

## Increasing soybean productivity by plant equidistant arrangements and modification of technology levels of cropping systems

*Incrementos produtivos na cultura da soja pelo uso de arranjos equidistantes e níveis tecnológicos nos sistemas de cultivo*

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### ABSTRACT

The arrangement of soybean plants defines their ability to recognize the environment and improve their abiotic and biotic interactions with it. This study aimed to evaluate the effect of planting arrangements associated to two distinct crop systems (high technological level or conventional crop level) for the soybean varieties 8473 RSF and AS 3730, to evaluate the plant performance and productivity in the field. A double factorial scheme was used with two varieties and six spatial arrangements. Two experiments were conducted during the 2017–2018 crop season: the first was based on standard cultivation of the Brazilian Cerrado (without irrigation systems and adequate fertilizer), whereas the second utilized high technology (irrigation systems and increased fertilizer). The morphophysiological parameters, normalized difference vegetation index (NDVI), leaf pigment content and crop yield parameters were evaluated. Our study pointed out the positive responses for both cultivated varieties and two technological levels adopted cultivated under equidistant arrangement and high density. The leaf area index, leaf pigment content, NDVI, and crop production showed substantial responses under different equidistant arrangements. Therefore, this procedure requires adjustments in the level of cultivation technology and identification of the most suitable soybean variety.

**KEYWORDS:** *Glycine max* L.; plant arrangement; technology; normalized difference vegetation index; crop production.

### RESUMO

O arranjo das plantas de soja define sua habilidade em reconhecer o ambiente e aprimorar suas interações bióticas e abióticas com este. Este estudo objetivou avaliar o efeito de diferentes arranjos de plantio associados com dois distintos sistemas de cultivo (alto nível tecnológico ou nível tecnológico convencional), nas variedades 8473 RSF e AS 3730, para avaliar a performance e produtividades das plantas à campo. O esquema fatorial duplo foi adotado, sendo: duas variedades e seis arranjos espaciais. Dois experimentos foram conduzidos durante o ano agrícola 2017-2018; no primeiro experimento considerou-se o padrão de cultivo da soja no Cerrado Brasileiro (sem sistema de irrigação a nível adequado de adubação), enquanto no segundo experimento foi adotado alto nível tecnológico (com sistema de irrigação e adubação superior aos padrões convencionados para o Cerrado). Foram avaliados parâmetros morfofisiológicos, índice de vegetação por diferença normalizada (IVDN), conteúdo de pigmentos foliares e parâmetros de produção da cultura. Nosso estudo apontou respostas positivas de ambos genótipos cultivados e sob os dois níveis tecnológicos sob os arranjos equidistantes e submetidos a alta densidade de plantio. Significativas respostas foram observadas para índice de área foliar, conteúdo de pigmentos foliares, IVDN e produção de soja, quando submetida aos diferentes arranjos equidistantes. Entretanto, este procedimento requer ajustes dentro dos níveis tecnológicos e identificação das variedades mais adequada a este modelo de cultivo.

**PALAVRAS-CHAVE:** *Glycine max* L.; arranjos espaciais; tecnologia; índice de vegetação por diferença normalizada; produção de cultivos.

## INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) crop has socioeconomic importance due to its magnitude of cultivated area (~122.69 million hectares) and production (339.7 million metric tons) during the 2019/2020 crop season (USDA 2021). It also shows wide adaptations in different geographical areas because of its characteristics of high phenotypic plasticity (BALBINOT JUNIOR et al. 2018), that is, it presents considerable distinctions in its morphological characteristics due to environmental and management factors. Thus, it is essential to understand the factors that provide better development conditions for soybean crops, allowing the achievement of its maximum productive potential.

From the beginning of the 20th century until present, studies on soybean arrangements have substantially demonstrated the changes in plant plasticity and crop production (AGUDAMU & SHIRAIWA 2016, KUMAGAI & TAKAHASHI 2020, RAHMAN & HOSSAIN 2011, SHIBLES & WEBER 1966, WERNER et al. 2021, WIGGANS 1939), as well as features directly linked to light interception efficiency (BOARD 2000) and radiation use efficiency (RUE) (PURCELL et al. 2002). In this sense, one of the strategies that has been studied, aimed at better usage of light radiation, is the use of different plant populations combined with different spatial arrangements (DUARTE et al. 2016, PURCELL et al. 2002).

To build a suitable environment for soybean cultivation with optimized light interception, several patterns of spatial distribution of plants must be considered, emphasizing the cultivation model that amplifies the interception of sunlight (SHIBLES & WEBER 1966). This model that can be achieved by a more homogeneous and equidistant distribution of plants (SHIBLES & WEBER 1965); thus, allowing a better expression of their physiological potential (EGLI 1994).

In the last decades, adjustments in the distribution of modern soybean genotypes prove that increasing plant density, that is, less spacing between crop rows or number of plants in the line, promote significant gains in crop yield (RAMBO et al. 2003), and these adjustments in the cultivation environment reflect physiological responses such as: greater homogeneity in the number of plants, increased RUE and leaf area index (LAI), and increments of dry matter (DM), which culminate in greater productivity of grains (ZHOU et al. 2011). However, returning to the works from the middle of the last century, researchers have already evaluated the impact of equidistant adjustments on soybean cultivation, with strong indications that soybean plants are able to adapt to intra- and interspecific competitions (BEUERLEIN et al. 1971, WILCOX 1974).

The soybean growth, yield and yield components under equidistant plant arrangement are variable according to genotype and geographical localization, nonetheless it is possible to consider that modern varieties of soybean can reach the higher productive index when subjected to high plant density, between 80 and 100 plants m<sup>-1</sup> (RAHMAN & HOSSAIN 2011).

For many years, sowing in an equidistant arrangement in a large-scale model was considered unattainable. However, the evolution of technologies applied to agribusiness, e.g., tractors equipped with GPS-assisted steering and high-precision seeders, allow the application of this cultivation model. This study aimed to evaluate different equidistant arrangements on two soybean varieties (Brasmax 8473 RSF and Agroeste 3730 IPRO) grown under two systems: high technology (HT), simulating conditions of high-productive potential, and conventional cultivation (CC), simulating crop conditions common in areas of the Brazilian cerrado, a Neotropical Savanna biome.

## MATERIAL AND METHODS

### Cultivation conditions and materials under analysis

The experiment was carried out throughout the 2017-2018 growing season in two field assays, HT and CC, in the Alto Paraíso Farm, municipality of Patos de Minas, MG, Brazil, during the 2017–2018 crop season under the cerrado biome (Fig. S1), considered adequate for soybean cultivation. Two varieties were chosen in this research: Brasmax 8473 RSF (Desafio RR) and Agroeste – AS 3730 IPRO, both belong to the maturity group: 7.4. They present similar phenological cycle, indeterminate growth habit, potential for lateral branching, resistance to lodging, and recommended optimal population when submitted to conventional planting (50 cm between rows). For both tests, HT and CC, each variety was subjected to five different arrangements, as follows: T1= 40,000 plants (arrangement equal to 50 x 50 cm); T2= 240,000 plants (arrangement equal to 20 x 20 cm); T3= 440,000 plants (arrangement equal to 15 x 15 cm); T4= 640,000 plants (arrangement equal to 12.5 x 12.5 cm); T5= 840,000 (arrangement equal to 10.9 x 10.9 cm); and compared with two controls plant arrangements T6= 440,000 or T6= 240,000 plants, equal to 50 x 4.5 cm

for 8473 RSF and 50 x 8.3 cm for 3730, respectively. The experimental plots had dimensions of 3 × 3 m, with a strip of 0.5 m as a border area and a useful area of 2 × 2 m (Figure 1).

