

REGULAR ARTICLE

Variability of oil and protein content in rapeseed cultivars affected by seeding date

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ABSTRACT

This investigation was undertaken in order to determine the variability of oil and protein content in winter rapeseed cultivars affected by seeding date. Four cultivars (Banačanka, Slavica, Express, Valesca) were sown at six seeding dates (SD1-21 August, SD2-31 August, SD3-10 September, SD4-21 September, SD5-1 October, SD6-9 October) in four replications across two cropping seasons. The trial was arranged as a randomized complete block design. The effect of year, cultivar, and seeding date were highly significant for oil and protein content. Increased oil content in the second year (2010/2011) was related to weather conditions which were favorable for rapeseed. However, protein content was significantly higher in the first year (2009/2010). Oil content ranged between 41.19% (Valesca) and 42.69% (Express). Significantly lowest oil content across seeding dates was found in SD6 (40.67%) and highest in SD4 (41.86%) and SD1 (41.61%). Valesca showed significantly highest mean protein content (21.54%). Protein content was highest in SD6 (20.18%). Oil content decreased with delayed seeding. Between oil and protein content highly significant negative correlation ($r = -0.730$) was stated. Seeding date showed a significant effect on oil and protein content in rapeseed. Cultivar Express and SD4 can be pointed out, based on the achieved oil content, and cultivar Valesca for high protein content. Study results may be helpful in recommending optimal rapeseed seeding date in the region.

Keywords: *Brassica napus*; Cultivar; Interaction; Seed quality; Seeding date

INTRODUCTION

Rapeseed (*Brassica napus* L.) represents an agronomically significant oilseed crop. It belongs to genus *Brassica*, family Brassicaceae. According to the total production of major oilseeds in the world, rapeseed takes the second place just after soybean (USDA, 2015), with the production of over 36 million hectares. Rapeseed is the most important source of vegetable oil in Europe. As winter and spring form it is cultivated on significant areas in moderate continental and continental climate. In the world main production areas of rapeseed are Canada, China, India, Northern and Western Europe (FAOSTAT, 2014). Rapeseed can be used as the alternative crop to sunflower in the increase of oil production. The growth of productivity is essential to further increase of the area under rapeseed in the regions where rapeseed is growing and its expansion into nongrowing regions (Seyis et al., 2004, Marjanović-Jeromela et al., 2014). Primarily rapeseed is grown for the production of seed with high oil and protein content. It is also used as animal feed and in biodiesel production

(Vujaković et al., 2014; Marjanović et al., 2016). Since it begins to bloom early in the spring and its flowering period lasts 15-25 days, it is also one of the best honey plants for bee grazing (Marinković et al., 2009). Rapeseed can be used as green manure in organic agriculture and also as a component in intercrops (Antanasović et al., 2012; Mikić et al., 2015). The seed has a high value of oil (40-48%) and protein (18-25%), (Marinković et al., 2009). The content of saturated fatty acids in rapeseed oil amounts to 7%. It has a high content of unsaturated fatty acids (oleic acid 61%, linoleic acid, 21% and linolenic acid 11%), as reported by Molazem et al. (2013).

For the successful cultivation of rapeseed, it is essential well aerated, deep soil with proper distribution of rainfall because of a long vegetation period of this crop. Proper field practices as crop rotation, plant density, and seeding date can influence oil and protein content in rapeseed (Mirrales et al., 2001; Crnobarac et al., 2002; Si, 2007). Rapeseed matures early, leaving enough time for quality soil preparation for the subsequent crop. Also, it leaves the

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field free of weeds. These characteristics make rapeseed a preferred component of crop rotation (Marjanović-Jeromela et al., 2006). The length of the vegetation period is affected by planting date, and then it has an impact on yield and yield components. Since seeding date has an influence on the physiological characteristics of the plant, it is important to determine proper seeding date in order to have optimum coordination between the plant growth and climatic conditions (Khajaj et al., 2014). The delay in sowing leads to a decrease in the period from planting time to flowering and maturity. With optimal seeding date, the degree of development can be regulated in which the plants will best overwinter.

Considering that in Southeast Europe the information on the impact of seeding date on quality characteristics in rapeseed is scarce, the objective of this investigation was to assess the variability of oil and protein content in winter rapeseed cultivars during two cropping seasons affected by seeding date.

