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CROP BREEDING AND
APPLIED BIOTECHNOLOGY

Adaptability and stability of soybean in terms of oil and protein content

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ABSTRACT - *The objective of this study was to evaluate the influence of genotype x environment interaction on the seed oil and protein content in soybean genotypes (*Glycine max *L. Merrill). Fifteen soybean genotypes were evaluated. The experiments were conducted in the growing seasons 2004/05, 2005/06 and 2006/07 in Viçosa and Frutal, Minas Gerais (MG), and Pindorama, São Paulo (SP), respectively, with different planting dates and soil types, in a total of 13 environments. The evaluation was based on the methods of Eberhart and Russell and Extended Centroid. The results showed that genotype UFV 18 is desirable in soybean breeding programs designed to increase the values of the two traits evaluated. The results of genotype PTN-Bio were different for each trait. Moreover, it was noted that the Extended Centroid method is more conservative than the methodology of Eberhart and Russell in the classification of genotypes.*

Key words: *Glycine max* (L.) Merrill, genotype x environment interaction, Eberhart and Russell, Extended Centroid.

INTRODUCTION

Due to the economic importance, the cultivation of soybean (*Glycine max* (L.) Merrill) is one of the crops with greatest increase in planted area and the agribusiness segment in Brazil (Nogueira et al. 2008). Soybean and its derivatives are highly promising raw material for the food industry, especially in cereal and meat-based products. The addition of appropriate soy products results in less caloric food products with lower lipid and high protein content, fit for the nutritional needs of adults, cheaper, besides preserving the physical and taste characteristics of the traditional product (McMindes 1991, Dhingra and Jood 2001).

The adaptability and stability of soybean genotypes have been evaluated in several studies. For example Oliveira et al. (2006) evaluated the adaptability and stability of 21 soybean lines/cultivars conducted in 78 environments in southern and central Brazil, between 2001 and 2003. Cucolotto et al. (2007) assessed the adaptability and stability of 30 soybean cultivars in the growing seasons 1999/2000, 2000/2001 and 2001/2002 in 30 environments in Paraná. Besides, Pelúzio et al. (2008) compared the performance, adaptability and stability of soybean genotypes sown on four dates in Gurupi, state of Tocantins.

Among the main traits of soybean genotypes, two particularly important ones are the oil and protein content. Soybean, rich in protein and oil, is one of the five main protein sources, and is also an energy source (Maehler et al. 2003). The characterization of the oil and protein content of soybean cultivars makes information available about the most appropriate cultivars for specific uses, aggregates quality value to these

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products and makes way for an increased market share. The evaluation of the effects of the sowing site on oil content allows the recommendation of locations that optimize the expression of this quality trait. For Brazilian conditions, there are few experimental data that can support soybean breeding programs for oil and/or protein content increases. The search for this information is important because it is known that in tropical and subtropical regions the potential production of oil and/or protein can be optimized.

The purpose of this study was to evaluated the adaptability and phenotypic stability in the oil and protein content of 15 soybean genotypes grown in different environments (locations, planting dates and soil types), based on methods of Eberhart and Russell (1966) and the Extended Centroid method (Nascimento et al. 2009). The results of the two methods were compared.

MATERIAL AND METHODS

The traits oil (OC) and protein content (PrC) were assessed in 15 soybean genotypes. The experiments were conducted in the growing seasons 2004/05, 2005/06 and 2006/07 in Viçosa-MG, Frutal-MG and Pindorama-SP, respectively, sown on different dates and soil types, in a total of 13 environments (described in Table 1).

Soil was conventionally tilled (plowing and harrowing) and fertilized according to soil analysis, as recommended for soybean. The planting was manual and shortly after the seeds were inoculated by spraying the inoculum into the furrow before covering the seeds.

The experiment in randomized blocks had a factorial design (genotypes x environments), with three replications. Each experimental unit consisted of one 5-m-long row. Rows were spaced 0.90 m apart and the density 14 plants per meter. The evaluated area was 3.6 m², since the central part of the rows was harvested, disregarding 0.5 m at either end. Four rows of soybeans were planted around each trial as another border. After plant emergence, seedlings were thinned to the pre-established population density. Weeds were controlled by hand weeding, and pests and diseases by spraying insecticides and fungicides as recommended for the crop.

