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Effect of soaking solution type on seed viability and seedling growth of Bonita and Loblita cayenne (*Capsicum frutescens* l.)

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Abstract

The problem with the inhibition of the germination process of cayenne seeds is the presence of capsaicin substances contained in cayenne seeds that can inhibit the germination process. This research aims to get varieties that have better quality between Bonita and Loblita based on the interaction that occurs between varieties and types of cayenne seed soaking solutions. This study used a factorial Randomized Group Design with 2 factors and 3 replications. The first factor was cayenne pepper varieties (Bonita and Loblita) while the second factor was the type of soaking solution (PGPR, IAA, KNO_3, hot water, and control). The results showed a very significant interaction between cayenne varieties and seed soaking solution. Bonita variety has a superior quality based on the variables of germination, speed of growth and uniformity of growth while the Loblita variety is superior in the variables of maximum growth potential and maximum growth potential. The second factor is the type of KNO3 soaking solution for all seed viability variables, while for seedling growth, hot water treatment is superior.

Keywords: Cayenne; Bonita; Loblita; Soaking solution; Germination; Seedling Growth

1. Introduction

Cayenne is included in the Indonesian horticultural crop subsector and is one of the leading commodities that is well known to all corners of the world. The potential of cayenne as a commodity for export is very large, but Indonesia is still relatively small in producing or exporting cayenne [1]. Seed quality is Farming activities in producing cayenne peppers are still constrained by the use of lowquality seeds [3]. The deterioration of seed quality can be seen biochemically and physiologically [4]. Indications related to the biochemical state of seeds include decreased enzyme activity and depleted food reserves [4]. The physiological activities of seeds that indicate seed deterioration are seed vigor and germination [4].

Germination is a complex series of changes in morphology, physiology, and biochemistry process [5]. There are two factors that can affect the germination process, namely external factors and internal factors. External factors such as hard skin, environmental moisture content, light, temperature, and seeding media, while factors from within the seed itself such as seed maturity level, seed size, and dormancy [5]. According to Laisbuke (2022), to accelerate the process of cayenne pepper germination, seeds require soaking treatment to accelerate germination [6]. Seed soaking treatment can be done using chemicals, as well as plant hormones (phytohormone) to break dormancy such as cytokinin, gibberellin, and auxin, in addition to using hormones, soaking using hot water can also facilitate water absorption by seeds [5].

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Research on seed soaking using soaking solutions has been carried out by Nahak et al, (2021) [7], who studied breaking the dormancy period of local cayenne pepper seeds by applying different PGPR concentrations and soaking times. Alqamari et al, (2021) reported that soaking seeds using IAA increased the percentage of germination, height, number of leaves, wet weight, and dry weight [8]. Research conducted by Laisbuke (2022) states that the use of KNO3 at a dose of 3% can increase the germination of cayenne pepper seeds [6]. Sriwigati et al, (2021) examined the soaking of seeds in hot water on the germination and growth of fennel seedlings (Foeniculum vulgare Mill) showing very significant results on germination, plant height, and number of leaf blades [9]. The previous research was still limited to the laboratory, not yet continued to the seeding test stage in the field. Therefore, this study was conducted to determine how the effect of soaking seeds using a soaking solution on seed viability such as germination, as well as seedling growth in the field.

2. Materials and methods

2.1. Place and time of research

The research was conducted at the Center for Tropical Horticulture Studies LPPM IPB University Laboratory and Tajur II experimental garden, located at IPB Baranangsiang Campus Jl. Raya Pajajaran, Bogor. The research was conducted from September to December 2023, and the laboratory of agricultural product technology, faculty of agricultural technology, Udayana University. This study used a 2-factor Randomized Group Design with 3 replications. The first factor was the variety (V) of cayenne which consists of 2 levels namely Vb (Bonita variety) and Vl (Loblita variety), while the second factor was the type of soaking solution (P) consisting of 5 levels namely Pp (with 1% PGPR soaking), Pi (with 22ppm IAA soaking), Pk (with 3% KNO_3 soaking), Pa (with hot water soaking 50°), and Pn (with normal temperature distilled water as control).

