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PLANT ABIOTIC STRESS TOLERANCE



Pantanal Editora

2020

Fábio Steiner
(Organizador)

PLANT ABIOTIC STRESS TOLERANCE



Pantanal Editora

2020

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Rua Abaete, 83, Sala B, Centro. CEP: 78690-000.
Nova Xavantina – Mato Grosso – Brasil.
Telefone (66) 99682-4165 (Whatsapp).
<https://www.editorapantanal.com.br>
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APRESENTAÇÃO

A obra “Plant Abiotic Stress Tolerance”, uma publicação da Pantanal Editora, apresenta, em seus 9 capítulos, uma ampla gama de assuntos sobre os recentes avanços e conhecimentos científicos nas áreas de ecofisiologia da produção vegetal e conservação dos recursos naturais e meio ambiente. Os temas abordados mostram algumas das ferramentas atuais que permitem o incremento da produção de alimentos, a melhoria da qualidade de vida da população, e a preservação e a sustentabilidade dos recursos disponíveis no planeta. A obra, vem a materializar o anseio da Editora Pantanal na divulgação de resultados e conhecimentos, que contribuem de modo direto no desenvolvimento humano.

Nas últimas décadas, a produção de alimentos tem sido frequentemente limitada por inúmeros fatores de estresse abióticos, dentre os quais, podemos citar a baixa disponibilidade de água (deficiência hídrica), temperaturas extremas (frio, geadas, calor e fogo), salinidade, deficiência de nutrientes minerais e toxicidade. Esses fatores são responsáveis por consideráveis perdas econômicas tanto para os pequenos agricultores quanto para os produtores de commodities como a cultura da soja, entre outras. Além disso, estes danos podem ser potencialmente agravados pelos efeitos das recentes mudanças climáticas globais, sendo, portanto, a sua mitigação um grande desafio para a comunidade científica. O foco principal das pesquisas abordadas neste e-book é compreender os mecanismos de defesa/tolerância dos estresses abióticos em plantas e apresentar tecnologias e práticas de manejo que possibilitem o aumento da tolerância das plantas a esses estresses abióticos.

Temas associados à identificação de cultivares de soja tolerantes à seca e o manejo da salinidade e da restrição hídrica nas culturas de soja, amendoim e pepino são abordados. A tolerância de plantas de pinhão-manso a toxicidade do alumínio (Al³⁺), a tolerância de quatro espécies hortícolas ao estresse térmico causado por altas temperaturas e a tolerância de mutantes de trigo ao estresse salino também é sugerido. Na área de recursos naturais é mostrado os efeitos fitotóxicos dos metais pesados nas plantas cultivadas e o estresse ambiental causado pelo fogo na região do Cerrado. Portanto, esses conhecimentos irão agregar muito aos seus leitores que procuram promover melhorias quantitativas e qualitativas na produção de alimentos e, ou melhorar a qualidade de vida da sociedade. Sempre em busca da sustentabilidade do planeta.

Aos autores dos diversos capítulos, pela dedicação e esforços sem limites, que viabilizaram esta obra que retrata os recentes avanços científicos e tecnológicos nas áreas de ecofisiologia da produção vegetal e conservação dos recursos naturais e meio ambiente, os agradecimentos do Organizador e da Pantanal Editora.

Por fim, esperamos que este e-book possa colaborar e instigar mais estudantes e pesquisadores na constante busca de novas tecnologias. Assim, garantir uma difusão de conhecimento fácil, rápido para a sociedade.

Fábio Steiner

PRESENTATION

The eBook “Plant Abiotic Stress Tolerance”, a publication by Pantanal Editora, presents in its 9 chapters a wide range of questions about recent advances and scientific knowledge in the areas of ecophysiology of plant production and conservation of natural resources and the environment. The topics presented show some of the current tools that allow the increase in food production, the improvement of quality of life in people's and the preservation and sustainability of the resources available on the planet. This eBook materializes Editora Pantanal's desire to disseminate results and knowledge, which directly contribute to the development of society.

In the last decades, food production has often been limited by numerous abiotic stress factors, among which, we can mention the low availability of water (water deficit), extreme temperatures (cold, frosts, heat and fire), salinity, mineral nutrient deficiency and toxicity. These factors are responsible for considerable economic losses, both for small farmers and for producers of commodities such as soybean, among others. In addition, these damages can potentially be aggravated by the effects of recent global climate changes, and therefore, mitigating these damages is a major challenge for the scientific community. The main objective of the research presented in this e-book is to understand the defense or tolerance mechanisms of abiotic stresses in plants and to present technologies and management practices that enable greater tolerance of plants to these abiotic stresses.

Topics associated with the identification of drought-tolerant soybean cultivars and the management of salinity and water restriction in soybean, peanut and cucumber crops are presented. The tolerance of physic nut plants to aluminum toxicity (Al^{3+}), the tolerance of four vegetable species to heat stress caused by high temperatures and the tolerance of wheat mutants to salt stress is also suggested. In the area of natural resources, the phytotoxic effects of heavy metals on plant growth and the environmental stress caused by fire in the Cerrado region are shown. Therefore, this knowledge can add much to its readers who seek to promote quantitative and qualitative improvements in food production and, or improve the quality of life in society. Always in search of the planet's sustainability.

To the authors of the chapters, for their dedication and efforts, that made this eBook possible, which exposes the recent scientific and technological advances in the areas of ecophysiology of plant production and conservation of natural resources and the environment, thanks to the Organizer and Pantanal Editora.

Finally, we hope that this e-book can collaborate and instigate more students and researchers in the constant search for new technologies. Thus, ensuring an easy and quick dissemination of knowledge to society.

Fábio Steiner

PRESENTACIÓN

El trabajo “Plant Abiotic Stress Tolerance”, publicación de Pantanal Editora, presenta, en sus 9 capítulos, una amplia gama de temas sobre avances recientes y conocimientos científicos en las áreas de ecofisiología de la producción vegetal y conservación de los recursos naturales y el medio ambiente. Los temas tratados muestran algunas de las herramientas actuales que permiten el aumento de la producción de alimentos, la mejora de la calidad de vida de la población y la preservación y sostenibilidad de los recursos disponibles en el planeta. El trabajo materializa el afán de Editora Pantanal por difundir resultados y conocimientos, que contribuyan directamente al desarrollo humano.

En las últimas décadas, la producción de alimentos se ha visto a menudo limitada por numerosos factores de estrés abiótico, entre los que podemos mencionar la baja disponibilidad de agua (deficiencia de agua), temperaturas extremas (frío, heladas, calor y fuego), salinidad, deficiencia, nutrientes minerales y toxicidad. Estos factores son responsables de considerables pérdidas económicas tanto para los pequeños agricultores como para los productores de commodities como la soja, entre otros. Además, estos daños pueden verse potencialmente agravados por los efectos de los cambios climáticos globales recientes y, por lo tanto, mitigarlos es un desafío importante para la comunidad científica. El foco principal de las investigaciones cubiertas en este libro electrónico es comprender los mecanismos de defensa / tolerancia contra el estrés abiótico en las plantas y presentar tecnologías y prácticas de manejo que permitan aumentar la tolerancia de las plantas a estos estreses abióticos.

