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Research Article

The Effect of Seed Priming to Improve Germination Parameters and Early Growth of Chickpea (*Cicer arietnum* L)

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Chickpea is one of the new crops being grown in Zimbabwe for its plethora of benefits in crop production and human diet. However, like most grain legumes preliminary research has shown that chickpea seed has a problem of poor germination hindering the realization of the crops full potential yield. Seed priming has a potential to improve germination of chickpea. Therefore, a laboratory experiment was carried out to determine the effects of seed priming on seed germination. The experiment was laid out as a 4×5 factorial in completely randomised design (CRD) with 20 treatments replicated three times. The treatments investigated were five seed priming methods viz hydro-priming, halo-priming (KNO_3), prechill, preheat, and no priming (control); and four chickpea varieties that were ICCV00305, ICCV03404, ICCV97105, and ICCV92944. Hydro-priming involves soaking seed for 24 hours and leaving it to dry in the laboratory for 24 hours at room temperature before it is planted. Halo-priming was done by soaking the seed in a solution with 2.4 g of potassium nitrate and 1.2 ml of distilled water. Prechill treatment involves subjecting seed at a temperature of 10°C for 7 days before planted. Preheating was done by subjecting the seed in an oven at 35° C for 30 minutes. The parameters measured were germination percent, speed of germination, radicle and plumule length, and seedling vigor index. The results showed that preheating and halo-priming chickpea seed significantly ($p < 0.05$) improved germination percentage, increased radicle and plumule length, and seedling vigor index. Hyro-priming and no priming reduced germination percentage, decreased radicle and plumule length with poor seedling vigor. Results also indicated that variety ICCCV92318 recorded the highest germination percentage, radicle and plumule length, and seedling vigor index, while variety ICCV97114 recorded the least figures on all tested parameters of chickpea. It can be concluded that preheating seed and halo-priming seed improved germination and seedling vigor in chickpea.

1. Introduction

The chickpea (*Cicer arietinum* L.) of the Fabaceae family, is currently one of the most widely cultivated cool-season food legume crops and is rated the third most important after common bean and field pea [1, 2]. The crop is being grown in over 50 countries across Africa, Asia, Europe, and Australia [1, 3]. Chickpeas originated in present-day South-Eastern Turkey and adjoining regions of Syria where three wild annual species of Cicer viz., *Cicer bijigum*, *Cicer echinospermum*, and *Cicer reticulatum* had been discovered [4]. However, in all the production areas, South-East Asia ranks higher in chickpea production with 80% of the global production. In Africa, Ethiopia dominates in both area grown (213187 ha/year) and total production (284640 t/year) of chickpea [5].

The crop is increasingly being produced successfully in drier parts of the world like in the Middle East and Ethiopia producing reasonable yields [6]. This is because the crop has varieties with a tap root system which allows for more nutrients and water uptake from deeper horizons, making it more adaptable in drier environments where other food legumes do not perform well [7]. With the increase in frequency and intensity of droughts because of climate change chickpea is the crop for the future food security. When compared with other legumes, chickpea grain is an excellent source of high-quality protein (23.3–28.9%), carbohydrates (61.5%), fats (4.5%), and minerals (phosphorus, calcium, magnesium, iron, and zinc) [8]. This makes the crop ideal for playing a significant nutritional role in the diet of millions of people in developing countries. Unlike other food legumes chickpea plant has diverse uses; the green leaves are used as leafy vegetable, and it has superior mineral content, making it an ideal crop to fight micronutrient deficiency which is rampant in developing countries [8]. The seed can also be consumed whilst immature as a snack or vegetable. The dry seed is used to make flour to prepare different types of nutritious baked dishes [9]. Besides its nutritional benefits, the chickpea plays a significant role in soil recapitalisation due to its nitrogen-fixing ability. It fixes up to 80% of its nitrogen needs, a benefit for most resource-poor farmers. Hence the crop is a panacea for most low-input agricultural systems practiced by resource-constrained farmers who cannot afford to buy fertilisers. Chickpea crop residue is richer in digestible crude protein compared with other cereal crops which makes it good for animal feed [10]. The poor condition of livestock during dry periods limits the contribution of livestock to economic household food security during the period. Supplementing with chickpea residues can improve the condition of the livestock, making them fit for draught power at the onset of the rains or fetching higher prices if taken to the market. Maize stover can also be ensiled with chickpea residues to improve silage quality for feeding livestock during the dry season.

