



# Sowing Dates and Seeding Rates Affect Soybean Grain Composition

Renan Caldas Umburanas<sup>1</sup> · Anderson Hideo Yokoyama<sup>2</sup> · Leonardo Balena<sup>2</sup> · Gabriela Caroline Lenhani<sup>2</sup> · Ângela Moraes Teixeira<sup>2</sup> · Roberta Letícia Krüger<sup>2</sup> · Klaus Reichardt<sup>3</sup> · Jackson Kawakami<sup>2</sup>

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## Abstract

The effect of agronomic practices in soybean grain composition lacks information. In addition, the importance of protein and oil contents in soybean grains is increasing due to the industry demand for grain quality. It is well known that soybean grain weight can change according to environmental conditions, like different sowing dates, however, the consequences in grain composition need to be better understood. The aim of this study was to evaluate the composition, oil and protein yield of soybean grains, in response to different sowing dates (early, mid, and late), seeding rates (15, 25, 35, and 45 seeds m<sup>-2</sup>), and two growing seasons. Late sowing reduced grain oil content, soybean oil yield, and protein yield. The increase in seeding rate from 15 to 45 seeds m<sup>-2</sup> increased grain protein content from 33.8% to 35.1%, oil yield increased 10% and protein yield, 17%. The results of this study show that in the management of soybean crops the sowing date and seeding rate can change grain composition. This information demonstrates that agronomic practices should be considered by growers and breeders when considering soybean grain quality.

**Keywords** *Glycine max* · Densities · Plant management · Sowing times · Lipid

## Introduction

Brazil is the largest soybean exporter and its soybean production goes mainly to China and Europe (Faostat 2017). Soybean is the main source of oil and protein in global agriculture. It is mainly used for feeding, but its use in non-feeding products is increasing. Nowadays soybean is commercialized taking only grain mass into consideration, but it is likely that in the future grain composition becomes considered. It is already well established that grain composition of soybean can change according to genotype (Jin et al. 2010; Rincker et al. 2014) and environmental conditions (Bianculli et al. 2016; Song et al. 2016). Therefore, knowing the factors that interfere in soybean grain composition is of interest to the agroindustry, to the final destination of this

raw material, since it is related to their production efficiency (Aguirrezábal et al. 2009).

Soybeans generally contain 18–23% oil and 38–42% protein. Higher protein content is of interest to the soybean meal industry because it can also reduce production costs. It has been reported that the soybean oil content is increasing year by year, while the protein content is declining in grains for the industry. During photosynthesis the synthesis “cost” of one oil unit is equivalent to 3.03 units of glucose, and the synthesis of one protein unit is 1.67–2.27 units of glucose (Penning de Vries et al. 1974).

Environmental conditions for a crop can be modified by management practices by choosing different sowing dates (SD) and seeding rates (SR). In the subtropical region of Brazil, the recommended period for soybean sowing comprises a range of approximately 70 days (Brasil 2016). Within these soybean sowing dates, there is a wide range of environmental conditions that can occur, due to variation in air temperature, precipitation, photoperiod and solar radiation. Such variations can change plant morphology (Pierozan Junior et al. 2015), phenology (Zanon et al. 2015) and yield (Meotti et al. 2012; Albrecht et al. 2008). Moreover, soybean grain weight is reduced in late sowings (Zheng et al. 2013), thus, a hypothesis arises that late sowings modify

✉ Renan Caldas Umburanas  
renan.umburanas@gmail.com

<sup>1</sup> Luiz de Queiroz College of Agriculture (ESALQ), University of São Paulo (USP), Piracicaba 13418-900, Brazil

<sup>2</sup> Midwestern State University (Unicentro), Guarapuava 85015-300, Brazil

<sup>3</sup> Center of Nuclear Energy in Agriculture (CENA), University of São Paulo (USP), Piracicaba 13400-970, Brazil

grain composition. Late sowing, compared to anticipated sowing, provides lower photothermic quotient during the reproductive stage (Zanon et al. 2016), anticipates flowering induction (Zheng et al. 2013), shortens crop cycle and plant size (Pierozan Junior et al. 2015).

SR influences plant size, biomass partition, plant grain number, microclimate, distribution of light, and intraspecific competition among plants. Higher seeding rates modify plant morphology, reducing branches per plant and leaf area index, increasing height, nod numbers, among other attributes (Bellaloui et al. 2015b). The impact of these changes on grain composition also needs further understanding. Also, the management of SR can lead to microclimatic changes within the plant canopy, such as shading, shifts in CO<sub>2</sub> availability, intraspecific competition, mainly. All of these changes can play a role in grain composition.