2.1.1. Preparation of cayenne seeds

Seed soaking is carried out in accordance with the germination test method for cultivated plants and vegetables (Seed Quality Testing Development Center for Food Crops and Horticulture, 2021) [10]. Seed soaking treatment is carried out in accordance with the treatment level, and is carried out for 12 hours (overnight). Germination of cayenne seeds using the Between paper (BP) method was rolled and established. The planting media used for seeding is a mixture of soil, goat manure, firewood, and Trichoderma in a ratio of 2:1:1/3 and 80gr Trichoderma, for 20 buckets of soil. Cayenne seeds will be planted in holes approximately 0.5 cm deep. Seedlings that have grown can be transferred into polybags measuring 25x25 cm. Transplanting seedlings is done when the cayenne enters the age of 4 weeks after sowing, or the seedlings already have 5-6 leaves.

2.2. Cayenne pepper seed analysis procedure

2.2.1. Germination (%)

Germination is calculated from the sprouts that grow normally in count II, calculated by separating normal sprouts with sprouts that grow abnormally and do not grow. Normal sprouts show the potential to live into a full plant when transferred to enlargement media with characteristics such as the growth of roots, stems, and leaves growing proportionally. Calculated at 14 days after seedling, as folows:

$$\text{Germination} = \frac{\sum seeds\ that\ germinated\ normally\ at\ day{-}14}{\sum number\ of\ seeds\ planted} \text{ x } 100\%$$

2.2.2. Growth uniformity (%)

Growth uniformity was calculated using normal sprouts at day 11. Growth uniformity was calculated using the formula:

$$\text{Growth uniformity} = \frac{\sum seeds\ that\ germinated\ normally\ at\ day{-}11}{\sum number\ of\ seeds\ planted} \text{ x } 100\%$$

2.2.3. Growth rate (%/etmals)

The growth rate was calculated using the sprouts that grew on the nth day, whether they grew normally or abnormally. Growth rate was calculated using the formula:

$$\text{Growth rate} = \frac{N1}{D1} + \frac{N2}{D2} + \frac{N3}{D3} + \ldots + \frac{Nn}{Dn}$$

Description
Nn = Number of seeds that germinate on the day
Dn = length of the germination day

2.2.4. Seedling height (cm)

Seedling height was observed once, at the time of transplanting into polybags. Seedling height was measured from the root tip to the tip of the seedling growth point, using a ruler.

2.2.5. Seedling root length (cm)

The root length of the seedlings was counted once when the seedlings were transplanted into polybags. Root length was calculated using a ruler, starting from the base of the stem to the tip of the root.

2.2.6. Number of seedling leaves (leaflets)

The number of leaves of the seedlings was counted once, collecting data on the number of leaves at the same time as collecting data on the height and root length of the seedlings. The number of seedling leaves was counted one by one from the lowest leaf to the top leaf.

2.2.7. Leaf glucose (%)

Samples of 2 grams obtained from the fine mashing of dried cayenne leaves were taken to the Laboratory of agricultural product technology, Faculty of Agricultural Technology, Udayana University, and analyzed. Analysis of leaf glucose content was carried out by the phenol-sulfur method.

2.2.8. Relative water content (%)

RWC measurements were taken twice. Samples were taken from leaves from the bottom, middle and top, each part of 1 leaf (total of 3 leaves) for each tree and carried out in all sub-trees experimental unit. In the laboratory, 30 leaf pieces of 1 x 1 cm each were taken from the six leaf samples and weighed for fresh weight. After weighing, it was put into a Petri dish cup containing water and irradiated with 40-Watt fluorescent light at room temperature for 5 hours. After it was removed, the water that was still attached was carefully cleaned using a tissue and then weighed the turgid weight. After that, the leaf pieces were oven dried at 70^{o} C for 24 hours and then weighed. KAR value (%) was calculated by the formula:

$$\text{RWC value (\%)} = \frac{\text{fresh weight leaves value} - \text{dry weight leaves value}}{\text{turgid weight leaves value} - \text{dry weight leaves value}} \text{ x } 100\%$$

3. Results

3.1. Seed viability parameters

3.1.1. Germination (%)

The germination of the combination of Bonita varieties with KNO_3 soaking solution showed the highest results of 90.00% (Figure 4.1), significantly different from the control Loblita (84.00%), Bonita with hot water soaking (83.00%), Bonita with IAA soaking (78.50%), and Bonita control (76.00%). The lowest combination treatment was owned by Bonita control (76.00%). The best combination of Bonita varieties in IAA soak (90.00%), but not significantly different from Bonita with PGPR soak (86.75%). The Loblita variety had the best germination in the hot water immersion treatment of 87.25%, but it was not significantly different from all immersion treatments except the control Loblita (84.00%). The decrease in the percentage of germination of the combination of varieties and types of soaking solutions that have the highest and lowest values amounted to 15.56%.