The following soybean genotypes were evaluated: Conquista, Elite, Garantia, PTN-Bio, M-Soy 8400, M-Soy 8001, Nambu, Sambaíba, Splendor, UFVS 2006, UFVS 2005, UFVTN 102, UFV 18, UFV 16 and Vencedora.

After harvest, samples were sent to the Laboratory of Animal Nutrition, Department of Animal Science of the Universidade Federal de Viçosa (UFV) where the concentrations of oil and protein in the grain-based dry matter (DM%) was determined. For the determination of oil (ether extract) the extraction of petroleum ether was used, according to the method of Goldfish and protein (raw) was determined by the Kjeldahl method (Silva and Queiroz 2002).

For each trait, the assessment of genetic variability among genotypes, the relative accuracy of each experiment and homogeneity of variances were verified by analysis of variance in each environment. After detection of differences between means of cultivars, the combined analysis of variance was performed.

Table 1. Description of the environments of evaluation of 15 soybean genotypes

Environment	Location, sowing date, soil type	Altitude	Latitude
1	Viçosa, growing season 1 (03/12/2004)	650m	20° 45' 20"S
2	Viçosa, growing season 2 (17/12/2004)	650m	20° 45' 20"S
3	Viçosa, growing season 3 (03/01/2005)	650m	20° 45' 20"S
4	Viçosa, growing season 4 (17/01/2005)	650m	20° 45' 20"S
5	Frutal, growing season 1 (14/11/2005), sandy soil	550m	20° 00' 45"S
6	Frutal, growing season 2 (02/12/2005), sandy soil	550m	20° 00' 45"S
7	Frutal, growing season 1 (14/11/2005), clay soil	550m	20° 00' 45"S
8	Frutal, growing season 2 (02/12/2005), clay soil	550m	20° 00' 45"S
9	Pindorama, growing season 1 (03/11/2006)	570m	21° 13' 10"S
10	Pindorama, growing season 2 (20/11/2006)	570m	21° 13' 10"S
11	Pindorama, growing season 3 (07/12/2006)	570m	21° 13' 10"S
12	Pindorama, growing season 4 (23/12/2006)	570m	21° 13' 10"S
13	Pindorama, growing season 5 (09/01/2007)	570m	21° 13' 10"S

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Once GE interaction had been detected, a detailed study of the performance of each cultivar in the changing environment became necessary. This study was performed using the methods of Eberhart and Russell (1966) and of the Extended Centroid Method (Nascimento et al. 2009).

The method of Eberhart and Russell (1966) is based on the analysis of simple linear regression and has the angular coefficient of the regression β_{1i} and the regression deviation σ^2_{di} respectively, as adaptability and stability parameters. Their estimators are given by:

$$\hat{\beta}_{1i} = \frac{\sum\limits_j Y_{ij} I_j}{\sum\limits_j I_j^2},$$

where

Y_{ij}: Mean of genotype i in environment j;

I_J: codified environmental index $\left(I_j = \dfrac{\sum\limits_i Y_{ij}}{g} - \dfrac{\sum\limits_i \sum\limits_j Y_{ij}}{ga} \right)$.

So that

$$\sum\limits_j I_j = 0;$$

$\dfrac{\sum\limits_i Y_{ij}}{g}$: Mean of all genotypes in environment j;

$\dfrac{\sum\limits_i \sum\limits_j Y_{ij}}{ga}$: General mean;

g: number of evaluated genotypes;
a: number of evaluated locations.

$$\hat{\sigma}^2_{d_i} = \frac{MSD_i - MSR}{r}$$

where:

MSD_i: Mean square deviation of genotype i;
MSR: Mean square of the residue;
r: number of replications.

The Extended Centroid method (ECM) is based on the comparison of values of Cartesian distance between the genotypes and seven centroids, from experimental data, whose mean values for each environment are given by: C_1: centroid 1 (maximum general adaptability), whose values in each environment, are represented by the maximum obtained by the set of cultivars studied; C_2: centroid 2 (maximum specific adaptability to favorable environments), whose values of the favorable environments, are represented by the maximum and in the unfavorable, by the minimum obtained in all cultivars studied; C_3: centroid 3 (maximum

specific adaptability to adverse environments), the values of the favorable environments are represented by the minimum and in unfavorable, by the maximum obtained by the set of cultivars; C_4: centroid 4 (minimum adaptability), whose values in each environment, are represented by the minimum obtained by the set of cultivars studied; C_5: centroid 5 (mean general adaptability), whose values in each environment, are represented by the means for the set of cultivars; C_6: centroid 6 (mean specific adaptability to favorable environments), the values of the favorable environments, are represented by the maximum and in the adverse, by the means of all cultivars studied; C_7: centroid 7 (mean specific adaptability to adverse environments), whose values, in favorable environments, are represented by the means and in the adverse, by the maximum obtained by the set of cultivars studied.