There is little information on the effect of SD and SR on grain composition, and even less on the combined effect of SD and SR on grain composition (Bellaloui et al. 2015a; Jaureguy et al. 2013), and yield of oil and protein. Therefore, this work analyzed the composition of soybean grains derived from different sowing dates (early, mid, and late sowings) and seeding rates (15, 25, 35, and 45 seeds m<sup>-2</sup>).

## Materials and Methods

Field experiments were conducted in Guarapuava, Parana, Brazil (25°23'S, 51°29'W, 1029 m) during the 2012/13 and 2013/14 growing seasons. The cultivar used was Brasmax Energia RR (MG 5.3, indeterminate, medium plant size, high branching index and high resistance to lodging). The soil of the area is classified as very Clayey Typic Hapludox (USDA taxonomy), with 3.6% organic matter content. The basic fertilization consisted of 80 kg ha<sup>-1</sup> P<sub>2</sub>O<sub>5</sub> and 70 kg ha<sup>-1</sup> K<sub>2</sub>O. Soybean seeds were inoculated and pests and diseases were adequately controlled. The experimental design was a randomized complete block in a split-plot arrangement with three replications. The main plot size was 96 m<sup>2</sup> (15 m × 6.4 m) and subplots of 24 m<sup>2</sup> (15 m × 1.6 m), oriented in a North–South direction. The row spacing was 0.4 m. The net area of the experimental unit was 6.4 m<sup>2</sup>. Sowing was carried out in a no-till system and cultural traits aimed at maximum yield. The initial seeding rate was 60 plants m<sup>-2</sup>, which was thinned at the V<sub>C</sub> – V<sub>1</sub> stage (Fehr and Caviness 1977) to the proposed seeding rates.

## Trial Description

Three sowing dates (SD) were used as main plots and four seeding rates (SR) as subplots. The SD were early sowing (SD<sub>1</sub>, in mid-October), mid-sowing (SD<sub>2</sub>, in mid-November), and late sowing (SD<sub>3</sub>, in mid-December). The SR

(plants m<sup>-2</sup>) were 15 (SR<sub>1</sub>); 25 (SR<sub>2</sub>); 35 (SR<sub>3</sub>); and 45 (SR<sub>4</sub>). Sowings in 2012/13 growing season were carried out on Oct. 18 (SD<sub>1</sub>), Nov. 19 (SD<sub>2</sub>), and Dec. 19 (SD<sub>3</sub>); and in 2013/14 growing season were carried out on Oct. 23 (SD<sub>1</sub>), Nov. 22 (SD<sub>2</sub>), and Dec. 20 (SD<sub>3</sub>).

Pod formation duration (R<sub>1</sub>–R<sub>5</sub>) and grain filling duration (R<sub>5</sub>–R<sub>7</sub>) were evaluated. At the R<sub>8</sub> growth stage plants from 3.6 m<sup>2</sup> of the center rows of each plot were harvested and the plant mortality rate was evaluated. Mature grain samples of each plot were collected, processed, and analyzed for grain composition.

## Evaluations

Water content, protein, oil, and ash determinations of each field replication were held in triplicate, and the results expressed on dry basis. Grain filling rate was evaluated as the ratio between the grain mass and the grain filling duration. Total nitrogen content (N) was quantified by the micro-Kjeldahl digestion method (Method 960.52 from AOAC, Horwitz 2005), and protein content was estimated using the transformation factor of N × 6.25. The oil content was determined using the Soxhlet extraction technique with the solvent petroleum ether p.a. (Method 945.16 from AOAC, Horwitz 2005). Oil content was expressed as percentage of total grain weight on dry basis. The protein content and oil content were considered in a water-free basis. Protein yield and oil yield were calculated as functions of the seed yield, protein content, and oil contents. The soybean oil density was considered as 0.9193 g mL<sup>-1</sup> at the temperature of 24 °C (Noureddini et al. 1992). The ash content was determined by incineration at 550 °C. (Method 923.03 from AOAC, Horwitz 2005). The carbohydrate content was estimated by the difference, from the total grain weight and the contents of oil, protein and ashes.

Average air temperature, precipitation and solar radiation data of the experimental period were collected in a meteorological station located 200 m far from the experimental area. A sequential water balance (Thornthwaite and Matter Thornthwaite and Matter 1955) was calculated to identify phases with water deficit during the crop cycle.