3.1.2. Growth uniformity (%)

Growth uniformity was calculated using data on the number of sprouts that grew normally on the 11th day after sowing (day-11). The percentage of growth uniformity of the combination of Bonita varieties with KNO3 soaking solution showed the highest results (90.00%) but was not significantly different from the combination of Loblita with hot water soaking (86.50%), Bonita with PGPR soaking (86.25%), and Loblita with KNO3 soaking (84.75%). The lowest combination treatment was owned by Bonita control (75.00%).

3.1.3. Growth rate (%/etmal)

The single factor of Bonita and Loblita varieties showed significantly different test results, the speed of seed growth of Loblita varieties was 22.25%/etmal higher than that of Bonita varieties at 19.38%. Type of soaking solution results were not significantly different at each level, but the highest seed germination speed was owned by the type of KNO_3 soaking solution (23.01%/etmal) and the lowest was owned by the type of IAA soaking solution at 19.92% (Table 4.3). The combination of Bonita with KNO_3 soaking solution had the highest growth speed of 23.08%/etmal.

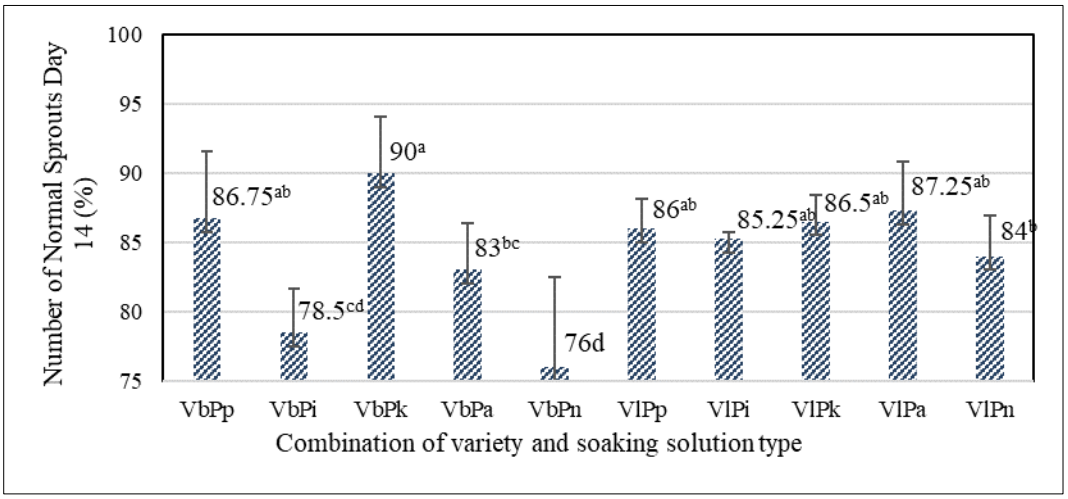

Figure 1 Number of normal sprouts on day 14, Bonita with PGPR (V_bP_p); Bonita with IAA (V_bP_i); Bonita with KNO_3 (V_bP_k); Bonita with Hot water (V_bP_a); Bonita Control (V_bP_n); Loblita with PGPR (V_lP_p); Loblita with IAA (V_lP_i); Loblita with KNO_3 (V_lP_k); Loblita with Hot water (V_lP_a); Loblita Control (V_lP_n); Numbers followed by the same letter show no significant difference in Duncan's multiple range test (DMRT) at the 5% level