After the establishment of the centroids, the principal component analysis was used, obtaining the scores used in a graphical representation. The position of the cultivars in the scatter plot, for the centroids and the values of Cartesian distance between the points (cultivars) to each of the seven centroids allow their classification. In addition, the configuration of the seven centroids generates an arrow-shaped geometric figure, allowing the spatial analysis of the set of genotypes. A measure of spatial probability is calculated using the inverse of the distance between a treatment and the seven points created as a reference, designated ideotypes:

$$P_{d(i,k)} = \frac{\left(\dfrac{1}{d_{ik}} \right)}{\sum\limits_i \dfrac{1}{d_{ik}}}$$

where:

$P_{d(i,k)}$ - is the probability that the standard stability is similar to the kth centroid and;

d_{ik} - is the distance from the ith genotype to the kth centroid.

The statistical analysis was performed using software (Cruz 2006).

RESULTS AND DISCUSSION

The coefficients of variation were 6.9% and 2.7% for the traits oil and protein content content,

respectively. The combined analysis of variance for each trait revealed significant effects of genetic variability among soybean genotypes, significant differences between the effects of environments assessed and the existence of significant interaction between genotypes and environments (GE). Significance was verified by the F test at 1% probability (Table 2).

The presence of significant GE interaction shows that the genotypes are influenced differentially by the environments, which makes the recommendation of new cultivars difficult and requires the use of techniques of adaptability and stability analysis.

Upon characterization by the centroids and the parameter estimates of adaptability and phenotypic stability, for each method and trait, the genotypes were ranked (Tables 4 and 5).

The ranking of genotypes for the trait oil content (OC) by the method of Eberhart and Russell (1966) showed that the genotypes UFV 18 and UFV 2006 may be considered adaptable to favorable environments and low stability. The genotypes Conquista, M-Soy 8400 and UFVTN 102 were considered as generally adaptable and stable and Splendor and Elite, which have good stability as recommendable for adverse environments. The results of the Extended Centroid Method (ECM) show that the general adaptability of 12 of the 15 genotypes was classified as medium (class V). Of these, the ones most likely to belong to the class are: Nambu, M-Soy 8400, Splendor, UFVTN 102 and Conquista. The adaptability of the genotypes PTN-Bio, Garantia and M-Soy 8001 was classified, respectively, as minimum adaptability, mean specific adaptability to favorable environments and mean specific adaptability to unfavorable environments.

For the oil content (OC) three components should be used for a good data interpretation since these components explain 75.70% of the total data variation (Table 3). Figure 1 shows the graphic dispersion of the first three principal components of the 15 genotypes in 13 environments. The graphical analysis, ECM-arrow, of the pattern of genotypic adaptability indicates that most genotypes are close to the center of the arrow formed by the centroids (Figure 2).

For the trait protein content (PrC), adaptability was classified as specific to favorable environments for the genotypes UFV 18, UFVS 2005, Elite, Nambu and Vencedora, all considered to have low stability. Only Splendor was classified as generally adaptable and highly stable. The genotypes M-Soy 8001 and 8400, Conquista and UFVTN 102 were classified as adaptable to adverse environments (Eberhart and Russell 1966). Based on the result obtained by the Extended Centroid methodology, the adaptability of genotype PTN-Bio was classified as maximum general, and of the cultivars M-Soy 8001 and UFVS 2005 as medium specific to favorable environments. The adaptability of the other genotypes was classified as moderate general. The five that are most likely to
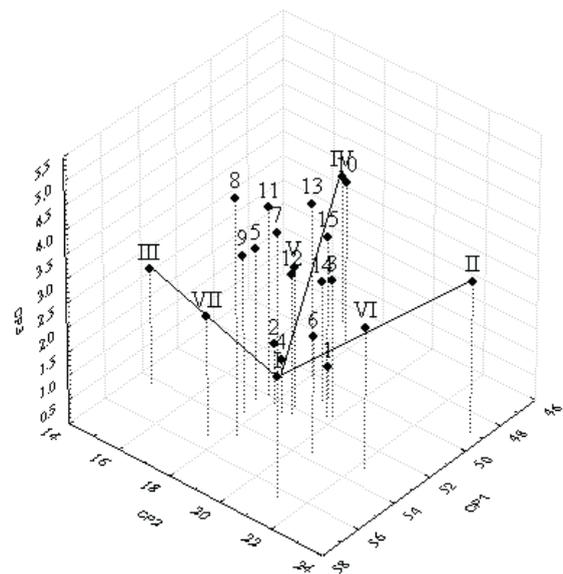
Figure 1. Graphic dispersion of the three first principal components (PC1, PC2 and PC3) of 15 soybean genotypes showing the trait oil content (OC). The seven points numbered with Roman algorisms represent the centroids: I – maximum general adaptability, II – maximum specific adaptability to favorable environments, III – maximum specific adaptability to unfavorable environments, IV – minimum adaptability, V - mean general adaptability, VI - mean specific adaptability to favorable environments, VII - mean specific adaptability to unfavorable environments