## Statistical Analysis

Sowing date and seeding rate were considered fixed, and growing season and replication were considered the random effect in ANOVA using PROC MIXED (SAS version 9.3, SAS Institute Inc., Cary, NC, USA). The Shapiro–Wilk statistic test indicated normality for all data. Qualitative data (sowing dates) were evaluated by the Tukey test and quantitative data (seeding rates) were assessed by the method of polynomial regression, selecting the highest coefficient of determination. The analysis of Pearson Correlation was

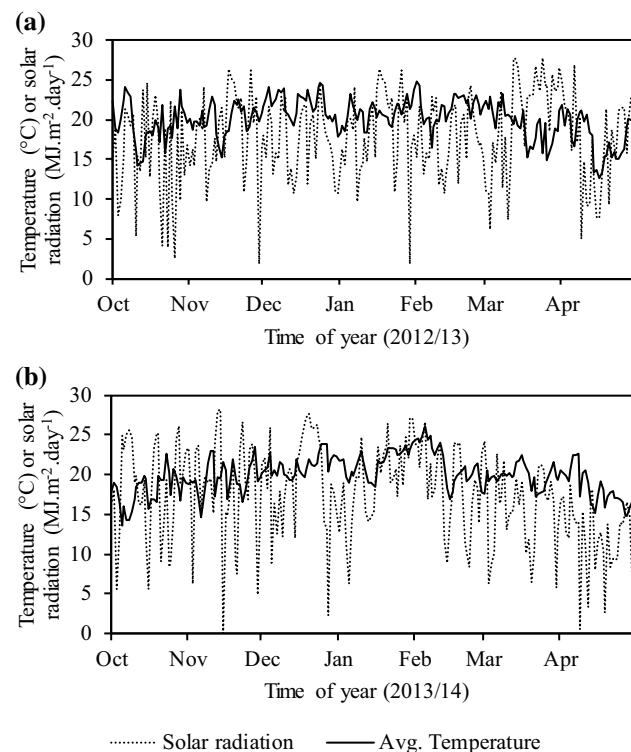
performed among evaluated attributes and final grain yield using PROC CORR option of SAS. Significance was determined at  $p = 0.05$ . The data were presented by the means of the growing seasons, and when there were three-way interactions, they were discussed in the text.

## Results

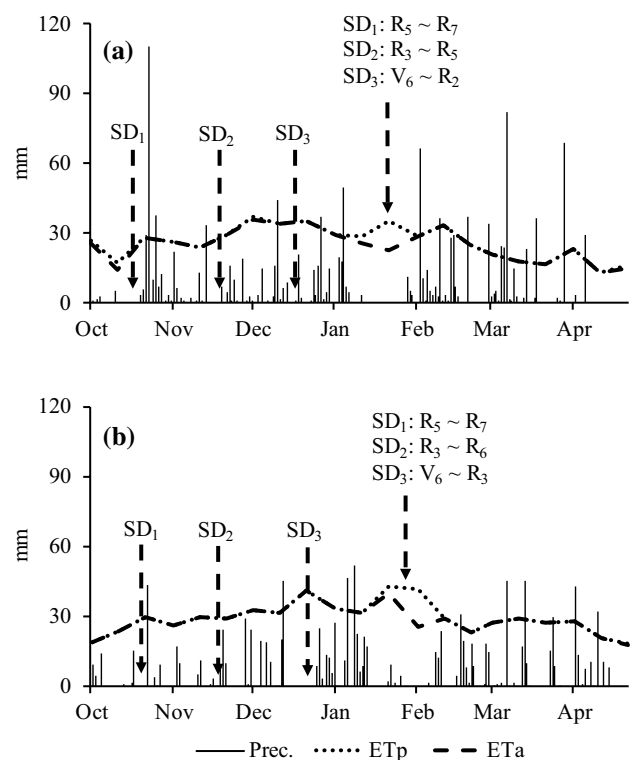
Total precipitation was 1271 mm during the 2012/13 season (Fig. 1a) and 1178 mm for 2013/14 season (Fig. 1b). Mean temperature was approximately 20 °C in both growing seasons (Fig. 1a, b). The accumulated solar radiation was 3553 MJ m<sup>-2</sup> during the 2012/13 season (Fig. 1a) and 3678 MJ m<sup>-2</sup> for 2013/14 season (Fig. 1b). For both growing seasons there was a small water deficit between the second half of January and the first half of February (Fig. 2).

