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# Differential tolerance of cowpea cultivars to osmotic stress in germinative phase: a multivariate approach

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VISUAL ABSTRACT

# DIFFERENTIAL TOLERANCE OF COWPEA CULTIVARS TO OSMOTIC STRESS IN GERMINATIVE PHASE: A MULTIVARIATE APPROACH

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*Vigna unguiculata* L.

Main sources of vegetable protein

Well adapted to different edaphoclimatic conditions

Water Deficit

Changes

Seed germination

Early development

2

## Cowpea Seeds

BRS-Pujante, BRS-Guariba, BRS-Potengi, BR 17-Gurgueia, BRS-Tumucumaque, BRS-Pajeú, BRS-Rouxinol, BRS-Novaera, BRS-Xiquexique, BRS-Milênio, BRS-Acauã, Patativa, BRS 3-Tracuateua e BRS-Aracê

## Levels of osmotic potential

(0, -0,1, -0,2 and -0,4MPa)

3

## Variables evaluated were:

Final germination percentage (G), Meangerminationtime (MGT), Lengths of the aerial part (LAP), Lengths of the root (LR), Germinationspeedindex (GSI), RDM/APDM ratio, Aerial part dry mass (APDM) and Root drymass (RDM).

Statistical analysis: Principal Component Analysis (PCA) R statistical package v. 4.0.2

4

## Conclusions

- BRS-Pujante as the most sensitive at the -0.4MPa level.
- BRS-Milênio, BRS-Acauã, and BRS-Aracê had difficulties in tolerating the stresses of -0.1 and -0.2 MPa imposed, with emphasis on the cultivar BRS-Milênio at the level -0.2MPa.
- BRS-Novaera and BRS-Pajeú were characterized with possible tolerance at both the -0.1 and -0.2MPa levels.

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# Abstract:

This study aimed to characterize the behavior of different cultivars of *Vigna unguiculata* L. Walp. to osmotic stress, from germination and vigor parameters. The experimental design used was completely randomized, in a 14x4 factorial arrangement, with fourteen cultivars and four levels of osmotic potential (0, -0.1, -0.2, and -0.4 MPa) of the germination solution. BRS-Novaera and BRS-Pajeú cultivars were characterized with possible tolerance at both the -0.1 and -0.2 MPa levels. The study pointed to the BRS-Pujante cultivar as the most sensitive to the -0.4 MPa level. The multivariate technique used allowed for a satisfactory characterization of the treatments adopted.



**Keywords:** *Vigna unguiculata* L. Walp; abiotic stress; germination; vigor; exploratory analysis.

# INTRODUTION

**FEIJÃO-CAUPI (*Vigna unguiculata* L. Walp.)**

Main sources of **vegetable protein**

Well **adapted** to **different edaphoclimatic conditions**

**Water deficit**

Changes

- Seed germination
- Early development

**In Brazil**

Regions

**North and Northeast**

Employment and income

(SILVA et al., 2010; VIJAYKUMAR et al., 2010; SILVA; NEVES, 2011; NASCIMENTO et al., 2011; FREITAS et al., 2017)

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Water limitation ⟷ **OSMOTIC STRESS** ⟷ Speed of germination

**Tends to compromise the initial establishment of the seedling stand**

**Water**

- The starting factor for germination
- Involved in all other stages of germination metabolism
- Shortage of water right after sowing can compromise the entire germination process

(MOŻDŻEŃ et al., 2015; OLIVEIRA; GOMES FILHO, 2010)

Studies with seeds

- Germination
- Vigor in low humidity conditions


✓ Studies related to the tolerance of different cultivars of cowpea to abiotic stresses at these developmental stages are still scarce in the literature

Interpretation of certain studied phenomena

**Multivariate analysis**

Simultaneous evaluation of several variables

- Carrying out studies to evaluate the performance of cultivars during germination and initial seedling growth.

(COLMAN et al., 2014; VICINI et al., 2018; MANLY; ALBERTO, 2019)

# OBJECTIVE

- The objective of this work was to **characterize**, through multivariate data analysis, the **behavior** of different cowpea bean cultivars to **osmotic stress**, based on **germination** and **vigor** variables

# MATERIAL AND METHODS

**Place:** Seed Analysis Laboratory

(Federal University of Ceara)

**Plant material:** Cowpea seeds ⇨ BRS-Pujante, BRS-Guariba, BRS-Potengi, BR 17-Gurgueia, BRS-Tumucumaque, BRS-Pajeú, BRS-Rouxinol, BRS-Novaera, BRS-Xiquexique, BRS-Milênio, BRS-Acauã, Patativa, BRS 3-Tracuateua e BRS-Aracê

