NOBRE, D. A. C.; SILVA, R. R.; NASSER, V. G.; SILVA, G. H.; AQUINO, L. A.; MACEDO, W. Plant physiological performance and seed chemical composition of soybean under different spatial arrangement. **Revista de Agricultura Neotropical**, Cassilândia-MS, v. 10, i. 1, e6850, jan./mar. 2023. ISSN 2358-6303. DOI: https://doi.org/10.32404/rean.v10i1.6850.

Plant physiological performance and seed chemical composition of soybean under different spatial arrangement

Danubia Aparecida Costa Nobre[1], Rodrigo Rocha Silva[2], Vinicius Guimarães Nasser[3], Geraldo Humberto Silva[3], Leonardo Angelo Aquino[4], Willian Macedo[3]

[1] Universidade Federal dos Vales do Jequitinhonha e Mucuri, Campus Diamantina, Diamantina, Minas Gerais. Brazil. E-mail: danubia.nobre@ufvjm.edu.br

[2] Rizosfera Tecnologia em Agricultura de Precisão Ltda, Patos de Minas, Minas Gerais. Brazil. E-mail: rodrigo.rizosfera@yahoo.com.br

[3] Universidade Federal de Viçosa, Campus Rio Paranaíba, Rio Paranaíba, Minas Gerais. Brazil. E-mail: vinicius.nasser@ufv.br, silvagh@ufv.br, wrmacedo@ufv.br

[4] Instituto de Pesquisa Agrícola do Cerrado, Rio Paranaíba, Minas Gerais, Brazil. E-mail: aquino.ufv@gmail.com

Received: 27/01/2022; Accepted: 07/11/2022

ABSTRACT

Few studies present interactions among plant physiological and seed chemical composition of soybean cultivates under different arrangements. Therefore, this study seeks to understand this relationship between space occupation and product quality. We grew two soybean cultivars (Tec 7849 Ipro and Monsoy 6972 Ipro) in four different spatial arrangements (50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 or 16.2 x 16.2 cm, respectively). During V4 and R2 phenological stages, we analyzed the gas exchange, and at the harvest were composed two different lots: freshly harvested seeds and the seeds subjected to accelerated aging; in both lots, we evaluated germination test, vigor, germination speed index, seedling fresh mass, and the determination of the chemical composition of the seeds. For experiment conducted to analyze physiological parameters we adopted the factorial scheme 2x2x4 (two evaluation periods x two soybean cultivars x four spatial arrangements), and for seed quality and chemical composition, we adopted the factorial scheme 2x4 (two soybean cultivars x four spatial crop arrangements). The vegetative stage was crucial for better gas exchange efficiency, and the spatial arrangements positively influenced the physiological and chemical quality of the seeds for soybean crop, where the fitted arrangement was observed in 40.0 cm between rows and 8.9 cm between plants for the cultivar Tec 7849 Ipro and 50.0 cm between rows and 5.3 cm between plants, for Monsoy 6972 Ipro. Adjusts in spatial arrangements can promote beneficial responses in seed quality.

Keywords: Gas exchange, Germination, Lipids, Gas chromatography.

Desempenho fisiológico e composição química de sementes de soja cultivadas em distintos arranjos espaciais

RESUMO

Poucos estudos apresentam interações entre a fisiologia das plantas e sua posterior composição química, quando cultivadas em diferentes arranjos. Esta pesquisa buscou compreender a relação entre o espaço de cultivo da soja e sua influência sobre a qualidade da semente. Foram estudadas duas cultivares de soja (Tec 7849 Ipro e Monsoy 6972 Ipro) em quatro arranjos espaciais distintos (50,0 x 7,1; 40,0 x 8,9; 30,0 x 11,9 e 18,9 x 18,9 ou 16,2 x 16,2 cm, respectivamente). Durante os estádios fenológicos V4 e R2, foram analisadas trocas gasosas e na colheita, foram constituídos 2 grupos, um de sementes frescas e outro de sementes envelhecidas, para ambos os grupos se analisaram: teste de germinação, vigor, índice de velocidade de germinação, massa fresca das plantas, além de determinar os constituintes químicos das sementes. Para o ensaio conduzido nas análises fisiológicas das plantas, adotou-se o esquema fatorial 2x2x4 (dois períodos de avaliação x duas cultivares x quatro arranjos espaciais), e para o ensaio de qualidade das sementes e composição química, adotou-se o esquema fatorial 2x4 (duas cultivares x quatro arranjos espaciais). O estágio vegetativo foi crucial para a melhor eficiência de trocas gasosas, e o arranjo espacial influenciou positivamente a qualidade química e fisiologia das sementes colhidas, onde, o arranjo espacial com melhor ajuste foi 40,0 x 8,9cm, para Tec 7849 Ipro, e 50,0 x 5,3 cm para Monsoy 6972 Ipro. Ajustes nos arranjos espaciais podem representa respostas benéficas na produção de sementes de melhor qualidade.

Palavras-chave: Trocas gasosas, Germinação, Lipídeos, Cromatografia gasosa.

1. Introduction

Due to the rich chemical composition of soybean (*Glycine max* L. Merrill) for human and animal nutrition, as well as the wide use of co-products, using high quality seeds is necessary to guarantee the rapid and uniform seedling emergence. Therefore, seed physiological quality is routinely used to determine field performance through feasibility and vigor (Peske et al., 2012). However, only these attributes are not enough to maintain the stand until the end of the crop cycle; the adequate environment and crop management (maturity group, sowing date, genotype, climate, and soil) ensures the final crop productivity (Carciochi et al., 2019).

