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# Osmopriming and its relationship with the quality of *Urochloa decumbens* seeds under water deficit

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## ABSTRACT

The use of high-quality *Urochloa decumbens* seeds is an essential factor for the establishment of pastures, and osmopriming under stress conditions may improve seed performance. This study was conducted at the Universidade Federal de Viçosa in a completely randomized design and aimed to evaluate the effects of osmopriming and its relationship with physical and physiological attributes of *U. decumbens* seeds subjected to water deficit. Seeds from ten lots were primed in $H_2O$ (0 MPa) and PEG (−0.8 MPa), $KNO_3$ (0.2%), and SNP (0.10 mmol $L^{-1}$) solutions for 24 hours. Unprimed seeds were used as control. The seeds were evaluated in two trials after osmopriming. Trial I consisted of the X-ray test to evaluate physical attributes and germination and vigor tests. In Trial II, the seeds were placed to germinate under favorable (0 MPa) and water deficit conditions (−0.2 MPa) to evaluate physiological attributes. In general, osmopriming of *U. decumbens* seeds assisted in increasing physical attributes such as area, gray scale, perimeter, and tissue density. Osmopriming does not contribute to the best physiological performance of *U. decumbens* seeds under ideal germination conditions (0 MPa). However, osmopriming with SNP (0.10 mmol $L^{-1}$) and PEG (−0.8 MPa) for 24 hours contributes to the better physiological performance of seeds under water deficit.

**Keywords:** Image analysis, Seed priming, Water stress, Forage species, Germination

## Osmocondicionamento e sua relação com a qualidade de sementes de *Urochloa decumbens* sob déficit hídrico

## RESUMO

O uso de sementes de *Urochloa decumbens* de alta qualidade é um fator essencial para o estabelecimento das pastagens e o condicionamento osmótico, sob condições de estresse pode auxiliar no melhor desempenho das sementes. O trabalho foi conduzido na Universidade Federal de Viçosa, em delineamento inteiramente casualizado e teve como objetivo avaliar os efeitos do condicionamento osmótico e sua relação com os atributos físicos e fisiológicos das sementes de *U. decumbens* submetidas ao déficit hídrico. Sementes de 10 lotes foram condicionadas em $H_2O$ (0 MPa) e em soluções de PEG (−0,8 MPa), $KNO_3$ (0,2%) e SNP (0,10 mmol $L^{-1}$) por 24 h. Sementes não condicionadas foram utilizadas como controle. Após o condicionamento, as sementes foram avaliadas em dois ensaios. No Ensaio I, foi realizado o teste de raios X para avaliação de atributos físicos, testes de germinação e vigor. No Ensaio II, as sementes foram colocadas para germinar em condições favoráveis (0 MPa) e em situação de déficit hídrico (−0,2 MPa), avaliando-se atributos fisiológicos. Em geral, o condicionamento osmótico de sementes *U. decumbens* auxiliou para o aumento de atributos físicos como área, escala de cinza, perímetro e densidade tecidual. Em condições ideais de germinação (0 MPa), o condicionamento osmótico não contribui para o melhor desempenho fisiológico de sementes de *U. decumbens*. No entanto, o condicionamento osmótico com SNP (0,10 mmol $L^{-1}$) e PEG (−0,8 MPa) por 24 h contribui para o melhor desempenho fisiológico das sementes sob déficit hídrico.

**Palavras-chave**: Análise de imagens, Condicionamento osmótico, Estresse hídrico, Espécie forrageira, Germinação.

## 1. Introduction

In Brazil, pasture areas occupy approximately 163 million hectares intended for raising beef and dairy cattle, standing out in the states of Minas Gerais, Mato Grosso, and Goiás (MAPA, 2019). It represents significant economic impacts, with a turnover of around 913.14 billion reais (ABIEC, 2022). *Urochloa* is the most representative among the forage genera, occupying 80% of the country's pasture areas. However, most pasture areas still have low-production technology despite this importance (ABIEC, 2018). In this sense, using seeds that have high pure live seed, in addition to germination and vigor, which are determining attributes for achieving success in commercializing seeds and implementing pastures, is essential (Zanuzo et al., 2015).

*Urochloa* seeds have physiological dormancy postharvest, which makes seedling emergence difficult, causing uneven pasture establishment. It can lead to the harvesting of seeds with low physical purity due to the large proportion of impurities and inert materials (Cardoso et al., 2015). Furthermore, soil and climate conditions are not always suitable for rapid establishment in the field, which justifies the use of treatments that reduce the time needed between sowing and the emergence of seedlings, resulting in higher security in obtaining desired plant populations per area.