### Analysis of Variance of Sowing Date and Seeding Rate

In this experiment, ANOVA (Tables 1 and 2, and Fig. 3) showed that growing seasons (Y) has significant effects on soybean oil content (Table 3). Sowing dates (SD) has significant effects on oil content. However, interactions of Y and SD were significant for oil content, grain weight, oil



**Fig. 1** Precipitation, average daily temperature, and solar radiation during the soybean growing seasons, **a** 2012/13; **b** 2013/14



**Fig. 2** Sequential water balance in Guarapuava, PR, Brazil, during the soybean growing seasons: **a** 2012/13; **b** 2013/14. *Prec* precipitation, *ETp* potential evapotranspiration, *ETA* actual evapotranspiration. The arrow indicates the phenological stage at the sowing dates in mid-October (SD<sub>1</sub>), mid-November (SD<sub>2</sub>) and mid-December (SD<sub>3</sub>)

yield, and protein yield, indicating that the response of oil is dependent on environmental factors in each growing season. Seeding rates (SR) has significant effects on protein content, oil yield and protein yield, while the interaction between SD and SR were significant only for oil yield.

### Effects of Sowing Date and Seeding Rate on Grain Composition

Protein content in grains increased from SR<sub>1</sub> to SR<sub>4</sub>, 33.8–35.1% respectively (Table 1). Oil content was reduced in plants from late sowing (SD<sub>3</sub>), while it ranged from 22.5% to 20.2% and there was no significant difference among different SRs (Table 1). Ashes and carbohydrate contents did not differ among different SD and SR (Table 1).

The grain weight from the 2012/13 growing season remained the same until mid-sowing (SD<sub>2</sub>) and was decreased in late sowing (SD<sub>3</sub>; Fig. 3a). In the 2013/14 growing season, the grain weight was similar among SDs, decreasing in early (SD<sub>1</sub>) and mid (SD<sub>2</sub>) sowings, when compared to first growing season (Fig. 3a).

Grain weight, oil, and protein content were analyzed as a function of grain weight (Fig. 4). It was observed that

**Table 1** Protein, oil, carbohydrate, and ashes content in grain composition of soybean originated from three sowing dates (SD), four seeding rates (SR), and two growing seasons in Guarapuava, PR, Brazil

SR (plants m <sup>-2</sup> )	SD <sub>1</sub> (early)	SD <sub>2</sub> (mid)	SD <sub>3</sub> (late)	Average
<b>Protein content (mg g<sup>-1</sup>)</b>				
SR <sub>1</sub> : 15	332	340	341	338
SR <sub>2</sub> : 25	343	342	350	345
SR <sub>3</sub> : 35	346	344	351	347
SR <sub>4</sub> : 45	350	349	353	351
Avg. 12/13	333 Bb	353 Aa	356 Aa	347
Avg. 13/14	353 Aa	334 Bb	342 Bab	343
Average	343	344	349	
<b>Oil content (mg g<sup>-1</sup>)</b>				
SR <sub>1</sub> : 15	222	222	205	216
SR <sub>2</sub> : 25	213	220	205	213
SR <sub>3</sub> : 35	213	220	206	213
SR <sub>4</sub> : 45	213	221	202	212
Avg. 12/13	210	217	205	210 B
Avg. 13/14	221	225	205	217 A
Average	215 a	221 a	205 b	
<b>Carbohydrates (mg g<sup>-1</sup>)</b>				
SR <sub>1</sub> : 15	401	389	405	398
SR <sub>2</sub> : 25	399	389	394	394
SR <sub>3</sub> : 35	397	389	393	393
SR <sub>4</sub> : 45	391	383	393	389
Average	397	387	396	
<b>Ashes (mg g<sup>-1</sup>)</b>				
SR <sub>1</sub> : 15	45.8	48.1	49.2	47.7
SR <sub>2</sub> : 25	57.0	48.7	50.6	52.1
SR <sub>3</sub> : 35	43.8	46.9	49.4	46.7
SR <sub>4</sub> : 45	44.8	47.3	52.2	48.1
Average	47.8	47.7	50.4	
<b>Significance</b>	<b>Protein content</b>	<b>Oil content</b>	<b>Carbohydrates</b>	<b>Ashes</b>
Growing season (Y)	ns	**	ns	ns
SD	ns	**	ns	ns
Y × SD	**	ns	ns	ns
SR	*	ns	ns	ns
Linear	0.0042 * SR + 33.254	ns	ns	ns
Y × SR	ns	ns	ns	ns
SD × SR	ns	ns	ns	ns
Y × SD × SR	ns	ns	ns	ns

