

EFFECT OF SEED INVIGORATION BY OSMOPRIMING ON SEED QUALITY PARAMETERS IN THREE *Capsicum species* UNDER AMBIENT HUMID CONDITIONS

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ABSTRACT

Slow and uneven germination of seeds of *Capsicum* species are undesirable to the seed producers and farmers. Seed osmo priming is one of the pre-treatments for enhancing seed quality. An experiment was conducted to determine the influence of osmopriming techniques on seed lots of three *Capsicum* species. The seeds were primed using seven different solutions for three different priming durations. Primed seed lots were evaluated for seed germination, seedling vigour index, seedling emergence and mean seedling emergence time using factorial arrangement in completely randomized design with three replications. Differences were found in seed germination and germination index among the three *Capsicum* species. Osmopriming agents and their treatment duration significantly affected seed germination and seedling emergence. Seeds primed in 2% KCl solution gave the highest seed germination while the seeds germinated faster when primed in 4% KCl solution. Longest priming duration of 36h adversely affected seed germination, germination index, seedling vigour, seedling emergence and mean seedling emergence time. Seeds of *C. frutescens* and *C. chinense* primed for 12h had significantly higher germination than those of *C. annuum*. The results showed that priming in distilled water, 2% KCl and 10 and 20% PEG 8000 can significantly improve seed germination in *Capsicum* species.

Keywords: Seed priming, seed vigour, treatment, priming solution and germination.

INTRODUCTION

Capsicum species popularly known as pepper is the world's second most important crop after tomato (Yoo *et al.*, 1989). According to Adamu *et al.* (1994), Nigeria is the largest producer of pepper in Africa, accounting for about 50% of the African production. Four different species of *Capsicum* are domesticated; *C. annuum*, *C. frutescens*, *C. chinense* and *C. pubescens*. Consumption of pepper accounts for about 20% of the average vegetable consumption per person per day in Nigeria (Alegbejo, 2002). It is used extensively in food flavouring in the daily diets of over 1,200 million Nigerians, irrespective of their socio-economic status. It is used in the preparation of soup and stew, which are among the major essential complements of staple based on cereals and root crops and also forms remedies for toothache and sore throat (Bosland, 1994; Leung and Foster, 1996).

Seed priming technology has been reported to have positive effect on seed germination and seedling growth in some species. Improved seed invigoration techniques are known to reduce emergence time, accomplish uniform emergence and give better stand in many horticultural and field crops. Such seed invigoration techniques include hydro priming, osmo conditioning, hardening and soaking before sowing (Ashraf and Foolad, 2005). Although, seed priming techniques have been found effective for better germination and seedling establishment in pepper under controlled conditions (Basra *et al.*, 2005), no comprehensive study has yet been

done to evaluate the effect of seed invigoration by osmopriming agents on germination and growth of different *Capsicum* species under ambient tropical conditions in Nigeria.

Osmo priming technique is a method of seed priming using salt solution in order to improve seed quality parameters. Here, the seed is pre-treated in an osmotic solution for enhancement of seed moisture content and speed of germination (Ehsanfar, 2006). This treatment consists of the controlled hydration of the seeds, initiating the preparatory processes for germination but avoiding radical protrusion (Ashraf and Foolad, 2005). The hydration of seeds is controlled by active osmotic solutions that influence the priming effect. Both the dosage and duration of treatment are important consideration in osmo priming. Although several studies have been carried out to determine the efficiency of seed osmo priming in some crops species and varieties, it is important to determine which dosage is more efficient to be used for a particular species. According to Reis *et al.* (2012), many factors can affect the duration of priming. Short duration may not always be an effective priming treatment (Reis *et al.*, 2012). On the other side, extended durations may allow seed germination during the treatment or adversely affect the seed vigour (Nascimento and Costa, 2009). This study was done to determine optimal seed priming conditions for three *Capsicum* species and to find out the most promising treatments for each of the species.