Table 2. Analysis of variance of the experiment in randomized blocks, in a factorial design, for the traits: oil content (OC) and protein content (PrC), evaluated in 15 soybean cultivars

Sources of variation	df	Mean squares	
		OC	**PrC**
Blocks/Environments	26	3.63	5.698
Genotypes	14	11.873**	137.632**
Environments	12	125.871**	209.226**
Genotypes x Environments	168	2.787**	4.619**
Error	364	1.484	1.253
Mean		17.691	40.982
CV(%)		6.886	2.731

**: significant at 1%, by the F test
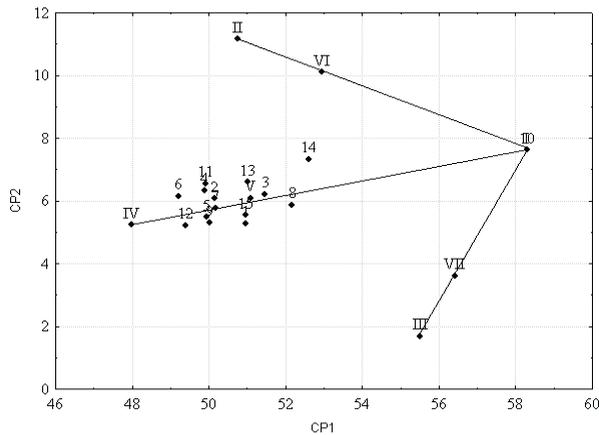
Figure 2. Graphic dispersion in the first two principal components (PC1 and PC2) of 15 genotypes for the trait protein content (PrC). The seven points numbered with Roman algorisms represent the centroids: I – maximum general adaptability, II – maximum specific adaptability to favorable environments, III – maximum specific adaptability to unfavorable environments, IV – minimum adaptability, V - mean general adaptability, VI - mean specific adaptability to favorable environments, VII - mean specific adaptability to unfavorable environments

belong to the class are: Splendor, Conquista, Vencedora, Elite, Garantia and Sambaiba (Table 5).

The first two components account for 91.16% of the total variation (Table 3). Therefore, the most relevant information is contained in these two components. Figure 2 shows the graphic dispersion of 15 genotypes in 13 environments. Again the figure shows a concentration of genotypes around Centroid V (mean general adaptability). Genotype PTN-Bio is located

exactly at the top of the arrow, showing that it is highly promising for higher protein content in view of its classification of maximum general adaptability.

The inclusion of genotype UFV 18 in breeding programs designed to increase the oil and protein contents is therefore indicated, since it is classified as adaptable to favorable environments (Eberhart and Russell) and as moderately generally adaptable (ECM). The results of genotype PTN-Bio were however different for each trait; for oil content the general adaptability was minimal and for protein content maximal (ECM), indicating its use for breeding to increase the protein content. Apart from genotype UFV 18, no genotype was found with an equivalent rating for the traits oil and protein content.