Lower case letters in rows and upper case letters in columns differ according to the Tukey test ( $p < 0.05$ )

\*\*, \*, ns are  $p$  value of  $< 0.01$ ;  $< 0.05$ , and not significant, respectively

oil content tends to increase with the increase in grain weight, while protein content tends to stabilize or decay. The first growing season has a greater variation in oil and protein content than second growing season, and its explained by the better environmental conditions that propitiated a greater grain weight among treatments. Pod formation duration varied from early to late sowing from 25

to 31 days in the 2012/13 season, and from 28 to 29 days in the 2013/14 season (Fig. 5a). Grain Filling duration varied from early to late sowing from 32 to 27 days in the 2012/13 season, and from 34 to 26 days in the 2013/14 season (Fig. 5b). Both pod formation duration and grain filling duration did not vary among SRs.

**Table 2** Yield of oil and protein of soybean originated from three sowing dates (SD), four seeding rates (SR), in two growing seasons in Guarapuava, PR, Brazil

SR (plants m <sup>-2</sup> )	SD <sub>1</sub> (early)	SD <sub>2</sub> (mid)	SD <sub>3</sub> (late)	Average
<b>Oil yield (L ha<sup>-1</sup>)</b>				
SR <sub>1</sub> : 15	862.5 a	771.8 b	578.5 c	737.6
SR <sub>2</sub> : 25	811.1 a	872.9 a	602.3 b	762.1
SR <sub>3</sub> : 35	862.5 a	893.4 a	663.5 b	806.4
SR <sub>4</sub> : 45	874.1 a	876.3 a	690.9 b	813.8
Avg. 12/13	866.7 aA	912.9 aA	688.2 bA	822.6
Avg. 13/14	838.4 aA	794.4 aB	579.3 bB	737.4
Avg. Seasons	852.5	853.6	633.8	
<b>Protein yield (kg ha<sup>-1</sup>)</b>				
SR <sub>1</sub> : 15	1402.4	1291.9	1045.1	1246.5
SR <sub>2</sub> : 25	1422.4	1476.6	1120.0	1339.7
SR <sub>3</sub> : 35	1523.5	1524.6	1233.6	1427.3
SR <sub>4</sub> : 45	1559.5	1510.4	1314.6	1461.5
Avg. 12/13	1495.5 bA	1617.8 aA	1303.0 cA	1472.1
Avg. 13/14	1458.4 aA	1284.0 bB	1053.6 cB	1265.3
Avg. Seasons	1476.9	1450.9	1178.3	
Significance	Oil yield		Protein yield	
Growing season (Y)	*		**	
SD	**		**	
Y × SD	*		**	
SR	**		**	
Linear	**		**	
Y × SR	ns		ns	
SD × SR	*		ns	
Y × SD × SR	ns		ns	

Lower case letters in rows and upper case letters in columns differ according to the Tukey test ( $p < 0.05$ )

\*\*, \*, ns are  $p$  value of  $< 0.01$ ;  $< 0.05$ , and not significant, respectively

The grain filling rate has been affected by the interaction between growing seasons and SDs (Fig. 5b). There was no difference in grain filling rate among SDs in the 2012/13 growing season, however, early (SD<sub>1</sub>) and mid (SD<sub>2</sub>) sowings has lower grain filling rate than late sowing in the 2013/14 growing season. Grain filling rate was 15% lower in early (SD<sub>1</sub>) and mid (SD<sub>2</sub>) sowings from the 2013/14 season in relation to the 2012/13 season.