**Experimental design:**

Completely randomized, in a 14 x 4 factorial arrangement, with four replications

Levels of osmotic potential ⇦ (**0**, **-0,1**, **-0,2** and **-0,4MPa**)

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## Germination test

Seed selection

Uniform seeds with intact integument

Distilled water

2.5 times the weight of the dry paper

Blotting paper

PEG 6000 solution

2.5 times the weight of the dry paper

**200** seeds → **4 replicates of 50 seeds**

BOD → 25°C

(BRASIL, 2009)

## Germination test

The count of germinated seeds → Daily → Nine days

- **Germination criterion:** the emission of a radicle with at least 2.0 mm (ROSA et al., 2005)

**The germination variables evaluated...**

Final germination percentage (G)

(LABORIAU, 1983)

Germination speed index (GSI)

(MAGUIRE, 1962)

Mean germination time (MGT)

(LABORIAU, 1983)

## Vigor evaluation

**Experimented with the same conditions GT**

**4 repetitions** of **20** seeds

(NAKAGAWA,1999)

## Nine days after sowing:

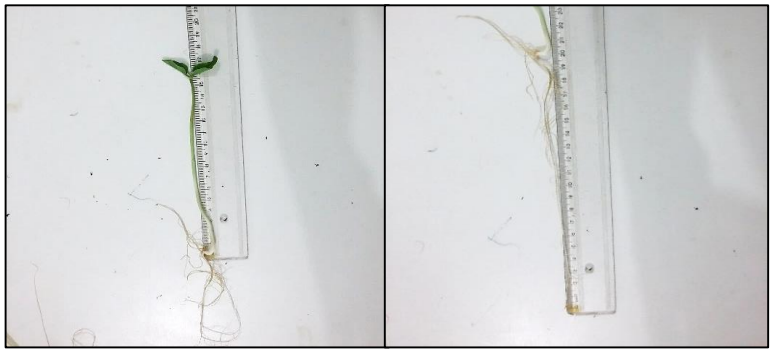
Lengths of the aerial part (LAP)

Lengths of the root (LR)

**Oven 65°C**

Aerial part dry mass (APDM)

Root dry mass (RDM)



The average results were expressed in grams per seedling

RDM/APDM ratio

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## Statistical analysis:

**Principal Component Analysis (PCA)**

Greater **correlation** between the original variables

**Spearman's correlation coefficient**

Shapiro-Wilk test

**p-value < 0.05**

**The data set was standardized ($\mu = 0$; $\sigma^2 = 1$)**

**Kaiser's criterion** → **Number** of main components

**R statistical package v. 4.0.2**

# RESULTS AND DISCUSSION

**Table 1.** Correlation matrix and their respective significance values (p-value) between the studied variables.

| | Variable | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **Variable** | G | GSI | MGT | LR | LAP | RDM | APDM | RDM/ APDM |
| G | 1.00 | | | | | | | |
| GSI | 0.93** | 1,00 | | | | | | |
| p-value | (<0.0001) | | | | | | | |
| MGT | -0.83** | -0.93** | 1.00 | | | | | |
| p-value | (<0.0001) | (<0.0001) | | | | | | |
| LR | 0.83** | 0.88** | -0.89** | 1,00 | | | | |
| p-value | (<0.0001) | (<0.0001) | (<0.0001) | | | | | |
| LAP | 0.87** | 0.94** | -0.87** | 0.86** | 1.00 | | | |
| p-value | (<0.0001) | (<0.0001) | (<0.0001) | (<0.0001) | | | | |
| RDM | 0.82** | 0.82** | -0.80** | 0.86** | 0.85** | 1.00 | | |
| p-value | (<0.0001) | (<0.0001) | (<0.0001) | (<0.0001) | (<0.0001) | | | |
| APDM | 0.88** | 0.92** | -0.86** | 0.88** | 0.95** | 0.91** | 1.00 | |
| p-value | (<0.0001) | (<0.0001) | (<0.0001) | (<0.0001) | (<0.0001) | (<0.0001) | | |
| RDM/ APDM | 0.33* | 0.25 | -0.28* | 0.35** | 0.23 | 0.42** | 0.22 | 1.00 |
| p-value | (0.013) | (0.062) | (0.039) | (0.008) | (0.090) | (0.001) | (0.108) | |

G= Final Germination Percentage; GSI= Germination Speed Index; MGT= Average Germination Time; LR= length of the root; LAP= Length of the Aerial Part; RDM= Root dry mass; APDM= Aerial Part Dry Mass; RDM/APDM= Root Dry Mass/Aerial Dry Mass Ratio. ** Significant correlation at the 0.01 level; * Significant correlation at the 0.05 level.