Se abordan temas relacionados con la identificación de cultivares de soja tolerantes a la sequía y el manejo de la salinidad y la restricción hídrica en cultivos de soja, maní y pepino. También se sugiere la tolerancia de las plantas de frutos secos a la toxicidad del aluminio (Al^{3+}), la tolerancia de cuatro especies hortícolas al estrés por calor causado por las altas temperaturas y la tolerancia de los mutantes del trigo al estrés por sal. El área de recursos naturales muestra los efectos fitotóxicos de los metales pesados en las plantas cultivadas y el estrés ambiental causado por los incendios en la región del Cerrado. Por tanto, este conocimiento aportará mucho a sus lectores que buscan promover mejoras cuantitativas y cualitativas en la producción de alimentos y, o mejorar la calidad de vida en la sociedad siempre en busca de la sostenibilidad del planeta.

A los autores de los distintos capítulos, por su dedicación y esfuerzo irrestricto, que hizo posible este trabajo, que retrata los recientes avances científicos y tecnológicos en las áreas de ecofisiología de la producción vegetal y conservación de los recursos naturales y el medio ambiente, gracias a la Organización y a Pantanal Editora.

Finalmente, esperamos que este libro electrónico pueda colaborar e instigar a más estudiantes e investigadores en la búsqueda constante de nuevas tecnologías. De esta forma, se garantiza una fácil y rápida difusión del conocimiento a la sociedad.

Fábio Steiner


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Selection indices to identify drought-tolerant soybean cultivars

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Rogério do Carmo Cabral¹ 

Simone Cândido Ensinas Maekawa² 

Maickon Alexandri Rezende Paulino¹ 

Lucila Silva Molina¹ 

Sara da Silva Abes¹ 

Fábio Steiner^{1*} 

INTRODUCTION

Soybean [*Glycine max* (L.) Merrill.] is one of the oilseed crops of greatest socioeconomic importance for world agribusiness. Brazil is one of the largest producers and exporters of soybeans in the world, in the 2019/2020 growing season, the crop occupied an area of 36.85 million hectares, with a production of 121.1 million tons, which represented an average productivity of 3,313 kg ha⁻¹ (Conab, 2020). Currently, the Cerrado region is the largest soybean producer in the country, representing about 60% of national production (Dickie et al., 2016). Soybean production in this region will certainly continue to be an important driver of economic growth in Brazil in the coming years.

Despite this favorable scenario for soybean cropping in the Midwest region of Brazil, the occurrence of climatic adversities is still a risk factor for the success of the cultivation of this crop. Among these climatic adversities, the occurrence of water deficiency is identified as the main factor that limits the development and grain yield of the crop (Mertz-Henning et al., 2018). Therefore, studies that aim to identify soybean genotypes with greater drought tolerance are important to increase agricultural production in regions with water deficiency.

Water restriction affects several biochemical, physiological and morphological processes in plants, and the responses of soybean plants to drought stress depend on the genotype, the stage of development of the plant, the severity and duration of water restriction, among other environmental factors (Kron et al., 2008; Catuchi et al. 2012; Zoz et al., 2013). Plants exposed to water restriction

¹ Curso de Agronomia, Universidade Estadual de Mato Grosso do Sul (UEMS), Rod. MS 306, km 6,4, CEP 795400-000, Cassilândia, MS, Brasil.

² Curso de Tecnologia em Produção Sucroalcooleira, Universidade Estadual de Mato Grosso do Sul (UEMS), Rua Projetada A, CEP 79730-000, Glória de Dourados, MS, Brasil.

* Autor de correspondência: steiner@uems.br

conditions have reduced stomatal conductance, transpiration rate, leaf area, photosynthetic rate, reduced relative growth rate and increased leaf and flower abscission rate (Kron et al., 2008; Fioreze et al., 2011; Vieira et al., 2017; Silva et al., 2019), in addition to changes in the activity of nitrogen and carbon metabolism enzymes and changes in antioxidant levels (Mantovani et al., 2015). Some of these responses are part of strategies that aim to reduce the negative effects of water restriction, thus constituting drought tolerance mechanisms.

The main characteristics for drought tolerance in soybean genotypes are related to the efficiency of water use by plants, the reduction in leaf area, the ability of cells to make the osmotic adjustment and the ability of roots to explore deeper layers of soil (Kron et al., 2008; Fioreze et al., 2011; Basu et al., 2016). Bahrami-Radb and Hajiboland (2017) reported that under conditions of water restriction, osmotic adjustment has a direct implication in maintaining stomatal conductance, leaf water content, photosynthetic rate, and consequently, plant growth rate. Genetic differences in drought tolerance under greenhouse conditions have been reported in Brazilian soybean genotypes (Zoz et al., 2013), which can be useful in identifying genotypes that are more adapted to adverse environmental conditions. However, the identification of drought-tolerant genotypes is not an easy task due to the fact that strong interactions between genotypes and the environment occur, in addition to the limited knowledge regarding the function and role of tolerance mechanisms (Naghavi et al., 2013).

The relative performance of grain production in optimal environmental conditions with adequate water availability or in water-restricted environments seems to be the beginning for the identification of desirable genotypes for cultivation in water-restricted conditions (Mohammadi et al., 2010). Therefore, the main conditions that must be considered during the selection and identification of drought-tolerant genotypes are cropping under optimal non-stressful conditions (irrigated system, for example) and under rain-fed conditions with water restriction (Naghavi et al., 2013; Menezes et al., 2014).

Several studies have proposed the use of different methods and/or selection indexes to assess genetic differences for drought tolerance. Some of these selection indices were used to assess genetic differences in genotypes of maize (Naghavi et al., 2013), sorghum (Menezes et al., 2014), wheat (Akçura et al., 2011; Farshadfar et al., 2013; El-Rawy; Hassan, 2014), sunflower (Gholinezhad et al., 2014) and common beans (Sánchez-Reinoso et al., 2020). However, these studies for soybeans are still unknown.

This research was carried out with the purpose of evaluating the response of 22 soybean genotypes grown under adverse environmental conditions (irrigated and rainfed systems), aiming to determine the best selection indexes to identify drought-tolerant genotypes.

MATERIAL AND METHODS

Location and Characterization of the Experimental Areas

Two field experiments were conducted during the 2018/2019 growing season in the municipality of Cassilândia, Mato Grosso do Sul, Brazil (Figure 1). The first experiment was carried out under rainfed conditions in the experimental area of the State University of Mato Grosso do Sul – UEMS (19°05'45" S, 51°48'51" W, and altitude of 520 m). The second experiment was conducted in an irrigated area with a central pivot system, located in a private area close to the Agricultural Experimental Station of UEMS/Cassilândia (19°05'16" S, 51°48'04" W, and average altitude of 470 m).