Soybean crop shows high phenotypic plasticity characteristics, thus adapting to the environmental and handling conditions, with modifications in plant morphology and yield components. Spatial arrangement of plants may favor this plasticity for influencing on intraspecific competition and, consequently, on the environmental conditions, such as: water, light and nutrients, in order to guarantee favorable responses to the final crop yield (Balbinot Junior et al., 2015; Pires et al., 2000).

The correct row spacing is an important agricultural practice to increase grain yield and increase profitability to the soybean farmers (Modolo et al., 2016). Performance responses of soybean crop to the spacing changes are dependent on the cultivar, the environmental conditions and the handling practices (Balbinot Junior et al., 2014). Considering the technological advances in the development of new soybean cultivars, as well as in crop management, it is essential to carry out constant research to recognize better soybean plant arrangements and thus allow the full genetic and physiological expression of the cultivated materials (Carciochi et al., 2019; Silva and Macedo, 2022).

However, little has been reported about analysis of seed quality (vigor and chemical composition) and their crop environment, mainly on analyzes of plant spacing and interspecific competition. Given the above, this study aimed to evaluate soybean physiological performance and seed quality and chemical composition in relation to the spatial arrangement.

2. Material and Methods

The experiment was conducted at the experimental station of the Cooperativa Agropecuária do Alto Paranaíba (COOPADAP), Rio Paranaíba, MG, Brazil. During 2016-2017 crop season, for study of two soybean cultivars (Tec 7849 Ipro and Monsoy 6972 Ipro). The soil fertility was corrected according pattern of cropping in the Cerrado Mineiro environment (Souza and Lobato, 2004). The soybean seeds had industrial treatment, with: Cruiser® + polymer seed coating + Nitragin Power + *Bradyrhizobium japonicum*.

The spatial arrangement for the cultivar Tec 7849 Ipro was 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm, between rows and between plants, with 28 plants m^{-2} (following the commercial recommendation), and for the cultivar Monsoy 6972 Ipro the spacing was 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8 and 16.2 x 16.2 cm, between rows and between plants, with 38 plants m^{-2} (following the commercial recommendation). Both cultivars were sown in three replicates. Handling and cultural dealings occurred according to the culture demands.

In the V4 and R2 phenological stages (Fehr and Caviness, 1997), we assessed the physiological parameters of gas exchange: photosynthesis rate (A), stomatal conductance (gs), transpiration (E) and CO_2 internal concentration (Ci) with the aid of an infrared gas analyzer (IRGA) LI-6400 (LI-COR, Lincoln, Nebraska, USA), coupled to a modular fluorometer (6400-40 LCF, LI-COR, Lincoln, Nebraska, USA), the measurements were taken in central leaves, of fully expanded trefoils, and located in the middle of the plants, under photosynthetically active radiation of 1,200 µmol photons m^{-2} s^{-1}. All measurements occur in the morning, between 8:30am to 11:30am, in four replicates for each cultivar and spatial arrangement, at the two evaluated periods.

After harvest, the seeds were taken to the Crop Physiology and Metabolism Lab (Laboratório de Fisiologia e Metabolismo da Produção Vegetal - LAFIMEPRO), at Federal University of Viçosa, *Campus* Rio Paranaíba, MG, Brazil, to evaluate seed quality, the experiment occurred in a completely randomized design, with four replicates of 50 seeds for each treatment (cultivar and spatial arrangement).

Seed quality was initially determined by the germination test. Where was used the Germitest® paper roll method, moistened with distilled water, 2.5 times the fresh dry mass of the paper. After the rolls were assembled, we put them in a germinator, at constant temperature (25ºC), with evaluations carried out on the fifth and eighth day after the test installation (Brasil, 2009); at the end of the test, the normal, abnormal and dead seedlings were calculated; the results were expressed as a percentage (%).

The first germination test (vigor), obtained by the number of normal seedlings, was carried out on the fifth day after the test assembly, and the results were also expressed as a percentage (Brasil, 2009). From each replicate, we removed 10 normal seedlings from the middle third of the paper and measured the fresh mass of the seedlings, using a precision scale (0.001 g). We also obtained the germination speed index (GSI), registering, daily at the same time, the number of seedlings that showed radicle protrusion during the eight days of evaluation. At the end of the test, with the daily data of the number of germinated seeds, we calculated the GSI (Maguire, 1962). Soybean seeds were also submitted to accelerated aging test, where the seeds were distributed in a single layer, taking the whole surface of the metal screen of the Gerbox plastic boxes with individual compartment containing 40 mL distilled water. The plastic boxes were maintained in a Biochemical Oxigen Demand (B.O.D.) germination chamber, regulated at 41ºC for 48 hours (Marcos Filho, 1999). Then, the seed quality parameters were again evaluated as described previously.

In order to evaluate seed chemical composition, all analyzes were performed in triplicate, according to the Adolfo Lutz Institute Methodology (Instituto Adolfo Lutz, 2008). We determined the lipid content in mass of 1.0 g ground sample of soybean seeds, packed in filter paper cartridge, by extracting the petroleum ether solvent in a Soxhlet extractor, the results were expressed in percentage (%). Subsequently, we obtained ash content from the mass residues within the cartridges. For protein, the Kjeldahl methodoly was applied, the evaluated mass was 0.4 g seeds previously dried and ground, digested in sulfuric acid; total nitrogen was transformed into protein via the conversion factor, order of 6.25, the results were expressed in percentage (%).