Regarding irrigation, only HT area was considered with this procedure, whereas the CC area was simulated with the conventional conditions of cultivation adopted under rainfed cultivation in the Cerrado. A center-pivot irrigation system was employed, and the irrigation frequency was determined from a mini meteorological station installed inside the farm to meet the water demands of the crop.

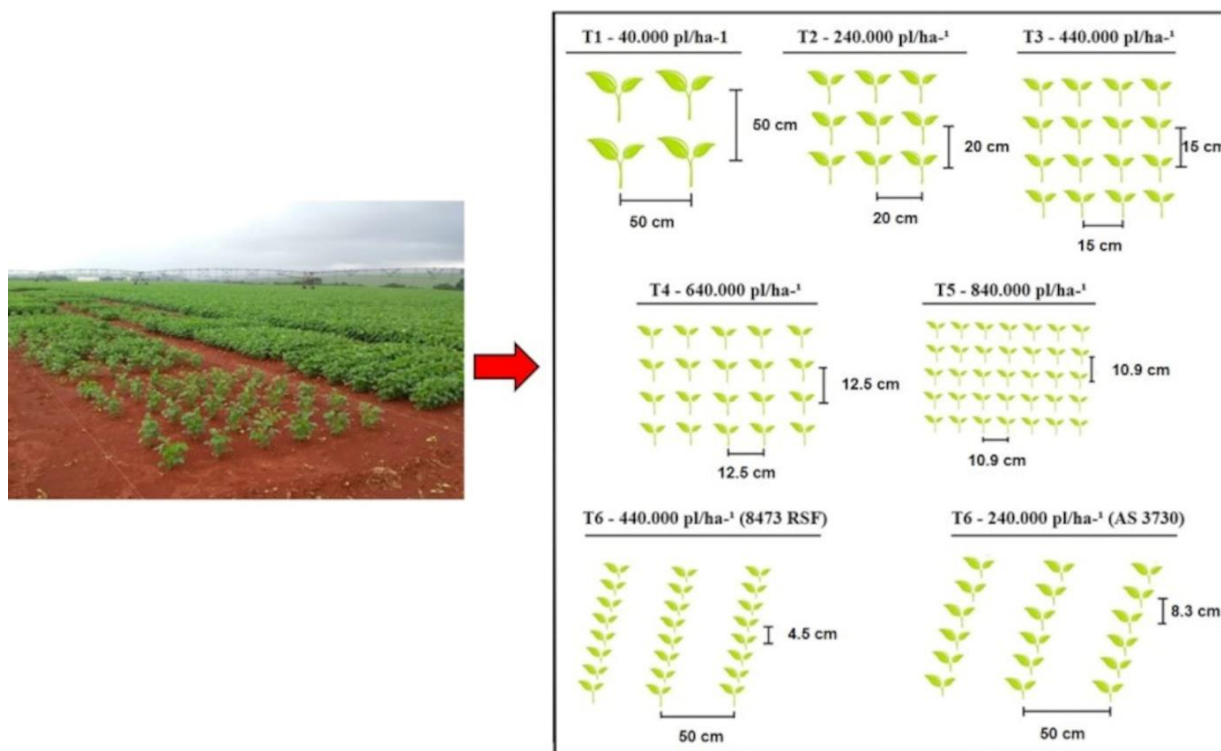


Figure 1. Experimental design in the field under 2017-2018 crop session. Patos de Minas, MG, Brazil.

### Soil preparation, fertilization, and planting

The soil of the experimental area was classified as clay loam oxisol (SOIL SURVEY STAFF 2014), and the fertility attributes evaluated in the upper layer of soil, at 0–20 cm depth, is available in Table S1. The soil of the experimental area was prepared with the aid of a medium harrow and, later, a leveling disk harrow, and manually with a rake, to remove small clods and vegetable residues (Fig. S2 A and B). For both HT and CC cultivation systems, fertilizer recommendation was considered based on the chemical analyses of each area. Doses higher than that recommended for each source of fertilizer were applied to HT system to ensure that nutrition is not a limiting factor for the crop development, whereas the recommended quantity of each source of fertilizer was applied in the CC system. This criterion for defining the dose was based on the estimated productivity estimated by area, 7.2 and 6 tons of grains per ha<sup>-1</sup> for HC and CC, respectively.

Thus, after soil preparation and before planting, fertilization with potassium chloride (60% K<sub>2</sub>O) was carried out manually in a haul, in each plot individually, without incorporating the fertilizer into the soil. In the HT system, 170 kg of KCl per ha<sup>-1</sup> was applied, whereas in the CC system, 106 kg ha<sup>-1</sup> of KCl was applied as 80% fertilizer efficiency was necessary (SFREDO 2008). After opening the furrows, 1000 kg ha<sup>-1</sup> and 590 kg per ha<sup>-1</sup> of a simple superphosphate fertilizer was applied manually in the HT and CC systems, respectively (SOUSA & LOBATO 2003).

Before planting, furrows were made along the plots with the aid of a mini furrow with a plot width of 3 m, where adjustments were made to the distance between the stems to adapt the distance between furrows in accordance with the treatment to be applied. The seeds used were previously inoculated with *Bradyrhizobium japonicum* and sowing was carried out from 23 to 26 October 2017 in HT plot, and from 14 to 16 November 2017 in CC.

For planting, PVC pipes (diameter 3/4"), with holes aligned along its length (1.5 m), with the distance between the holes varying according to the distances of the different treatments (10.9 to 50 cm) (Fig. S2 C and D). Thus, with the aid of an adapted vacuum cleaner, the seeds were sucked along the pipe by the

vacuum generated in the holes, and deposited along the furrows and covered with soil.

### Phytosanitary procedures

In the HT system, phytosanitary control was more rigorous with increased pesticide application and control of invasive plants in the phenological stages V1 (first node) and V4 (fourth node) (FEHR & CAVINESS 1977) with a glyphosate-based product (1.2 kg ha<sup>-1</sup> of commercial product), whereas in the CC system, invasive plant control was performed only at the V3 stage with a glyphosate-based product (1.68 kg ha<sup>-1</sup> of commercial product). In HT, together with the second application of glyphosate (V4), and in CC, coupling with application of glyphosate (V3), insecticide applications based on profenofos + lufenuron were carried out (350 ml ha<sup>-1</sup> of commercial product). In addition to the HT system, fungicide was also applied (500 ml ha<sup>-1</sup> of azoxystrobin + difenoconazole of commercial product).

In the reproductive phase in both experiments, fungicide was applied thrice. In R2 (full bloom) and R4 (ful pod) (FEHR & CAVINESS 1977), azoxystrobin + benzovindiflupir was applied (0.2 kg ha<sup>-1</sup> of commercial product), besides mancozeb 750 (1.5 kg ha<sup>-1</sup> of commercial product), and lastly R5 (FEHR & CAVINESS 1977) was applied with products peobased on azoxystrobin 500 (0.12 kg ha<sup>-1</sup> of commercial product) and propiconazole + difenoconazole (0.15 l ha<sup>-1</sup> of commercial product). In addition, insecticide applications were also carried out, with thiamethoxan + clorantpraniliprole (200 ml ha<sup>-1</sup> of commercial product) in the R4 phase and thiametoxan + lambda-cyhalothrin (350 ml ha<sup>-1</sup> of commercial product) in the R5 stage (FEHR & CAVINESS 1977).