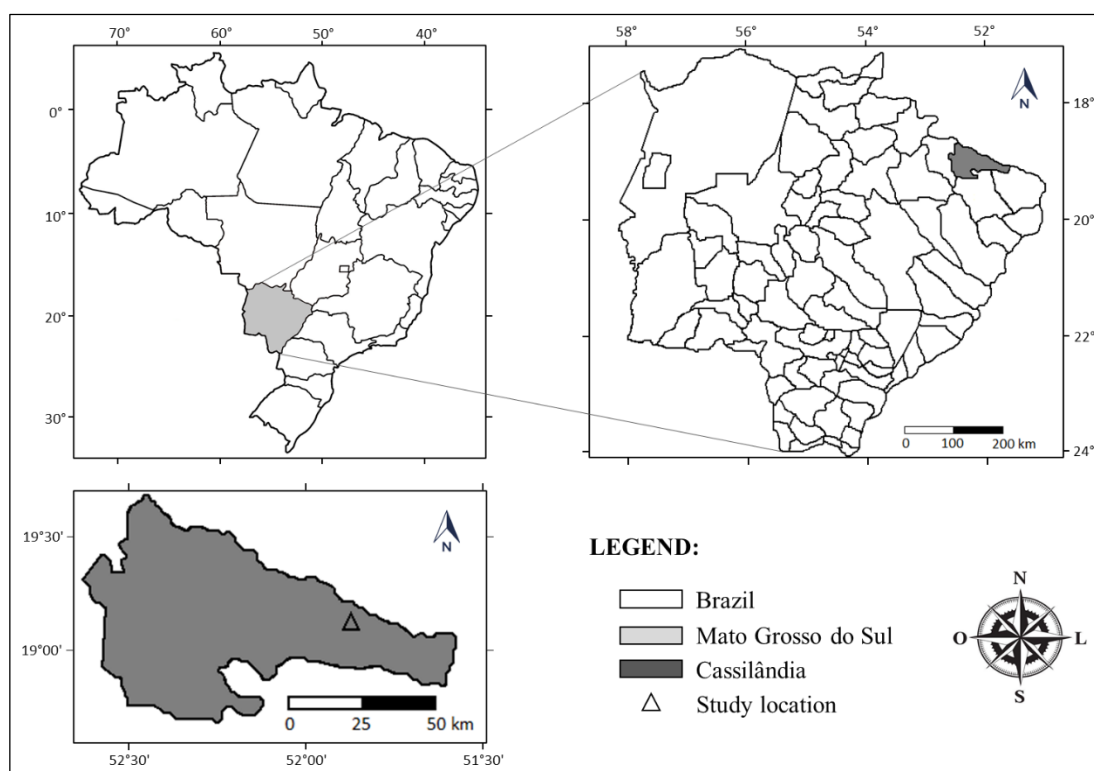


Figure 1. Location of the study area in the municipality of Cassilândia, State of Mato Grosso do Sul, Brazil. Source: The authors.

The region's climate, according to Köppen's classification, is tropical rainy (Aw), with rainy summer and dry winter between the months of May and September (winter rainfall less than 60 mm), with annual rainfall and an average annual temperature of 1,520 mm and 24.1 °C, respectively. The rainfall data collected during the conduction of the experiments are shown in Figure 2. The total rainfall accumulated during the cultivation of soybean genotypes in the dry area was 520 mm, with monthly

rainfall of 143, 108, 149 and 72 mm during the months of December, January, February, and March (Figure 2).

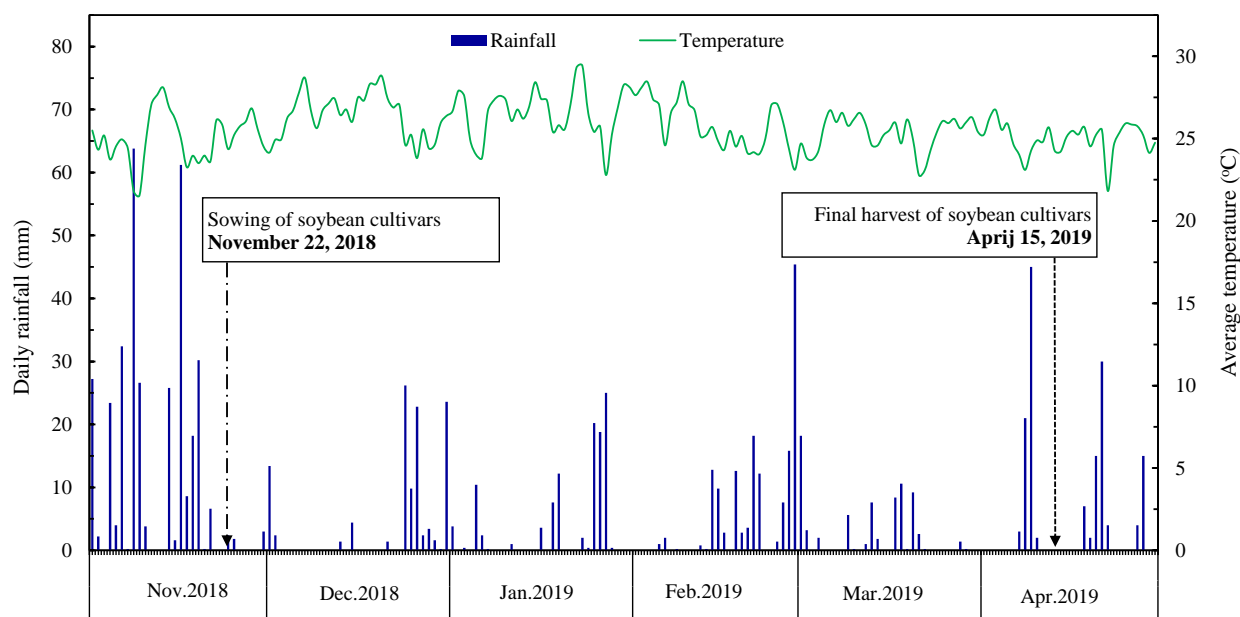


Figure 2. Daily rainfall (mm) and average temperature (°C) during the period of conducting soybean experiments in Cassilândia, MS, Brazil. Source: The authors.

The soil of the two experimental areas was classified as Neossolo Quartzarênico Órtico latossólico (NQo), with 120 g kg⁻¹ of clay, 40 g kg⁻¹ of silt and 840 g kg⁻¹ of sand). Before the implementation of the experiments, soil samples were collected in layers 0.0–0.20 in depth, and the main chemical properties of the soils are shown in Table 1.

Table 1. Main chemical properties of soils in the 0.0–0.20 m depth layer before the implementation of the experiments.

Field	pH	P	OM	H + Al	Al	K	Ca	Mg	CEC	V
		mg dm ⁻³	g dm ⁻³	----- cmol _c dm ⁻³ -----						%
Rainfed	5.5	12.3	19.0	2.10	0.00	0.15	2.50	1.10	5.6	64
Irrigated	5.2	10.7	14.4	2.30	0.00	0.12	2.10	0.90	5.4	57

pH in CaCl₂. P Mehlich-1. OM: organic matter. CEC: cation exchange capacity. V: soil base saturation.

Experimental Design and Treatments

The experimental design used was randomized blocks, in a 2×22 factorial scheme with four replications. The treatments consisted of two cropping systems (rainfed and irrigated) and 22 soybean genotypes. The management of irrigation in the experimental area with a central pivot system was carried out by applying a daily irrigation depth of 15 mm, which was applied every day when there was no rainfall. The seeds of the 22 soybean genotypes were purchased directly from the companies holding their registration with the Ministry of Agriculture, Livestock and Supply. The agronomic characteristics of soybean genotypes are shown in Table 2. Each experimental unit consisted of 5.0 m in length and 2.25 m in width (5 rows with 0.45 m spacing). For the measurement of grain yield, the three central rows were considered, disregarding 1.0 m from the ends of each row of plants, totaling 4.05 m^2 ($3.0 \times 1.35 \text{ m}$).

Table 2. Agronomic characteristics and germination rate of the 22 soybean genotypes [*Glycine max* (L.) Merrill.] used in the study.

