{-2}) than SARIGOL (2.698 g.day^{-1}.m^{-2}). Plant density levels were categorized in four different statistical classes. Mean comparison of plant density revealed that 150(plant.m^{-2}) had higher net assimilation rate (5.11 g.day^{-1}.m^{-2}) but Mean comparison of plant density revealed that 125(plant.m^{-2}) had lower net assimilation rate (1.046 g.day^{-1}.m^{-2}). Mean comparison of nitrogen*cultivar interaction indicated that different plant density levels and cultivars fell into different statistical classes. Highest net assimilation rate (6.406 g.day^{-1}.m^{-2}) in genotype RGS003 was recorded when 175(plant.m^{-2}) was added. The least value (0.406 g.day^{-1}.m^{-2}) was recorded in SARIGOL, when 125 (plant.m^{-2}) was applied(Tables 8, 9, 10). However, plant photosynthesis, hence NAR, is known to be greatly affected also by other factors such as radiation, temperature, nutrient availability.

Crop growth rate (CGR) at stem elongation: Variance analysis indicated significant effect for plant density, cultivar and plant density *cultivar interactions on CGR at probability level of 1%. Mean comparison of plant density showed that the highest crop growth rate (40.32 g.day^{-1}.m^{-2}) was recorded in 150(plant.m^{-2}) followed by 175(plant.m^{-2}) (32.52 g.day^{-1}.m^{-2}). Mean comparison of cultivar showed that the highest crop growth rate (28.772 g.day^{-1}.m^{-2}) was recorded in RGS003 followed by SARIGOL (16.739 g.day^{-}1.m^{-2}). Different plant density levels fell into different statistical classes. Results obtained from mean comparison on plant density *cultivar interaction that genotype RGS003 exhibited the highest CGR (57.22 g.day^{-1}.m^{-2}), when 175(plant.m^{-2}) was applied. Also, the least CGR value (1.910 g.day^{-1}.m^{-2}) was obtained when SARIGOL with 125(plant.m^{-2}) was added (Tables 8, 9, 10). Some researchers reported that crop growth rate is affected by plants photosynthetic area directly (HabibZadeh et al., 2006; Shilbes and Weber, 1995)[10, 31].

Table 8. Variance analysis of LWR, NAR, CGR, and RGR

SOV	df	LWR in stem elongation (m^2.g^{-1})	NAR in stem elongation (g.day^{-1}.m^2)	CGR in stem elongation (g.day^{-1}.m^2)	RGR in stem elongation (g.day^{-1}.m^2)
Replication	2	0.136^{ns}	0.037^{ns}	4.838^{ns}	4.123^{ns}
Density(D)	4	214.344**	13.137**	1211.068**	2948.777**
error	8	0.082	0.080	6.135	15.046
Cultivars (V)	1	572.907**	4.693**	1085.936**	401.868**
N* D	4	286.344**	14.058**	719.956**	2649.859**
error	10	0.096	0.073	3.388	14.259
Total	29				
CV%		0.52	2.91	8.09	7.82

**, ** and ^{ns}: significantat5%, 1% probability levels, and Non-significant.*

Table 9. Mean comparison of effects density and cultivars on NAR, CGR, LWR and RGR

Plant density	LWR in stem elongation (m^2.g^{-1})	NAR in stem elongation (g.day^{-1}.m^2)	CGR in stem elongation (g.day^{-1}.m^2)	RGR in stem elongation (g.day^{-1}.m^2)
100	0.53d	3.003c	11.05d	0.053b
125	0.56c	1.046d	6.450e	0.016d
150	0.68a	5.110a	40.32a	0.077a
175	0.56c	3.645b	32.52b	0.051b
200	0.61b	2.660c	23.43c	0.043c
RGS003	0.54b	3.489a	28.772a	0.052a
SARIGOL	0.63a	2.698b	16.739b	0.044b

Means in each column having similar letter (s), are not significantly at the 5% level

Relative growth rate (RGR) at stem elongation: Variance analysis indicated that significant plant density, cultivar and plant density *cultivar interactions on RGR at probability level of 1%. Mean comparison of cultivar showed that the highest relative growth rate (0.052 g.day^{-1}.m^{-2}) was recorded in RGS003 followed by SARIGOL (0.044 g.day^{-1}.m^{-2}). Different plant density levels fell into different statistical classes. The highest and least relative growth rates were obtained (0.077 and 0.016 g.day^{-1}.m^{-2}) when 150 and 125(plant.m^{-2}) were applied respectively. Results obtained from mean comparison on plant density *cultivar interaction that genotype RGS003 exhibited the highest RGR (0.086 g.day^{-1}.m^{-2}), when 175 (plant.m^{-2}) but the lowest relative growth rate (0.006 g.day^{-1}.m^{-2}), when 125 (plant.m^{-2}) with SARIGOL (Tables 8, 9, 10). RGR is a complex parameter determined by a number of physiological, morphological and biomass allocation components. In addition, some researchers reported that crop growth rate is

affected by plants photosynthetic area directly (HabibZadeh et al., 2006; Shilbes and Weber, 1995)[10, 31]. Increased plant density significantly increased crop growth rate (CGR) during early stage and reduced the net assimilation rate (NAR) and CGR during later part of crop growth. Higher CGR at vegetative stage originates from which high leaf area index (LAI) and that CGR at reproductive and ripening stages is controlled by NAR. There was an increase relationship between leaf area and NAR. The increase in CGR was ascribed to the increased in NAR and leaf area. Plant growth analysis decomposes RGR into net assimilation rate (NAR, rate of dry matter production per unit leaf area) and leaf area ratio (LAR, leaf area per unit total plant mass), where RGR=NAR x LAR (Evans, 1972; Causton and Venus 1981)[4, 8]. NAR is determined primarily by the ratio of carbon gained through photosynthesis and carbon lost through respiration. LAR reflects the amount of leaf area a plant develops per unit total plant mass and, therefore, depends on the proportion of biomass allocated to leaves relative to total plant mass (leaf mass ratio, LMR) and how much leaf area a plant develops per unit leaf biomass (specific leaf area, SLA), where LAR = LMR x SLA. Most work evaluating RGR variation among species has compared species from habitats differing in fertility or productivity. The ecological advantage of high RGR is very clear. Due to high RGR, a plant will rapidly increase in size and is able to occupy a large space, both below and above ground. A high RGR may also facilitate rapid completion of life cycle of a plant.

Table10. Mean comparison of density * cultivars interaction on NAR, CGR, LWR and RGR

Plant density	Cultivar	LWR in stem elongation ($m^2.g^{-1}$)	NAR in stem elongation ($g.day^{-1}.m^2$)	CGR in stem elongation ($g.day^{-1}.m^2$)	RGR in stem elongation ($g.day^{-1}.m^2$)
100	**RGS003**	0.40i	1.552cde	7.264g	0.029d
100	**SARIGOL**	0.67b	4.455b	14.83e	0.076b
125	**RGS003**	0.55g	1.687cde	10.99f	0.025d
125	**SARIGOL**	0.56f	0.406e	1.910h	0.006f
150	**RGS003**	0.70a	5.029ab	4.60b	0.074b
150	**SARIGOL**	0.67b	5.191ab	39.04b	0.081ab
175	**RGS003**	0.46h	6.406a	57.22a	0.086a
175	**SARIGOL**	0.66c	0.885de	7.827fg	0.17e
200	**RGS003**	0.62d	2.769c	26.79c	0.043c
200	**SARIGOL**	0.60e	2.551cd	20.08d	0.042

Means in each column having similar letter (s), are not significantly at the 5% level.

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