### Oil Yield and Protein Yield

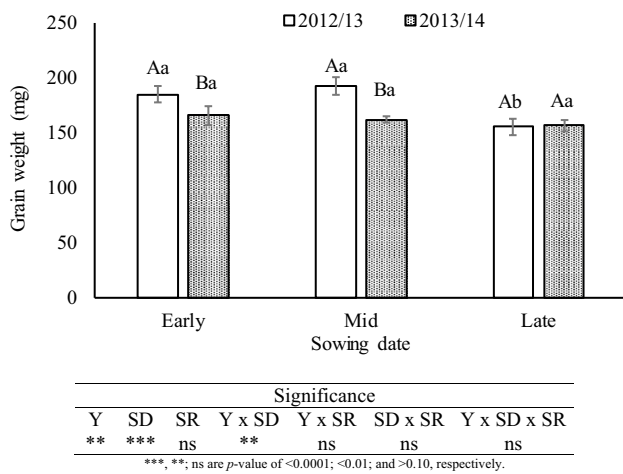
The oil yield was higher in the first season in early (SD<sub>1</sub>) and mid (SD<sub>2</sub>) sowings when compared to the second season and the late sowing (SD<sub>3</sub>) (Table 2). Oil yield and protein yield were lower in late sowing in both growing seasons. In the second growing season the oil yield and protein yield were lower in the mid (SD<sub>2</sub>) and late sowings (SD<sub>3</sub>).

In the first growing season, protein yield was higher in the mid-sowing (SD<sub>2</sub>) than early sowing (SD<sub>1</sub>) and decays

in the late sowing (SD<sub>3</sub>) (Table 2). In the second growing season, the highest protein yield was in early sowing (SD<sub>1</sub>), and also decays in the late sowing (SD<sub>3</sub>). The mid (SD<sub>2</sub>) and late sowings (SD<sub>3</sub>) were higher in the first season compared to the second season.

### Correlation Between Grain Components

There were positive correlation between grain weight versus protein yield and oil yield; protein content versus protein yield; oil content versus oil yield; and oil yield versus protein yield (Table 3). There were a negative correlation between grain weight versus ashes; oil content versus carbohydrates; ashes versus carbohydrates, oil yield, and protein yield (Table 3).



**Fig. 3** Grain weight of soybean originated from three sowing dates (SD), four seeding rates (SR), and two growing seasons (Y) in Guarapuava, PR, Brazil. Values are mean ± standard deviation. Bars with different letters within each experiment are significantly different ( $p < 0.05$ ). Capital letters for growing season and lowercase for sowing date

**Discussion**

This study shows that sowing soybean until the middle of the recommended period results in higher oil content in grain (Table 1), and also higher oil yield (Table 2). The protein content was stable across all SDs, however, protein yield varied across SD values due to differences in grain yield.

The increase in SR from 15 to 45 plants per square meter, increased grain protein content in 1.3% (Table 1). This information corroborates other studies (Bellaloui et al. 2015a, b, c). However, these authors found that this response could be quadratic or in plateau form, rather than linear like our study, meaning that in higher SRs than the ones used in this research, the increase in protein content could decrease or stabilize (Bellaloui et al. 2015a). Therefore, we confirm our argument that oil and protein contents are sensitive to agronomic practices and environmental factors.

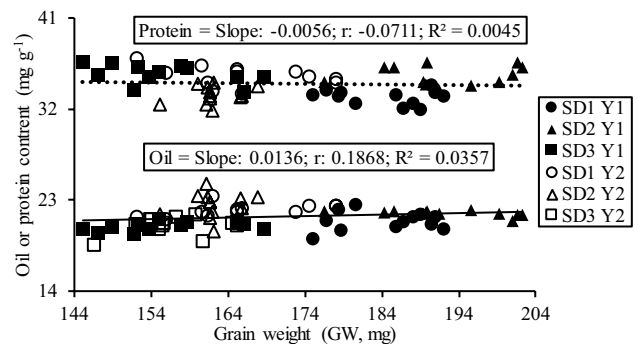
**Table 3** Correlation coefficients among analyzed attributes of soybean originated from three sowing dates (SD), four seeding rates (SR), and two growing seasons in Guarapuava, PR, Brazil

	Grain Weight	Protein content	Oil content	Ashes	Carbohydrate	Oil yield (g)
Protein	-0.07 ns					
Oil	0.19 ns	-0.18 ns				
Ashes	-0.44 ***	0.06 ns	-0.09 ns			
Carbohydrate	0.08 ns	-0.19 ns	-0.63 ***	-0.35 ***		
Oil yield	0.72 ***	0.03 ns	0.49 ***	-0.46 ***	-0.09 ns	
Protein yield	0.70 ***	0.31 *	0.18 ns	-0.45 ***	0.06 ns	0.92 ***