Before the harvest, small differences were observed between the treatments in relation to leaves' senescence and grain moisture content. For this reason, a pre-harvest desiccant was applied at stage R8 (2 L ha<sup>-1</sup> of paraquat), 6 days before the harvest, to obtain more homogeneous plants at the time of harvest.

### Normalized difference vegetation index (NDVI)

For NDVI, two analyses were conducted in the CC experiment (V5 and R5 phenological stages) and one in the HT experiment (R5 phenological stage). These images were obtained by drone remote sensing using a multispectral sensor (Parrot, Sequoia), mounted on a Phantom 3 drone (DJI®). Through this evaluation, it was also possible to obtain the NDVI through the relationship between the reflectance of the red wavelength (R) and near infrared wavelength (IR) spectra. The following model was used to calculate this index (ROUSE et al. 1974) (Equation 1):

$$NDVI = (IR - R) \div (IR + R) \quad (1)$$

Where, the IR range comprises the wavelength between 730 to 740 nm and R is from 640 to 680 nm.

### Plant biometric analysis

In stage R5, the evaluation of plant heights (cm), and leaf area (cm<sup>2</sup>) per plant was conducted. One plant per plot was collected for the last variable, which underwent manual and total defoliation. Subsequently, the leaflets were spreaded on a flat surface, with a background color contrasting the green of the leaves, together with a metric scale. Thereafter, images were obtained using a conventional RGB digital camera and subjected to processing by the free software ImageJ (SCHNEIDER et al. 2012), which through isolation of the colors of the leaflets and dimensioning of the image components calculated the leaf area from the images of each plot. After obtaining the leaf area values (cm<sup>2</sup>), LAIs were calculated using Equation 2:

$$LAI = PLA \div SA \quad (2)$$

Where, PLA represents the plant leaf area and SA represents the soil area, typically reported as square meters per square meter (NGUY-ROBERTSON et al. 2012).

Only for the experiment under HT conditions, leaves samples were collected and stored in ice contained in Styrofoam, for further analysis of photosynthetic pigments (chlorophyll a, b, total chlorophyll, and carotenoids), the solvent extraction methodology (80% acetone) was used without macerating the plant tissues (MACEDO et al. 2013). The reading was taken using a UV-Vis spectrophotometer (Perkin-Elmer, Lambda 25), at wavelengths 645, 652, and 663 nm for chlorophyll a, b, and total chlorophyll, respectively (WITHAM et al. 1971), and at 470 nm for carotenoids (LICHTENTHALER & WELLBURN 1983). The results were expressed in milligrams of pigment per gram fresh weight of leaf tissue (mg g<sup>-1</sup>).

In the plant harvest (stage R9), the lodging of plants was evaluated following the scale proposed by BERNARD et al. (1965), where: 1 - almost all plants are erect; 2 - all plants are leaning slightly or a few plants are down; 3 - all plants are leaning moderately or 25% to 50% of the plants are down; 4 - all plants are leaning considerably or 50% to 80% of the plants down; and 5 - almost all plants are down. Next, the plants were harvested for production analysis: height of insertion of the first pod (cm), total number of pods per plant, quantity of grains per plant, and quantity of grains per pod.

## Yield analysis

To obtain productivity per plant, as many plants as possible were collected from within the useful area of each plot, with separation and collection of grains, and subsequent peining, to minimize the amount of waste. Next, grain mass was weighed individually, and its moisture percentage was analyzed using portable grain moisture meter model AL-102 (Agrologic). Thus, we proceeded with the calculations for moisture discount to obtain a standardized dry weight at 14% humidity. Thereafter, the calculations for thousand grain weight were performed by counting the grains and weighing on a digital scale.

## Statistical analysis

For both experiments, a randomized block design with four replications in a  $2 \times 5 + 2$  factorial scheme, where: two varieties; five different arrangements; and two treatments control (8473 RSF with arrangement equal to  $50 \times 4.5$  cm, and AS 3730 with arrangement equal to  $50 \times 8.3$  cm) were applied. The physiological and biometric parameters had their means submitted to analysis of variance and were subsequently compared by the SNK test at 95% level of significance, and orthogonal contrast (Holm-Bonferroni test) was applied to compare each special arrangement with control, of respective varieties (8473 RSF or AS 3730), with aid the SpeedStat Spreadsheet 2.6 Program (CARVALHO et al. 2020). The parameters of individual plant production and productivity were subjected to multivariate analysis through principal component analysis (PCA), using the PAST software 4.06 (HAMMER et al. 2001).

## RESULTS

For the CC experiment, the effect of interaction between varieties and spatial arrangements was observed for the variables: LAI, NDVI (in stage R5), plant height, chlorophyll a, chlorophyll b; total chlorophyll and carotenoids. For NDVI (in stage V5) was observed significance isolated for variety and plant arrangement, and for plant lodging (PL) index only the varieties showed significant effect (Table S2). The HT experiment showed a significant interaction effect on the variables LAI and PL, whereas the variable NDVI (R5) was influenced only by the experimental arrangement, and PH showed isolated effects for varieties and plant arrangements (Table S3).

### LAI under different crop arrangements

Significant interaction of factors was observed in both cultivation environments (HT and CC), where plants responded to the number of plants per hectare and to the genotype, so we observed that the population of 840,000 plants had the highest LAI. The variety AS 3730 showed the highest LAI means when compared with the variety 8473 RSF, under both crop conditions (HT and CC), unless arranged under low plant density, under 40,000 plants per hectare (Figure 2). In the analysis of orthogonal contrasts, it was possible to verify that the control treatment of each genotype, for HT conditions, only 840,000 plants showed significant differences for AS3730, and for CC environment, the AS3730 increased the leaf area index by 440,000; 640,000 and 840,000 plants per hectare, when compared to control plants (Figure 2).