Despite the present importance and rising demand for chickpea across the globe, the yields of this crop are still very low especially in arid and semi-arid areas. This is because the crop often fails to establish quickly and uniformly, leading to decreased yield because of low plant. Both Desi and Kabuli the two most cultivated types of chickpea in the world, have low germination percentages, and the reasons behind this phenomenon are not yet explored [11]. Low germination percentages of chickpea lead to poor stand which results in reduced yields, and this has impacted its adoption in most countries. According to Khan et al. [12], good stand establishment is of paramount importance because patchy stands due to uneven germination result in low yields and often crop failure. Presowing seed treatments have been employed to enhance the germination of legumes, and this as well can be used to enhance the germination of chickpeas and increase their productivity. According to McDonald [13] and Farooq et al. [14], presowing seed treatments can improve seedling emergence, and one of the efficient practices is seed priming. Seed priming is a simple presowing treatment that triggers uniform germination and growth in different plant species [15]. In simple terms, seed priming is a controlled hydration treatment, which improves the germination rate, uniformity of germination, and sometimes total germination percentage [16]. In the opinion of Farooq et al. [14], this hydration is only adequate to permit pre-germinative metabolic processes but not sufficient to enhance germination. The most common priming methods employed in crop production are not limited to hydro-priming (soaking) but also include the use of osmotic agents such as PEG (osmo-priming) and the use of specific salts such as KNO_3, NaCl, and KCl (halo-priming). The use of growth regulators such as GA (Homo-priming) is another priming method. In addition, some physical treatments such as UV, cold (prechill) or heat (preheat) are being used for enhancing germination in many crops, thus suggesting that priming effects are not necessarily related to seed imbibition only [14]. Several authors have ascribed the priming technique to being efficient in improving the germination and growth of several crops [17, 18]. Nonetheless, even though priming is reported to improve germination and early growth of seedlings, its efficacy varies under different conditions and in different crop species [19]. This research evaluated the effects of different priming methods on the germination properties and seedling vigor of chickpea seeds.

2. Material and Methods

The research was done in a laboratory at the Harare research station (HRS) of the Department of Research and Specialist Services of Zimbabwe in the Seed services division. HRS is situated in the North Eastern part of the city of Harare. It is situated at a longitude of 30°32′E and a latitude of 17°41′S at an altitude of 1506 m above sea level. The laboratory has an automated system that regulates all the weather parameters (humidity, moisture, and temperature). The temperature range alternates between 20 and 30°C.

2.1. Experimental Design and Treatments. The experiment was laid out as a 4×5 factorial in completely randomised design (CRD) with 20 treatments replicated three times. Treatments investigated were five seed priming methods: hydro-priming, halo-priming with potassium nitrate (KNO_3), prechill, preheat, and control and four varieties: ICCV00305, ICCV03404, ICCV97105, and ICCV92944.

2.2. Seed Priming. Seeds for the hydro-priming treatment were soaked in distilled water for 24 hours and left to dry in the laboratory for 24 hours at room temperature before they were planted. In the prechilling treatment, the seeds were prechilled at a temperature of 10°C for 7 days before being planted [20]. For halo-priming, 2.4 g of potassium nitrate was dissolved in 1.2 ml of distilled water and the solution was used to saturate the germination substrate at the beginning of the test. For preheat, seeds were heated in a controlled oven at 35° C for 30 minutes. Distilled water was then used to moisten the substrate thereafter [20]. In the control treatment, no priming was done on the seeds.