MATERIALS AND METHODS

Plant materials and experimental conditions

The field trial was carried out to study the response of oil and protein content of four rapeseed cultivars, two originating from Serbia (Banačanka, Slavica), and two from Germany (Express, Valesca), to six seeding dates (SD1-21 August, SD2-31 August, SD3-10 September, SD4-21 September, SD5-1 October, SD6-9 October). The two-year trial was carried out from 2009 to 2011 at Rimski šančevi (45°19'51" N; 19°50'59" E; 84 m altitude), on the experimental field of the Institute of Field and Vegetable Crops, Novi Sad, Serbia. Sowing was done on 6 m² plots for each cultivar and seeding date in four replications on August 25, 2009, and on August 26, 2010. The distance between rows was 25 cm and within rows 5 cm. Harvest date was on June 30, 2009, and on June 23, 2010. Rapeseed plants were harvested at the time of the second technical maturity level (Harper and Berkenkamp 1975). In both investigated seasons optimal agricultural practices were carried out.

Climatic data on precipitations and temperatures were taken from the Hydrometeorological Service of the Republic of Serbia (<http://www.hidmet.sr.gov.rs>).

Chemical analyses

Magnetic resonance analyzer (Newport 4000 NMR analyzer), using the NMR (Nuclear Magnetic Resonance) method by Granlund and Zimmerman (1975), was applied for the determination of oil content. It was expressed as a percentage of seed. The content of total protein was

determined using standard Kjeldahl (1883) procedure after harvest. The analyses were carried out in the chemical laboratory of the Institute of Field and Vegetable Crops, Oil Crops Department, Novi Sad, Serbia.

Statistical analyses

Randomized complete block design with four replications was applied. Collected data were analyzed using three-way analysis of variance, and correlation analysis in the STATISTICA 12.0 package computer program (*StatSoft*). LSD test at 5% and 1% level of probability was used.

RESULTS AND DISCUSSION

Oil content

Oil content, except seed yield, is one of the very important criteria in creating rapeseed cultivars (Marinković et al., 2003; Si and Walton, 2004; Marjanović-Jeromela et al., 2007). High seed yield together with high oil content will result in high oil yield (Pospíšil et al., 2014). Oil content and oil quality depend on the genetic potential of the cultivar, respectively by its expression in specific agro-ecological conditions.

Highly significant differences were stated for all main sources of variation (year, cultivar, seeding date) for oil content. Only year × seeding date interaction was highly significant. Oil content in rapeseed was mostly under the influence of the cultivar (50.7%). Year of growing amounted to 34.4% of the variability of oil content and seeding date to 4.2% (Table 1).

Our results have a close similarity with the reports of Fanaei et al. (2007). These authors also stated that year was the main source of variation for oil content and equal significant contribution gave cultivar and seeding date. On the basis of 19 hybrids and cultivars, Pospíšil et al. (2008) concluded that in rapeseed oil content, seed yield and oil yield were significantly caused by genotype and by the conditions of production (year, site). Ma et al. (2016) amounted that oil content in rapeseed was significantly

Table 1: ANOVA for oil content in rapeseed cultivars (2009/2010 and 2010/2011)

Source of variation	df	SS (%)	MS	P
Replication	3	1.1	3.50	0.162
Year (Y)	1	34.4	162.70	0.000**
Cultivar (C)	3	50.7	161.00	0.000**
Seeding date (SD)	5	4.2	7.80	0.002**
Y×C	3	1.2	3.90	0.124
Y×SD	5	3.9	7.30	0.004**
C×SD	15	3.3	2.10	0.409
Y.x C×SD	15	1.2	0.70	0.984
Error	141		2.00	

*P<0.05; **P<0.01

caused by seeding date. Genotype, year of production and quantity of the N-fertilizer applied by the top dressing were highly significant for oil and protein content, as mentioned by Vujaković et al. (2010). Kahrarian et al. (2014) reported highly significant differences for year and cultivars, but interaction year \times cultivar was not significant. Some authors (Modares and Daneshgar, 2007) have found only significant differences in oil content between rapeseed cultivars while seeding dates and the interaction between cultivars and seeding dates were non-significant. They concluded that late planting date reduced seed oil percent through decreasing 1000-grain yield.

Oil content was significantly higher (42.51%) in the second year of investigation (2010/2011) in relation to 2009/2010 (39.89%), caused by different climatic conditions. It was a long period of rain in May and June in 2009/10. The temperatures were low and frost appeared in January 2010. These conditions were unfavorable (Vujaković et al., 2015) for rapeseed in comparison with 2010/11 (Figs. 1 and 2). It could be the main reason for the lower oil content in 2009/10 (Table 2). Our results are similar to studies reported by Pospišil et al. (2014). These authors also reported that the highest values of oil content were in 2010/11 (48.04%) in relation to previous 2009/10 (41.40%) vegetation period. These authors stated that the highest oil content in seed was obtained in 2010/11 year, as a result of moderate air temperature