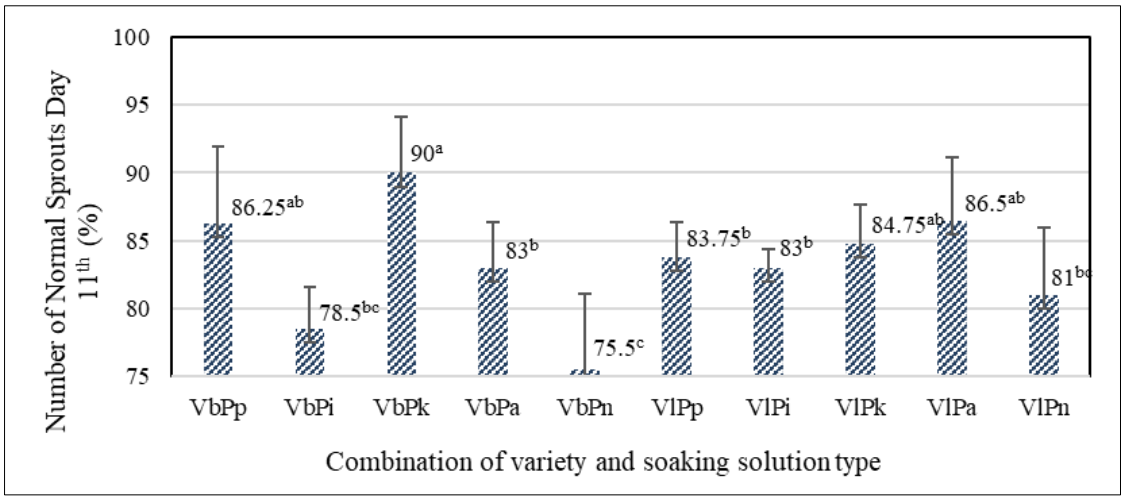

Figure 2 Number of normal sprouts on day 14, Bonita with PGPR (V_bP_p); Bonita with IAA (V_bP_i); Bonita with KNO_3 (V_bP_k); Bonita with Hot water (V_bP_a); Bonita Control (V_bP_n); Loblita with PGPR (V_lP_p); Loblita with IAA (V_lP_i); Loblita with KNO_3 (V_lP_k); Loblita with Hot water (V_lP_a); Loblita Control (V_lP_n); Numbers followed by the same letter show no significant difference in Duncan's multiple range test (DMRT) at the 5% level

3.2. Seedling growth parameters

3.2.1. Seedling height (cm)

The highest seedling height was owned by the combination of Bonita with hot water immersion which had a length of 19.10 cm (Figure 4.5) but was not significantly different from the control Bonita (16.80 cm), Loblita with IAA immersion

(16.43 cm), Loblita with PGPR (16.10 cm), Loblita with KNO_3 immersion (15.50 cm), and Bonita with PGPR immersion (14.23 cm). Bonita variety had the highest seedling length in hot water immersion treatment, significantly different from KNO_3 and IAA. Loblita variety has the best seedling length in IAA soaking with a value of 16.43 cm but not significantly different in all types of soaking solutions. The types of PGPR solution, IAA, KNO_3, and control each differed not significantly in both varieties. The percentage reduction in the highest and lowest seedling length values amounted to 39%.

3.2.2. Seedling root length (cm)

The root length of the seedlings was calculated with a ruler, starting from the base of the stem with roots, to the tip of the longest root. The data obtained had a fairly large coefficient of variation, so data transformation was carried out using the square root transformation method (root X + 0.5). Based on Figure 4.6, the combination treatment that has the longest seedling root length is Bonita with hot water immersion, with a root length of 13.43 cm, significantly different from the combination treatment of Bonita IAA (7.57 cm) and lobita control (7.17 cm).

3.2.3. Number of seedling (leaflets)

The number of seedling leaves was calculated using samples of untransplanted seedlings, the same samples were used for the variables of seedling height, seedling stem length, primary root length, root length, and number of secondary roots. Based on Figure 4.8, the combination of Bonita varieties and hot water soaking has the highest average number of leaves at 5.67 leaves, which is not significantly different from the Bonita control combination. Bonita variety is highest in the type of hot water soaking solution (5.67), significantly different from the type of PGPR soaking solution, IAA, KNO3, and control. The highest Loblita variety in the type of PGPR and KNO3 soaking solution, not significantly different in all types of soaking solutions.

3.2.4. Leaf glucose (%)

The results of the single factor test on the variable leaf glucose content showed results that were not significantly different at each level of variety and the type of seed soaking solution factor. Bonita variety has a higher leaf glucose content of 20.37% compared to Loblita variety of 19.02%. The type of seed soaking solution factor, the control treatment gave the highest leaf glucose content value of 26.27% and the lowest was owned by KNO3 which amounted to 23.74%.