2.3. Germination Test. Treated and untreated seeds were sown in 30×30 cm trays filled with moistened sterilized sand. The trays were placed in the germination room, and the temperature was maintained at 20–30 °C [20]. The sand method was used because it improves accuracy in seedling evaluation and is the most recommended media by ISTA rules [20]. Each tray contained 10 seeds which were treated

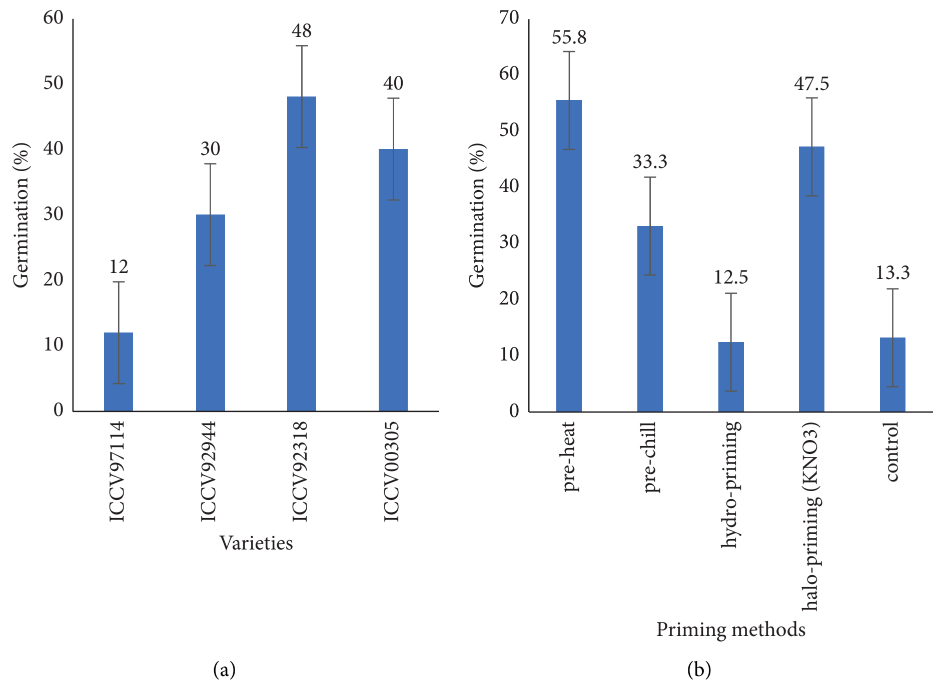


Figure 1: Effect of seed priming and variety on the germination percentage of chickpea. Vertical bars represent standard error bars of means.

with thiram (Dimethyl-carbamothioic) at 2.5 g/kg of seed to control surface borne fungi, and the seeds were uniformly spread on the substrate to ensure optimum spacing. After sowing, trays were covered with clear polythene plastics to conserve moisture.

2.4. Data Collection and Analysis. Seed germination was determined according to the rules of ISTA [20]. The covered trays were kept in an incubator at 20–30 °C temperature for germination. The germination lasted for 16 days with data being collected on a daily basis. Seeds were considered germinated when the radicle appeared for at least 2 mm [20]. The germination percentage was calculated by dividing the number of germinated seeds and the total number of sowed seeds. Speed of germination which is an expression of vigor was calculated by dividing the highest cumulative germination percentage by the number of days taken to reach that germination percentage [21, 22]. The shoot and root lengths of seedlings were measured after the final count in the standard germination test. Five normal seedlings were selected randomly from each replicate. The shoot length was measured from the point of attachment of the cotyledon to the tip of the seedling. Similarly, the root length was measured from the point of attachment to the tip of the root. The seedling vigor index was calculated by multiplying the standard germination with the average sum of shoot length (cm) and root length (cm) on the 8^{th} day of germination [23]. Data collected were subjected to analysis of variance (ANOVA) using Genstat 18^{th} edition. Where significant differences were detected, means were compared at $p < 0.05$ using Fisher's protected least significant difference (LSD) test [24].