Oils extracted from each cultivar and spatial arrangement was characterized by gas chromatography coupled to mass spectrometry (GC-MS) for their composition in fatty acids methyl esters (Instituto Adolfo Lutz, 2008). We used the equipment QP2010 (Shimadzu) with a fused silica capillary column (30m length and 0.25mm internal diameter) with stationary phase RTX®-5MS (0.25 µm film thickness) and gas-filled helium with flow rate of 1.0 mL minute^{-1}. The temperature of the injector was at 220 ºC and initial column temperature at 60 ºC, with programming to increase from 2ºC per minute until reaching 200 ºC, and from 5ºC per minute until reaching maximum 250ºC. The mass spectra were obtained by electron impact at 70 eV, with a scan from 29 to 400 (m/z). Was injected 1µL oil solution with 10 mg L^{-1} preparation at 1:20 split ratio into three replicates per treatment.

Compound identification occurred by comparing mass spectra with those in the NIST library, visual interpretation of mass spectra and retention index comparison. The relative percentage of each compound was calculated by the ratio of area of each peak to the total area of all sample constituents. The experiment was carried in randomized block design and four spatial arrangement forms for both cultivars (Tec 7849 Ipro and Monsoy 6972 Ipro). For physiological parameters, we applied the 2x2x4 factorial scheme (two periods of evaluation x two soybean cultivars x four spatial crop arrangements). For seed quality and chemical composition, data were submitted to analysis of variance in a 2x4 factorial scheme (two soybean cultivars x four spatial crop arrangements). When the *F test* was significant ($p < 0.05$), the *Tukey test* was performed at a 95% probability level.

3. Results and Discussion

The physiological parameters evaluated in the soybean plants to which the triple factorial scheme (2x2x4) was applied did not show interactions among the factors. There was a difference ($p<0.05$) only for the evaluated periods (Table 1). For both cultivars were observed highest means of photosynthesis for vegetative stage (V4), when compared with reproductive stage (R2) (Figure 1A). However, for treatments 3 (30.0 cm between rows and 11.9 cm between plants), 7 (30.0 cm between rows and 8.8 cm between plants) and 8 (spacing equidistant from 16.2 cm) the means not differ in statistical analysis.

For stomatal conductance higher values were observed in the V4 stage (Figure 1B), except for the cultivar Tec 7849 Ipro in spacing 1 (50.0 x 7.1 cm) and for the Monsoy 6972 Ipro in the spacings 5 and 6 (50.0 x 5.3 cm and 40.0 x 6.6 cm, respectively), and for transpiration, in which the highest averages were also observed in the V4 only for the cultivar Tec 7849 Ipro in spacings 1 and 3 (50.0 x 7.1 cm and 30.0 x 11.9 cm, respectively); the others treatments did not showed differences (Figure 1C). Distinct performance was visualized for internal CO_2 concentration (Figure 1D), where the means of R2 were higher than V4. Only the Monsoy 6972 Ipro showed a significant difference for the spacings 5 and 6 (50.0 x 5.3 cm and 40.0 x 6.6 cm, respectively).

These results showed the importance of to carry out an adequate management of soybean crops, allowing the environment to be a positive factor in yield gains in the culture. Similar conclusions were observed in researches with soybean in different regions of Brazil and in the world (Carciochi et al., 2019; Kumagai and Takahashi, 2020; Silva and Macedo, 2022). It was possible to verify the significance of development stages on soybean plants for means observed for photosynthesis, stomatal conductance, transpiration and internal CO_2 concentration, in two phenological stages, for two cultivars and different spatial arrangements. According Jiang et al. (1993) as plant development begins to occur, the ability to synthesize photoassimilates increases to the maximum point of leave maturity, a period that coincides with the differentiation between the vegetative and reproductive phase, in which the decrease of the photosynthetic rates begins.

Table 1. Summary of analysis for the photosynthesis (*A*), stomatal conductance (*gs*), transpiration (*E*) and internal CO_2 concentration (C*i*), in two phenological stages (periods V4 and R2), for two cultivars (Tec 7849 Ipro and Monsoy 6972 Ipro) in the spacings (S) between rows and between plants (1, 2, 3 and 4)

	DF	*A*	*gs*	*E*	C*i*
Periods (P)	1	873.09^{**}	0.081^{**}	13071.46^{**}	35.88^{**}
Spacings (S)	3	33.53^{ns}	0.002^{ns}	1135.78^{ns}	0.02^{ns}
Cultivars (C)	1	13.43^{ns}	0.006^{ns}	1298.78^{ns}	2.25^{ns}
P x S x C	3	2.74^{ns}	0.001^{ns}	223.33^{ns}	0.45^{ns}
Error	39	19.96	0.004	827.62	0.70
CV (%)	-	25.53	18.36	10.95	14.36