The results also suggest that the Extended Centroid method tends to classify the genotypes as having mean general performance, i.e., mean adaptability for both traits under study. This classification is biologically coherent, since the performance of most variables in nature tends to follow a mean value. Figures 1 and 2 corroborate these results showing that most genotypes are grouped around the Ideotype V (mean general adaptability). Besides, the graphical analysis is an extremely important tool for a combined evaluation of the performance of genotypes. The method of Eberhart and Russell (1966) on the other hand provides a more detailed classification of genotypes. As an example

Table 3. Estimates of the variances (eigenvalues) obtained by principal component analysis and cumulative fraction of variance explained by the components, for the two traits

Oil content (OC)		Protein content (PrC)	
Eigenvalues (%)	% accumulated	Eigenvalues (%)	% accumulated
5.310	40.844	8.169	62.842
3.562	68.245	3.681	91.160
0.969	75.700	0.506	95.056
0.902	82.638	0.215	96.708
0.668	87.775	0.135	97.745
0.495	91.584	0.108	98.573
0.408	94.723	0.059	99.025
0.226	96.463	0.053	99.432
0.181	97.857	0.035	99.702
0.115	98.743	0.027	99.910
0.088	99.418	0.007	99.965
0.054	99.831	0.004	99.997
0.022	100.000	0.000	100.000

Table 4. Classification and probability associated to the genotypes in one of seven groups characterized by the Extended Centroid method (ECM) and estimates of the parameters of adaptability and phenotypic stability, by the method of Eberhart and Russell (1966), for the trait oil content (OC)

Cultivars	Mean	ECM		Eberhart and Russell	
		Classif.	Prob.	β_{i1}	σ^2_{di}
1 - Vencedora	17.552	V	0.225	0.733[+]	0.784[**]
2 - UFVS 2006	17.755	V	0.228	1.271[+]	0.774[**]
3 - Splendor	17.771	V	0.290	0.754[+]	-0.150[ns]
4 - UFV 16	17.866	V	0.253	1.137[ns]	0.419[*]
5 - Nambu	17.645	V	0.299	1.061[ns]	0.076[ns]
6 - M-Soy 8001	18.530	VII	0.217	0.930[ns]	0.663[**]
7 - Sambaiba	18.256	V	0.211	1.001[ns]	0.569[*]
8 - Garantia	18.344	VI	0.213	1.111[ns]	0.885[**]
9 - UFV 18	17.947	V	0.239	1.339[++]	0.032[ns]
10 – PTN-Bio	16.207	IV	0.406	0.952[ns]	0.328[ns]
11 - M-Soy 8400	17.509	V	0.298	1.001[ns]	0.109[ns]
12 - UFVS 2005	17.806	V	0.257	0.895[ns]	0.405[*]
13 - Conquista	17.071	V	0.263	1.048[ns]	0.029[ns]
14 - UFVTN 102	17.467	V	0.278	1.082[ns]	0.212[ns]
15 - Elite	17.639	V	0.265	0.685[++]	0.166[ns]

Classif.: Classification; Prob.: Probability ; [ns]: non-significant; [++] and [+] differ significantly from 1, by the t test, at 5% and 1% probability (H_0: β_{t1} =1); [**] and [*] significant at 5% and 1% probability, by the F test, respectively (H_0: σ^2_{di}=0)

Table 5. Classification and probability associated to the genotypes in one of the seven groups characterized by the Extended Centroid method (ECM) and estimates of parameters of adaptability ($\hat{\beta}_{1i}$) and phenotypic stability ($\hat{\sigma}^2_{di}$) by the method of Eberhart and Russell (1966), for the trait protein content (PrC)

Cultivars	Mean	ECM		Eberhart and Russell	
		Classif.	Prob.	β_{i1}	σ^2_{di}
1 - Vencedora	40.750	V	0.389	1.248[++]	0.768[**]
2 - UFVS 2006	40.204	V	0.358	0.943[ns]	0.878[**]
3 - Splendor	41.270	V	0.470	0.903[ns]	0.252[ns]
4 - UFV 16	40.193	V	0.327	1.005[ns]	1.541[**]
5 - Nambu	39.927	V	0.332	1.212[+]	0.405[*]
6 - M-Soy 8001	39.557	IV	0.317	0.803[+]	0.747[**]
7 - Sambaiba	40.140	V	0.363	1.026[ns]	0.689[**]
8 - Garantia	41.814	V	0.365	1.157[ns]	0.427[*]
9 - UFV 18	39.927	V	0.307	1.324[++]	1.001[**]
10 – PTN-Bio	47.091	I	1.000	0.957[ns]	2.000[**]
11 - M-Soy 8400	40.295	V	0.297	0.723[++]	2.257[**]
12 - UFVS 2005	39.467	IV	0.337	1.171[+]	0.375[*]
13 - Conquista	40.987	V	0.407	0.726[++]	0.595[**]
14 - UFVTN 102	42.462	V	0.265	0.626[++]	0.687[**]
15 - Elite	40.647	V	0.368	1.176[+]	1.379[**]