Evidence of a correlation between the characteristics

The use of multivariate analysis techniques is adequate

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**Table 2.** Weight coefficients (eigenvectors), eigenvalues, explained variance (EV), and accumulated explained variance (AEV) for each principal component, based on the studied variables.

| Variable | CP1 | CP2 | CP3 | CP4 | CP5 | CP6 | CP7 | CP8 |
|---|---|---|---|---|---|---|---|---|
| G | 0.366 | 0.270 | -0.012 | 0.688 | -0.212 | -0.085 | -0.466 | -0.225 |
| GSI | 0.397 | -0.042 | -0.157 | 0.401 | -0.060 | 0.105 | 0.735 | 0.321 |
| MGT | -0.359 | 0.050 | 0.829 | 0.358 | 0.164 | 0.011 | 0.149 | 0.065 |
| LR | 0.394 | 0.050 | 0.100 | -0.110 | 0.610 | -0.666 | -0.012 | 0.061 |
| LAP | 0.340 | -0.444 | 0.108 | 0.028 | 0.460 | 0.570 | -0.090 | -0.361 |
| RDM | 0.382 | 0.145 | 0.395 | -0.374 | -0.437 | -0.094 | 0.284 | -0.507 |
| APDM | 0.362 | -0.342 | 0.330 | -0.179 | -0.308 | 0.050 | -0.356 | 0.622 |
| RDM/ APDM | 0.177 | 0.765 | 0.070 | -0.230 | 0.234 | 0.449 | -0.068 | 0.254 |
| eigenvalues | **5.98** | **1.34** | 0.29 | 0.19 | 0.10 | 0.05 | 0.03 | 0.02 |
| EV (%) | **74.77** | **16.73** | 3.61 | 2.36 | 1.31 | 0.62 | 0.38 | 0.22 |
| AEV (%) | **74.77** | **91.50** | 95.11 | 97.47 | 98.77 | 99.39 | 99.78 | 100 |

Kaiser (1958)

**Table 2.** Weight coefficients (eigenvectors), eigenvalues, explained variance (EV), and accumulated explained variance (AEV) for each principal component, based on the studied variables.

| Variable | CP1 | CP2 |
|---|---|---|
| G | 0.366 | 0.270 |
| GSI | 0.397 | -0.042 |
| MGT | -0.359 | 0.050 |
| LR | 0.394 | 0.050 |
| LAP | 0.340 | -0.444 |
| RDM | 0.382 | 0.145 |
| APDM | 0.362 | -0.342 |
| RDM/APDM | 0.177 | 0.765 |
| eigenvalues | **5.98** | **1.34** |
| EV (%) | **74.77** | **16.73** |
| AEV (%) | **74.77** | **91.50** |