Osmopriming stands out among the techniques that have been studied for this and other purposes. It consists of controlled hydration of seeds without allowing the radicle to be emitted. The use of osmopriming has stood out as a pre-sowing treatment that aims to provide rapid and uniform germination and emergence, in addition to increasing seed tolerance levels to unfavorable environmental conditions such as water deficit and others (Lei et al., 2021). The technique has been widely applied to small-sized seeds such as lavender (Benadjaoud et al., 2022), tomato (Silveira et al., 2023), and carrot (Rosińska et al., 2023). Therefore, it also has the potential to be applied to forage grass seeds, helping to increase the speed and uniformity of seed germination with positive impacts on pasture formation (Batista et al., 2016). Osmopriming can be performed using different osmotic solutions, such as spermidine (Lopes et al., 2018), gibberellin (Li et al., 2019), sodium nitroprusside (SNP) (Kaiser et al., 2016), polyethylene glycol (PEG-6000) (Lei et al., 2021), and sodium selenate (Silveira et al., 2023). Oliveira et al. (2021) evaluated the effect of osmopriming with SNP in *U. brizantha* seeds and found an improvement in the physiological performance of seeds under water and saline stress conditions.

Another promising technique for evaluating the effects of this type of treatment is X-ray image analysis. This technique allows the physical quality to be characterized by providing information on damage and other internal characteristics of the seeds, which can be associated with the physiological potential (Medeiros et al., 2019). It is a non-destructive, fast, accurate analysis that does not present subjectivity in the interpretation of results. Medeiros et al. (2020) evaluated the analysis of X-ray images on *U. ruziziensis* seeds and observed that physical attributes obtained by the X-ray test such as tissue density and seed filling were highly correlated with physiological quality, efficiently helping to carry out their large-scale phenotyping. The physical attributes obtained by the X-ray technique in *U. decumbens* seeds were highly correlated with germination and vigor, showing the potential of the technique for this species (Ramos et al., 2022).

However, studies related to osmopriming of *U. decumbens* seeds have been still scarce and inconclusive, especially when evaluating the effects of the technique on seeds subjected to water deficit conditions and associating the results obtained from the evaluation of physiological quality with those obtained from X-ray tests. In this context, this study aimed to evaluate the effects of osmopriming and its relationship with the physical and physiological attributes of *U. decumbens* seeds subjected to water deficit.

## 2. Material and Methods

This research was conducted at the Laboratory of Seed Analysis of the Department of Agronomy at the Universidade Federal de Viçosa (UFV). Two trials were conducted using seeds of *Urochloa decumbens*, which were supplied by the Brazilian Agricultural Research Corporation (EMBRAPA).

The trial I consisted in the relationship between osmopriming, the physical attributes of seeds by the X-ray test, and the physiological potential of *U. decumbens* seeds. Initially, three replications of 20 seeds from two lots were used and subjected to determination of the degree of moisture using the oven method at 105 ± 3 °C for 24 hours. The results were expressed as a percentage (wet basis) (Brasil, 2009). Once the initial moisture content of the seeds was known, two replications of 50 seeds from each lot were used to obtain the imbibition curve in water and a 0.2% $KNO_3$ solution. The seeds were weighed to obtain the initial weight of the samples and then placed to soak between two sheets of paper towel moistened with a volume of water or $KNO_3$ solution equivalent to 2.5 times the weight of the dry paper in gerbox boxes. The boxes were covered and remained in a BOD incubator at 25 °C for five days.

The water absorption rate by the seeds was monitored using the successive weighing method every 2 hours during the first 10 hours and, subsequently,

every 12 hours until root protrusion in approximately 50% of the seeds. The seeds were removed from the substrate at each interval and weighed to obtain an estimate of their final degree of moisture, calculated based on their initial weight and initial degree of moisture (Hampton and TeKrony, 1995). In the application of treatments, the imbibition curve allowed observing that root protrusion occurred around 100 hours after assembly (Figure 1). Thus, a 24-hour soaking time was defined to carry out osmopriming, which allowed seed hydration until Phase II of the germination process, when the seeds had active metabolism but without radicle emission.

After carrying out the imbibition curve, 40 g of 10 lots of seeds were primed in osmotic solutions using the aerated solution method. The seeds were placed in Erlenmeyer flasks containing 250 mL of each osmopriming solution: distilled water ($H_2O$) (0 MPa), polyethylene glycol (PEG 6000) at −0.8 MPa, 0.2% potassium nitrate ($KNO_3$) (Cardoso et al., 2015), and 0.10 mmol $L^{-1}$ sodium nitroprusside (SNP) (Oliveira et al., 2021). The Erlenmeyer flasks were sealed, supplied with constant aeration using an air pump, and kept in a BOD incubator at 25 °C for 24 hours (Pereira et al., 2018). Unprimed seeds were used as control. The seeds were washed in running water after osmopriming and then dried with paper towels for 72 hours in a laboratory environment at a temperature of 20 ± 2 °C.

*X-ray test:* After osmopriming, 100 seeds from each lot were chosen at random and were positioned with the embryonic axis facing downwards and orderly fixed on adhesive paper to enable individual identification in subsequent analyses. Then, the seeds were placed inside an MX-20 Faxitron digital X-ray equipment (Faxitron X-ray Corp., Wheeling, IL, USA). The equipment was set with a radiation exposure time of 10 seconds, voltage of 23 kV, focal length of 41.6 cm, and image contrast calibrated at 1.6383 (width) x 3.124 (center) to generate the radiographic images. The generated digital images were saved on a computer in TIFF format, processed, and then analyzed. Subsequently, the seeds were processed and evaluated using ImageJ® software.