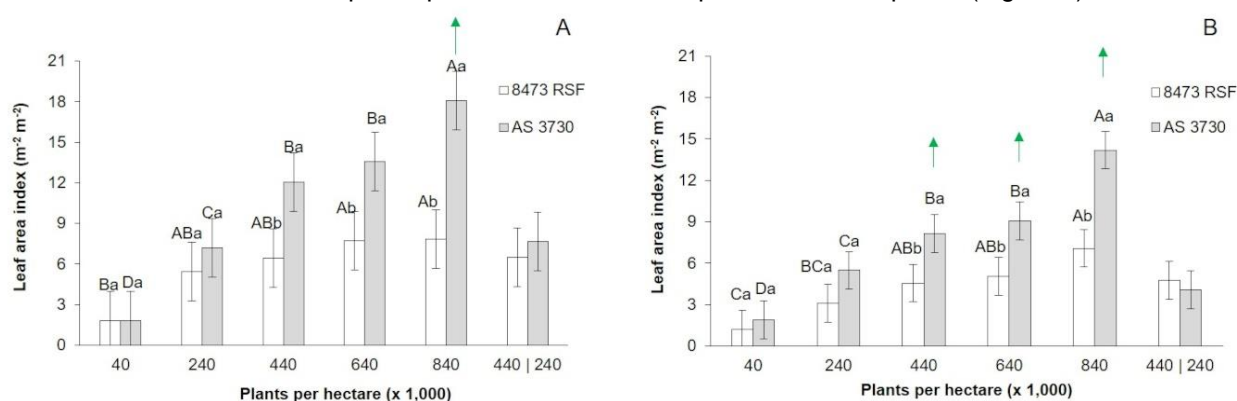


Figure 2. Leaf area index (LAI) in HT (A) and CC (B) systems. Where: 40,000 plants (arrangement equal to  $50 \times 50$  cm); 240,000 plants (arrangement equal to  $20 \times 20$  cm); 440,000 plants (arrangement equal to  $15 \times 15$  cm); 640,000 plants (arrangement equal to  $12.5 \times 12.5$  cm); 840,000 (arrangement equal to  $10.9 \times 10.9$  cm); and 440,000|240,000 plants (arrangement equal to  $50 \times 4.5$  cm for 8473 RSF and  $50 \times 8.3$  cm for 3730). Capital letters indicate the comparison of each soybean variety among all plant arrangements, and small letters indicate the comparison of soybean varieties inside of each plant arrangement, where different letters show significant differences by SNK test ( $P < 0.05$ ). The arrows indicate significant difference in orthogonal contrast of each plant arrangement against the pattern of plant arrangement (440,000 or 240,000 plants per hectare).

### Visualization of crop status by NDVI

Regarding the analysis of spectral images, we adopted NDVI as an indicator tool for density and vigor. The results demonstrate a positive effect on interaction between plant arrangement and soybean varieties (Figure 3). Major differences were observed in soybean plants in V5 stage under CC environment (Figure 3A), where the variety 8473RSF showed higher value of NDVI, when compared to AS3730 in all plant arrangements of this study. For analysis of orthogonal contrast were verified increase of NDVI means for 440,000; 640,000 and 840,000 equidistant plant arrangements when compared to both control arrangements (8473RSF and AS3730). However, the 8473RSF under 40,000 plants showed lower NDVI, if compared to control arrangement (440,000 plants - 8473RSF).

Increases in the NDVI for high-density equidistant arrangement for CC and HT environments, under R5 phenological stage, were observed. From 240,000 plants until 840,000 plants, no differences were found for varieties 8473RSF and AS3730, only treatment with 40,000 plants showed less means of NDVI. Nonetheless, evaluating the orthogonal contrast in the AS3730 variety, in CC environment and subjected to 440,000; 640,000 and 840,000 equidistant plant arrangements, is verify a slow gain in NDVI, and 40,000 plants of same variety reduced significantly their NDVI means (Figure 3B). Similar behavior was observed in the 8473RSF, subjected to 40,000 and 240,000 plants per hectare (Figure 3B).

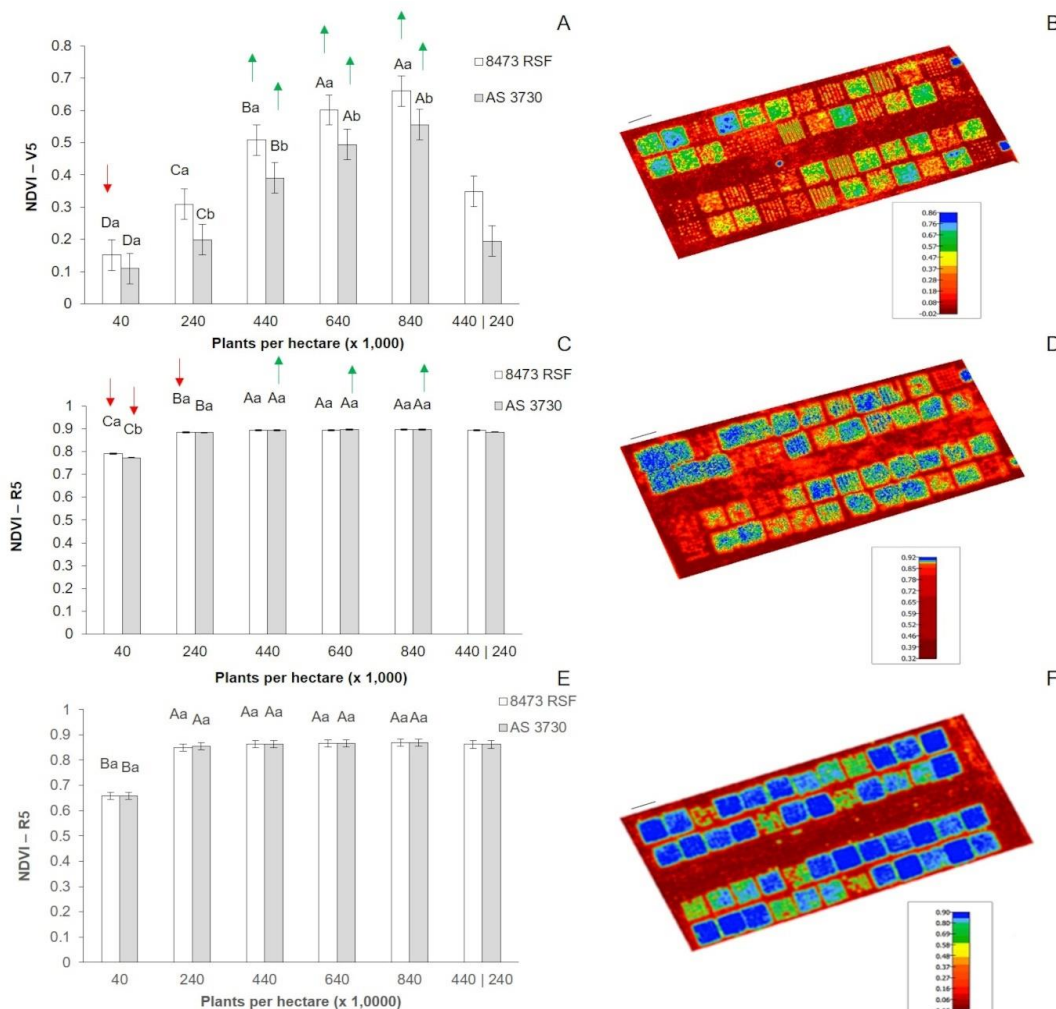


Figure 3. Analysis of NDVI and orthomosaic images (field experimental area) for soybean plants under phenological stages V5 (A, B) and R5 (C, D) for experiment CC and R5 (E, F) for experiment HT, the black bar in orthomosaic image indicate 3 meters of length. Where: 40,000 plants (arrangement equal to 50 x 50 cm); 240,000 plants (arrangement equal to 20 x 20 cm); 440,000 plants (arrangement equal to 15 x 15 cm); 640,000 plants (arrangement equal to 12.5 x 12.5 cm); 840,000 (arrangement equal to 10.9 x 10.9 cm); and 440,000|240,000 plants (arrangement equal to 50 x 4.5 cm for 8473 RSF and 50 x 8.3 cm for 3730). Capital letters indicate the comparison of each soybean variety among all plant arrangements, and small letters indicate the comparison of soybean varieties inside of each plant arrangement, where different letters show significant differences by SNK test ( $P < 0.05$ ). The arrows indicate significant difference in orthogonal contrast of each plant arrangement against the pattern of plant arrangement (440,000 or 240,000 plants per hectare).