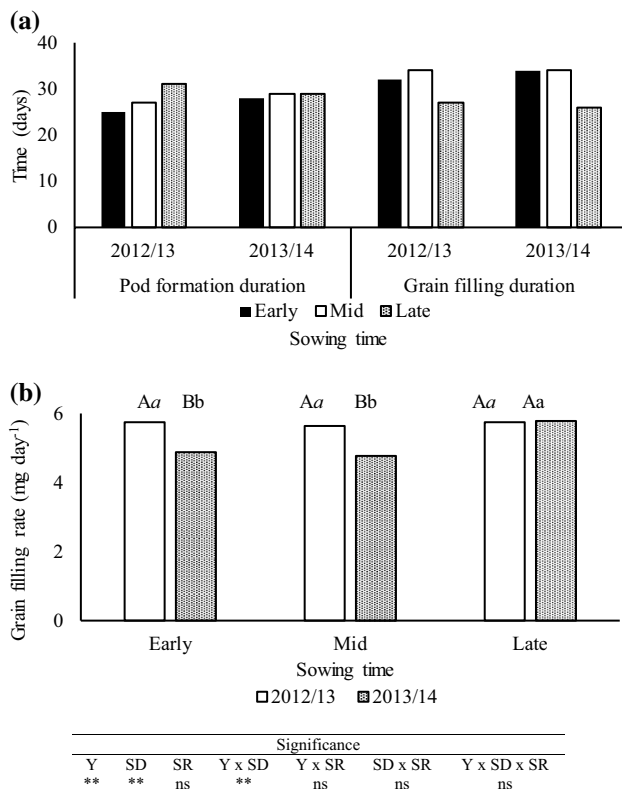
\*\*\*, \*, ns are p value of < 0.0001; < 0.05, and not significant, respectively

In this experiment the increase of SR did not cause lodging, consequently allowing normal redistribution of photosynthates to grains. The higher protein content in soybean grains grown under higher SR is possibly due to a higher ratio of source to sink, since the increase in SR reduces the biomass per plant, the leaf area per plant and, the number of seeds per plant, but increases the relation between leaf area and seeds per plant (Cox and Cherney 2011).

It was expected that the increase in SR would reduce grain protein content, since protein is related to N availability (Table 1). However, we hypothesize that soybean nitrogen (N) assimilation, as a plant that has an extra source of N from biological fixation, is less or not affected at all by plant competition in relation to non-nitrogen fixing species. There are evidences that soybeans cultivated under lower SR, cannot result in higher N content per plant than higher SR, also that plants from lower SR can accumulate less protein in their grain (Luca and Hungria 2014). The lowest SR had a higher number of grains per plant, possibly, this caused a greater redistribution of the source, that limited the potential of the grain to accumulate protein.



**Fig. 4** Dependence of oil and protein content on grain weight of soybean originated from three sowing dates (SD), four seeding rates (SR), and two growing seasons in Guarapuava, PR, Brazil. Grain weight (a), oil (solid line), and protein (dotted line) weight per grain (b) are plotted as a function of grain weight



**Fig. 5** Pod formation duration, grain filling duration (a), and grain filling rate (b) of soybean originated from three sowing dates (SD), four seeding rates (SR), and two growing seasons in Guarapuava, PR, Brazil. Values are mean  $\pm$  standard deviation. Bars with different letters within each experiment are significantly different ( $p < 0.05$ ). Capital letters for growing season and lowercase for sowing date

Climatic variations in the grain filling period ( $R_5$ – $R_{5.5}$ ) can alter the protein content of grains (Albrecht et al. 2008; Bellaloui et al. 2015a, b, c; Bianculli et al. 2016). This environmental effect might be more related to periods of severe hydric and/or temperature stress, conditions that were not observed in the two growing seasons of this study. Temperature and precipitation can affect protein content because these factors interfere on biological N fixation (Song et al. 2016).

Soybean grain oil content (Table 1) was not affected by rainfall (Fig. 2), it was also positively correlated with the photoperiod, and it present a peak at mean daily temperature of 19.7 °C (Fig. 1) (Song et al. 2016). Lower temperatures during the grain filling stages decrease the oil content in grains (Bellaloui et al. 2016). This information corroborates the results of this work, that for the late sowing ( $SD_3$ ), which has lower oil content, the plants were subject to lower temperatures, shorter grain filling duration and shorter photoperiod in relation to the early ( $SD_1$ ) and mid ( $SD_2$ ) sowings.

The temperature increase within a range of approximately 25–30 °C increases protein content of the grain (Song et al.

2016; Zuil et al. 2012). In this experiment the temperature range among SDs may not have been enough for plants to express difference in protein content.