3. Results

3.1. Effects of Seed Priming and Variety on the Germination Percentage of Chickpea. There was no interaction ($p > 0.05$) between variety and priming methods on the germination percentage of chickpea. However, varieties were significantly different ($p < 0.05$) on germination percentage (Figure 1(a)). Variety ICCV92318 had the highest germination percentage, although it was not significantly different from variety ICCV00305. The least germination percentage was recorded on variety ICCV97114 which was significantly lower ($p < 0.05$) than the rest of the varieties. The priming method was also significantly different ($p < 0.05$) on germination percentage (Figure 1(b)). Across all the priming methods, preheat had the highest germination percentage; however, it was not significantly different from halo-priming. Although hydro-priming gave the least germination percentage it was not significantly different from no priming (control).

3.2. Effects of Seed Priming and Variety on the Speed of Germination of Chickpea. There was a significant interaction ($p < 0.05$) between the priming method and variety on the speed of germination (Figure 2). The highest speed of germination was recorded on all varieties that were preheated. No priming (Control) led to a sharp decrease in the speed of germination of all varieties, and it was not significantly different form the varieties that were hydroprimed (Figure 2). Across all the priming methods, variety ICCV92318 recorded the highest speed of germination, although it was not statistically different from variety ICCV00305. However, variety ICCV97114 germinated slowly on all priming methods.

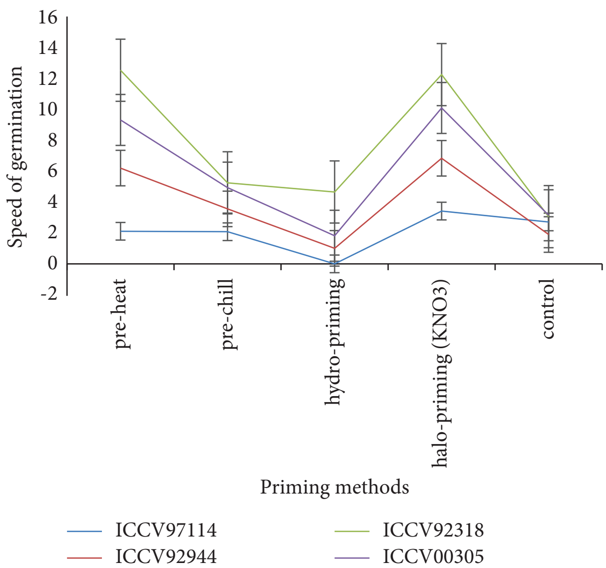


Figure 2: Effect of seed priming × variety on the speed of germination of chickpea. Vertical bars represent standard error bars of means.

3.3. Effect of Seed Priming and Variety on Radicle Length of Chickpea. There was no significant interaction ($p > 0.05$) between variety and priming methods on the radicle length of chickpea. However, significant differences were observed on the main effects. Variety ICCV 92318 had the longest radicles; however, it was not significantly different from varieties ICCV00305 and ICCV92944 (Figure 3(a)). Variety ICCV97114 had significantly the shortest radicle length compared to the other three varieties. Halo-priming recorded the longest radicles; however, it was not significantly different from preheating (Figure 3(b)). The shortest radicles were recorded on hydro-priming which was also not significantly different from no -priming (control).

3.4. Effect of Seed Priming and Variety on Plumule Length of Chickpea. There was no significant interaction ($p > 0.05$) between variety and priming methods with respect to plumule length of chickpea. However, a significant difference ($p < 0.05$) was observed on the main effects. Variety ICCV92318 recorded the longest plumule, although it was not significantly different from ICCV00305 (Figure 4(a)). ICCV 97114 recorded significantly shorter plumule than the other three varieties. Preheat seeds scored significantly the longest plumule; however, it was not significantly different from halo-priming (Figure 4(b)). The shortest plumule was observed on hydro-primed seeds which were also not significantly different from no priming (control).