**Germinal performance and root growth**

$$CP1 = 0.366X_1 + 0.397X_2 - 0.359X_3 + 0.394X_4 + 0.340X_5 + 0.382X_6 + 0.362X_7 + 0.177X_8$$

**Differences in seedling size**

$$CP2 = 0.270X_1 - 0.042X_2 + 0.050X_3 + 0.050X_4 - 0.444X_5 + 0.145X_6 - 0.342X_7 + 0.765X_8$$

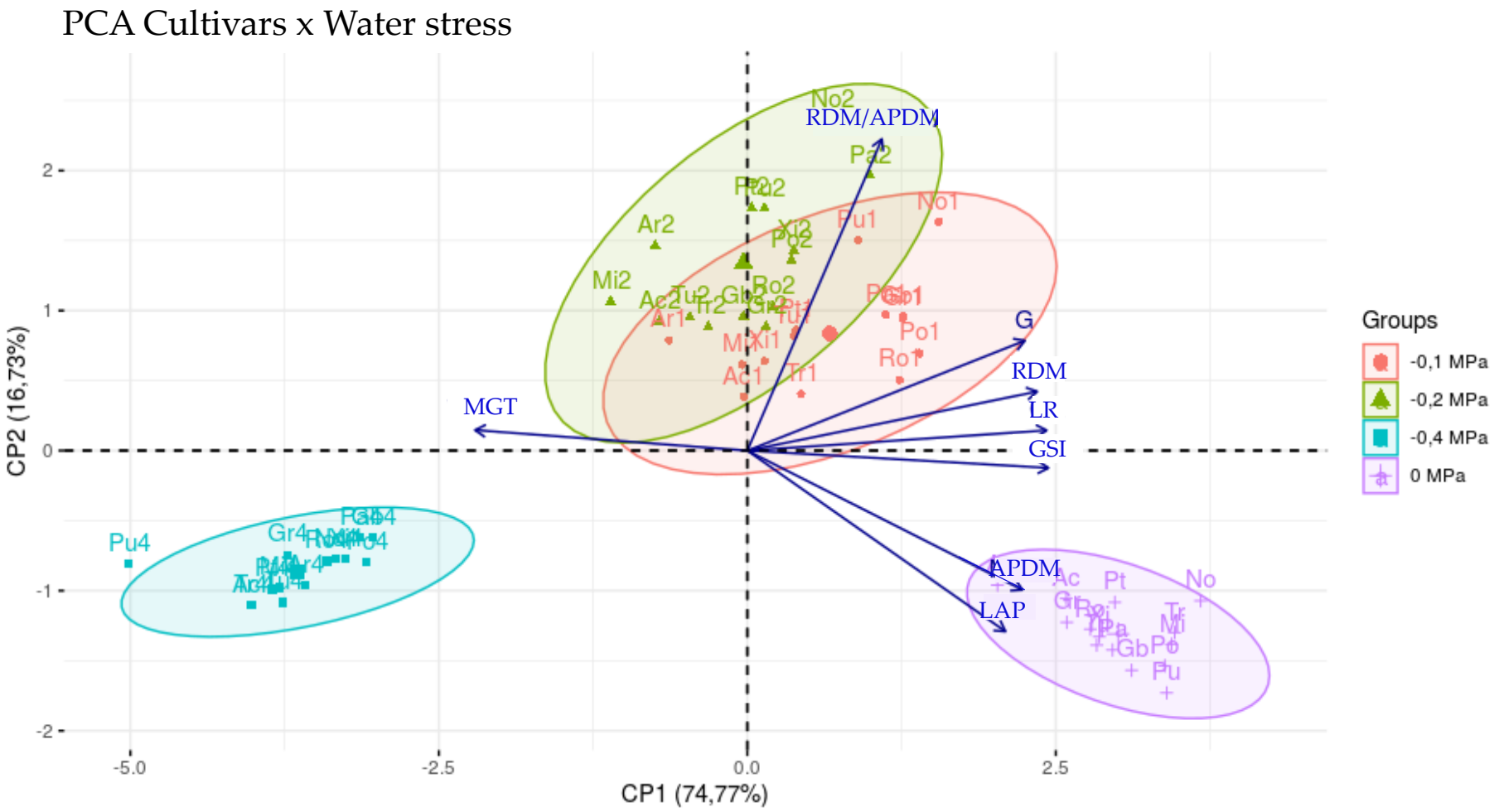


**Figure 1.** Biplot showing the relationship between variables and treatments for the first two main components (CP1 e CP2). Pu = BRS-Pujante; Gb = BRS-Guariba; Po = BRS-Potengi; Gr = BR 17-Gurgueia; Tu = BRS-Tumucumaque; Pa = BRS-Pajeú; Ro = BRS-Rouxinol; No = BRS-Novaera; Xi = BRS-Xiquexique; Mi = BRS-Milênio; Ac = BRS-Acauã; Pt = Patativa; Tr = BRS-Tracuateua; Ar = BRS-Aracê.

# RESULTS AND DISCUSSION

Bewley; Black (1994) and Taiz et al. (2017)

Reductions in seedling LAP values can be explained by the decrease in seed metabolism since there is **less** water availability for the **digestion of reserves** and **translocation of metabolized products.**

Agostini et al. (2013)

**These decreases in the GSI and increases in the MGT**, due to the increase in the expressiveness of stress, are widely reported in the literature

Bewley et al., 2013 and Pelegrini et al., 2013

Answer shows adequate water availability.

Scalon et al. (2011)

The water deficit condition suggests a prioritization of root growth, an important feature in the escape from this type of stress, where it can favor water absorption precisely by **increasing the surface of contact** with the substrate.

Machado Neto et al. (2004)

When seeds are subjected to water deficiency by osmotic solutions, vigor is more affected than germination.

Soares et al. (2015)

Ferreira et al. (2017)

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# CONCLUSIONS

- The Principal Component Analysis allowed the characterization of the treatments, pointing the cultivar **BRS-Pujante** as the most sensitive at the **-0.4MPa level**.
- The technique showed that the cultivars **BRS-Milênio, BRS-Acauã, and BRS-Aracê** had difficulties in tolerating the stresses of **-0.1 and -0.2 MPa** imposed, with emphasis on the cultivar **BRS-Milênio** at the level **-0.2MPa**.
- The cultivars **BRS-Novaera and BRS-Pajeú** were characterized with **possible tolerance at both the -0.1 and -0.2MPa levels**.

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# ACKNOWLEDGMENTS


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