Positive Effects of Priming with Glucose and Sucrose on Seed Dormancy and Germination Parameters of Some Varieties of Chickpeas (*Cicer Arietinum* L.)

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Abstract: Chickpea seeds are rich in protein and have an excellent balance of essential amino acids. In addition, it is also rich in calcium, phosphorus, vitamins B1, B2, and dietary fiber. Seed priming is a simple and effective practice to improve the expression of the physiological potential of seeds. The technique consists in synchronizing and reducing the germination time of the seeds by controlled hydration. The experimental design was composed of priming using glucose and sucrose with four concentrations (0, 3, 6, 10 $g.L^{-1}$). The parameters studied were the percentage of seed germination, the MGT, and the T50. The priming of seeds of four varieties of chickpeas with glucose and sucrose gave positive results on the studied germination parameters. Priming with sugar allowed the lifting of the dormancy of seeds of most of the varieties and especially with the concentration of 10 $g.L^{-1}$ in the variety Farihane (more than 60% compared to the control). While the priming with glucose allowed the lifting of the dormancy of seeds of the variety Mubarak of 15% compared to its control with the concentration of 6 $g.L^{-1}$. For the variety Zahor, the T50 and MGT were significantly improved (20% and 33%, respectively) for the Priming with a glucose concentration of 3 $g.L^{-1}$ compared to the control. Priming with a sucrose concentration of 10 $g.L^{-1}$ gave the highest value of PG (98.33%, Farihane variety), and the concentration of 6 $g.L^{-1}$ improved T50 and MGT by 14 and 8%, respectively in the Bouchra variety compared to the control.

Key-Words: Chickpea, Germination, Glucose, osmopriming, Storage, Sucrose.

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1 Introduction

Chickpea is a plant mainly intended for human consumption, especially its grains, which are rich in protein (26.77%) [1], and without cholesterol. The chickpea has an excellent balance of essential amino acids. Moreover, it is also rich in calcium, phosphorus, vitamins B1, B2, and dietary fiber [2, 3]. Seed germination is the whole of the events, which begins with the absorption of water by the seed and ends with the elongation of the embryonic axis and the emergence of the radicle.

Seed germination is important biological processes in the plant life cycle. Seed life is a key feature of agriculture, directly affecting seed germination and ultimately determining crop productivity and thus food security. Numerous studies have shown that seed deterioration is regulated by complex interactions between various genetically controlled endogenous factors and exogenous environmental factors such as temperature and relative humidity during seed storage [4]. Germination and dormancy depend on the balance between radicle growth and embryonic tissue resistance. The activity of cell wall remodeling proteins affects the resistance of these tissues. Seeds rapidly lose their dormancy during absorption under certain conditions [5]. Seed priming can be used to achieve uniform germination and to lift seed dormancy in different plants. Several studies have reported that seed priming helps to accelerate and synchronize germination. In fact, seed priming is an effective technology to improve rapid and uniform emergence and to achieve a high level of vigor, leading to better crop establishment and yield [6,7,8].

Seed priming is a technique that is performed before sowing. It is a method that involves soaking the seeds in different solutions for a specified period under controlled conditions but before the radicles emerge [9,10,7]. The seeds are then dried until they reach their initial moisture content. This stimulates several metabolic processes that enhance seed germination and emergence. Seed priming is an easy, effective, and inexpensive technique. Seed priming

provides many benefits such as uniformity, early emergence, speed, germination in a wide temperature range, crop establishment, and germination of dormant seeds [11,12]. Abscisic acid is the plant hormone most involved in maintaining seed dormancy and delaying germination [13]. Seed priming induces different physiological changes in seeds such as protein, nucleic acid synthesis, enzyme activation, increased antioxidant activity, and reduced lipid peroxidation level in plants [14,15]. Several seed priming techniques have been developed including hydro-priming, halo-priming, glucopriming, etc. Its various techniques have proven effective in improving germination and growth parameters in different plants [7,8,10]. The aim of our study is to evaluate the effect of priming with glucose and sucrose on seed dormancy and germination parameters of chickpea seeds.

2. Material and methods

The experimental work was conducted at the Laboratory of Biotechnology and Agrophysiology of Symbiosis- Faculty of Science and Technology - Marrakech, with the aim of studying the effect of osmopriming (glucose and fructose) on the germination behavior of four varieties of chickpea (Rizki - Farihane - Zahor - Moubarak). During this work, we performed a control with distilled water alone (UP) and three concentrations (3, 6, 10 $g.l^{-1}$) of glucose (priming glucose concentration *PGC*) and sucrose (priming sucrose concentration *PSC*). For each variety, there are 3 replicates and 20 seeds in each petri dish.