Genotype	Agronomic characteristics			Germination rate
	Cycle (days) ¹	RMG	Growth type	(%)
TMG 2383 IPRO	120	8,3	Semi-determined	87
TMG 2381 IPRO	120	8,1	Indeterminate	100
TMG 2378 IPRO	125	7,8	Semi-determined	96
TMG 7067 IPRO	112	7,2	Semi-determined	100
TMG 7063 IPRO	110	7,0	Indeterminate	86
TMG 2165 IPRO	112	6,5	Indeterminate	98
TMG 7061 IPRO	110	6,1	Indeterminate	98
97R50 IPRO	115	7,5	Indeterminate	100
98R31 IPRO	130	8,3	Indeterminate	97
98R35 IPRO	130	8,3	Indeterminate	98
HO Cristalino IPRO	125	8,3	Indeterminate	100
HO Maracaí IPRO	120	7,7	Indeterminate	87
HO Paranaíba IPRO	115	7,4	Indeterminate	93
BMX Foco IPRO	110	7,2	Indeterminate	83
BMX Bônus IPRO	120	7,9	Indeterminate	100

ST 777 IPRO	108	7,7	Indeterminate	100
ST 797 IPRO	110	7,9	Indeterminate	100
RK 8115 IPRO	120	8,1	Indeterminate	96
RK 6719 IPRO	105	6,7	Indeterminate	100
RK 7518 IPRO	112	7,5	Indeterminate	100
RK 8317 IPRO	125	8,3	Indeterminate	88
M 5917 IPRO	95	5,9	Indeterminate	93

¹ Average cycle, in days, from plant emergence to harvest. RMG: Relative maturity group. Source: The authors.

Implementation and Conduction of Experiments

The soil preparation was carried out using two harrows, leaving the land level and suitable for soybean cropping and free of weeds. Sowing of soybean genotypes was carried out on November 22, 2018, in rows spaced 0.45 m apart. The sowing density was defined based on the technical recommendations for each genotype. Soybean seeds previously treated with pyraclostrobin + methyl thiophanate + fipronil (Standak Top[®]) in a rate of 2 mL kg⁻¹ were inoculated with *Bradyrhizobium japonicum*, using a commercial liquid inoculum Simbiose Nod Soja[®] (Symbiosis: Biological Agrotechnology) containing the strains SEMIA 5079 and SEMIA 5080 (minimum concentration of 7.2 x 10⁹ colony-forming units per mL), at a rate of 3 mL kg⁻¹ of seed.



Figure 3. Illustration of two soybean genotypes during the beginning bloom stage – R1 (at the left) and during the beginning seed - R5 (at the right) in the 2018/2019 growing season, in Cassilândia, Mato Grosso do Sul, Brazil. Source: The authors.

The base fertilization was carried out with the application of 600 kg ha⁻¹ of the fertilizer formulation NPK 04-22-09 in the sowing furrow. Topdressing fertilization was carried out, at 30 and

50 days after the emergence of the plants, with the application of 200 kg ha⁻¹ of the fertilizer formulation NPK 20-00-20. During the development of soybean genotypes, the management of weeds, pests and diseases was carried out according to the needs of the crop and technical recommendations (Embrapa, 2011). The optimal phytosanitary level of soybean plants during the reproductive stage can be seen in Figure 3.

Yield and Calculation of Drought Tolerance Indexes

Harvesting of soybean genotypes was carried out manually at the R8 development stage (95% of mature pods). All plants contained in 3.0 m of the three central rows of each plot were harvested, placed in the shade for drying for 5 days, and then mechanically traced. Grain yield was converted to kg ha⁻¹, correcting for 13% moisture (dry basis). From the grain yield data recorded for each genotype, in each production environment, drought tolerance indexes were calculated (Table 3).

Table 3. Drought tolerance indexes to assess the grain yield response of the 22 soybean genotypes grown under adverse environmental conditions (rainfed and irrigated systems)

Drought tolerance index	Equation†	Reference
1. Tolerance	$TOL = Y_P - Y_S$	Rosielle & Hamblin (1981)
2. Mean productivity	$MP = (Y_S + Y_P)/2$	Rosielle & Hamblin (1981)
3. Yield stability index	$YSI = Y_S/Y_P$	Bousslama & Schapaugh (1984)
4. Drought resistance index	$DI = [Y_S \times (Y_S/Y_P)] / \bar{Y}_S$	Blum (1988)
5. Stress tolerance index	$STI = (Y_S \times Y_P) / (\bar{Y}_P)^2$	Fernández (1992)
6. Geometric mean productivity	$GMP = \sqrt{Y_S \times Y_P}$	Fernández (1992)
7. Yield index	$YI = Y_S / \bar{Y}_S$	Gavuzzi et al. (1997)
8. Modified stress tolerance (k ₁)	$k_1STI = Y_P^2 / \bar{Y}_P^2$	Farshadfar & Sutka (2002)
9. Modified stress tolerance (k ₂)	$k_2STI = Y_S^2 / \bar{Y}_S^2$	Farshadfar & Sutka (2002)
10. Stress susceptibility percentage index	$SSPI = [Y_P - Y_S/2 \times \bar{Y}_P] \times 100$	Moosavi et al. (2008)
11. Abiotic tolerance index	$ATI = [(Y_P - Y_S) / (\bar{Y}_P / \bar{Y}_S)] \times \sqrt{Y_P \times Y_S}$	Moosavi et al. (2008)
12. Harmonic mean	$HM = [2 \times (Y_S \times Y_P)] / (Y_S + Y_P)$	Jafari et al. (2009)

† In the above equations, Y_S, Y_P, represent the soybean grain yield under water restricted conditions (rainfed system) and under conditions of adequate water availability (irrigated system) for each genotype, respectively, whereas \bar{Y}_S and \bar{Y}_P represent the average grain yield under conditions of water restriction (rainfed system) and under conditions of adequate water availability (irrigated system) of all soybean genotypes, respectively.

In this study, 12 drought tolerance indexes proposed by several researchers were used to evaluate the grain yield response of the 22 soybean genotypes, grown under optimal environmental conditions (irrigated system) and under water restriction conditions (rainfed system). The drought tolerance indexes used in this study are shown in Table 3.

Statistical Analysis

The data were subjected to analysis of variance (ANOVA), and the averages were grouped by the Scott-Knott test at 5% probability level, in order to discriminate soybean genotypes within adverse production environments (irrigated and rainfed systems) and between environments by drought tolerance indexes. The analyzes were performed using the Sisvar software version 5.6 for Windows (Ferreira, 2014).

The identification of drought-tolerant and/or susceptible genotypes was carried out based on all drought tolerance indexes, using the three multivariate analysis methods (ranking method, hierarchical clustering method and principal component analysis).

The ranking method was used as proposed by Farshadfar et al. (2012), with modifications. In this method, a genotype with the highest value for each of the YP, YS, MP, YSI, DI, S₁TI, GMP, YI, k₁S₁TI, k₂S₁TI, SSPI, ATI and HM tolerance scores received a ranking score of 1, whereas for the genotype with the lowest value for the TOL tolerance index it received a ranking score equal to 22. The average ranking score (\bar{R}) and the ranking standard deviation (RSD) were calculated for all drought tolerance indexes of the 22 soybean genotypes under irrigated or rainfed conditions. The discrimination of soybean genotypes regarding the level of drought tolerance was performed based on the average ranking score of each genotype, considering the quartile value that divides the 22 possible ranking positions into four equal parts. Therefore, a genotype with an average ranking score below the value of the first quartile (<6.25 points) is classified as drought tolerant (I); a genotype with an average score between the value of the first and second quartiles (6.25 to 11.50 points) is classified as moderately tolerant (MI) to drought; a genotype with an average ranking score between the value of the second and third quartiles (11.51 to 16.75 points) is classified as moderately susceptible (MS) to drought; and, in turn, the group of drought susceptible (S) genotypes is represented by genotypes with an average ranking score above the value of the third quartile (> 16.75 points).