### Alterations in the leaf photosynthetic pigments

Significant differences were observed in the interaction of plant arrangements and soybean varieties for contents of leaf pigments, under low densities (40,000 and 240,000 plants per hectare) and high density (840,000 plants per hectare) the variety 8473RSF was superior to the AS 3730 variety (Figure 4). The highest pigment content for 8473RSF was observed under 840,000 plants per hectare, and the highest pigment content for AS3730 was observed under 440,000 plants per hectare. Only AS3730 variety under 440,000 plants per hectare showed significant effect for orthogonal contrast, where chlorophyll b and carotenoids increased their content in control treatment (240,000 plants per hectare) (Figure 4).

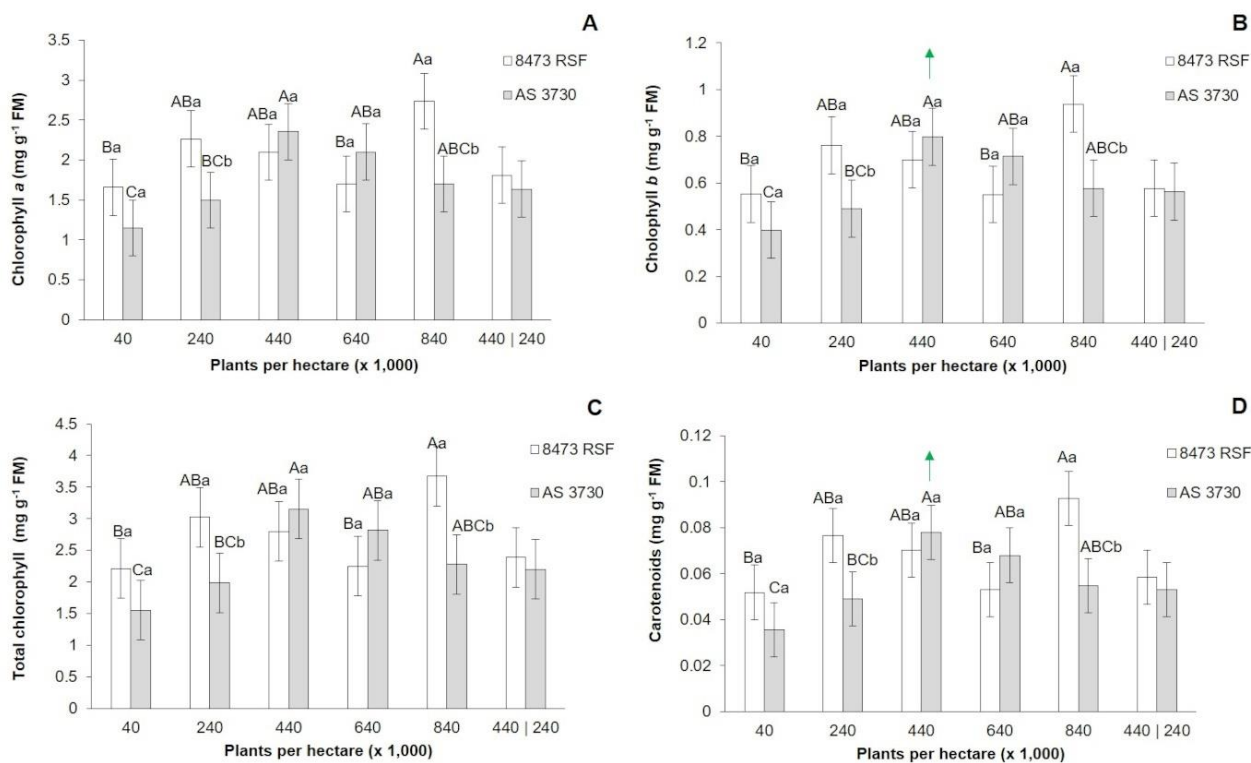


Figure 4. Content of chlorophyll a (A), chlorophyll b (B), total chlorophyll (C), and carotenoids (D) in soybean leaves, grown in an HT cultivation. Where: 40,000 plants (arrangement equal to 50 x 50 cm); 240,000 plants (arrangement equal to 20 x 20 cm); 440,000 plants (arrangement equal to 15 x 15 cm); 640,000 plants (arrangement equal to 12.5 x 12.5 cm); 840,000 (arrangement equal to 10.9 x 10.9 cm); and 440,000|240,000 plants (arrangement equal to 50 x 4.5 cm for 8473 RSF and 50 x 8.3 cm for 3730). Capital letters indicate the comparison of each soybean variety among all plant arrangements, and small letters indicate the comparison of soybean varieties inside of each plant arrangement, where different letters show significant differences by SNK test ( $P < 0.05$ ). The arrows indicate significant difference in orthogonal contrast of each plant arrangement against the pattern of plant arrangement (440,000 or 240,000 plants per hectare).

### Plant growth analysis

Evaluating the data of soybean growth, during R5 phenological stage, was verified a distinct response between the crop environments (HT and CC), plants under irrigation and increase fertilization showed greater growth and consequently higher rates of lodging (Figure 5).

For soybean lodging scale in the HT system, there was a significant interaction between the varieties and the different arrangements, where no significant differences in the lodging of plants between the spatial plant distributions for the variety 8473 RSF were observed. We also observed that the plants of the variety AS 3730 grew more than the plants of the variety 8473 RSF. There was no effect of varieties or spatial plant arrangements for the CC system. In this scenario, the variety 3730 proved to be more suitable for high density, preventing the plants from lodging later.

For further analysis of orthogonal contrast, has been proven that 40,000 plants per hectare in HT, grew less than control arrangement 440,000 and 240,000 plants per hectare, for both varieties, and the variety AS3730 under 440,000 and 640,000 plants per hectare lodging more than the respective control treatment (Figure 5).