2.1. Germination percentage (GP) = (n/N) × 100 with n: number of seeds germinated on the day i and N: total number of seeds.

2.2. Mean germination time (MGT) calculated according to the equation of [16] and expressed in days. The equation is as follows: MGT = $\sum$ D n /$\sum$ n Where "D" is the number of days counted from the beginning of the test and "n" is the number of seeds that germinate on day 'D'.

2.3. Time to 50% germination (T50) calculated according to the formula of [17], T50 is defined as the days needed to reach 50 percent of the final germination percentage.

Statistical analysis

Statistical analysis was conducted using SPSS (21.0) software. A two-way analysis of variance was carried out (ANOVA II) using three replicates per combination and per treatment for almost all the parameters studied. Mean values and standard errors were determined. The Tukey test was used to compare the means of the parameters considered.

3 Results

3.1. Effect of seed priming with glucose on germination percentage, MGT, and T50

3.1.1. Effect of seed priming with glucose on percent germination (PG)

The percentage of germination (PG) showed highly significant variation ($p<0.001$, Table 1) in most of the chickpea varieties studied compared to the control. The PG of treated seeds was improved by (15%) for seeds treated with priming glucose concentration 6 $g.L^{-1}$ compared to the control in the Mubarak variety. The latter showed the lowest PG in both treated and untreated seeds (56.7%). The percentage of germination decreased slightly (1.7%) in the treated seeds of the variety Rizki for the PGC 6 $g.L^{-1}$ and those not treated. For the variety Farihane, the germination percentage increased by (12%) in seeds treated with PGC 10 $g.L^{-1}$ compared to the control. This variety showed the highest PG in seeds treated with PGC 3, 6, and 10 g.L-1 (91.7, 91.7, and 96.7%, respectively). The PG of seeds primed with PGC 3 $g.L^{-1}$ showed a slight decrease (2%) compared to untreated seeds.

3.1.2. Effect of seed priming with glucose on MGT

The germination parameter MGT showed a significant improvement ($p<0.001$, Table 1) in some chickpea varieties studied compared to the control. Indeed, for the Mubarak variety, the MGT decreased (19%) for the priming glucose concentration (*PGC*) of 6 $g.L^{-1}$ compared to the control. For the variety Zahor, the MGT has experienced a decrease of 2% for the PGC 3 $g.L^{-1}$ compared to the control. For the variety Rizki, MGT increased slightly (0.1%) for the PGC 3 $g.L^{-1}$ compared to the control. The MGT increased slightly (5%) also for the PGC 10 $g.L^{-1}$ compared to the control in the Farihane variety.

3.1.3. Effect of seed priming with glucose on T50

The germination time T50 showed a significant decrease ($p<0.001$, Table 1) in the studied chickpea varieties compared to the control. Indeed, the priming with glucose decreased the T50 in Bouchra and Farihane varieties by 9% and 12% with a concentration of PGC 6 $g.L^{-1}$ and for the Zahor variety by 12% with the treatment of 3 $g.L^{-1}$. The highest value of T50 was noted in the variety Moubarak (4.73d) with PGC 3 $g.L^{-1}$ and for its control with a value of 4.7

Table 1: Effect of seed priming with glucose: PGC (UP, 3, 6 and 10 $g.L^{-1}$) on germination parameters (GP, MGT and T50) of four chickpea varieties (Moubarak, Rizki, Farihane and Zahor).

Varieties	PGC (g.L^{-1})	GP (%)	MGT (d)	T50 (d)
Moubarak	**UP**	56.67 ± 2.9^{h}	4.53 ± 0.055^{ab}	4.70 ± 0.01^{ab}
	3	56.67 ± 2.9^{h}	4.51 ± 0.049^{abc}	4.73 ± 0.05^{a}
	6	66.67 ± 2.9^{jh}	4.40 ± 0.046^{cde}	$4.54 \pm 0.02^{a-e}$
	10	65 ± 5^{h}	$4.48 \pm 0.036^{a-d}$	$4.56 \pm 0.03^{a-d}$
Rizki	**UP**	100 ± 0^{a}	4.13 ± 0^{f}	4.36 ± 0^{de}
	3	$90 \pm 10^{a-e}$	4.15 ± 0.04^{f}	4.34 ± 0.03^{e}
	6	98.33 ± 2.5^{ab}	4.50 ± 0.015^{abc}	4.42 ± 0.02^{cde}
	10	$86.67 \pm 2.9^{b-e}$	4.33 ± 0.045^{e}	$4.53 \pm 0.03^{a-e}$
Farihane	**UP**	85 ± 5^{cde}	4.38 ± 0.005^{de}	$4.56 \pm 0.01^{a-d}$
	3	$91.67 \pm 6.3^{a-d}$	4.46 ± 0.06^{bcd}	4.41 ± 0.02^{cde}
	6	$91.67 \pm 2.9^{a-d}$	4.51 ± 0.01^{abc}	4.37 ± 0.01^{de}
	10	96.67 ± 2.9^{abc}	4.40 ± 0.05^{cde}	4.49 ± 0.01^{cde}
Zahor	**UP**	80 ± 0^{def}	4.51 ± 0.02^{abc}	4.58 ± 0.02^{abc}
	3	78.33 ± 2.9^{efj}	4.41 ± 0.04^{cde}	4.39 ± 0.03^{cde}
	6	66.67 ± 2.9^{jh}	4.60 ± 0.04^{a}	$4.52 \pm 0.02^{b-e}$
	10	68.33 ± 2.9^{fjh}	4.45 ± 0.45^{bcd}	4.46 ± 0.01^{cde}