The multivariate analysis using the hierarchical clustering method was performed based on Euclidean distance and Ward's minimum variance method, to classify the 22 soybean genotypes in different levels of drought tolerance (tolerant, moderately tolerant, moderately sensitive, and sensitive).

Principal component analysis (PCA) based on the correlation matrix of drought tolerance indexes and Biplot analysis were performed using the statistical software Action Stat Pro[®] version 3.6 for Windows.

RESULTS AND DISCUSSION

Grain yield and drought tolerance indexes

The grain yield in the irrigated system (Y_p) allowed to separate soybean genotypes in seven groups; genotypes RK 8317 IPRO and 98R35 IPRO represented the two groups with the highest grain yield, and genotypes TMG 7067 IPRO, TMG 7061 IPRO 97R50 IPRO, RK 6719 IPRO and M5917 IPRO represented the group with the lowest grain yield (Table 4). Under rainfed conditions with drought stress, grain yield (Y_s) separated the soybean genotypes into six groups, in which the genotypes RK 8115 IPRO, RK 8317 IPRO, 98R35 IPRO and TMG 2381 IPRO were classified into the two groups with the highest grain yield, whereas the group lower grain yield was represented by genotypes TMG 7067 IPRO, TMG 7061 IPRO, 97R50 IPRO, HO Maracaí IPRO, RK 6719 IPRO and M5917 IPRO (Table 4).

The average grain yield in the irrigated system was $2,620 \text{ kg ha}^{-1}$, and under dry conditions it was $1,150 \text{ kg ha}^{-1}$, which represents a loss of grain yield of approximately 56% (Table 4). The average grain yield obtained in the irrigated system was lower than the average yield of soybeans $2,960 \text{ kg ha}^{-1}$, recorded for the state of Mato Grosso do Sul in the 2018/2019 growing season (Conab, 2019). Of the 22 soybean genotypes tested in the municipality of Cassilândia, MS, Brazil, only seven genotypes had grain yields higher than the average grain yield of the state of Mato Grosso do Sul for the 2018/2019 season. Therefore, based on the above, it appears that the genotypes TMG 2378 IPRO, 98R31 IPRO, 98R35 IPRO, ST 777 IPRO, ST 797 IPRO, RK 8115 IPRO and RK 8317 IPRO are the genetic materials most adapted to the edaphoclimatic conditions of Cassilândia, MS, Brazil. It should be noted that due to the drought and excess temperature during the vegetative phases and during flowering and grain filling, between the months of December 2018 and January 2019 (Figure 2), the grain yield of all soybean genotypes in the rainfed system was lower than the average grain yield of crop in the 2018/2019 growing season for the state of Mato Grosso do Sul.

The tolerance index (TOL) separated the genotypes into nine distinct groups, with the group with the best index being represented by the genotypes TMG 7067 IPRO, RK 7518 IPRO and M 5917 IPRO, and the genotype RK 8317 IPRO was classified in the group of lowest TOL index (Table 4). The drought tolerance index of mean productivity (MP) classified soybean genotypes in eight groups; genotypes 98R50 and RK 8317 IPRO represented the two groups with the highest MP index, while the

group with the lowest MP index was represented by genotypes TMG 7067 IPRO, TMG 7061 IPRO, 97R50 IPRO, RK 6719 IPRO and M 5917 IPRO (Table 4).

Table 4. Grain yield and drought tolerance indexes for the 22 soybean genotypes under optimum environmental conditions (irrigated system) and under water restriction conditions (rainfed system), during the 2018/2019 growing season, in the municipality of Cassilândia, Mato Grosso do Sul, Brazil.

Genotype	Y _p (kg/ha)	Y _s (kg/ha)	TOL	MP	YSI	DI	STI	GMP	YI	k ₁ STI	k ₂ STI	SSPI	ATI	HM
TMG 2383 IPRO	2240f	1423c	817b	1832f	0.64b	0.79b	0.47c	1785e	1.24c	0.73e	1.54c	15.6h	642910f	1740c
TMG 2381 IPRO	2500e	1683b	817b	2092e	0.67a	0.99a	0.62d	2051d	1.46b	0.91e	2.16b	15.6h	733731f	2011b
TMG 2378 IPRO	3.487c	1123d	2363g	2305d	0.32g	0.32f	0.57d	1979d	0.98d	1.77c	0.96e	45.1c	2055328d	1699c
TMG 7067 IPRO	1220g	737f	483a	978h	0.60b	0.39e	0.13g	948g	0.64f	0.22f	0.41f	9.2i	200816g	918f
TMG 7063 IPRO	1883f	1177d	707b	1530g	0.63b	0.64c	0.32f	1488f	1.02d	0.52f	1.05d	13.5h	463025g	1447d
TMG 2165 IPRO	2277f	1300c	977c	1788f	0.57c	0.65c	0.43e	1720e	1.13c	0.76e	1.29d	18.6g	741840f	1655c
TMG 7061 IPRO	1487g	733f	753b	1110h	0.49d	0.32f	0.16g	1044g	0.64f	0.32f	0.41f	14.4h	345104g	982f
97R50 IPRO	1560g	740f	820b	1150h	0.47e	0.31f	0.17g	1074g	0.64f	0.36f	0.42f	15.6h	388141g	1004f
98R31 IPRO	3537c	1417c	2120f	2477d	0.40f	0.49d	0.73c	2238c	1.23c	1.82c	1.52c	40.5d	2083048d	2023b
98R35 IPRO	4237b	1693b	2543g	2965b	0.40f	0.59c	1.05b	2678b	1.47b	2.62b	2.18b	48.5c	2996440b	2419a
HO Cristalino IPRO	2833d	1070d	1763e	1952f	0.38f	0.35e	0.44e	1741e	0.93d	1.18d	0.87e	33.7e	1354237e	1553c
HO Maracá IPRO	2197f	853f	1343d	1525g	0.39f	0.29f	0.27f	1369f	0.74f	0.70e	0.55f	25.6f	807998f	1229e
HO Paranaíba IPRO	2153f	973e	1180d	1563g	0.45e	0.38e	0.31f	1448f	0.85e	0.68e	0.72e	22.5f	751089f	1340d
BMX Foco IPRO	2907d	900e	2007f	1903f	0.31g	0.24f	0.38e	1617e	0.78e	1.24d	0.62f	38.3d	1430068e	1374d
BMX Bônus IPRO	2637e	983e	1653e	1810f	0.37f	0.32f	0.38e	1610e	0.86e	1.02d	0.73e	31.6e	1171278e	1432d
ST 777 IPRO	3403c	990e	2413g	2197e	0.29g	0.25f	0.49e	1835e	0.86e	1.69c	0.74e	46.1c	1951363d	1534c
ST 797 IPRO	3773c	1020e	2753h	2397d	0.27g	0.24f	0.56d	1962d	0.89e	2.08c	0.79e	52.6b	2377613c	1606c
RK 8115 IPRO	3590c	1880a	1710e	2735c	0.52d	0.86b	0.98b	2598b	1.63a	1.88c	2.68a	32.6e	1949428d	2467a
RK 6719 IPRO	1547g	667f	880b	1107h	0.43e	0.25f	0.15g	1015g	0.58f	0.35f	0.34f	16.8h	393709g	932f
RK 7518 IPRO	2077f	1430c	647a	1753f	0.69a	0.86b	0.43e	1723e	1.24c	0.63e	1.55c	12.3i	494134g	1692c
RK 8317 IPRO	4770a	1827a	2943i	3298a	0.38f	0.61c	1.27a	2952a	1.59a	3.32a	2.53a	56.2a	3818061a	2641a
M 5917 IPRO	1320g	680f	640a	1000h	0.51d	0.31f	0.13g	947g	0.59f	0.26f	0.36f	12.2i	266035g	896f
Mean	2620	1150	1470	1885	0.46	0.47	0.48	1719	1.00	1.14	1.11	28.0	1246154	1572
CV (%)	6.82	8.87	7.28	7.18	5.37	12.90	16.76	7.57	8.87	14.78	19.98	7.28	14.88	8.02

For abbreviation of drought tolerance indices, see Table 3. CV: Coefficient of variation. Source: The authors.