** significant at 1%; ^{ns} not significant

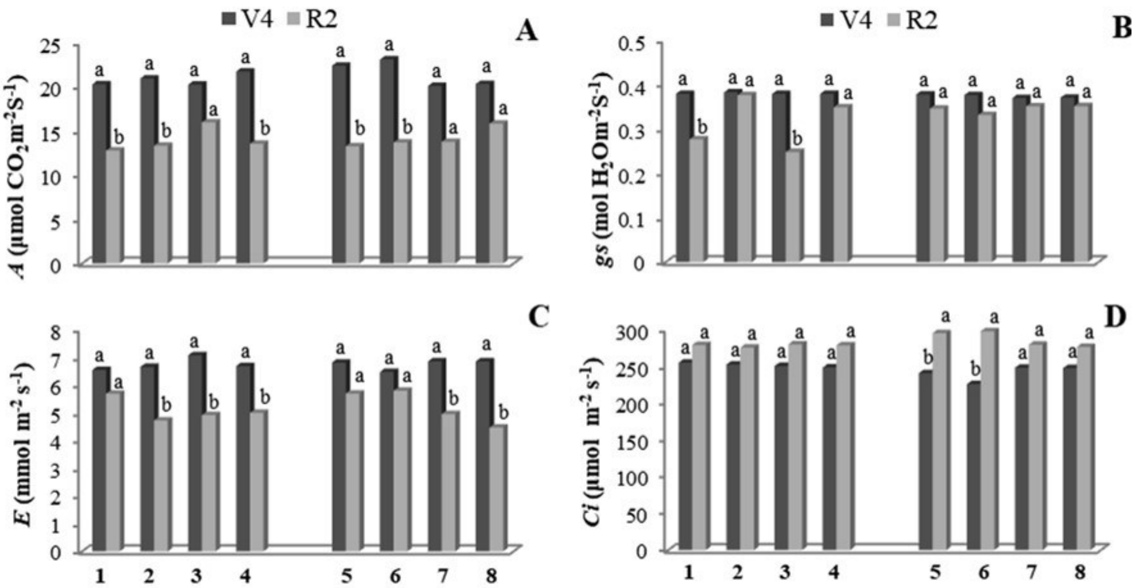

Figure 1. Photosynthesis (*A*, A), stomatal conductance (*gs*, B), transpiration (*E*, C) and internal CO_2 concentration (C*i*, D), in two phenological stages: V4 (dark gray column) and R2 (light gray column), for cultivars Tec 7849 Ipro (50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4) and Monsoy 6972 Ipro (50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and between plants, respectively, for spacings 5, 6, 7 and 8) grown in four different spatial arrangements. Equivalent letters, for the evaluation periods (V4 and R2), do not differ by Tukey test (5%), n=4

Our results present a major physiologic performance in soybean plants during vegetative phenological stage, when compared with reproductive phenological stage (Figure 1). However, these differences observed in soybean plants, can be related to the soybean maturity group and its environmental conditions (Gordon et al., 1982). Under few spatial arrangements we confirm significate similar responses among these treatments. In this study we consider that photosynthesis reduction in the R2 stage is linked to the development process of the reproductive organs, in which photoassimilates are allocated to the strong drains in development, and the reductions in stomatal conductance and transpiration in the V4 stage can be described as natural mechanisms of tissue maturity in order to initiate their senescence.

This justifies the increase of the CO_2 concentration of plants in the R2 stage, proving the reduced capacity that this has to carboxylate (Jiang et al., 1993). During the development and maturation of leguminous seeds, the demand for assimilated carbon comes mainly from young pods, although the leaves are the primary source (Zhang et al., 2017). Photosynthesis through the pods recycles the respiratory carbon, released by the seed in the internal cavity of the fruit, thus the formation of storage reserves in the seed depends on the assimilated formed during the fruiting itself; seed yield is also sensitive to adverse environmental influences during this filling (Bewley and Black, 1985), consequently, the allocation of photoassimilates regulate the ability of each variety to reserve its compounds, as seen in Table 7.

Therefore, we consider that the results in Figure 1, during the R2 stage, confirm the greater contributions of the pods in relation to the leaves, to form the seeds. There is an impact dilemma caused by plant spacing and population in a given area on the seed qualitative characteristics. It is possible that there is no population effect on seed quality (Rossi et al., 2017) or that constitutive change is possible in the chemical composition under different planting spacings (Bellaloui et al., 2014).

For seed vigor analysis, the cultivars differed only in spacing 2, in which the Tec 7849 Ipro showed greater vigor when compared to the Monsoy 6972 Ipro (Table 2). The comparison between the arrangements showed no significant difference when the seed vigor for the cultivar Tec 7849 Ipro was evaluated; however, for the cultivar Monsoy 6972 Ipro smaller averages occurred for the spacing 2, which differed from the others (Table 2). The GSI compared among cultivars showed higher averages in all the spatial arrangements evaluated to the cultivar Tec 7849 Ipro.

Meanwhile, a parallel of the GSI within the spatial arrangements showed that cultivar Tec 7849 Ipro presented lower average for the spacing 2, which differs only from 1. For the cultivar Monsoy 6972 Ipro, only the spacing 4 showed the lowest mean and did not differ from the spacing 2 (Table 2). Concerning the fresh mass of soybean seedlings, the cultivar Monsoy 6972 Ipro showed higher averages when compared to the Tec 7849 Ipro. To compare the spatial arrangements, the Tec 7849 Ipro did not show any difference, whereas the Monsoy 6972 Ipro showed a higher fresh seedling mass for those seeds originated from spacing 2, differing from the others (Table 2).

The Monsoy 6972 plants (normal maturity cycle) spaced between rows (arrangements 3 and 4) showed lower averages when compared to the conventional and equidistant planting model, presenting better adaptation under these conditions. The cultivar Tec 7849 (late maturity cycle) showed no divergence between the spacings. According to Egli (2010), the seed vigor of soybean seeds is also linked to the period of plant exposure to solar radiation; since the production of pods and seeds is directly related to the environmental adjustment of soybean plants; furthermore, for soybean crops the yield reductions are, partially, due to reductions in capture of cumulative irradiance, resulting from slowed canopy development during vegetative and early reproductive stages (Kumagai and Takahashi, 2020).