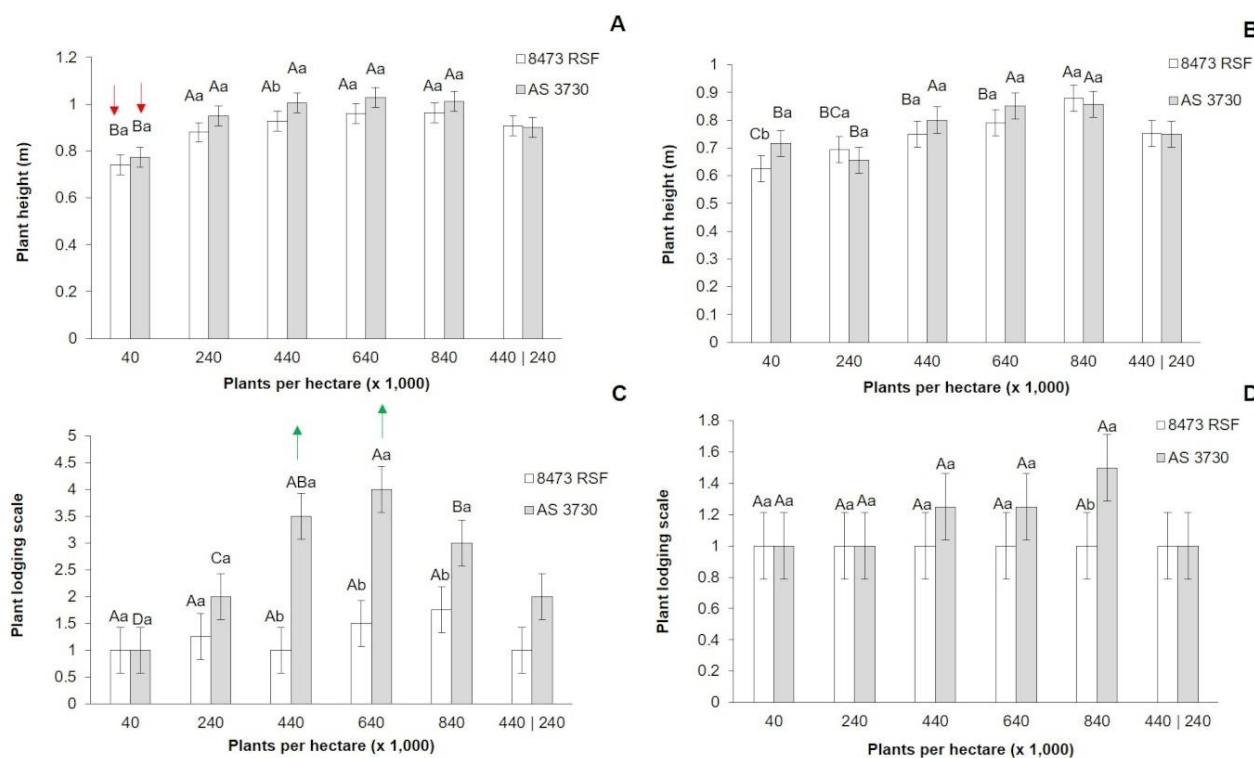


Figure 5. Soybean plant height (cm) in HT systems (A) and CC Systems (B), and lodging scale in HT systems (C) and CC Systems (D). Where: 40,000 plants (arrangement equal to 50 x 50 cm); 240,000 plants (arrangement equal to 20 x 20 cm); 440,000 plants (arrangement equal to 15 x 15 cm); 640,000 plants (arrangement equal to 12.5 x 12.5 cm); 840,000 (arrangement equal to 10.9 x 10.9 cm); and 440,000|240,000 plants (arrangement equal to 50 x 4.5 cm for 8473 RSF and 50 x 8.3 cm for 3730). Capital letters indicate the comparison of each soybean variety among all plant arrangements, and small letters indicate the comparison of soybean varieties inside of each plant arrangement, where different letters show significant differences by SNK test ( $P < 0.05$ ). The arrows indicate significative difference in orthogonal contrast of each plant arrangement against the pattern of plant arrangement (440,000 or 240,000 plants per hectare).

### Production analysis: multivariate and descriptive results

PCA was adopted to evaluate the production indexes to identify crop production patterns for both varieties and in both cultivation systems. It was established that the CC cropping system and HT cropping system could explain 85.84% and 79.57%, respectively, of the total variances observed (Table S4). It was possible to identify the major effect of individual plant production for Principal Components 1 (PC1), and the major effect of plant productivity for Principal Components 2 (PC2), from the loading values observed (Table S5).

For high-level technology (HT), the biplot graph presented the existence of an independent gradient, characterized by positive grouping among productivity ( $\text{kg ha}^{-1}$ ) and high density of crops (10.9 x 10.9 cm; 12.5 x 12.5 cm; 15 x 15 cm) for AS 3730 variety, whereas the crop density for 8473 RSF presented a grouping in the lower left quadrant, and was associated with only the height of the first pod. In contrast, soybean crop with lower density showed high affinity with production parameters per plant (e.g., grains per plant, grains per pod, one grain per pod, two grains per pod, three grains per pod, four grains per pod, and pods per plant) (Figure 6).

As in the PCA model observed for HT, the CC system also presents a pattern very similar to the HT system, where it can be seen that the AS 3730 variety grown under high densities (10.9 x 10.9 cm, 12.5 x 12.5 cm, and 15 x 15 cm) showed a high association with productivity ( $\text{kg ha}^{-1}$ ), and similarly the variety 8473 RSF presented an antagonistic response when compared to the other variety. It was also possible to verify that the less dense plants (50 x 50 cm) of both varieties had a high association with production parameters per plant (Figure 7).



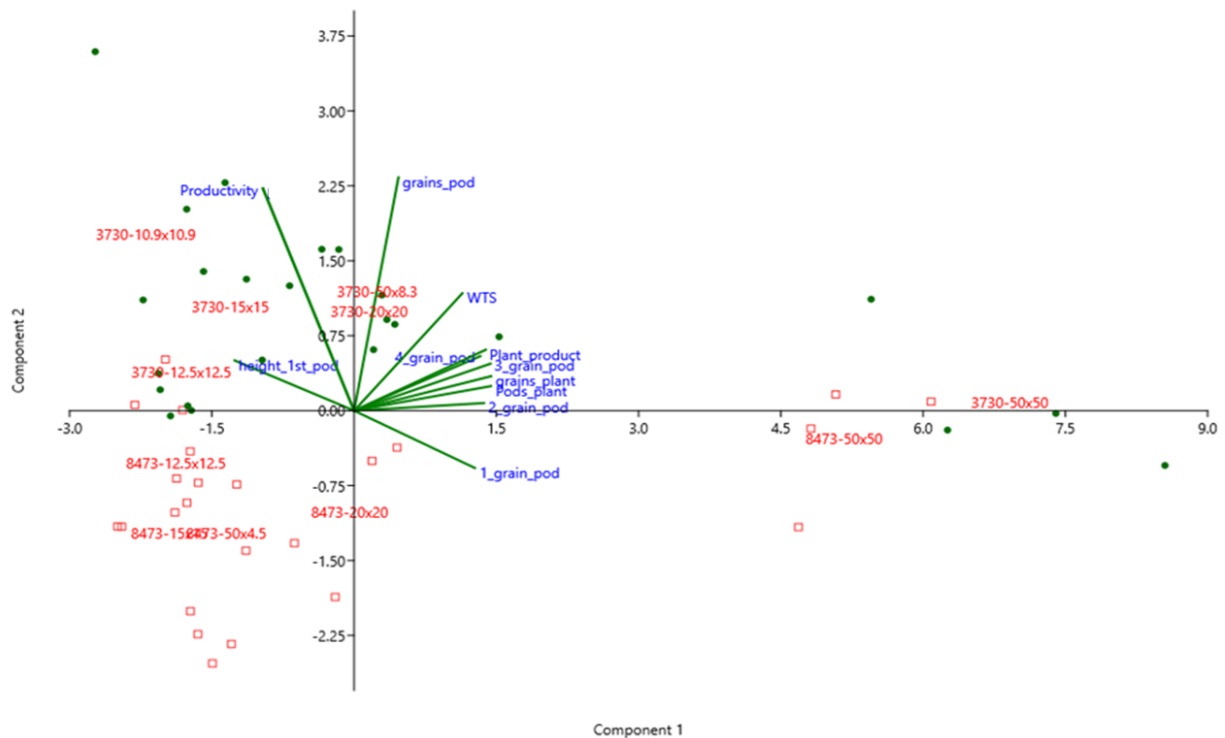


Figure 6. Scatter plot of PCA for the experiment in high-tech cropping (HT), of varieties 8473 RSF (red squares) and AS 3730 (green dots), during 2017-2018 crop session (n=3).