(18.1 - 21.6°C) and average rainfall (122.4 mm) in the stage of silique (pod) development and oil synthesis in the seeds. In 2010/11 year oil content in seeds ranged from 44.06% in the variety Robust to 50.80% in the variety Ricco. In our experiments, oil content in average for two years ranged between 41.19% (cv. Valesca) and 42.69% (cv. Express). In both years Valesca showed significantly lower oil content in relation to other investigated cultivars (Table 2). Marjanović-Jeromela et al. (2007) reported that the seed oil content was highest in cultivar Oktavija (47.77%). Other cultivars included in their experiment (Jana, Banačanka,

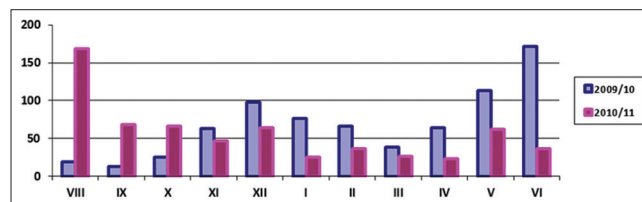


Fig 1. Precipitation in 2009/2010 and 2010/2011 (Rimski šančevi).

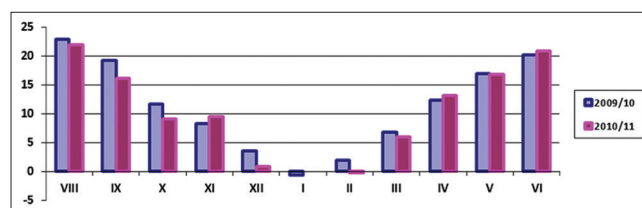


Fig 2. Temperatures in 2009/2010 and 2010/2011 (Rimski šančevi).

Table 2: Mean values and variability for oil content in rapeseed cultivars

Year (Y)	Cultivar (C)	Seeding date (SD)						Mean (Y \times C)	Mean (Y)	
		SD1	SD2	SD3	SD4	SD5	SD6			
2009/2010	Banačanka	40.87	39.50	40.53	40.82	41.10	39.92	40.46	39.89	
	Slavica	41.02	39.28	40.49	41.90	41.41	40.35	40.74		
	Express	41.27	39.99	41.11	42.02	41.46	40.73	41.09		
	Valesca	37.25	35.75	37.13	38.65	37.10	37.73	37.27		
	Mean	40.10	38.63	39.82	41.58	41.32	39.68			
2010/2011	Banačanka	44.58	44.01	43.31	43.52	43.22	41.40	43.34	42.51	
	Slavica	42.76	43.05	43.62	42.42	42.32	41.49	42.61		
	Express	44.99	44.72	44.16	45.25	43.24	43.30	44.28		
	Valesca	40.15	39.07	40.49	40.28	38.22	40.51	39.79		
	Mean	43.12	42.71	42.90	42.87	41.75	41.68			
Mean (2 years)	Banačanka	42.72	41.76	41.92	42.17	42.16	40.66	Mean (C)	41.90	
	Slavica	41.89	41.16	42.08	42.16	41.86	40.87			41.67
	Express	43.13	42.36	42.63	43.64	42.35	42.02			42.69
	Valesca	38.70	37.41	38.81	39.46	37.66	39.12			41.19
	Mean	41.61	40.67	41.35	41.86	41.01	40.67			
V (%) 3.3										
LSD	Y	C	SD	Y \times C	Y \times SD	C \times SD	Y \times C \times SD			
	0,05	0.40	0.56	0.68	0.80	0.98	1.96			
	0,01	0.52	0.74	0.90	1.05	1.28	2.57			
LSD	2009/2010			2010/2011						
	C	SD	C \times SD	C	SD	C \times SD				
	0.05	0.65	0.80	1.60	0.92	1.13	2.26			
	0.01	0.87	1.06	2.12	1.24	1.50	2.99			

UM-11, B-009, Jet Neuf, Samourai, K-571, and Valesca) showed significantly higher seed oil content in relation to the mean over cultivars, which amounted to 44.87%. Marjanović-Jeromela et al. (2011) communicated that oil content in the period 2007-2010 varied between 37.5% (in 2009/10) and 47.2% (in 2007/08). UM-13 (41.97%) cultivar showed lowest oil content. In the experiment 40 winter and 9 spring rapeseed cultivars were included. The seed oil content in all tested rapeseed varieties ranged from 34.49 to 43.01% in the 2007/2008 growing season (Vujaković et al., 2014).