The following variables were determined after processing: Area: Selection obtained in square pixels and later in units of square millimeters ($mm^2$); Perimeter: Length of the outer boundary of the selection, in millimeters (mm); Mean gray scale: Refers to the sum of the gray values of all pixels in the selection area divided by the number of pixels in the selection, obtaining the mean seed density (gray $mm^{-1}$); Integrated density: Product of the area by the mean gray scale (gray $mm^2$ $pixel^{-1}$).

The seeds from each lot described above were subjected to the following tests to assess physiological quality after acquisition and analysis of the radiographic images. The germination, conducted in gerbox boxes with sheets of germitest paper moistened with distilled water at a proportion of 2.5 times the weight of the paper and maintained in a BOD with alternating temperatures of 20–35 °C; four replications of 50 seeds were used. Assessments were performed daily to obtain the percentage of root emission (≥ 2 mm) and normal seedlings at seven (first germination count) and 21 days (Brasil, 2009). The results were expressed as a percentage. The radicle emission speed index (RESI) and germination speed index (GSI) were obtained based on the daily count of germinated seeds, calculated using the equation proposed by Maguire (1962).

Four replications of 50 seeds were used for each treatment. First, the seeds from each replication were weighed on a scale with precision to four decimal places (0.0001 g). They were then immersed in 50 mL of distilled water and kept at a temperature of 25 °C in a germinator. Readings were taken after 24 hours. The obtained weight was divided by the reading and the result was expressed in µS $cm^{-1}$ $g^{-1}$ (Melo et al., 2019).

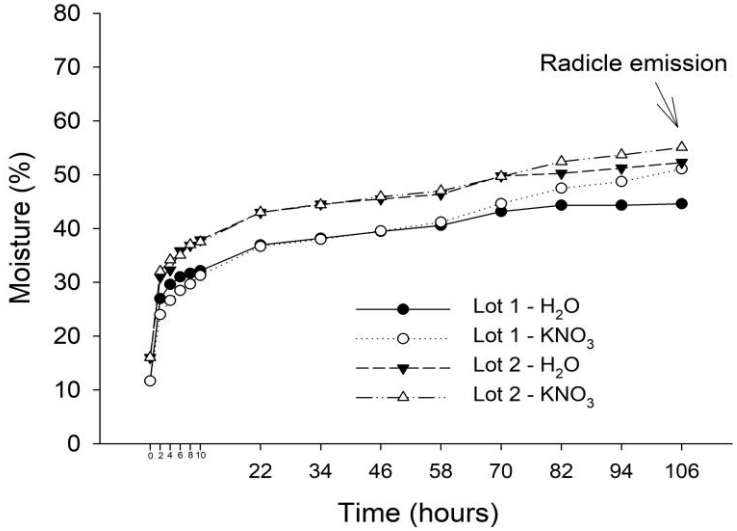


**Figure 1.** Imbibition curve of two lots of *U. decumbens* seeds.

For tetrazolium test, three replications of 20 seeds were used for each treatment. The seeds were pre-primed between paper moistened with distilled water of 2.5 times the weight of the paper for 16 hours to allow slower and more uniform water absorption (Custódio and Aguiar, 2020). Subsequently, the seeds were longitudinally cut to visualize the embryo, separating each of the halves, being then immersed in a 0.1% tetrazolium solution for 3 hours at a temperature of 30 °C without the presence of light. Subsequently, the tetrazolium solution was drained and the seeds were washed under running water. A stereoscopic microscope was used to differentiate viable and non-viable regions (Custódio and Aguiar, 2020).

The seedling length was measured using four replications of 20 seeds for each treatment, distributed on a dashed line in the upper third of the germination paper in the longitudinal direction. Subsequently, each replication was placed in a BOD at 25 °C and then evaluated after seven days. After this period, the seedlings were placed on a blue photographic base made of ethylene-vinyl acetate (EVA) along with a ruler graduated in cm, and measurements were taken using ImageJ® software. Firstly, the 10-mm horizontal line was calibrated to define the values in the *Set scale*, using the measurement value of the graduated ruler as a basis. After calibration, the *Segmented line* tool was used to measure the shoot and root of each seedling. The data were saved in a spreadsheet and analyzed using the Seedcalc package in the R software (Silva et al., 2019).

The seedling endosperms were removed using a scalpel after carrying out the growth test, and the seedlings from each replication were placed in paper bags and maintained in a forced-air circulation oven at 65 °C for 72 hours to determine dry matter. Subsequently, the samples were removed from the oven and placed in a desiccator. Then, weighing was conducted on a scale with precision to four decimal places (0.0001 g), and the weight obtained was divided by the number of seedlings of the respective replication. The result was expressed in mg seedling$^{-1}$ (Nakagawa, 2020).

The Trial II consisted in the relationship between osmopriming and the performance of *Urochloa decumbens* seeds under water stress. Seeds from the 10 lots were placed to germinate under water deficit conditions. To this end, four replications of 50 seeds, distributed on paper towels moistened with PEG 6000 solution at −0.2 MPa in gerbox boxes, were used per treatment. The control treatment was moistened only with distilled water (0 MPa). The boxes were maintained in a germinator at alternating temperatures of 20–35 °C with an 8-hour photoperiod (Brasil, 2009). Analyses of radicle emission, radicle emission speed index, germination, germination speed index, and seedling length were carried out as described in Trial I.