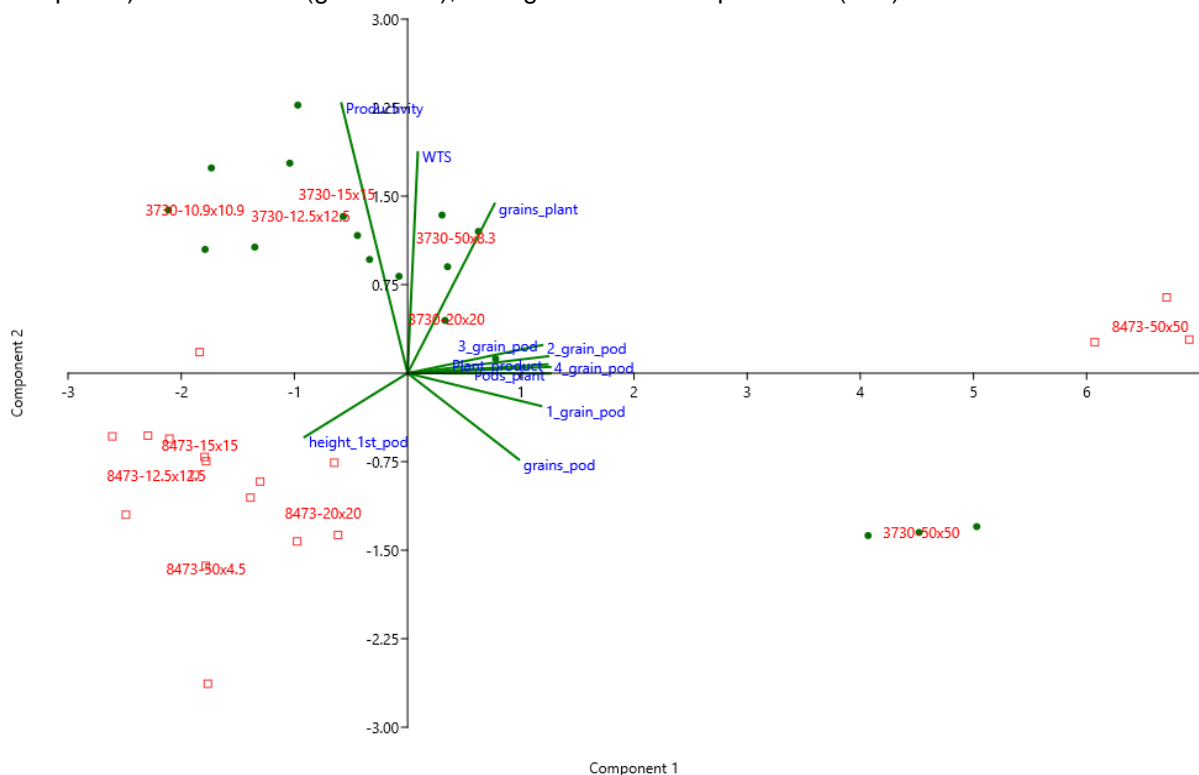


Figure 7. Scatter plot of PCA for the experiment in conventional cropping (CC), of varieties 8473 RSF (red squares) and AS 3730 (green dots), during 2017-2018 crop session (n=4).

Biologically, it was possible to verify the impact of low plant density (50 cm × 50 cm) on individual plant gains through characteristics such as pods per plant, grains per plant, grains per pod, pods with one grain, pods with two grains, pods with three grains, pods with four grains, weight of total seeds (WTS, g), and individual plant production (g) for both varieties. Similar results were found by HERMANN DERETTI et al. (2022), where reduction on plant density increased the number of pods on the branches per plant. However, the productivity analysis showed that low plant density exerts a negative impact on the field, such that the gains from production would hardly compensate for the costs of planting. For variety 8473 RSF, a reduction

of 55.64% in soybean productivity, compared to equidistant low-density seeds (T1) under conventional row seed distribution treatment (T6), was observed for the treatments with the same plant population, whereas in T3 (equidistant seed distribution) and T6 (row seed distribution), reduction in productivity by 21.91% was observed. It was possible to obtain 12.08% gain in productivity in the equidistant treatment with high-density seeds (T5). This scenario changed when variety AS 3730 was studied, where only low plant density treatment (50 cm × 50 cm) showed reduction in productivity (61.99%), whereas the treatments with same plant population (T2) and equidistant high density (T5) presented an increase by 1.91% and 21.73%, respectively.

## DISCUSSION

To maintain agricultural sustainability, it is necessary to develop new crop cultivation systems, especially those classified as commodities. In this case, soybean can be considered essential for human and animal nutrition. In view of the above, studies that contemplate gains in crops' physiological and productive performance are essential. Therefore, studies on plant-environment interactions, such as analysis of plant arrangements, promote new fronts in sustainability research. Thus, we consider that a better understanding of the phenotypic plasticity of soybean plants cultivated under equidistant arrangements or under row cultivation (standard cultivation model) becomes a tool for the establishment of new criteria for soybean cultivation.

Since in the current model of soybean cultivation, the distribution of seeds in rows allows spacing between the rows up to eight times greater than the distance between the plants (usually between 6 and 10 cm) in (sub)tropical conditions, revealing a very heterogeneous distribution relationship, which, for certain varieties, may not allow the expression of its maximum productive potential. This fact should not be attributed only to the physiological and environmental needs of the crop but primarily to the great need for gains in production.

### Relationship between LAI, cropping arrangement, and varieties

Among the parameters studied, we verified that LAI has substantial relevance in cropping science because it provides information regarding the development of the crop, with a typical pattern of delay in the beginning of the cultivation followed by a rapid increase until reaching a maximum value, and subsequent reduction in the physiological maturity of the plants (SETIYONO et al. 2008). The highest LAIs were observed in crops with larger populations and smaller spacing between rows (HEIFFIG et al. 2006). Our results show that the equidistant arrangement with the highest density of plants enable a quick coverage of the soil surface, and according to HEIFFIG et al. (2006), it allowed the critical LAI, when the canopy intercepted 95% of the light radiation at noon, to be obtained in advance by the plants under this treatment, for both varieties (Figure 2).

This study demonstrates that in the HT condition, only the spatial arrangement of 50 cm × 50 cm (40,000 plants per hectare) did not provide LAI above the critical level, similar to the values described by HOLSHOUSER & WHITTAKER (2002), i.e., between 3.5 e 4 for soybean crop, regardless of the variety studied. In the CC experiment, a behavior similar to the HT system is found, where the varieties AS 3730 and 8473 RSF under equidistant arrangement of 50 × 50 cm presented values in a range considered minimum critical LAI (Figure 2), demonstrating that the increase in plant density can benefit the percentage of solar radiation interception and the rate of plant DM accumulation, as proposed by (SHIBLES & WEBER 1965).

The high-density arrangement of AS 3730 variety showed greater phenotypic plasticity in relation to the 8473 RSF variety. While the AS 3730, from 240,000 plants, did not present gains in the average leaf area index for the cultivation environments under HT and CC.

### Spectral analysis by NDVI interpretation and explanation of leaf pigments content

NDVI allows to obtain the vegetation reflectance in the visible range (400–700 nm). This parameter is derived from tissue pigments, whereas the NIR range (700–1300 nm) is determined by cell structure and tissue biomass (TUCKER 1979). Therefore, this parameter is associated with plant photosynthetically-active pigments and plant biomass and is indicative of growth and adaptability to biotic or abiotic stresses in cultivation (SYTAR et al. 2016). It is expressed by the range of values between -1 and 1, with comparison characteristics, and not by interpreting its absolute value (SOTILLE et al. 2020). In addition to having a high correlation with LAI (NGUY-ROBERTSON et al. 2012) and leaf pigments (TUCKER 1979), parameters that corroborate the results of this research.