3.2.5. Relative water content (%)

Table 1 Effect of soaking solution type (P) and cayenne pepper varieties (V) Bonita and Loblita on variables seedling height, seedling root length, number of seedling leaves, and relative water content

Combination	Seedling height (cm)	Seedling root length (cm)	Number of seedling leaves (leaflets)	Relative water content (%)
V_bP_p	14,23^{ab}	9,07 (3,08) ^{ab}	4,00^{bc}	70%^{ab}
V_bP_i	12,83^{b}	7,57 (2,79) ^{b}	3,67^{bc}	59%^{de}
V_bP_k	13,37^{b}	8,90 (3,04) ^{ab}	3,67^{bc}	67%^{bcd}
V_bP_a	19,10^{a}	13,43 (3,73) ^{a}	5,67^{a}	53%^{e}
V_bP_n	16,80^{ab}	12,43 (3,58) ^{a}	4,67^{ab}	74%^{ab}
V_lP_p	16,10^{ab}	9,73 (3,19) ^{ab}	4,00^{bc}	80%^{a}
V_lP_i	16,43^{ab}	9,93 (3,21) ^{ab}	3,67^{bc}	61%^{cde}
V_lP_k	15,50^{ab}	9,33 (3,13) ^{ab}	4,00^{bc}	76%^{ab}
V_lP_a	13,50^{b}	9,10 (3,10) ^{ab}	2,33^{c}	77%^{ab}
V_lP_n	11,73^{b}	7,17 (2,76) ^{b}	3,00^{c}	74%^{ab}

Notes Bonita with soaking PGPR (V_bP_p); Bonita soaking with IAA (V_bP_i); Bonita soaking with KNO_3 (V_bP_k); Bonita soaking with Hot water (V_bP_a); Bonita Control (V_bP_n); Loblita soaking with PGPR (V_lP_p); Loblita soaking with IAA (V_lP_i); Loblita soaking with KNO_3 (V_lP_k); Loblita soaking with Hot water (V_lP_a); Loblita Control (V_lP_n); Numbers followed by the same letter indicate not significantly different in Duncan's multiple range test (DMRT) at 5%.

The relative water content of the leaves was measured twice and averaged. Leaf sampling for KAR test was conducted before noon to avoid excessive evaporation. The highest KAR was owned by the combination of Loblita varieties soaked with PGPR with a value of 80.31%, which was not significantly different from the combination of Loblita with PGPR (77.07%), Loblita with KNO3 soak, (75.85%), Loblita control (74.05%), Bonita control (73.63%), and Bonita with PGPR soak (70.15%).

Table 2 Effect of cayenne pepper variety (V) and type of soaking solution (P) on variable growth speed

Treatment	Leaf glucose	Growth rate (%/*etmals*)
Varieties of cayenne papper		
$V_{(b)}$	20,37 a	19,38 b
$V_{(L)}$	19,02 a	22,25 a
LSD 5%	3,57	0,73
Type of soaking solution		
$P_{(p)}$	24,42 a	20,68 b
$P_{(i)}$	25,00 a	19,92 b
$P_{(k)}$	23,74 a	23,01 a
$P_{(a)}$	25,33 a	20,31 b
$P_{(n)}$	26,27 a	20,15 b
LSD 5%	5,64	1,15

Notes the numbers followed by the same letter are not significantly different at the least significant difference (LSD) 5% level, Bonita variety (V_b), Loblita variety (V_l), with PGPR soaking (P_p), with IAA soaking (P_i), with KNO_3 soaking (P_k), with hot water soaking (P_a), control treatment (P_n).

4. Discussion

Soaking seeds using several types of solutions can produce germination up to 90%. The treatment combination of Bonita varieties with the type of control soak, showed the lowest results among all treatment combinations. The combination of Bonita with KNO_3 soaking solution has the highest germination of 90.00%, in line with Laisbuke's research (2022) which states that soaking cayenne pepper seeds using KNO_3 is the best treatment to increase seed germination [6], besides that, KNO_3 is classified as a chemical substance used for dormancy breaking treatment according to the Center for the Development of Seed Quality Testing of Food Crops and Horticulture (2021) [10].