*Experimental design and statistical analysis:* The two trials were conducted in a completely randomized design with four replications. The data were tested for normal distribution of errors using the Shapiro-Wilk test and homogeneity of variances using the Bartlett test. The means obtained for each treatment in Trial I were compared using the Tukey test at a 5% probability. The data from Trial II were analyzed in a 2 (with and without water deficit) x 5 (osmopriming methodologies) double factorial scheme. The means obtained for each osmopriming treatment were compared using the Tukey test at a 5% probability, while the means for water deficit treatments were compared using the F-test at a 5% probability.

## 3. Results and Discussion

For the Trial I, the analysis of physical attributes obtained by the X-ray test on seeds of lot 1 showed that the unprimed seeds (control) presented lower values for the attributes of area (Figure 2A), gray scale (Figure 2B), perimeter (Figure 2C), and integrated density (Figure 2D). It probably occurred because the unprimed seeds had not previously soaked in water, which contributed to their smaller size. The radiographic images also indicated a larger size of primed seeds, which probably occurs because osmopriming led to the hydration of tissues and reactivation of seed metabolism, resulting in higher translocation and consumption of their reserves, thus reflecting an increase in seed size (Marcos-Filho, 2015).

Seeds from lot 2 were similar to lot 1, with a difference between treatments for all physical variables obtained by the X-ray test. The area of the control seeds did not differ from the $KNO_3$ treatment. The treatments with $H_2O$ and SNP presented higher values for this variable (Figure 3A). The variables gray scale and perimeter (Figures 3B and 3C) showed that the control seeds had lower values, followed by PEG and $H_2O$, and superior results in seeds osmoprimed with $KNO_3$ and SNP. The integrated seed density (Figure 3D) varied in the seeds of all treatments, with the control seeds being inferior and SNP being superior to the other treatments.

Figure 4 shows a similar trend to that observed in the X-ray test of the seeds, with lower filling and tissue density of unprimed seeds (Figure 4A) than primed ones (Figure 4B). Colors closer to red in the three-dimensional representation of density represent higher density and colors closer to blue represent lower density, reinforcing the observed results (Figure 4). Similarly, osmopriming with PEG provided higher tissue density and mean of gray in tomato seeds than unprimed ones (Silveira et al., 2023).

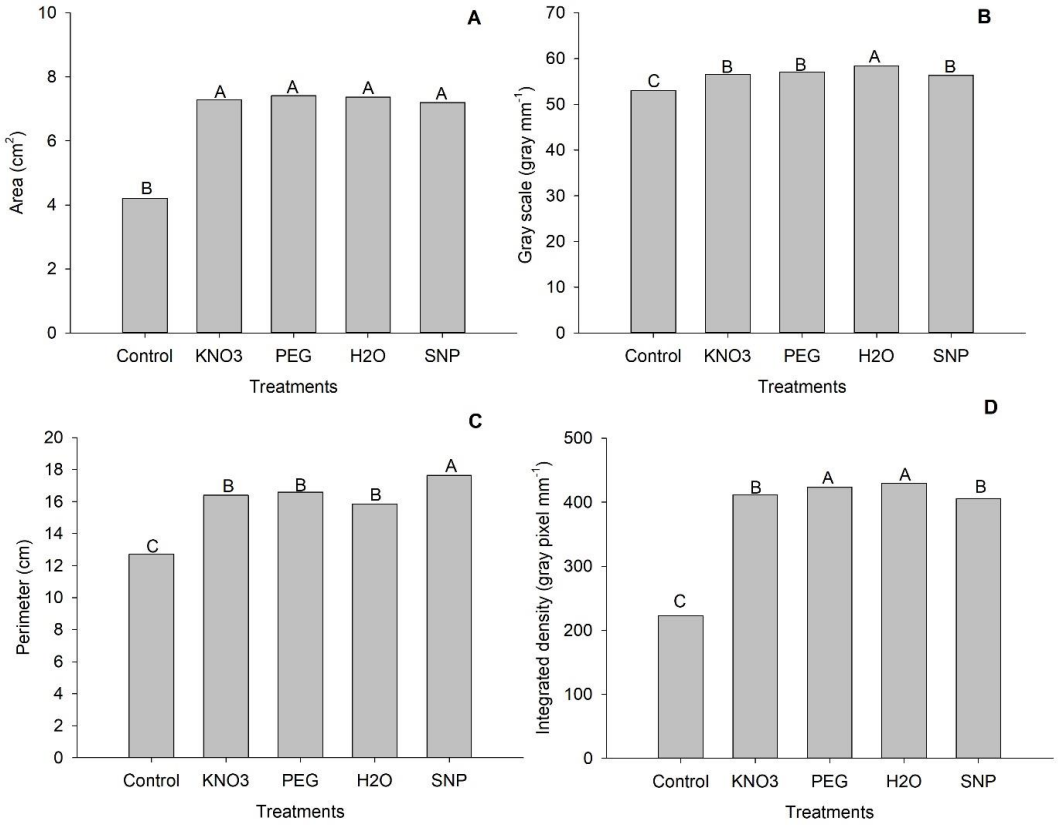


**Figure 2.** Area (A), gray scale (B), perimeter (C), and integrated density (D) of lot 1 of *Urochloa decumbens* seeds subjected to different osmopriming treatments. Different letters differ from each other using the Tukey test at a 5% probability.