In this study, the modifications in germination and vigor of the seed were directly dependent of the environment cultivation systems. For the variables that quantify the vigor portrayed is in Table 2 and for the germination results is in Table 3. For the variables that quantify the vigor portrayed in Table 2, we confirm that the effect of spatial arrangement manifests under the quality of the seeds, these observations corroborate with Finch-Savage and Bassel (2016).

Table 2. Means of vigor (%), germination speed index (GSI) and fresh seedling mass (g), from seeds of two soybean cultivars, in different spatial arrangements.

Variables	Cultivar	Spatial Arrangement (cm)*			
		1	2	3	4
Vigor	Tec 7849 Ipro	87.50 Aa	81.00 Aa	83.50 Aa	89.50 Aa
	Monsoy 6972 Ipro	88.50 Aab	66.00 Bc	78.00 Ab	89.50 Aa
CV (%) = 6.69					
GSI	Tec 7849 Ipro	39.04 Aa	36.83 Ab	38.50 Aab	38.87 Aab
	Monsoy 6972 Ipro	36.16 Ba	34.04 Bab	35.75 Ba	32.71 Bb
CV (%) = 3.05					
Fresh mass	Tec 7849 Ipro	6.73 Ba	6.66 Ba	6.63 Ba	6.57 Ba
	Monsoy 6972 Ipro	9.06 Abc	9.82 Aa	9.21 Ab	8.67 Ac
CV (%) = 3.10					

Cultivar Tec 7849 Ipro: 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Cultivar Monsoy 6972 Ipro: 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Means followed by the same letter, upper case in the column and lower case in the row, do not differ by Tukey test (5%), n=4.

Table 3. Means of germination and abnormal seedlings (%) results from two cultivars of soybean seeds, grown in different arrangements.

Variables	Cultivar	Spatial Arrangement (cm)*			
		1	2	3	4
Germination	Tec 7849 Ipro	97.50 A	97.00 A	97.00 A	95.50 A
	Monsoy 6972 Ipro	94.00 A	90.50 B	94.50 A	96.00 A
CV (%) = 3.06					
Abnormal Seedlings	Tec 7849 Ipro	2.00 A	2.50 B	2.50 A	4.00 A
	Monsoy 6972 Ipro	5.50 A	9.00 A	5.50 A	4.50 A
CV (%) = 28.56					

Cultivar Tec 7849 Ipro: 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Cultivar Monsoy 6972 Ipro: 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Means followed by the same letter, upper case in the column, do not differ by Tukey test (5%), n=4.

Finch-Savage and Bassel (2016), claim that the environment in which a seed develops affects its behavior in subsequent generations, that is, seed quality is influenced by the maternal environment. For the germination results (Table 3), there was a significant difference between the cultivars only for the spatial arrangement 2, in which the Tec 7849 Ipro showed higher germination, which can also be verified by the higher average number of abnormal seedlings for Monsoy 6972 Ipro.

Evaluating the seed germination and number of abnormal seedlings we were able to verify how much the spatial arrangement was incisive on the development of the seedlings, mainly for the cultivar Monsoy 6972, not recommending its cultivation outside the one preconized for the cultivation on Brazilian Cerrado. Seed vigor, when comparing the cultivars, showed lower average only for the Tec 7849 Ipro in the spacing 4, for the other spacings, the cultivars did not present significant difference (Table 4). For the spatial arrangements within each cultivar, it is again noted that the cultivar Tec 7849 Ipro showed lower strength result for the spacing 4, differing from the others.

For Monsoy 6972 Ipro, smaller averages were expressed in spacings 1 and 3 (Table 4). The average germination, for the cultivars Tec 7849 Ipro and Monsoy 6972 Ipro, in the different spatial arrangements, showed an average reduction of 25 and 20%, respectively, and after accelerated aging. To compare the cultivars within each spatial arrangement, the lowest averages were expressed for the cultivar Tec 7849 Ipro in the spacings 3 and 4 (Table 4). The spatial arrangements within each cultivar allowed us to infer that the Tec 7849 Ipro had lower germination for the spacing 3, which did not differ from 4. There was no difference in germination for the Monsoy 6972 Ipro in the different spacings (Table 4).

When comparing the cultivars within each spatial arrangement, the Tec 7849 Ipro showed higher mean values of abnormal seedlings in spacing 3 and 4 (Table 4), which confirms the low germination results mentioned above. The same occurred for the comparison of the spatial arrangements within each cultivar. The cultivar Tec 7849 Ipro showed greater percentage of abnormal seedlings for the spacing 3 that did not differ from 4; there was no difference for the Monsoy 6972 Ipro (Table 4).

Seedling fresh mass are the only results after aging similar to the previous ones. When comparing the cultivars in each spatial arrangement, the cultivar Monsoy 6972 Ipro showed higher averages when compared to the Tec 7849 Ipro, which is a characteristic of the cultivar itself, because the seeds have larger size and give rise to larger and heavier seedlings (Table 4). The comparison between the spatial arrangements for each cultivar shows that the Tec 7849 Ipro did not show any difference. However, the Monsoy 6972 Ipro showed higher fresh seedling mass for those seeds originating from spacing 1, which did not differ from 4 (Table 4).

The accelerated aging test was more sensitive than the freshly harvested seeds to detect differences in seed physiological potential. After aging, the seeds showed different results and lower than those previously presented, except for seedling fresh mass. Accelerated seed aging generally causes complete metabolic inactivation, especially in less vigorous seeds, it is thus possible to provide information on the potential for emergence in the field and seed storage. According to Delouche and Baskin (1973) and Marcos Filho (2015), the lots of higher vigor tolerates better these aging conditions, these observations corroborated with our results, where seeds originated from arrangements 1 and 2 showed minor damage when subjected to accelerated aging test.