Germination is closely related to growth uniformity (r = 0.96**), maximum growth potential (r = 0.91**), and growth speed (r = 0.82**) as evidenced by the correlation table which shows a value of positive and very significant effect. The highest growth uniformity is owned by the combination of Bonita varieties with KNO_3 soaking (90.00%), but it is not significantly different from the combination of Loblita with hot water soaking (86.50%), Bonita with PGPR soaking (86.25%), and Loblita with KNO_3 soaking (84.75%). The interaction of growth speed variables has no significant effect, but the test using LSD 5%, shows that the Loblita variety has a higher growth speed of 22.25%/etmal compared to the Bonita variety which is 19.38%/etmal, for the single factor of the type of soaking solution, KNO_3 solution has the highest growth speed of 23.01%/etmal, in accordance with research conducted by Ardi et al. (2018) on the provision of KNO_3 which has a significant effect on papaya (*Carica papaya* L.) seed viability test [11]. The element of potassium in KNO_3 (Potassium Nitrate) is a macro nutrient that is absorbed in the form of K^+ cations. The function of the element Potassium itself includes helping the formation of proteins, carbohydrates and sugars, strengthening plant tissue, increasing photosynthate transportation from the leaves to all plant tissues, in the cell, the element K^+ plays a role in regulating cell turgor related to osmosis pressure (Rai, 2023) [12].

Growth variables of cayenne pepper seedlings such as seedling height, number of seedling leaves, and seedling root length. The Bonita hot water combination treatment has the highest seedling height (19.10 cm), but it is not significantly different from Bonita PGPR (14.23 cm), Loblita PGPR (16.10 cm), Loblita IAA (16.43 cm), Loblita KNO_3 (15.50 cm), and Bonita control (16.80 cm). The control Loblita treatment had the lowest height of 11.73 cm, in line with research conducted by Sriwigati et al. (2021) on the effectiveness of seed soaking using relatively high temperature water on the germination and growth of fennel seedlings [9].

Seedling height is significantly affected by root length (r = 0.92**), this is because seedling height is measured from the tip of the root to the shoot. The combination treatment of Bonita with hot water immersion has the highest root length, amounting to 13.43 cm, which is not significantly different at all levels of combination treatment except the control Loblita. Seedling growth is also influenced by the number of leaves, because the more the number of leaves, the higher the photosynthesis rate (Sari et al., 2019) [13]. The Bonita hot water combination treatment has the highest number of leaves (Table 4.8) which is 5.67 leaflets, significantly different from Bonita PGPR (4.00 leaflets), Loblita PGPR (4.00 leaflets), Bonita IAA (3.70 leaflets), Loblita IAA (3.67 leaflets), Bonita KNO3 (3.67 leaflets), Loblita KNO3 (4.00 leaflets), Loblita hot water (2.33 leaflets), and Loblita control (3.00 leaflets), almost all significantly different in each combination treatment.

The relationship between water absorption and mineral nutrients with seedling growth is very important, because water and nutrients play an important role in the photosynthesis process [14]. Relative water content is a parameter of plant resistance facing drought stress (Quilambo.2004), because it describes the condition of the cells (the amount of water and turgor pressure) of the plant when the water potential decreases [15]. The combination treatment of Loblita with PGPR soaking has the highest KAR value of 80.31%, significantly different from Bonita IAA (58.99%), Loblita IAA (61.41%), Bonita KNO_3 (67.13%), and Bonita hot water.

Leaf glucose levels represent the results of photosynthesis. Single factors and combinations between varieties and types of soaking solutions showed results that had no significant effect on leaf glucose levels. At the level of each single factor showed results that were not significantly different based on the 5% DMRT test, which means that Bonita and Loblita varieties do not respond to the type of soaking solution on leaf glucose. Bonita control has the highest glucose content value with a level of 20.37%, and for the control soaking treatment gives a glucose level of 26.27%.

5. Conclusion

The combination of Bonita varieties with KNO3 soaking is effective in increasing germination, growth uniformity and growth speed of cayenne pepper seeds. Seedling growth variables showed that the combination of Bonita with hot water soaking was superior in the variables of seedling height, seedling root length, and number of seedling leaves. The highest leaf glucose variable is owned by the combination of Loblita varieties with PGPR soak, and the highest leaf KAR is owned by the combination of Bonita control varieties with a KAR value of 44.50%.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declared that there is no conflict of interest.

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