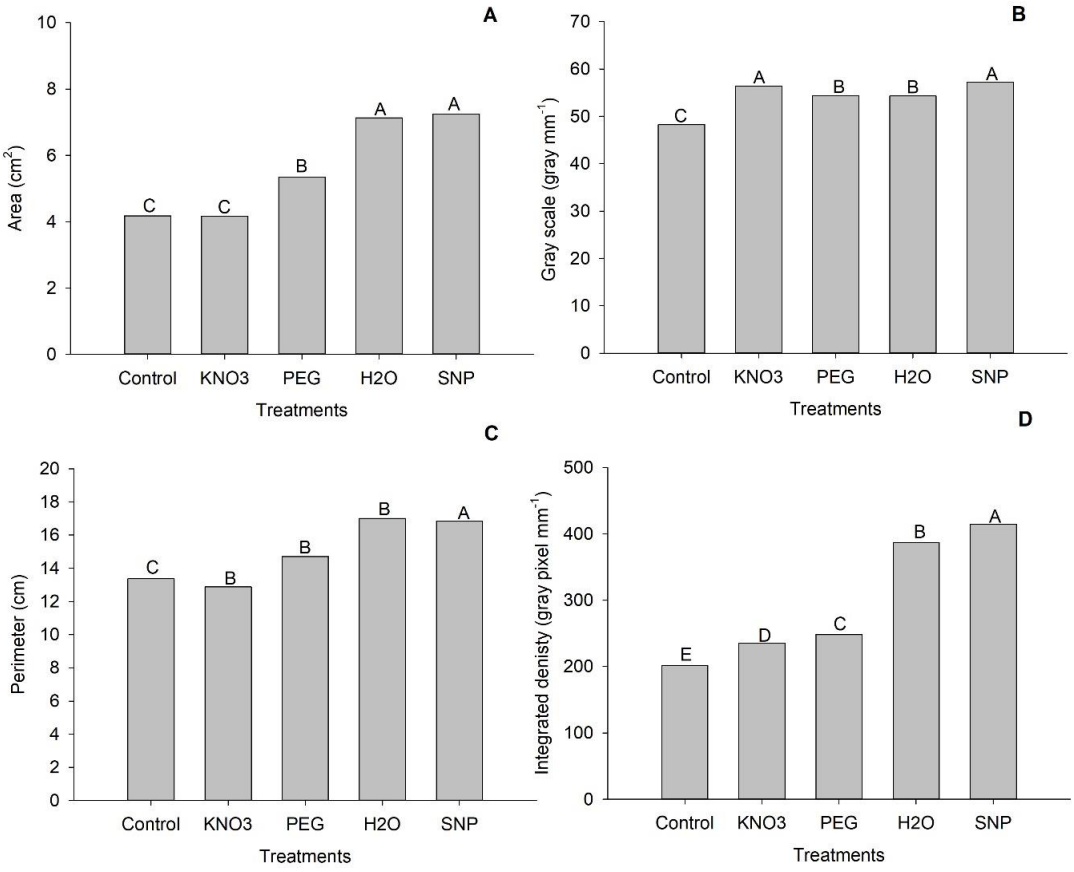


**Figure 3.** Area (A), gray scale (B), perimeter (C), and integrated density (D) of lot 2 of *Urochloa decumbens* seeds subjected to different osmopriming treatments. Different letters differ from each other using the Tukey test at a 5% probability.

Considering the physiological variables of lot 1, unprimed seeds (control) performed better in most variables compared to seeds primed with $KNO_3$, PEG, $H_2O$, and SNP (Table 1). In general, and different from what was observed in this study, osmopriming has been used and shown physiological benefits in seeds of different species (Batista et al., 2016; Lopes et al., 2018; Lei et al., 2021; Silveira et al., 2023). However, when considering only the osmopriming treatments ($KNO_3$, PEG, $H_2O$, and SNP), SNP provided better performance for radicle emission (RE), germination (G), seedling lengths (SL), and dry matter (DM).

SNP stands out for being a donor of nitric oxide (NO), an inorganic radical synthesized from L-arginine (Kolbert et al., 2021), which acts in several cellular processes, stimulating germination and breaking dormancy of several species, such as mustard (Rather et al., 2020), tomato (Khan et al., 2021), and pea (Sekita et al., 2022). Similarly, Oliveira et al. (2021) observed positive results in the germination of *U. brizantha* when primed with SNP, showing the positive role of this compound. In contrast, the variables of germination speed index (GSI) and tetrazolium test (TZ) stand out for the osmopriming carried out with PEG. Treatments with $H_2O$ and $KNO_3$ provided lower seed performance than the other osmopriming treatments (Table 1).