The yield stability index (YSI) separated soybean genotypes into seven groups; genotypes TMG 2383 IPRO, TMG 2381 IPRO, TMG 7067 IPRO, TMG 7063 IPRO and RK 7518 IPRO were classified in the two groups with the highest YSI indices, and genotypes TMG 2378 IPRO, BMX Foco IPRO, ST 777 IPRO and ST 797 IPRO were classified in the group with the lowest YSI index. The drought resistance index (DI) classified soybean genotypes into six groups, in which the genotypes TMG 2383 IPRO, TMG 2381 IPRO, RK 8115 IPRO and RK 7518 IPRO were classified in the two groups with the highest DI indices, while whereas the group with the lowest DI index was represented by genotypes TMG 2378 IPRO, TMG 7061 IPRO, 97R50 IPRO, HO Maracaí IPRO, BMX Foco IPRO, BMX Bonus IPRO, ST 777 IPRO, ST 797 IPRO, RK 6719 IPRO and M 5917 IPRO (Table 4).

The stress tolerance index (STI) and the geometric mean productivity (GMP) classified soybean genotypes into seven distinct groups, with the two groups with the highest STI and GMP indexes being represented by the genotypes 98R35 IPRO, RK8115 IPRO and RK 8317 IPRO, while the group with the lowest STI and GMP indexes were represented by genotypes TMG 7067 IPRO, TMG7061 IPRO, 97R50 IPRO, RK 6719 IPRO and M5917 IPRO. The yield index (YI) separated soybean genotypes into six groups; the two groups with the highest YI indexes were represented by the genotypes TMG 2381 IPRO, 98R35 IPRO, RK 8115 IPRO and RK 8317 IPRO, while the group with the lowest YI index was represented by the genotypes TMG 7067 IPRO, TMG 7061 IPRO, 97R50 IPRO , HO Maracaí IPRO, RK 6719 IPRO and M 5917 IPRO (Table 4).

The k_1 modified stress tolerance index (k_1 STI) classified soybean genotypes into six groups; the two groups with the highest k_1 STI indexes were represented by the genotypes 98R50 IPRO and RK 8317 IPRO, and the genotypes TMG 7067 IPRO, TMG 7063 IPRO, TMG 7061 IPRO, 97R50 IPRO, RK 6719 IPRO and M 5917 IPRO were classified in the group with the lowest k_1 STI index. The k_2 modified stress tolerance index (k_2 STI) separated soybean genotypes into six groups, and the two groups with the highest k_2 STI indexes were represented by genotypes TMG 2381 IPRO, 98R50 IPRO, RK 8115 IPRO and RK 8317 IPRO, while the group with the lowest k_2 STI index was represented by the genotypes TMG 7067 IPRO, TMG 7061 IPRO, 97R50 IPRO, HO Maracaí IPRO, BMX Foco IPRO, RK 6719 IPRO and M 5917 IPRO (Table 4).

The stress susceptibility percentage index (SSPI) classified soybean genotypes into nine distinct groups, with the two groups with the highest SSPI indices being represented by the genotypes ST 797 IPRO and RK 8317 IPRO, and the genotypes TMG 7067 IPRO, RK 7518 IPRO and M 5917 IPRO were classified in the group with the lowest SSPI index. The abiotic tolerance index (ATI) separated soybean genotypes into seven groups, in which the genotypes 98R50 IPRO and RK 8317 IPRO were classified in the two groups with the highest ATI index, while the group with the lowest ATI index was represented by genotypes TMG 7067 IPRO, TMG 7063 IPRO, TMG 7061 IPRO, 97R50 IPRO, RK

6719 IPRO, RK 7518 IPRO and M 5917 IPRO. The tolerance index based on the harmonic mean (HM) classified soybean genotypes into six groups, with genotypes TMG 2381 IPRO, 98R31 IPRO, 98R35 IPRO, RK 8115 IPRO and RK 8317 IPRO being grouped in the two groups with the largest HM indexes, and the genotypes TMG 7067 IPRO, TMG 7061 IPRO, 97R50 IPRO, RK 6719 IPRO and M 5917 IPRO were grouped in the group with the lowest HM index (Table 4).

The HM, YI, DI, k_1 STI and k_2 STI indexes separated the soybean genotypes into six groups, while the STI, GMP, YSI and ATI indexes separated genotypes into seven different groups. These results indicate that these drought tolerance indices were less sensitive to differentiate soybean genotypes in terms of drought tolerance. In turn, the MP index separated soybean genotypes into eight groups, and the TOL and SSPI indices separated genotypes into nine groups (Table 4). These results indicate that these tolerance indices are the most sensitive to identify and differentiate soybean genotypes in terms of drought tolerance. Menezes et al. (2014) evaluating eight drought tolerance indices, reported that the TOL and YSI indices were not adequate to differentiate drought tolerant grain sorghum genotypes. In another study, Naghavi et al. (2013) found that the STI, YI, SSPI, k_1 STI and k_2 STI indices were the most appropriate, and can be used to identify drought-tolerant corn genotypes.

Ranking Method

The ranking of the 22 soybean genotypes based on the different drought tolerance indexes calculated based on grain yield in irrigated system (Y_I) and under rainfed system with water restriction (Y_S) are shown in Table 5. The discrimination of the level of tolerance, or susceptibility of soybean genotypes to drought stress based on only a single criterion or drought tolerance index can be contradictory (Table 5). For example, according to the YSI index, genotypes RK 7518 IPRO, TMG 2381 IPRO and TMG 2383 IPRO were considered the most drought tolerant, while according to the MP, STI, GMP, YI, k_2 STI and HM indices, the genotypes RK 8317 IPRO, 98R35 IPRO and RK 8115 IPRO were considered the most drought tolerant. Therefore, differentiation and separation of genotypes at different levels of drought tolerance must be carried out considering all tolerance indices (Naghavi et al., 2013). In this sense, the ranking method has been used to classify genotypes in different levels of drought tolerance (Farshadfar et al., 2012).

Considering all drought tolerance indexes, soybean genotypes 98R35 IPRO, RK 8115 IPRO and RK 8317 IPRO were classified in the best average classification in the ranking method, receiving scores between 4.2 and 4.9 (Table 6) and, therefore, these genotypes were classified as drought tolerant. The genotypes TMG 7067 IPRO, TMG 7061 IPRO, RK 6719 IPRO and M 5917 IPRO

received the highest score in the ranking method and were then classified as susceptible to drought stress for cropping in the region of Cassilândia, State of Mato Grosso do Sul, Brazil (Table 6).