	dF	F	F	F
varieties	3	1.7^{***}	1.82^{***}	1.23^{***}
Priming	3	1.82^{***}	1.38^{***}	0.76^{***}
Error	90			

3.2. Effect of seed priming with sucrose on germination percentage, MGT, and T50

3.2.1. Effect of seed priming with sucrose on germination percentage (GP)

Germination percentage increased significantly ($P< 0.001$, Table 2) in the treated varieties compared with their respective controls. The highest PG values in most genotypes were those corresponding to the 10 g.L^{-1} treatment concentrations. The variety Rizki showed the highest PG (98%) in the treated seeds and the untreated control while the variety Farihane recorded the lowest PG in the control (38%). Seeds of the Farihane variety subjected to osmopriming (3, 6, and 10 g.L^{-1}) had a significant increase in germination percentage (88.3%, 81.66%, and 98.33%, respectively) compared to the control (38.33%). The priming treatment that gave the best results varied from one variety to another: PSC 3 g.L^{-1} for the Rizki variety (98%) and PSC 10 g.L^{-1} for the Zahor variety (91.7%).

Table 2: Effect of seed priming with sucrose: PSC (UP, 3, 6, and 10 g.L^{-1}) on germination parameters (GP, MGT, and T50) of four chickpea varieties (Moubarak, Rizki, Farihane, and Zahor).

Varieties	PGC (g.L^{-1})	GP (%)	MGT (d)	T50 (d)
Moubarak	**UP**	75 ± 5^{c}	4.7033 ± 0.025^{gh}	4.4267 ± 0.0251^{de}
	3	73 ± 7.63^{b}	4.7533 ± 0.0208^{fg}	4.49 ± 0.01^{bc}
	6	88 ± 2.88^{ab}	4.6467 ± 0.00577^{h}	4.3667 ± 0.0152^{e}
	10	88.33 ± 9.07^{ab}	4.7833 ± 0.0152^{f}	4.37 ± 0.01^{e}
Rizki	**UP**	98.33 ± 2.88^{a}	4.9133 ± 0.0321^{e}	4.4233 ± 0.0577^{de}
	3	98.33 ± 2.88^{a}	4.9167 ± 0.0158^{e}	4.39 ± 0.001^{e}
	6	83 ± 15^{ab}	5.0567 ± 0.0404^{bc}	4.52 ± 0.03^{b}
	10	91.25 ± 12.45^{a}	5.09 ± 0.0360^{bc}	4.5333 ± 0.0321^{b}
Farihane	**UP**	38.33 ± 2.88^{c}	4.71 ± 0.02^{fgh}	4 ± 0.001^{f}
	3	88.33 ± 7.63^{ab}	5.2233 ± 0.0115^{a}	4.5333 ± 0.0152^{b}
	6	81.66 ± 10.41^{ab}	5.0167 ± 0.0513^{cd}	4.5133 ± 0.0152^{bc}
	10	98.33 ± 2.88^{a}	4.9533 ± 0.0152^{de}	4.4533 ± 0.0251^{cd}
Zahor	**UP**	88.33 ± 7.63^{ab}	5.1 ± 0.02^{b}	4.52 ± 0.0264^{b}
	3	90 ± 00^{ab}	5.1 ± 0.0264^{b}	4.4533 ± 0.0152^{cd}
	6	86.66 ± 7.63^{ab}	4.9033 ± 0.0152^{e}	4.6367 ± 0.0305^{a}
	10	91.66 ± 7.63^{ab}	4.89 ± 0.01^{e}	4.4833 ± 0.0208^{bcd}

	dF	F	F	F
varieties	3	1.607^{***}	1.282^{***}	1.223^{***}
Priming	3	1.282^{***}	1.638^{***}	0.716^{***}
Error	90			

3.2.2. Effect of seed priming with sucrose on MGT

The mean germination time showed a highly significant increase ($P<0.001$, Table 2) in the treated

seeds compared to their respective controls in most of the varieties studied. The highest value was noted in the PSC 3 $g.L^{-1}$ treatment of the variety Farihane (5.22 d), and the highest speed of the untreated seeds was recorded in the variety Zahor (5.1 d). In the variety Bouchra we noted a minimum value of MGT for the PSC 10 $g.L^{-1}$ (4.7 d). In the variety Zahor, the best result was recorded with the PSC 3 $g.L^{-1}$ (5.1 d).