The physiological variables obtained from seeds of lot 2 show that unprimed seeds (control) had better physiological performance through the variables of radicle emission (RE), radicle emission speed index (RESI), germination (G), germination speed index (GSI), tetrazolium (TZ), and seedling length (SL), as observed for lot 1 (Table 2).

Therefore, a similar interpretation can be made relative to lot 1, as osmopriming with $KNO_3$, PEG, $H_2O$, and SNP showed no positive effects on the physiological performance of the seeds compared to the control. However, seeds primed with PEG had better physiological performance when than the other treatments ($KNO_3$, $H_2O$, and SNP), differently from what was observed for lot 1. Similarly, Armondes et al. (2016) evaluated cabbage seeds and reported that osmopriming with PEG provided an increase in the percentage and speed of germination.

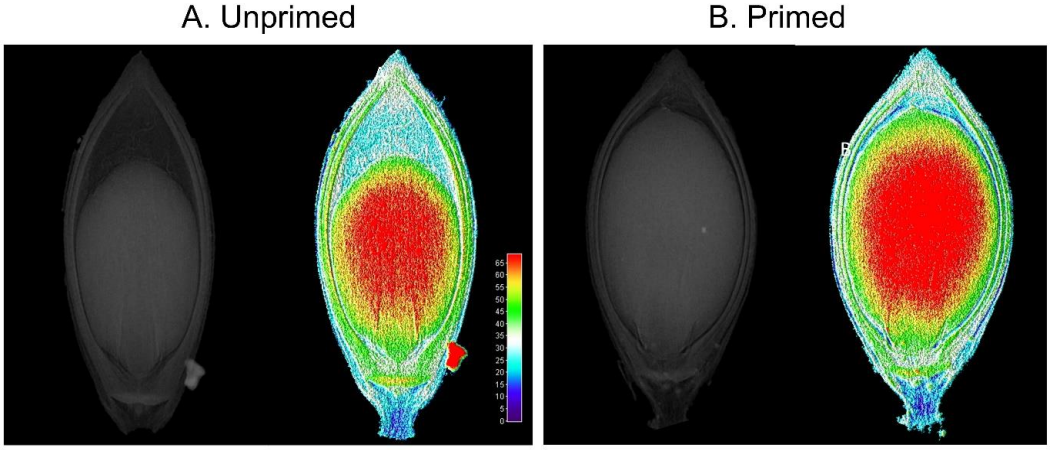


**Figure 4.** Radiographic images and three-dimensional representation of the density of unprimed (A) and primed (B) *Urochloa decumbens* seeds.

**Table 1.** Radicle emission (RE), radicle emission speed index (RESI), germination (G), germination speed index (GSI), electrical conductivity (EC), tetrazolium (TZ), seedling length (SL), and dry matter (DM) of lot 1 of *U. decumbens* seeds subjected to different osmopriming treatments.

| Treatment | RE (%) | RESI (index) | G (%) | GSI (index) | EC ($\mu S\ cm^{-1}\ g^{-1}$) | TZ (%) | SL (mm seedling$^{-1}$) | DM (mg seedling$^{-1}$) |
|---|---|---|---|---|---|---|---|---|
| Unprimed | 71 A | 9.60 A | 71 A | 9.61 A | 51.12 B | 74 A | 8.88 A | 0.057 A |
| $KNO_3$ | 48 C | 4.41 C | 48 C | 3.80 C | 55.03 A | 45 D | 3.39 D | 0.034 C |
| PEG | 47 C | 6.41 B | 47 C | 5.30 B | 41.47 B | 63 B | 4.61 C | 0.035 C |
| $H_2O$ | 37 D | 4.59 C | 37 D | 3.32 C | 36.75 C | 54 C | 5.14 B | 0.039 C |
| SNP | 55 B | 5.88 B | 59 B | 4.26 C | 34.44 C | 56 C | 8.20 A | 0.048 B |
| $F_{0.05}$ | 17.5* | 45.1* | 27.8* | 99.7* | 40.8* | 29.7* | 339.1* | 100.1* |
| CV (%) | 9.5 | 9.8 | 9.3 | 9.7 | 6.3 | 6.7 | 3.8 | 7.4 |

* = significant by F-test at a 5% probability; F = calculated F-value; CV = coefficient of variation. Means followed by uppercase letters in the column compare the osmopriming treatments by the Tukey test at a 5% probability.

Dantas et al. (2021) evaluated osmopriming with PEG-6000 at a potential of −0.2 MPa in coriander seeds for different periods and observed higher root growth and seedling dry matter after 24 hours. Osmopriming with $H_2O$ and SNP on seeds from lot 2 showed intermediate results, as osmopriming with $KNO_3$ was less effective for the physiological variables of radicle emission (31%), radicle emission speed index (1.92), germination (28%), and germination speed index (1.92) (Table 2).