Table 5. Ranking, average ranking score (\bar{R}) and ranking standard deviation (RSD) for grain yield under irrigated (Y_P) and rainfed system (Y_S) and drought tolerance indices of 22 soybean genotypes under optimal environmental conditions (irrigated system) and under drought stress conditions (rainfed system) during the 2018/2019 growing season, in the municipality of Cassilândia, Mato Grosso do Sul, Brazil.

Genotype	Y_P	Y_S	TOL	MP	YSI	DI	STI	GMP	YI	k_1 STI	k_2 STI	SSPI	ATI	HM	$\bar{R}(\pm SD)$	Tolerance level [†]
TMG 2383 IPRO	13	6	7	11	3	4	9	9	6	13	6	17	15	6	8,9 ($\pm 3,5$)	MT
TMG 2381 IPRO	11	4	6	8	2	1	5	5	4	11	4	16	14	5	6,9 ($\pm 3,7$)	MT
TMG 2378 IPRO	6	10	18	6	19	15	6	6	10	6	10	5	5	7	9,2 ($\pm 3,8$)	MT
TMG 7067 IPRO	22	19	1	22	5	10	22	21	19	22	19	22	22	21	17,6 ($\pm 5,3$)	S
TMG 7063 IPRO	17	9	4	16	4	6	15	15	9	17	9	19	17	13	12,1 ($\pm 4,6$)	MS
TMG 2165 IPRO	12	8	10	13	6	5	11	12	8	12	8	13	13	9	10,0 ($\pm 2,3$)	MT
TMG 7061 IPRO	20	20	5	19	9	14	19	19	20	20	20	18	20	19	17,3 ($\pm 3,4$)	S
97R50 IPRO	18	18	8	18	10	17	18	18	18	18	18	15	19	18	16,5 ($\pm 2,4$)	MS
98R31 IPRO	5	7	17	4	13	9	4	4	7	5	7	6	4	4	9,9 ($\pm 2,7$)	MT
98R35 IPRO	2	3	20	2	14	8	2	2	3	2	3	3	2	3	4,9 ($\pm 3,9$)	T
HO Cristalino IPRO	9	11	15	9	17	12	10	10	11	9	11	8	9	11	10,9 ($\pm 1,7$)	MT
HO Maracá IPRO	14	17	12	17	15	18	17	17	17	14	17	11	11	17	15,3 ($\pm 2,1$)	MS
HO Paranaíba IPRO	15	15	11	15	11	11	16	16	15	15	15	12	12	16	13,9 ($\pm 1,8$)	MS
BMX Foco IPRO	8	16	16	10	20	21	13	13	16	8	16	7	8	15	13,4 ($\pm 3,8$)	MS
BMX Bónus IPRO	10	14	13	12	18	13	14	14	14	10	14	10	10	14	12,9 ($\pm 1,8$)	MS
ST 777 IPRO	7	13	19	7	21	19	8	8	13	7	13	4	6	12	11,2 ($\pm 4,5$)	MT
ST 797 IPRO	3	12	22	5	22	22	7	7	12	3	12	2	3	10	10,1 ($\pm 5,8$)	MT
RK 8115 IPRO	4	1	14	3	7	3	3	3	1	4	1	9	7	2	4,4 ($\pm 2,8$)	T
RK 6719 IPRO	19	22	9	20	12	20	20	20	22	19	22	14	18	20	18,4 ($\pm 2,9$)	S
RK 7518 IPRO	16	5	3	14	1	2	12	11	5	16	5	20	16	8	9,6 ($\pm 5,4$)	MT
RK 8317 IPRO	1	2	21	1	16	7	1	1	2	1	2	1	1	1	4,2 ($\pm 4,6$)	T
M 5917 IPRO	21	21	2	21	8	16	21	22	21	21	21	21	21	22	18,5 ($\pm 4,2$)	S

[†] T = refers to a drought-tolerant soybean genotype, receiving an average ranking score (\bar{R}) of 1 to 6.25; MT = moderately tolerant genotype with an average ranking score (\bar{R}) of 6.26 to 11.50; DM = moderately susceptible genotype with an average ranking score (\bar{R}) of 11.51 to 16.75; S = drought-sensitive soybean genotype with an average ranking score (\bar{R}) of 16.76 to 22. Source: The authors.

Multivariate Analysis of Hierarchical Clustering

The multivariate analysis of hierarchical clustering of the 22 soybean genotypes, based on grain yield in irrigated and rainfed conditions and on the 12 drought tolerance indexes, separated soybean genotypes in four groups with 5, 5, 4 and 8 genotypes, respectively (Figure 3). The first group was represented by the genotypes with the lowest drought tolerance indexes and, therefore, was considered the group most susceptible to drought stress. The second and fourth groups represented the genotypes with the intermediate values of drought tolerance indexes and, therefore, the genotypes belonging to these groups were classified as moderately susceptible and moderately tolerant to drought stress, respectively. In turn, the third group represented the genotypes with the highest drought tolerance indexes and, thus, classified as the most tolerant to the adverse effects of drought.

Therefore, soybean genotypes 98R35 IPRO, RK 8317 IPRO, 98R31 IPRO and RK 8115 IPRO were identified as the most drought tolerant, whereas genotypes TMG 7067 IPRO, RK 6719 IPRO, M 5917 IPRO, TMG 7061 IPRO and 97R50 IPRO were classified as the most sensitive to drought for cultivation conditions in the region of Cassilândia, Mato Grosso do Sul, Brazil (Figure 4).