3.2.3. Effect of seed priming with sucrose on T50

The T50 time is a germination parameter that informs about the time required for 50% of the seeds to germinate. The osmopriming results showed a highly significant decrease ($P<0.001$, Table 2) in T50 time in treated seeds compared to their respective controls in most of the varieties studied. In the Farihane variety, the untreated seeds could not reach 50% of seed germination after 7 days, however, the PSC 10 $g.L^{-1}$ recorded the highest T50 (4.45d). The Bouchra variety also experienced a decrease in T50 for the PSC 6 and 10 $g.L^{-1}$ (4.37d).

4 Discussion

Sugar is the main component of storage organs. Sugar and its derivatives are energy and signal molecules involved in various plant development processes such as seed dormancy and plant growth [18]. Seed priming enhances the antioxidant system, and increases sugar metabolism and the activities of enzymes involved in carbohydrate metabolism in germinating seeds [19]. Seed priming treatment had a positive impact on germination percentage this may be explained by the increase in seed water content, which allows the activation of enzymes responsible for embryo development and exploitation of starch-rich reserves. In addition, this priming increases cell RNA content, improves DNA replication, and strengthens defense systems by increasing the activity of antioxidant enzymes such as catalase and superoxide dismutase [20]. In the same sense, this improvement in germination percentage can be explained by the reduction of the effect of ABA (senescence hormone), such a reduction of ABA is essential for germination to occur [21]. The beneficial effects of priming are associated with various physiological, biochemical, cellular, molecular, and genetic changes such as reserve mobilization, osmolyte biosynthesis, antioxidant process, and cell cycle regulation [22].

Seed priming has allowed the lifting of the dormancy seed in most of the chickpea seeds studied. Indeed, the results showed that priming with glucose (6 and 10 $g.L^{-1}$) increased the germination percentage by 15% in the Moubarak variety and by 12% in the Farihane variety compared to their respective controls. Similarly, priming with sucrose (6 and 10 $g.L^{-1}$) increased the PG by 14.8 and 61% in the varieties Bouchra and Farihane compared to their respective controls. Several authors have reported the effective effects of osmopriming on seed dormancy and vigor. Seed priming leads to the breaking of seed dormancy, reduces seedling emergence time, and improves germination parameters and growth of various plants [23]. Indeed, after priming, different phytohormones are involved in the lifting of seed dormancy especially abscisic acid (ABA), gibberellic acid (GA), and cytokinins [23,24].Most importantly, the balance between the levels and pathways of abscisic acid (ABA) and gibberellin is essential for germination enhancement and dormancy stimulation [25]. Pretreatments such as osmopriming help increase the germination rate, vigor index, and dry weight of seedlings; these improvements are particularly pronounced when plants are subjected to abiotic stress. This is in agreement with the work of [7,8 and 10] who noted that osmopriming improved seed germination, growth, antioxidant responses, and membrane stability in alfalfa and chickpea under water deficit.

The use of glucose priming improved the MGT in some chickpea varieties compared to the control. In fact, the MGT decreased slightly in the seeds of the Mubarak variety primed with glucose (6 $g.L^{-1}$). Seed priming allows the increase of cell division in treated seeds and the stimulation of several metabolic activities involved in the first phase of germination [26, 27]. The results showed an improvement in the T50 with both primings. Indeed, Priming with 6$g.L^{-1}$ of glucose and sucrose improved T50 by 3.4% and 14% in Mubarak and Bouchra varieties compared to their respective controls. The improvement of germination parameters like T50 and MGT is associated with the concentration of soluble sugars [28,29]. In addition, seed priming improves germination and seedling growth due to protein synthesis, membrane repair mechanisms, and increased availability of substrates for germination [28, 30].

5 Conclusion

This study showed that the applied priming treatments improved the different germination parameters studied. Our study also showed that seed priming significantly lifted seed dormancy and improved the germination rate. Thus, different seed priming treatments can potentially be used in agricultural production to increase germination uniformity and better plant growth and development.

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