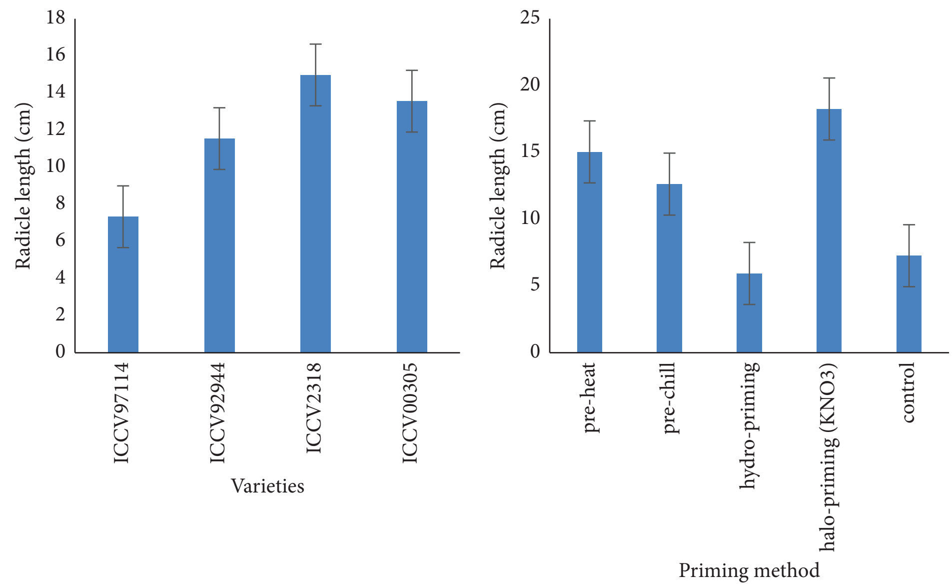
3.5. Effect of Seed Priming and Variety on Seedling Vigor Index 1 of Chickpea. Although there were no significant interactions ($p > 0.05$) between variety and priming methods on seedling vigor index 1 of chickpea, significant differences ($p < 0.05$) were observed on the main effects. Variety ICCV92318 had the highest seedling vigor index 1, nonetheless, it was not significantly different from ICCV00305 (Figure 5(a)). ICCV 97114 recorded significantly the lowest seedling vigor index 1. Preheated seeds and halo-priming had significantly the highest seedling vigor index followed by prechilled seeds (Figure 5(b)). Halo-priming and no priming gave significantly the least seedling vigor index 1 compared to the other three methods.

4. Discussion

In this study, preheating and halo-priming increased the germination percentage of chickpea seed. The positive response to germination of preheating chickpea seed might be due to its role in influencing the permeability of the membranes which ultimately leads to the activation of enzymes involved in protein synthesis and carbohydrate metabolism [26]. Sharma et al. [27] revealed that enzymes such as superoxide dismutase (SOD) are activated at optimal temperatures, around 20°C and this increases seed germination. Improved germination of chickpea seed due to halo-priming can be attributed to the ability of KNO_3 to break dormancy in seeds of many species [28]. Although it is not clear of how KNO_3 improves germination, some authors have reported the role on potassium nitrate in modulating abiscisic acid (ABA) metabolism or ABA signalling in developing seeds [29]. Furthermore, a higher germination percentage on preheated and halo-primed seeds is related to the capabilities of these treatments in repairing and building up of nucleic acid, enhancing the synthesis of RNA and proteins and respiratory activities of seeds membranes [14].