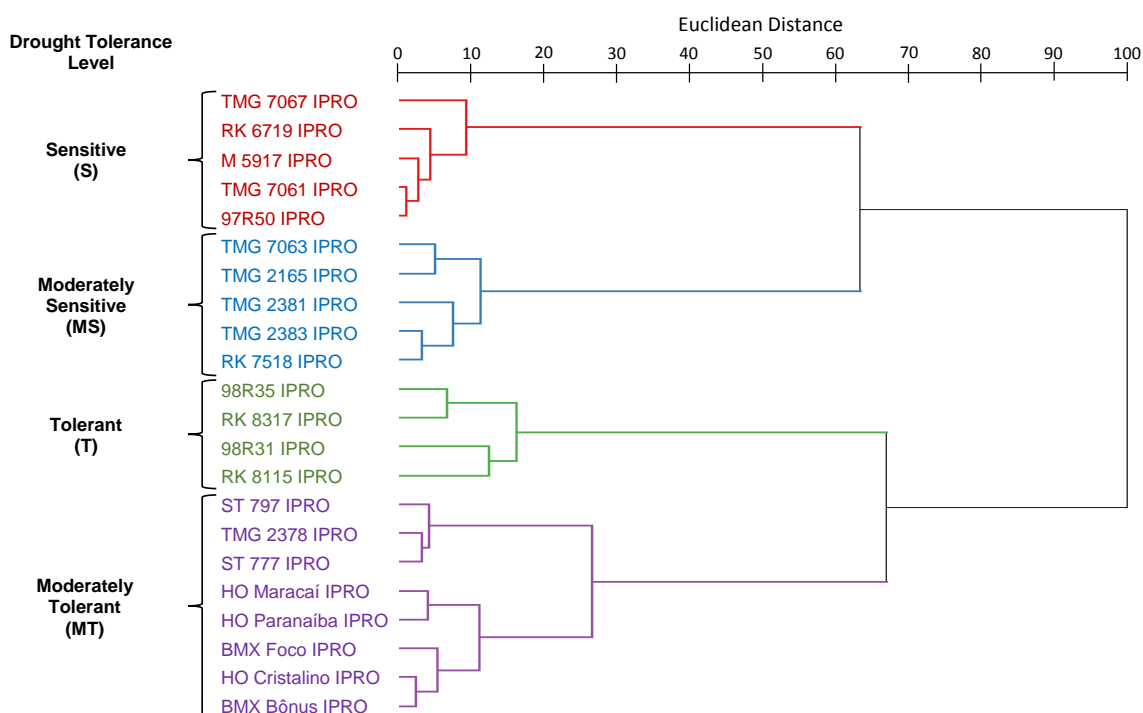


Figure 4. Dendrogram of the hierarchical cluster analysis of the 22 soybean genotypes based on Euclidean distance and Ward's minimum variance method using grain yield in irrigated system (Y_p) and rainfed system (Y_s) and drought tolerance indexes (TOL, MP, YSI, DI, STI, GMP, YI, k_1 STI, k_2 STI, SSPI, ATI and HM) during the 2018/2019 growing season, in the municipality of Cassilândia, Mato Grosso do Sul, Brazil. Source: The authors.

Principal Component Analysis (PCA)

The first principal component explains 61.25% of the total variance, while the second principal component explains 37.15% of the variation (Figure 5). According to the eigenvector value, the weights of the MP (-0.318), GMP (-0.315), STI (-0.313), YP (-0.311), k_1 STI (-0.308), HM (-0.301), ATI (-0.300), SSPI (-0.265), YS (-0.261), YI (-0.261) and k_2 STI (-0.258) are negative for this principal component. This indicates that the higher the value of these drought tolerance indexes, the lower the score of the first main component. Therefore, the higher the score of these drought tolerance indexes, the lower the score of the first main component, and then the genotype can be considered tolerant to water restriction.

The first principal component can be interpreted as a global performance index of the tolerance of soybean genotypes to drought. As the weights are negative, the higher the drought tolerance indexes, the lower the value of this component and the better the global tolerance index of the soybean genotype. Therefore, a lower score on the first principal component indicates that the genotype's tolerance index is better. The soybean genotypes RK 8317 IPRO, 98R35IPRO and RK 8115 IPRO had the best overall performance indexes, respectively, and, therefore, these genotypes were classified as tolerant to drought stress. In turn, the lowest global performance indices were observed in soybean genotypes TMG 7067 IPRO and M 5917 IPRO, and thus these genotypes were classified as sensitive to drought stress.

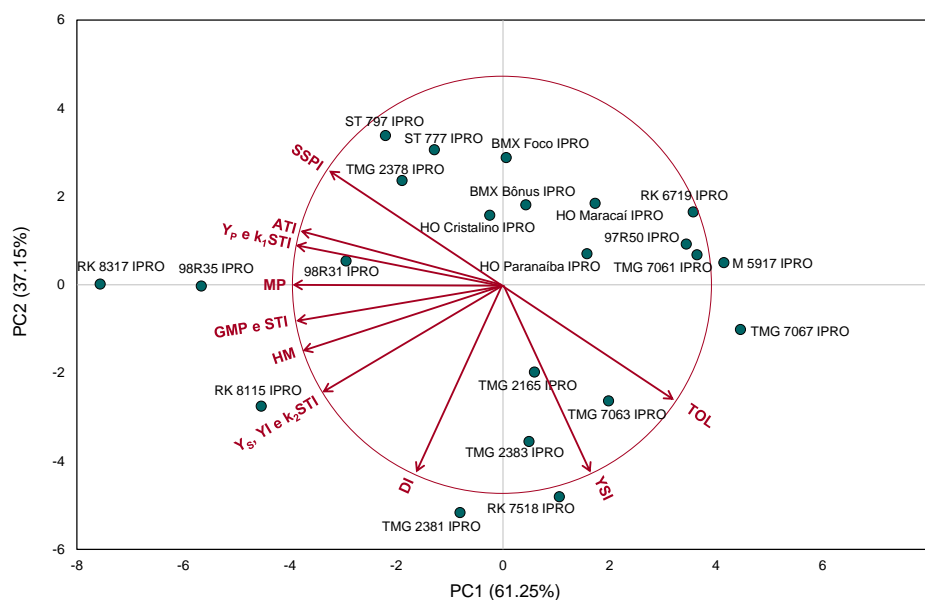


Figure 5. Biplot diagram based on the first and second principal components (PC) for grain yield of the 22 soybean genotypes under irrigated system (Y_P) and rainfed system (Y_S) and drought tolerance indexes (TOL, MP, YSI, DI, STI, GMP, YI, k_1 STI, k_2 STI, SSPI, ATI and HM) during the 2018/2019 growing season, in the municipality of Cassilândia, Mato Grosso do Sul, Brazil. Source: The authors.

In summary, the three multivariate analysis methods used in this study (ranking method, hierarchical cluster analysis and principal component analysis) grouped soybean genotypes RK 8115 IPRO, RK 8317 IPRO and 98R35 IPRO as drought tolerant (Table 5, Figures 4 and 5), whereas the genotype 98R31 IPRO was classified as drought tolerant only by the hierarchical cluster analysis method (Figure 4). Therefore, these soybean genotypes are the most suitable to be recommended for cropping in conditions with high probability of occurrence of water restriction in the Cerrado region.

The ranking, hierarchical cluster analysis and principal component analysis methods grouped 4, 5 and 2 soybean genotypes, respectively, as sensitive to drought stress (Table 5, Figures 4 and 5). The soybean genotypes TMG 7067 IPRO and M5917 IPRO were classified as sensitive to drought by the three methods of multivariate analysis. In turn, the genotypes TMG 7061 IPRO and RK 6719 IPRO were classified as sensitive to drought stress by the methods of ranking and hierarchical cluster analysis (Table 5 and Figure 5). Therefore, when soybean sowing is carried out in the Cassilândia region at a time with high probability of occurrence of water restriction during cultivation, these genotypes should not be recommended for sowing.

FINAL CONSIDERATIONS

The soybean genotypes 98R35 IRPO, RK 8317 IPRO and RK 8115 IPRO have greater tolerance to drought, and are the most suitable genotypes to be cultivated in Cerrado regions with occurrence of water restriction. In contrast, the genotypes TMG 7067 IPRO, M 5917 IPR, TMG 7061 IPRO and RK 6719 IPRO are more susceptible to water restriction, and should not be recommended for cultivation in the Cassilândia region under rainfed conditions with high probability of occurrence of water restriction.

The tolerance indexes MP, STI, GMP and HM were the most suitable to identify soybean genotypes with greater drought tolerance and with high grain yield potential in irrigated and rainfed systems in the region of Cassilândia, State of Mato Grosso do Sul, Brazil.

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Fábio Steiner



Doutor em Agronomia - Agricultura (UNESP - Botucatu). Mestre em Agronomia (Produção Vegetal) e Graduado em Agronomia (UNIOESTE - Marechal Cândido Rondon). Professor, Universidade Estadual de Mato Grosso do Sul em Cassilândia.

Contato: steiner@uems.br

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Rua Abaete, 83, Sala B, Centro. CEP: 78690-000
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Telefone (66) 99682-4165 (Whatsapp)
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