Loof (2000) reported that a significant factor in the production of seed yield and high oil in the autumn rapeseed belongs to the timely cultivation. Seeding date has a significant influence on oil content. In our experiments, the highest oil content was stated in SD4 (41.86%) and SD1 (41.61%). Significantly lower oil content across seeding dates was found in SD6 (40.67%), Table 2. To late seeding date decreased oil content in rapeseed. The obtained result can be explained by the shortened vegetative period of growth and increase of the temperature in the period of flowering. Our results are similar to the report of Zeng et al. (2007). These authors also reported that late sowing reduces the oil content in the rapeseed.

Protein content

Protein is the main requirement for growth and development of all organisms. The protein content of rapeseed depends on various factors, including genetical and environmental factors, as well as the interaction between them. The variability of protein content in rapeseed, in addition to genetic factors, is greatly influenced by many environmental factors as location (Marinković et al., 2010), temperature (Piljuk, 2006), year (Kulikovskij, 2006), and seeding date (Kapilović, 2006).

Highly significant differences were found for all main effects (year, cultivar, seeding date) for protein content. Concerning interactions only year \times seeding date was significant. Protein content was predominantly influenced by the year of growing (70.5%). The influence of cultivar on protein content amounted to 9.7% and of sowing date to 5.3% (Table 3). The results obtained in our investigation are in agreement with the report of Turhan et al. (2011). These authors reported that in the two-year experiment with eight German winter rapeseed genotypes, and four seeding dates, highly significant differences occurred for years, genotypes, seeding dates and year \times seeding date interaction, both for oil and protein content.

The rapeseed cultivars responded differently to different seeding dates. On the contrary to oil content, protein content was significantly higher in 2009/2010 (22.15%)

Table 3: ANOVA for protein content in rapeseed cultivars (2009/2010 and 2010/2011)

Source of variation	df	SS (%)	MS	P
Replication	3	1.7	4.32	0.060
Year (Y)	1	70.5	537.58	0.000**
Cultivar (C)	3	9.7	24.56	0.000**
Seeding date (SD)	5	5.3	8.10	0.000**
Y \times C	3	1.0	2.44	0.239
Y \times SD	5	4.6	7.07	0.002*
C \times SD	15	3.5	1.79	0.415
Y \times C \times SD	15	3.7	1.88	0.366
Error	141		1.72	

* $P < 0.05$; ** $P < 0.01$

than in 2010/2011 (18.78%), (Table 4). Seed protein content generally shows an adverse relationship to oil content and both are under genetic and environmental control. Therefore factors increasing seed protein content are also decreasing oil content (Šidlauskas and Rife, 2000). Protein content in two-year average varied between 19.97% in Express and 21.54% in Valesca (Table 4). Vujaković et al. (2014) mentioned similar results. In their experiment protein content ranged from 18.06% to 20.00% in 2007/2008 growing season. In mutant lines of winter rapeseed, Ahmad et al. (2013) presented similar results for this trait.

Different factors affect directly or indirectly protein content of rapeseed. One of the important factors is seeding date. Protein content was the highest in SD6 (21.41%) and it was highly significant in relation to all other seeding dates. Between other seeding dates, no significant differences were detected (Table 4). Experiments conducted by Lääniste et al. (2016) also showed that both sowing date and weather conditions during the period of growth affected the seed quality (oil and protein content) of winter oilseed rape. Locations have also an important influence on protein content in rapeseed (Marinković et al., 2010). These authors informed that protein content at Rimski šančevi location varied between 19.60% in NS-L-74 and 25.93% in JR-NS-36). At Sombor location the values of protein content ranged from 19.26% in NS-L-7 to 24.06% and 24.09% in NS-L-46 and cultivar Mira.

Variability and correlation between oil and protein content

Variability of oil content in the two-year experiment was $V=3.3\%$ and for protein content $V=6.1\%$ (Tables 2 and 4). Ahmad et al. (2013) determined lower values for both oil (1.69%) and protein content (3.76%). On the contrary, Yousuf (2011) reported higher values for oil content (4.19%) and also for protein content (7.73%).