[1] Classif.: Classification; Prob.: Probability ; [++] and [+] significant by the t test, at 5% and 1% probability (H_0: β_{1i}=1); [**] and [*] significant at 5% and 1% probability, by the F test, respectively (H_0: σ^2_{di}=0)

the performance of the genotypes M-Soy 8001 and UFVS 2005 for the trait protein content (PrC) can be cited; the adaptability of these genotypes was classified as minimum by the Enhanced Centroid method, while by the method of Eberhart and Russell (1966) they were classified as adaptable to adverse and to favorable environments, respectively. This shows that the Extended Centroid method is more conservative than the methodology of Eberhart and Russell (1966) in the classification of genotypes.

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Adaptabilidade e estabilidade de soja quanto ao teor de óleo e proteína

RESUMO - *Objetivou-se neste trabalho avaliar a influência da interação genótipos x ambientes quanto ao teor de óleo e proteína em genótipos de soja (*Glycine max *L. Merrill). Foram avaliados 15 genótipos de soja. Os experimentos foram conduzidos nos anos agrícolas 2004/05, 2005/06 e 2006/07 nos municípios de Viçosa-MG, Frutal-MG e Pindorama-SP, respectivamente, em diferentes épocas de plantio e tipos de solo, perfazendo um total de 13 ambientes. A avaliação foi feita com base nos métodos de Eberhart e Russell e do Centróide Ampliado. Os resultados mostraram que o genótipo UFV 18 é desejável em programas de melhoramento de soja, nos quais se pretende aumentar os valores associados às duas características avaliadas. Já o genótipo PTN-Bio apresentou resultados divergentes para cada caractere. Além disso, percebeu-se que o método Centróide Ampliado é mais conservador que a metodologia de Eberhart e Russell, quando se trata de classificar genótipos.*

Palavras chave: *Glycine max* L. Merrill, interação genótipo x ambiente, Eberhart e Russell, Centróide Ampliado.

REFERENCES

Cruz CD (2006) **Programa Genes: biometria.** Universidade Federal de Viçosa, Viçosa, 382p.

Cucolotto M, Pipolo VC, Garbuglio DD, Fonseca Junior NS, Destro D and Kamikoga MK (2007) Genotype x environment interaction in soybean: evaluation through three methodologies. **Crop Breeding and Applied Biotechnology 7**: 270-277.

Dhingra S and Jood S (2001) Organoleptic and nutritional evaluation of wheat breads supplemented with soybean and barley flour. **Food Chemistry 77**: 479-488.

Eberhart SA and Russell WA (1966) Stability parameters for comparing varieties. **Crop Science 6**: 36-40.

Maehler AR, Costa J, Pires JLS and Rambo L (2003) Qualidade de grãos de duas cultivares de soja em função da disponibilidade de água no solo e arranjo de plantas. **Ciência Rural 33**: 213-218.

McMindes MK (1991) Applications of isolated soy protein in low-fat meat products. **Food Technology 45**: 61-64.

Nascimento M, Cruz CD, Campana ACM, Tomaz RS, Salgado C and Ferreira RP (2009) Alteração no método centroide de avaliação da adaptabilidade genotípica. **Pesquisa Agropecuária Brasileira 44**: 263-269.

Nogueira APO, Sediyama T, Cruz CD, Reis MS, Pereira DG and Jangarelli M (2008) Novas características para diferenciação de cultivares de soja pela análise discriminante. **Ciência Rural 38**: 1-7.

Oliveira MAR, Pípolo VC, Schuster I, Vicente D, Dellagostin M and Oliveira EF (2006) Soybean stability and adaptability in Southern and Central Brazil. **Crop Breeding and Applied Biotechnology 6**: 55-64.

Pelúzio JM, Fidelis RR, Giongo P, Silva JC, Cappellari D and Barros HB (2008) Adaptabilidade e estabilidade de cultivares de soja em quatro épocas de semeadura no sul do Estado do Tocantins. **Revista Ceres 55**: 4-40.

Silva DJ and Queiroz AC (2002) **Análise de alimentos: métodos químicos e biológicos**. Editora UFV, Viçosa, 235p.