Table 4. Means of vigor, germination, abnormal seedlings (%) and seedling fresh mass (g), after accelerated aging, from two soybean cultivars cultivated in different arrangements.

Variables	Cultivar	Spatial Arrangement (cm)*			
		1	2	3	4
Vigor	Tec 7849 Ipro	45.00 Ab	61.00 Aa	53.00 Aab	27.50 Bc
	Monsoy 6972 Ipro	48.50 Ab	65.00 Aa	45.00 Ab	73.50 Aa
CV (%) = 12.00					
Germination	Tec 7849 Ipro	78.50 Aa	78.00 Aa	64.00 Bb	69.50 Bab
	Monsoy 6972 Ipro	74.50 Aa	77.50 Aa	76.5 Aa	81.00 Aa
CV (%) = 6.86					
Abnormal seedlings	Tec 7849 Ipro	21.00 Ab	21.00 Ab	36.00 Aa	28.50 Aab
	Monsoy 6972 Ipro	25.00 Aa	21.50 Aa	23.00 Ba	18.50 Ba
CV (%) = 10.32					
Fresh mass	Tec 7849 Ipro	6.08 Ba	5.87 Ba	6.18 Ba	5.75 Ba
	Monsoy 6972 Ipro	8.87 Aa	9.18 Aa	8.23 Ab	8.72 Aab
CV (%) = 3.47					

Cultivar Tec 7849 Ipro: 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and plants, respectively, - spacings 1, 2, 3 and 4. Cultivar Monsoy 6972 Ipro: 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and plants, respectively - spacings 1, 2, 3 and 4. Means followed by the same letter, upper case in the column and lower case in the row, do not differ by Tukey test (5%), n=4.

These results revalidate the best seed performance with greater vigor, under the best spatial arrangement, for this study. In the Table 5 was measured GSI after accelerated aging, we did not find significant interaction among the treatments, only different arrangements presented significant effect. For Tec 7849 Ipro the best result was observed for spacing 2, which differed only from spacing 1, and the cultivar Monsoy 6972 Ipro did not show differences between the spacings. In this study, the simple modification in the spatial arrangement, maintaining the same population density, favored the quality in soybean seeds. In general, before and after accelerated aging, the spatial arrangement 2 (40.0 cm between rows and 8.9 cm between plants) was the best outcome for the cultivar Tec 7849 Ipro and, arrangement 1 (50.0 cm between rows and 5.3 cm between plants) for the Monsoy 6972 Ipro.

For seed chemical composition, when comparing the two cultivars and in each spatial arrangement (Table 6), there was difference only in the spacing 2, in which the cultivar Tec 7849 Ipro showed higher lipid content. However, the comparison of the spatial arrangements for each cultivar allows us to infer that the Tec 7849 Ipro again showed higher lipid content in spacing 2, which did not differ from the spacing 3. The Monsoy 6972 Ipro did not express significance for the spatial arrangements.

Table 5. Means of germination speed index (GSI), after accelerated aging, from two soybean cultivars in different arrangements.

Variables	Cultivar	Spatial Arrangement (cm)*			
		1	2	3	4
GSI	Tec 7849 Ipro	24.71 b	29.14 a	25.00 ab	26.00 ab
	Monsoy 6972 Ipro	26.12 a	28.18 a	27.47 a	27.70 a
CV (%) = 8.07					

Cultivar Tec 7849 Ipro: 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Cultivar Monsoy 6972 Ipro: 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Means followed by the same letter, lower case in the row, do not differ by Tukey test (5%), n=4.

Table 6. Means of lipids (%) and proteins (%) of seeds of two soybean cultivars in different arrangements.

Variables	Cultivar	Spatial Arrangement (cm)*			
		1	2	3	4
Lipid	Tec 7849 Ipro	18.53 Ab	25.33 Aa	19.17 Aab	17.60 Ab
	Monsoy 6972 Ipro	20.90 Aa	15.80 Ba	17.33 Aa	20.76 Aa
CV (%) = 14.74					
Protein	Tec 7849 Ipro	35.03 Aa	34.17 Aa	33.38 Aa	35.42 Aa
	Monsoy 6972 Ipro	31.72 Aa	30.60 Ba	28.23 Ba	29.81 Ba
CV (%) = 6.19					

Cultivar Tec 7849 Ipro: 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Cultivar Monsoy 6972 Ipro: 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Means followed by the same letter, upper case in the column and lower case in the row, do not differ by Tukey test (5%), n=4.

Table 7. Means of soybean constituints oil (relative percentage, %) of the relative area peaks of the major constituents of seed oil of two soybean cultivars, grown in different arrangements.

Variables	Cultivar	Spatial Arrangement (cm)*			
		1	2	3	4
Methyl palmitate	Tec 7849 Ipro	8.87 Aa	8.95 Aa	8.25 Aa	8.11 Ba
	Monsoy 6972 Ipro	7.92 Ab	7.93 Ab	8.00 Aab	9.37 Aa
CV (%) = 7.25					
Methyl linoleate	Tec 7849 Ipro	51.28 Aa	51.64 Aa	53.14 Aa	53.82 Aa
	Monsoy 6972 Ipro	50.65 Ab	52.33 Aab	53.91 Aa	54.11 Aa
CV (%) = 2.64					
Methyl oleate	Tec 7849 Ipro	37.06 Aa	34.75 Aa	37.01 Aa	36.18 Aa
	Monsoy 6972 Ipro	38.76 Aa	37.32 Aa	35.57 Aab	32.62 Bb
CV (%) = 5.18					

Cultivar Tec 7849 Ipro: 50.0 x 7.1; 40.0 x 8.9; 30.0 x 11.9 and 18.9 x 18.9 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Cultivar Monsoy 6972 Ipro: 50.0 x 5.3; 40.0 x 6.6; 30.0 x 8.8; 16.2 x 16.2 cm between rows and between plants, respectively, for spacings 1, 2, 3 and 4. Means followed by the same letter, upper case in the column and lower case in the row, do not differ by Tukey test (5%), n=4.