Unlike what was observed in this study, Ramos et al. (2022) observed that *U. decumbens* seeds with higher germination potential and vigor showed significant correlations with some of the physical variables obtained by the X-ray test, such as area, perimeter, and tissue density. Furthermore, as already mentioned, several other studies have demonstrated the benefit of osmopriming when compared to unprimed seeds. Therefore, primed seeds were expected to present better physiological performance than the control. However, some factors can hinder the efficiency of this technique, such as the species, seed deterioration level, drying, aeration, seed size, and absorption rate (Marcos-Filho, 2015).

In the trial II, regarding the effects of water deficit on the −0.2 MPa potential (Table 3) for unprimed seeds, there was a reduction in physiological performance for most of the analyzed variables (Table 3). The comparison of the effects of osmopriming for both potentials shows a positive effect of osmopriming on the physiological potential of seeds subjected to water deficit, especially in treatments with SNP and PEG. Osmopriming with SNP shows an increase of 17 percentage points in radicle emission and 1.07 in radicle emission speed index (Table 3).

An increase of 27 percentage points in radicle emission was found when considering the positive effects of osmopriming with PEG, which is higher than that observed for SNP. Furthermore, an increase of 6 percentage points was observed in germination and 1.77 mm in seedling length. Therefore, osmopriming with PEG provided better performance of seeds from this lot under water deficit. Osmopriming with $H_2O$ and $KNO_3$ showed inferior results compared to SNP and PEG. However, seeds subjected to water deficit (−0.2 MPa) had, in general, superior physiological performance compared to seeds at 0 MPa potential (Table 3).

Water deficit for seeds from lot 2 showed similar effects to that of seeds from lot 1, and seeds from the control treatment had lower physiological performance for most variables under water deficit. The water deficit simulated with PEG 6000 for *U. decumbens* and *U. ruziziensis* (Pereira et al., 2012) and quinoa seeds (Barbieri et al., 2019) also negatively affected their performance. The seed germination process directly depends on water absorption for metabolic activation and conversion/transport of energy reserves to the embryo (Jain et al., 2019). In this sense, water deficit causes several cellular changes that result in a reduction in germination speed, delay in seedling development, and cell death at more drastic levels (Możdżeń et al., 2015).

In the present study, among the osmopriming treatments that contributed to mitigating water deficit, treatment with SNP increased root emission by 20 percentage points, radicle emission speed index by 0.25 percentage points, and germination by 2 percentage points. Similar results were observed for the treatment with PEG, with an increase of 2 percentage points in germination and 1.28 mm, also similar to the control. Unlike lot 1, osmopriming with $H_2O$ for seeds from lot 2 provided better seed performance under water deficit conditions considering radicle emission, in addition to an increase of 0.87 in the radicle emission speed index and 14 percentage points of germination (Table 4).

**Table 2.** Radicle emission (RE), radicle emission speed index (RESI), germination (G), germination speed index (GSI), electrical conductivity (EC), tetrazolium (TZ), seedling length (SL), and dry matter (DM) of lot 2 of *U. decumbens* seeds subjected to different osmopriming treatments.

| Treatment | RE (%) | RESI (index) | G (%) | GSI (index) | EC ($\mu S\ cm^{-1}\ g^{-1}$) | TZ (%) | SL (mm $seedling^{-1}$) | DM (mg $seedling^{-1}$) |
|---|---|---|---|---|---|---|---|---|
| Unprimed | 68 A | 6.82 A | 67 A | 9.02 A | 49.23 B | 61 A | 6.76 A | 0.033 A |
| $KNO_3$ | 31 D | 1.92 D | 28 D | 1.92 D | 33.96 A | 54 B | 4.23 B | 0.020 B |
| PEG | 53 B | 5.47 B | 47 B | 5.60 B | 34.91 C | 58 B | 4.66 B | 0.027 B |
| $H_2O$ | 42 C | 4.82 C | 38 C | 3.91 C | 51.87 B | 34 D | 6.17 A | 0.030 A |
| SNP | 47 C | 5.35 B | 47 B | 3.92 C | 38.20 C | 45 C | 6.20 A | 0.031 A |
| $F_{0.05}$ | 62.6* | 111.1* | 77.0* | 142.3* | 137.5* | 55.5* | 51.4* | 49.8* |
| CV (%) | 7.2 | 7.1 | 7.5 | 9.1 | 3.2 | 5.9 | 4.7 | 6.5 |

* = significant by F-test at a 5% probability; F = calculated F-value; CV = coefficient of variation. Means followed by uppercase letters in the column compare the osmopriming treatments by the Tukey test at a 5% probability.