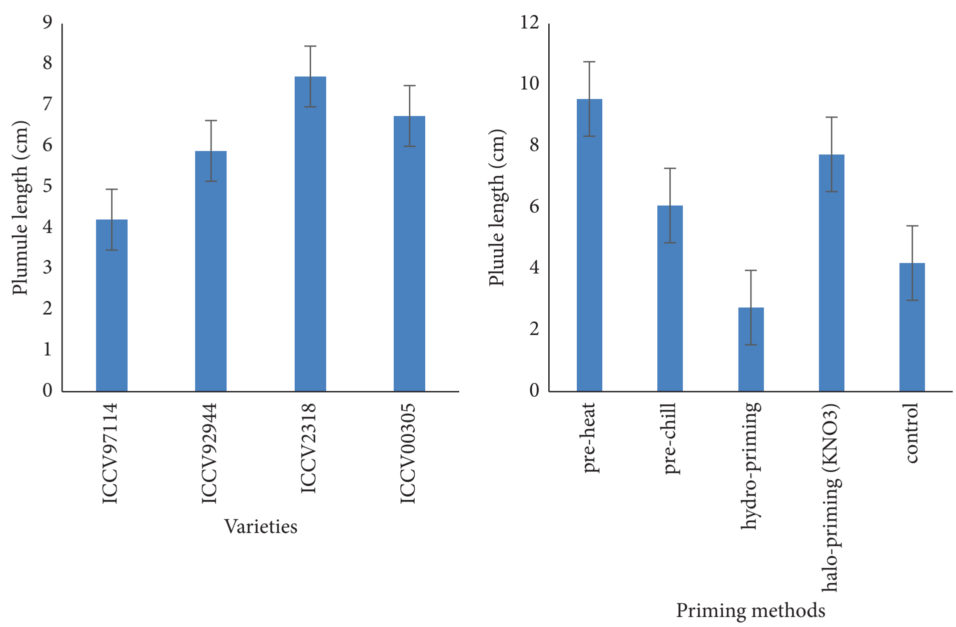
Hydro-priming and no priming of chickpea seed resulted in a poor germination percentage. It is possible that hydro-priming was prolonged which could have caused an oxygen deficiency and the build-up of inhibitors which hinders germination. Adhikari et al. [30] revealed that the time of soaking the seed when hydro-priming is critical to seed germination as it may delay or improve germination. Some authors have reported an improved germination after hydro-priming chickpea seed [31].

In this study, preheating and halo-priming chickpea seed increased the speed of germination. Preheating chickpea seed before germination may have scarified the seed so that the growth of plumule and radicle had less hindrance around the seed testa [32]. In addition, preheating enhances absorption of water and gases, which ultimately leads to increased speed of germination of the seeds [33]. The results of this study corroborated findings by Shah et al. [34] who reported that preheating seed at a high temperature for a short duration can improve the speed of germination of chickpea. The increased speed of germination after halo-priming chickpea seed can be explained by potassium nitrate which induces embryo response and promotes seed viability [35]. Potassium nitrate reduces the rate of water imbibition and this prevents the disruption of cell membrane to facilitate further biochemical and physiological processes for seed germination [35]. The present study observed that no priming and hydro-priming (control) led to a sharp decrease in the speed of germination of all varieties. Perhaps chickpea has the typical hard, impermeable seed coat like other



(a) (b)

Figure 3: Effect of variety and priming method on radicle length of chickpea. Vertical bars represent standard error bars of means.



(a) (b)

Figure 4: Effect of variety and priming method on plumule length of chickpea. Vertical bars represent standard error bars of means.

leguminous species which can reduce the speed of germination, especially in nonprimed seeds.

Variations in radicle length can be attributed to genotypic differences among chickpea varieties [36]. Longer radicles recorded on halo-priming can be ascribed to its ability to act as a mode of scarification which enhances rapid embryo growth. This is because priming with KNO_3 can significantly enhance the activities of hydrolytic enzymes which bring about the breakdown of food material available in the endosperm and deliver energy to the living embryo for further growth of both the roots and shoots. Moreover, halo-priming leads to changes in macromolecular synthesis and increased enzyme activities, which overcome dormancy and boast rapid radicle growth [37]. These results corroborate the findings of Ahmadvand et al. [38] who conducted two laboratory and green house experiments to evaluate the effects of seed priming with KNO_3 on germination and emergence traits of two soybean cultivars *cv*. Gorgan −3 and *cv*. Sahar. They reported that seed priming with KNO_3 caused a significant increase in germination and emergence