For protein (Table 6), when comparing the two cultivars, the Tec 7849 Ipro showed the highest averages, except for the spacing 1, which did not show a significant difference. The comparison between the different arrangements in each cultivar did not express any significance. The results of chemical composition (Table 6), again, indicated arrangements 1 and 2 for the Monsoy 6972 Ipro and Tec 7849 Ipro, respectively, as those of better performance for soybean seeds.

In Table 7 the results showed major constituents of soybean oil, expressed in relative percentage, in their different spatial arrangements. A comparison between the cultivars showed that in the cultivar Monsoy 6972 Ipro the methyl palmitate expressed better in arrangement 4, whereas the same arrangement showed lower values for the methyl oleate, differing from the averages in the arrangements 1 and 2. The Tec 7849 Ipro showed a linear pattern of lipid constituent expression in the different spatial arrangements (Table 6). For the methyl linoleate compound, there was no significant effect between the cultivars and between the spatial arrangements for the cultivar Tec 7849 Ipro. As for the Monsoy 6972 Ipro, there were better averages for arrangements 3 and 4, differing only from 1 (Table 7).

Oil compositions in soybean seeds were influenced by the different spatial arrangements; however, genetic and environmental factors must be considered in the formation of these compounds in seeds (Cherry et al., 1985). These methyl esters found in soybean oil confer quality on soybean and may eventually have physiological function because of the functionality they show in seeds, since lipids are essential for cell structure, storage and signaling (Ohlrogge et al., 2015). So, it is proved that differential arrangements modified seed quality, by environment influence during all phonologic phases of crop on the field. In addition, seed lipid composition may affect the germination performance, due to changes in the water absorption dynamics, proving that these constituents have a great impact on crop establishment (Izquierdo et al., 2017), for this reason, the specific fatty acid would have a salient function in the physiological quality, and, consequently, can alter the vigor and germination of soybean seeds.

In this study, the use of a spatial arrangement for soybean culture justifies the alterations showed in the physiological quality of plants and seeds, as propused by Bellaloui et al. (2015), these authors argue that agricultural practices such as sowing rate and row spacing, can alter constituents in soybean seeds, affecting their chemical composition. Then, the chemical composition of the seeds observed in this assay can be linked to greater photosynthetic gain, dry matter maintenance and better distribution of these reserves for the formation of a new seedling.

4. Conclusions

The plant gas exchange was stable in both genotypes under distinct arrangement, under vegetative phenological stage, however, a decrease in gas exchange was observed in reproductive stage (R2), expected result, taking into account leaf senescence. Keeping the number of plants per hectare and loosening the row and plants spacing, it is possible to obtain gains in seed physiology and quality, with best results in plants growing under treatments 1 and 2, respectively, 50.0 x 7.1 and 40.0 x 8.9 cm, for both genotypes.

Authors' Contribution

Danubia Aparecida Costa Nobre contributed to the experiment setup, manuscript writing, data analysis and elaborated the graphs. Rodrigo Rocha Silva assisted in the evaluations, data collection, tabulation. Vinicius Guimarães Nasser assisted in the evaluations and data collection. Geraldo Humberto Silva writing and data reviewing, Leonardo Angelo Aquino writing and data reviewing. Willian Rodrigues Macedo experiment setup, manuscript writing, data analysis and guided the first and second authosr.

Acknowledgments

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001". And Cooperativa Agropecuária do Alto Paranaíba (COOPADAP), Rio Paranaíba - MG, for allowing us to use the field experiment area.

Bibliographic References

Balbinot Junior, A.A., Procópio, S.O., Debiasi, H., Franchini, J.C., 2014. Redução do espaçamento entre linhas na cultura da soja. Embrapa Soja, Londrina.

Balbinot Junior, A.A., Procópio, S.O., Debiasi, H., Franchini, J.C., Panison, F., 2015. Semeadura cruzada em cultivares de soja com tipo de crescimento determinado. Semina, 36, 1215-1225. DOI: http://dx.doi.org/10.5433/1679-0359.2015v36n3p1215

Bellaloui, N., Mengistu, A., Walker, E.R., Young, L.D., 2014. Soybean seed composition as affected by seeding rates and row spacing. Crop Science, 54, 1782-1795. DOI: https://doi.org/10.2135/cropsci2013.07.0463

Bellaloui, N., Bruns, H.A., Abbas, H.K., Mengistu, A., Fisher, D.K., Reddy, K.N., 2015. Effects of row-type, row-spacing, seeding rate, soil-type, and cultivar differences on soybean seed nutrition under US Mississippi delta conditions. PLoS One, 10, 1-23. DOI: https://doi.org/10.1371/journal.pone.0129913

Bewley, J.D., Black, M., 1985. Seeds: physiology of development and germination. Plenum Press, New York.

Brasil. 2009. Ministério da Agricultura, Pecuária e Abastecimento. Regras para Análise de Sementes. Secretaria de Defesa Agropecuária. Mapa/ACS, Brasília.