Based on this interpretative analysis, we consider that the results of this research reinforce the theory concerning the increase in plant density and the gain in LAI indices (Figure 3). In phenological stage V5 under CC environment, the plants presented greater difficulty to establish better soil coverage naturally, when reaching the reproductive stage R5 in the HT and CC systems, all equidistant arrangements and the pattern cultivation in rows (control treatments) reached the highest limits of coverage of the soil surface, except for 40,000 plants per hectare (lower population density), in which the soil coverage by the plants was not properly constituted (Figure 3).

NDVI presents responses influenced by the growth of canopy of the plant (where there is overlapping of leaves) and by the amount of photosynthetic pigments, and consequently by the absorption of light and the reduction of reflectance (PONZONI et al. 2012); thus, proving the relationship between NDVI and leaf pigment content. Specifically, for photosynthetic pigments, we consider that the photon flow itself, more homogeneous over the canopy in equidistant plants, is a better signal of pigment biosynthesis (YUAN et al. 2017), as shown in the treatment of 840,000 plants per hectare of variety 8473 RSF (Figure 4).

Upon careful observation of the content of chlorophylls and carotenoids in soybean leaves, under the HT cultivation system, it was noted that the varieties strongly influence these parameters, where 8473 RSF presented higher amounts of chlorophylls and carotenoids in most of its tissues, than in AS 3730. Furthermore, the arrangement of plants also played an important role in the biosynthesis of these compounds, where the variety 8473 RSF showed a linear growth when subjected to equidistant planting, significantly superior to cultivation in rows, whereas the variety AS 3730 had its highest concentration in the not quite dense equidistant treatments (Figure 3).

#### **Soybean biometric analysis and lodging scale**

The results of this research show that the different cultivation arrangements were preponderant for obtaining different phenotypes, where the equidistant arrangement of 50 × 50 cm (40,000 plants per hectare) for both varieties and CC production systems did not show expressiveness in the growth of the plants, as well as in the lodging scale, while for HT crop environment and AS 3730 and 8473 RSF varieties showed minor growth when compared to control treatments. An opposite response was observed in treatments with high-density equidistant arrangements, showing linear plant height increments for both varieties and production systems (HT and CC).

It can be considered that the greater plant heights correlate positively to higher rates of lodging and plant growth for determinate soybean, process defined primarily during the vegetative growth (BOARD 2001). In this study, specifically for the HT crop system, the variety AS3730 subjected to 840,000 plants per hectare did not present a high plant lodging rate, unlike observed in the CC crop system where high-density arrangement culminated in greater plant lodging.

The occupation of physical space by plants in a less dense arrangement allowed for the best distribution of solar radiation on the canopy of the plant, not favoring interspecific competition between the plants, in contrast to this growth model, the dense arrangements led to the etiolation of the stem of the plants, and resulted in a progressive index of plant lodging, primarily for the variety AS 3730 (Figure 5C and 5D). Soybean lodging is a factor that is closely related to this spatial distribution of plants, and can negatively affect soybean productivity (BOARD 2001). Considering the spatial arrangements mentioned, the reduced spacing of lines shows a better relationship between the distance of plants in the row and in the line. However, this relationship may still be unsatisfactory, depending on the plant population used.

The AS 3730 variety is more sensitive to lodging, which showed no significant difference in the higher population (840,000 plants ha<sup>-1</sup>) compared to smaller populations (640,000 and 440,000 plants ha<sup>-1</sup>), proving that the genetic factor is essential in choose varieties for cultivation in high-density model.

#### **PCA and descriptive analyses of soybean production parameters**

The analysis of PCA seeks to recognize patterns of clusters or dispersion of the samples under analysis (LYRA et al. 2010). In this research, we adopted PCA to perform the evaluation of two soybean varieties grown under six different spatial arrangements and in systems of HT and CC soy cultivation. For the purpose of a holistic interpretation of the impact of these treatments, the following were evaluated: height of first pod, pods per plant, grains per plant, grains per pod, number of pods with one grain, number of pods with two grains, number of pods with three grains, number of pods with four grains, weight of total seeds (WTS), production per plant, and productivity. These 11 parameters evaluated are directly related to the soybean yield indexes.

The components of PCA can explain the variation in the whole dataset in a certain sense, and the technique is able to identify some phenomena (BRO & SMILDE 2014). Thus, this multivariate analysis

technique is suitable for identifying the variability of productive indexes between different factors, such as: varieties, cultivation system, and agricultural practices, as already studied for wheat culture (FERREIRA et al. 2019), maize (BHUSAL et al. 2016), and oat (UARROTA et al. 2017).

In both cultivation systems, HT and CC, the responses of productive characters are intimately linked to the variety and the arrangement of plants at the environment of cultivation. The traits of soybean plants make it able to compensate the yield components in the plant individually, causing reduction in its population level, which in many cases do not generate differences in production by area (BÜCHLING et al. 2017). Via PCA, the areas where the best-adjusted clusters linked to the lower density arrangements had better production rates per plant were identified, whereas the more densely packed arrangements were characterized as having better productivity rates, considering production by cultivation area (Figure 6 and 7).

It is important to point out that the population density of soybean influences the whole phenotype of soybean plants. The characteristic height of insertion of the first pod of soybean can influence the efficiency of mechanized harvesting, because the insertion of the first pod above 10 cm provides better performance and reduces losses during mechanical harvest (MOTOMIYA et al. 2017).

The results of this work reveal a decrease in the number of pods as the population increases for all varieties and in the two cultivation systems evaluated. However, under TA conditions, a stabilization in the reduction of pods per plant was found in a population of 440,000 plants ha<sup>-1</sup> (T3, T4, and T5), demonstrating a potential for gains in productivity under treatments with higher density, corroborating with (RAMBO et al. 2003), who claimed that the number of pods per plant is one of the main components of production in soybean culture. We also consider that the highest number of pods per plant for the variety AS 3730 was possibly obtained due to the higher LAI values observed for this variety. Another relevant fact refers to the claim that the number of grains per pod and the variation in the plant population are not much related (KUSS et al. 2008), a statement that contradicts our research because we found that the lower density of plants allowed significant gains in the increase of grains per pod.

Grain weight is another characteristic strongly related to the productive performance of soybean crops (DE LUCA & HUNGRÍA 2014), and in modern soybean varieties, this larger grain formation is linked to a small characteristic influenced by the crop environment but with a strong action of genetic factors (MAUAD et al. 2010). Again, it was observed that low plant density (50 × 50 cm), for both cultivation systems, the strongest tendency to produce larger grains.

However, when the population density of soybean increases, productivity per plant stabilized as it remained balanced when the plant population increased from 640,000 plants ha<sup>-1</sup> to 840,000 plants ha<sup>-1</sup>. Thus, at these higher levels of population, it is expected that the increase in productivity per area becomes more expressive due to the gain in the number of plants per cultivated area, under a smaller spacing.

## CONCLUSION

The equidistant arrangement with lower population density (50 × 50 cm) for both varieties, better reflected in the individual plant productive indexes, reinforcing the effect of soybean phenotypic plasticity in adapting under low-density cultivation conditions, allowing the best use of natural resources (light, water, and nutrients) by these plants. In contrast, the high cultivation density contributed to increased productivity when the number of grains per area was analyzed. However, management of these plants needs to be better understood for the effective application of this technique, as the results are highly dependent on the varieties and technologies adopted.

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