Study of the correlation between oil and protein contents reveals that when oil increases under favorable

Table 4: Mean values and variability for protein content in rapeseed cultivars

Year (Y)	Cultivar (C)	Seeding date (SD)						Mean (YxC)	Mean (Y)	
		SD1	SD2	SD3	SD4	SD5	SD6			
2009/2010	Banaćanka	22.28	22.96	22.94	20.65	20.98	22.82	22.10	22.15	
	Slavica	21.21	22.77	22.05	21.91	21.34	21.85	21.85		
	Express	19.57	22.52	22.50	20.93	21.73	23.32	21.76		
	Valesca	23.52	22.35	23.81	23.08	21.95	22.68	22.90		
	Mean	21.65	22.65	22.83	21.64	21.50	22.67			
2010/2011	Banaćanka	18.30	17.75	18.24	18.04	17.85	19.93	18.35	18.78	
	Slavica	18.54	17.98	17.13	18.03	18.25	20.36	18.38		
	Express	17.86	18.46	17.30	17.85	18.40	19.26	18.19		
	Valesca	20.15	20.27	19.19	19.50	20.94	21.08	20.19		
	Mean	18.71	18.62	17.96	18.35	18.86	20.16			
Mean (2 years)	Banaćanka	20.29	20.35	20.59	19.34	19.42	21.38	Mean (C)	20.23	
	Slavica	19.88	20.37	19.59	19.97	19.80	21.10			20.12
	Express	18.71	20.49	19.90	19.39	20.06	21.29			19.97
	Valesca	21.84	21.31	21.50	21.29	21.44	21.88			21.54
	Mean	20.18	20.63	20.39	20.00	20.18	21.41			
V (%) 6.1										
LSD	Y	C	SD	YxC	YxSD	CxSD	YxC x SD			
0.05	0.37	0.53	0.65	0.71	0.90	1.27	1.82			
0.01	0.49	0.69	0.85	0.98	1.18	1.67	2.39			
LSD	2009/2010			2010/2011						
	C	SD	CxSD	C	SD	CxSD				
0.05	0.70	0.85	1.71	0.72	0.89	1.77				
0.01	0.93	1.13	2.27	0.96	1.18	2.36				

environments protein concentration decreases (Si et al., 2003). In our two-year experiment (Fig. 3) oil and protein content were in highly significant negative correlation ($r = -0.730$). The results obtained in our experiment are in agreement with the report of Kennedy et al. (2011). These authors also observed a highly significant negative correlation between oil and protein content ($r = -0.848$) in rapeseed, suggesting that oil content increases on account of protein content because of competition for the source of carbon. Vujaković et al. (2014) found a significant negative correlation between oil and protein content in rapeseed. It was $r = -0.481$ in the 2007/2008, and $r = -0.391$ in the 2008/2009 growing season. Other researchers (Ping et al., 2003; Hao et al., 2004; Ahmad et al., 2013) also published significant negative correlation between oil and protein content in rapeseed. In the spring rapeseed there existed also a high negative correlation ($r = -0.980$) between oil and protein content, as communicated by Kuht et al. (2015).

CONCLUSION

The following conclusions can be drawn, on the basis of the two-year experiments: Years, cultivars, seeding dates, and year \times seeding date interactions were significant sources of variation for oil and protein content in rapeseed. Oil content ranged between 41.19% (Valesca) and 42.69% (Express).

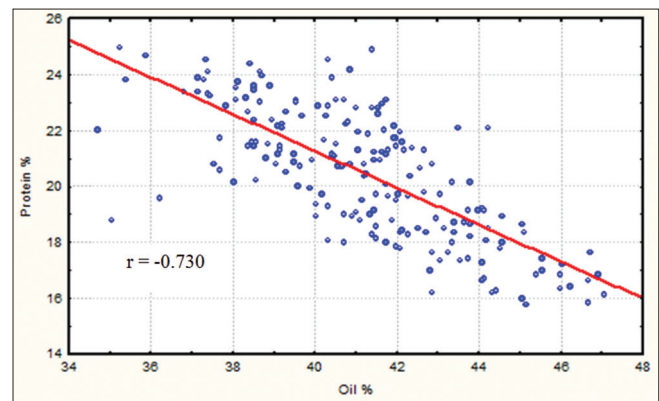


Fig 3. Correlation between oil and protein content in rapeseed.

Significantly lowest oil content across seeding dates was found in SD6 (40.67%) and highest in SD4 (41.86%) and SD1 (41.61%). Valesca showed significantly highest mean protein content (21.54%). Protein content was highest in SD6 (20.18%). Cultivar Express can be pointed out, based on the achieved oil content, and cultivar Valesca for high protein content. The correlation between oil and protein content was highly significant negative ($r = -0.730$). Seeding cultivar Express until the middle of September (SD4) could be suggested according to the results of this research, in order to achieve maximum oil content. The obtained study results may be helpful in recommending optimal rapeseed seeding date in the region.

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Author's contribution

I.B. Wrote the manuscript, did statistical analysis and analyzed the results. A.M.-J. Was the project leader and designed the study. J.C. Contributed in research planning. S.T. Carried out field experiments. V.R. Was involved in literature collection. V.M. Was involved in manuscript preparation. D.J. Contributed in chemical analysis.

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