Some stability in protein content among SDs (Table 1), especially in late sowing ( $SD_3$ ), showed that regardless of the grain weight reduction, the grain protein content was stable, i.e., not altered by the grain sink. Protein content was mainly dependent on genotype (Albrecht et al. 2008; Pierozan Junior et al. 2017), although it was also related to environmental conditions.

The effect of shading on the soybean grain oil content, has contradictory results in the literature, in which some authors have observed that the light restriction reduces the oil content, protein content and grain weight (Bianculli et al. 2016) or increases oil content and reduces protein content (Bellaloui et al. 2012). This difference may be because Bianculli et al. (2016) installed a shading treatment at  $R_5$  and Bellaloui et al. (2012) in the whole cycle. The shading intensity was different and also the temperature was not constant in these experiments.

Although the increase in SR increased the shading between the plants in this study, and also according to Bellaloui et al. (2015b), with the increase in SR no difference was found in the oil content, and the protein content increased (Table 1). The shading with the increase of SR under field conditions was probably smaller or less expressive in relation to the treatments applied by Bianculli et al. (2016) and Bellaloui et al. (2012).

To better understand the effect of shading on the composition of soybeans, further studies should be performed with different shading intensities. It was also possible that there is interaction between shading and temperature.

In this study the oil content in the grains was reduced in late sowings in the same way as in the study by Bellaloui et al. (2015a). These authors attributed the reduction of oil content to drought and reduction of temperature. However, another factor that possibly affects grain composition was the photoperiod, which was lower during the grain filling period of late sowings in relation to anticipated sowings.

Table 3 shows that grain oil content is positively correlated with the photoperiod and its peak appears at mean daily temperatures of 19.7 °C (Fig. 1), but it was less sensitive to environmental changes than protein (Song et al. 2016). The reduction in oil content in late sowings ( $SD_3$ ) evidences that when the environment provides lower temperatures during the grain filling period, as observed in late sowing ( $SD_3$ ), the plant reduces the production of higher energy demanding compounds, like oil, and tends to increase the production of lower demanding energy compounds, like carbohydrates.

Some authors suggest an inverse relationship between oil and protein content (Albrecht et al. 2008; Bianculli et al. 2016; Bellaloui et al. 2016), however, this relationship was not observed in this study, in which the correlation

coefficient between these attributes was not significant. Such difference may be related to the genotype used. This result shows that the relationship between protein and oil were more complex, and cannot be considered directly proportional in any situation.

Early ( $SD_1$ ) and mid ( $SD_2$ ) sowings tend to have heavier grain weight. In the early sowing, the period of grain filling occurred in a more favorable environmental condition range, with higher average temperatures and higher photoperiod, which, under adequate conditions of water availability, favor the photosynthetic activity and the production of photo-assimilates in the plant.

Both oil and protein yields showed interaction between growing season and SD, as grains from mid-sowing ( $SD_2$ ) had lower oil and protein yield on the 2013/14 season in relation to the 2012/13 season (Table 2). This may have occurred due to the hydric stress period on the 2013/14 growing season (Fig. 2b), that occurred when mid-sowing ( $SD_2$ ) plants were on their grain filling periods. Since on both growing seasons, a small water deficit happened in the middle of the grain filling period on early sowing ( $SD_1$ ) (Fig. 2), it was possible that the potential of oil and protein yield on early sowing ( $SD_1$ ) has been limited. Also, the level of total incident solar radiation during the months of March and April of first growing season was higher than in the second growing season. Higher oil yield in the 2012/13 growth season compared to 2013/14 may be explained by the higher average temperature and precipitation during the grain filling period on the first growing season.

Ashes contents were negatively correlated with grain weight and carbohydrates content (Table 3). Our results point out that ashes contents are, apparently, not influenced by the environment.

The higher seeding rate favored a faster canopy closure, greater interception of solar radiation, lower incidence of radiation in the pods, and higher sink-source ratio.

## Conclusions

The current research showed that management practices can affect grain oil and protein content of soybean grains. Late sowing reduced grain oil content and oil yield. The late sowing decreased the grain oil content, oil yield, and protein yield due to smaller grain filling duration and also due to the change in environmental conditions in the period of grain filling, such as lower temperature and smaller photoperiod. The increase in seeding rate from 15 to 45 seeds  $m^{-2}$  increased grain protein content, oil yield and protein yield. Further research is needed to better understand the effect of shading and photoperiod interactions with grain composition. The findings of this work help breeders and growers to

consider the sowing date and seeding rate in the production of better quality of soybean grains.

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