**Table 3.** Radicle emission (RE), radicle emission speed index (RESI), germination (G), germination speed index (GSI), and seedling length (SL) of lot 1 of *U. decumbens* seeds subjected to different osmopriming treatments under water stress.

| Treatment | Osmotic potential | RE (%) | RESI (index) | G (%) | GSI (index) | SL (mm seedling$^{-1}$) |
|---|---|---|---|---|---|---|
| Unprimed | -0.2 MPa | 75 Aa | 6.70 Ab | 57 Ab | 3.38 Ab | 7.66 Ab |
| | 0 MPa | 71 Aa | 9.60 Aa | 71 Aa | 9.61 Aa | 8.8 Aa |
| $KNO_3$ | -0.2 MPa | 61 Ba | 5.29 Ba | 46 Ba | 2.63 Bb | 5.30 Ca |
| | 0 MPa | 48 Bb | 4.41 Cb | 48 Ca | 3.80 Da | 3.39 Cb |
| PEG | -0.2 MPa | 74 Aa | 6.37 Aa | 53 Aa | 3.57 Ab | 6.38 Ba |
| | 0 MPa | 47 Ba | 6.41 Ba | 47 Cb | 5.30 Ba | 4.61 Bb |
| $H_2O$ | -0.2 MPa | 64 Ba | 4.90 Ba | 44 Ba | 2.34 Bb | 2.74 Db |
| | 0 MPa | 37 Cb | 4.59 Ca | 37 Db | 3.32 Da | 5.14 Ba |
| SNP | -0.2 MPa | 72 Aa | 6.95 Ba | 45 Bb | 2.65 Bb | 6.86 Bb |
| | 0 MPa | 55 Bb | 5.88 Bb | 59 Ba | 4.26 Ca | 8.20 Aa |
| $F_{0.05}$ | - | 6.3* | 15.8* | 11.1* | 55.5* | 147.7* |
| CV (%) | - | 9.1 | 8.6 | 8.7 | 10.2 | 5.7 |

* = significant by F-test at a 5% probability; F = calculated F-value; CV = coefficient of variation. Means followed by uppercase letters in the column compare the osmopriming treatments and lowercase letters compare the stress by the Tukey test at a 5% probability.

**Table 4.** Radicle emission (RE), radicle emission speed index (RESI), germination (G), germination speed index (GSI), and seedling length (SL) of lot 2 of *U. decumbens* seeds subjected to different osmopriming treatments under water stress.

| Treatment | Osmotic potential | RE (%) | RESI (index) | G (%) | GSI (index) | SL (mm seedling$^{-1}$) |
|---|---|---|---|---|---|---|
| Unprimed | -0.2 MPa | 65 Ba | 5.43 Ab | 51 Ab | 3.23 Ab | 5.56 Ab |
| | 0 MPa | 68 Aa | 6.82 Aa | 67 Aa | 9.02 Aa | 6.76 Aa |
| $KNO_3$ | -0.2 MPa | 63 Ba | 5.97 Aa | 51 Aa | 3.42 Aa | 5.66 Aa |
| | 0 MPa | 31 Db | 1.92 Cb | 28 Db | 1.92 Db | 4.23 Ba |
| PEG | -0.2 MPa | 59 Ca | 4.71 Bb | 49 Aa | 3.31 Ab | 5.94 Aa |
| | 0 MPa | 53 Ba | 5.47 Ba | 47 Ba | 5.60 Ba | 4.66 Bb |
| H2O | -0.2 MPa | 70 Aa | 5.59 Aa | 52 Aa | 3.24 Ab | 5.55 Ab |
| | 0 MPa | 42 Cb | 4.72 Bb | 38 Cb | 3.91 Ca | 6.17 Aa |
| SNP | -0.2 MPa | 67 Ba | 5.60 Aa | 49 Aa | 2.73 Bb | 3.18 Bb |
| | 0 MPa | 47 Cb | 5.35 Ba | 47 Ba | 3.92 Ca | 6.20 Aa |
| $F_{0.05}$ | - | 21.3* | 47.6* | 22.2* | 98.0* | 102.7* |
| CV (%) | - | 8.1 | 8.4 | 9.2 | 9.5 | 4.8 |

* = significant by F-test at a 5% probability; F = calculated F-value; CV = coefficient of variation. Means followed by uppercase letters in the column compare the osmopriming treatments and lowercase letters compare the stress by the Tukey test at a 5% probability.

In summary, the results observed in this study reinforce that, despite not improving the performance of *U. decumbens* seeds under favorable conditions for germination, osmopriming clearly acted to attenuate the deleterious effects of water deficit and helped to improve the physiological potential of *U. decumbens* seeds. These effects have also been reported for seeds of several species, such as *U. ruziziensis* (Oliveira et al., 2021) and tomato (Aghaei et al., 2023), subjected to water deficit.

## 4. Conclusions

Osmopriming of *Urochloa decumbens* seeds helps to increase physical attributes such as area, gray scale, perimeter, and tissue density. Osmopriming does not contribute to the better physiological performance of *U. decumbens* seeds under ideal germination conditions. However, osmopriming with SNP (0.10 mmol $L^{-1}$) and PEG (−0.8 MPa) for 24 hours contributes to the better physiological performance of seeds under water deficit conditions.

**Authors' Contribution**

Gabriel Cordeiro de Oliveira Peris, Lucas Rodrigues de Souza, and Rafaela Martins Alves Ramos carried out data collection, interpretation, and writing of the first version of the manuscript. Daniel Teixeira Pinheiro and Marcelo Augusto Rocha Limão supervised the experiment, analyzed and interpreted the data, and wrote and corrected the final version of the manuscript. Denise Cunha Fernandes dos Santos Dias supervised and guided all stages and critically reviewed the final version.

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