Carciochi W.D., Schwalbert R., Andrade F.H., Corassa G.M., Carter P., Gaspar A.P., Schmidt, J., Ciampitti, I. A., 2019. Soybean seed yield response to plant density by yield environment in North America. Agronomy Journal, 111(4), 1923–1932. DOI: https://doi.org/10.2134/agronj2018.10.0635

Cherry, J.H., Bishop, L., Hasegawa, P.M., Leffler, H.R., 1985. Differences in the fatty acid composition of soybean seed produced in northern and southern areas of the U.S.A. Phytochemistry, 24(2), 237-241. DOI: https://doi.org/10.1016/S0031-9422(00)83527-X

Delouche, J.C., Baskin, C.C., 1973. Accelerated aging techniques for predicting the relative storability of seed lots. Seed Science and Technology, 1(2), 427-452.

Egli, D.B., 2010. Soybean reproductive sink size and short-term reductions in photosynthesis during flowering and pod set. Crop Science, 50(5), 1971-1977. DOI: https://doi.org/10.2135/cropsci2009.09.0518

Fehr, W., Caviness, R.H., 1997. Stages of soybean development. Special Report 80. Iowa State University, Iowa. https://crops.extension.iastate.edu/soybean/production_growth stages.html. (Accessed May 01, 2021).

Finch-Savage, W.E., Bassel, G.W., 2016. Seed vigour and crop establishment: extending performance beyond adaptation. Journal of Experimental Botany, 67(3), 567-591. DOI: https://doi.org/10.1093/jxb/erv490

Gordon, A.J., Hesketh, J.D., Peters, D.B., 1982. Soybean leaf photosynthesis in relation to maturity classification and stage of growth. Photosynthesis Research. 3(2), 81-93. DOI: https://doi.org/10.1007/BF00040706

IAL. INSTITUTO ADOLFO LUTZ, 2008. Métodos físico-químicos para análise de alimentos. 4ed. IAL, São Paulo.

Izquierdo, N., Benech-Arnold, R., Batlla, D., Belo, R.G., Tognetti, R., 2017. Seed composition in oil crops: its impact on seed germination performance. In: Ahmand, P., (Ed) Oilseeds crops: yield and adaptations under environmental stress. John Wiley & Sons Ltd. Hoboken, p.34-51.

Jiang, C.Z., Rodermel, S.R., Shibles, R.M., 1993. Photosynthesis, rubisco activity and amount, and their regulation by transcription in senescing soybean leaves. Plant Physiology, 101(1), 105-112. DOI: https://doi.org/10.1104/pp.101.1.105

Kumagai, E., Takahashi, T., 2020. Soybean (*Glycine max* (L.) Merr.) yield reduction due to late sowing as a function of radiation interception and use in a cool region of northern Japan. Agronomy 10(1): 66. DOI: https://doi.org/10.3390/agronomy10010066

Maguire, J.D., 1962. Speed of germination-aid in selection and evaluation for seedling emergence and vigor. Crop Science, 2(2), 176-177. DOI: http://dx.doi.org/10.2135/cropsci1962.0011183X000200020033x

Marcos Filho, J., 2015. Seed vigor testing: an overview of the past, present and future perspective. Scientia Agricola, 72(4), 363-374. DOI: https://doi.org/10.1590/0103-9016-2015-0007

Marcos Filho, J., 1999. Testes de vigor: importância e utilização, in: Krzyzanowski, F.C., Vieira, R.D., França Neto, J.B., (Ed.) Vigor de sementes: conceitos e testes. Londrina, Associação Brasileira de Tecnologia de Sementes. p. 1-21.

Modolo, A.J., Schidlowski, L.L., Storck, L., Benin, G., Vargas, T.O., Trogello, E., 2016. Rendimento de soja em função da descompactação do solo. Revista de Agricultura, 91(3), 216-229.

Ohlrogge, J., Browse, J., Jaworski, J., Somerville, C., 2015. Lipids. In: Buchanan, B.B., Gruissem, W., Jones, R.L., (Ed), Biochemistry & molecular biology of plants. Hoboken, Wiley Blackwell. p. 337-400.

Peske, S.T., Villela, F.A., Meneghello, G.E., 2012. Sementes: fundamentos científicos e tecnológicos. 3.ed. Editora Universitária/UFPel, Pelotas.

Pires, J.L.F., Costa, J.A., Thomas, A.L., Maehler, A.R., 2000. Efeito de populações e espaçamentos sobre o potencial de rendimento da soja durante a ontogenia. Pesquisa Agropecuária Brasileira, 35(8), 1541-1547. DOI: https://doi.org/10.1590/S0100-204X2000000800006

Rossi, R.F., Cavariani, C., França-Neto, J.B., 2017. Vigor de sementes, população de plantas e desempenho agronômico de soja. Revista de Ciências Agrárias. 60(3), 215-222. DOI: https://doi.org/10.4322/rca.2239

Silva, R.R.. Macedo, W.R., 2022. Increasing soybean productivity by plant equidistant arrangements and modification of technology levels of cropping systems. Revista de Ciências Agroveterinárias. 21 (3): 182-195. DOI: https://doi.org/10.5965/223811712132022182

Souza, D.M.G., Lobato, E., 2004. Cerrado: correção do solo e adubação. Embrapa Cerrados, Planaltina.

Zhang, W., Mao, P., Li, Y., Wang, M., Xia, F., Wang, H., 2017. Assessing of the contributions of pod photosynthesis to carbon acquisition of seed in alfalfa (*Medicago sativa* L.). Scientific Reports, 7, e42026. DOI: https://doi.org/10.1038/srep42026