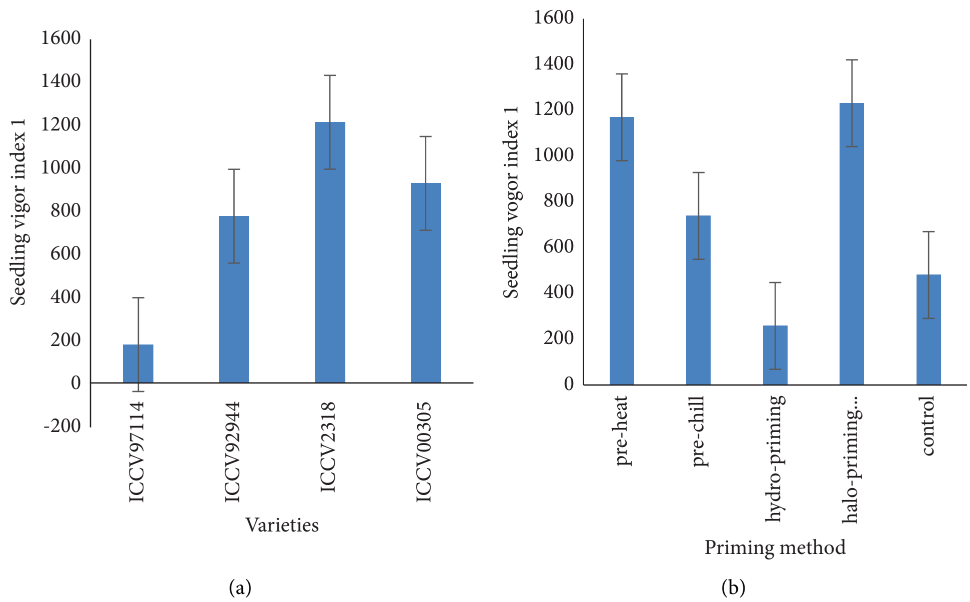


Figure 5: Effect of variety and priming method on seedling vigor index 1 of chickpea. Vertical bars represent standard error bars of means.

percentage which then translated to increased radical and plumule length.

The longer plumule recorded on preheated seeds can be attributed to the ability of preheating to energize seeds and activate various enzymes and stimulate the metabolic activity of seed, resulting in rapid sprouting of seeds and protrusion of radicle and plumule. Moreover, preheating can enhance earlier metabolic activities and a lesser mechanical restriction of seed coat as a result of the softening of the seed coat consequences which leads to rapid sprouting of the seeds and protrusion of the plumule [13]. Perhaps hydro-priming may have reduced plumule length due to the lethal effect of prolonged time of soaking. Prolonged soaking inhibits nuclear replication in the shoot tips of fresh seed.

The significant differences between varieties on the seedling vigor index when primed using different methods can be attributed to different genetic make ups which render varieties. Since seedling vigor index is a parameter that depends on plumule and radicle length. The higher radicle and plumule lengths recorded on ICCV92318 could have resulted in the higher seedling vigor index 1 of the variety [15]. The improved seedling vigor index in halo-primed seeds can be attributed to the osmotic advantage that K^+ has in improving cell water saturation which acts as a cofactor in the activities of numerous enzymes which enhance seedling vigor [39]. Furthermore, KNO_3 is believed to increase the activity of total amylase, proteases, nitrate reductase (NR), glutamate dehydrogenase (GDH), and glutamine synthetase (GS) enzymes that participate in nitrogen metabolism and GOGAT cycle, which, in turn, increase the nitrogen and protein production, increasing seedling vigour [40]. Several authors have suggested that seed priming with nitrate salts (halo-priming) could manipulate the yield-determining parameters successfully such as vigorous seedlings in many diverse environments and various crops [28, 40, 41]. Preheating of chickpea seeds increases seedling vigor can be ascribed to the ability of preheating to enhance oxygen uptake and increase efficiency in mobilizing nutrients from the cotyledons to the embryonic axis which boasts seedling vigor [42]. Heat treatments significantly improve not only germination rate and percent but also further improve the number of secondary roots, radicle, and plumule length which translate to a higher seedling vigor index [43]. Reduced seedling vigor index in hydro-primed and no priming was due to the lower germination percentage, shorter radicles, and smaller plumules observed for these treatments in this experiment [25].

5. Conclusions

Preheating and halo-priming improved germination percentage, speed of germination, and increase radicle and plumule lengths as well as seedling vigor index. Hydro-priming and no priming significantly reduced the germination percentage and seedling growth of chickpea. Variety ICCCV92318 recorded the highest germination percentage, radicle and plumule length, and seedling vigor index.

Data Availability

All the necessary data are included in the manuscript. If additional data are required, the corresponding author can be contacted.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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