MATERIALS AND METHODS

Seed materials

Newly harvested seeds of three species of pepper (*C. frutescens*, *C. chinense* and *C. annum*) used for the study were sourced from National Institute of Horticultural Research and Training (NIHORT), Ibadan, South western Nigeria. The experiment was carried out both at the seed laboratory and teaching and research farms, Plant Breeding and Seed Technology Department, FUNAAB, Ogun State, South western Nigeria between February and May, 2010.

Seed Priming Solution

Seven priming solutions were investigated in the study, viz., distilled water (control), potassium chloride (KCl; 2% and 4% w/v), potassium dihydrogen phosphate (KH_2PO_4 ; 0.5% and 1% (w/v)) and polyethylene glycol (PEG 800; 10% and 20% w/v). All the chemicals were dissolved in 100 ml of distilled water. The water potential of an aqueous solution of PEG 8000 has been reported to be -0.8 MPa (Fanan and Novembre, 2007; Nascimento and Lima, 2008).

Seed priming treatment

Fifty seeds of each *Capsicum* species were placed in individual net bags and immersed in each of the priming solutions for 12, 24 and 36 h. Distilled water served as the control. All seeds were then removed from the priming solutions, rinsed thoroughly with distilled water and surface dried lightly using blotting paper. While still damp, the seeds were treated with a fungicide (*apron plus* -trade name, with thiamethoxan, methylaxyl and difenocanazole as active ingredients at the recommended rate of 5g/1kg seed) and then allowed to dry for 3h on paper towel at room temperature (28-29°C).

Experimental design

The treatments were arranged in a completely randomized design according to a factorial arrangement in three replications. There were 189 experimental units consisting of three factors (priming solution (7), pepper species (3) and priming duration (3)) which were replicated three times.

Seed assessment

Seed samples were taken from each primed seed lots and tested for the following seed quality traits;

Germination percentage: One hundred seeds replicated three times from each genotype were placed in Petri dishes lined with moistened paper towels at 25⁰C temperature in an incubator (ISTA, 1995) and recorded using the formula below.

$$\frac{\text{Seedling germination after 8 days}}{\text{No of seeds sown}} \times 100$$

Germination Index: This was calculated as $\sum GT/Tt$

Where Gt is the number of seed germinated on day t and
Tt is the number of days.

Seedling length: This was assessed by measuring the length in centimetres (cm) from the tip of the cotyledon to the point of radicle emergence with a metric ruler.

Seedling fresh weight: This was determined by weighing 10 randomly selected seedlings in grammes at 8 days after germination (ISTA, 1995).

Seedling dry weight: This was measured using 10 randomly selected seedlings using oven dry method at 130⁰C for one hour at 8 days after germination (ISTA, 1995).

Seedling Vigour Index (SVI): This was determined by multiplying percentage seed germination by the average of seedling length after 8 days of germination and divided by 100 (Kim *et al.*, 2002; Adebisi, 2004).

Seedling emergence

Top soil was collected from the Teaching and Research Farm of FUNAAB into medium sized polythene bags and labelled after which 50 seeds from each seed lots were placed separately in each bag and covered lightly with the soil. Each bag was given equal volume of water daily until no further emergence occurs.

Seedling emergence was determined as $\frac{\text{number of seeds at 14 days}}{\text{Number of seeds sown}} \times 100$

Mean emergence time: This was measured as time taken for seeds to emerge from soil.

Data analysis

Data obtained from the trial were subjected to analysis of variance with the significant level tested at 1 and 5 % probability levels. Significant treatment means were separated using Tukey HSD test at 5 % probability level.

RESULTS

Result in Table I shows the summary of ANOVA for the seed quality characteristics evaluated in three *Capsicum* species. Germination percentage and germination index varied significantly between the species. The priming solution had a significant effect on seedling emergence. The effects of priming duration were significant with regard to germination percentage, mean seedling emergence time and germination index. The interaction of priming media x duration significantly affected the mean seedling emergence time whereas the interaction of species and priming duration significantly affected the germination percentage and mean seedling emergence time. The interaction of species x priming media and species x priming media x priming duration did not have any significant effect on any of the seed quality characteristics examined.

The main effect of species, priming solution and duration on the seed quality of three *Capsicum* species is presented in Table 2. *Capsicum frutescens* gave the highest germination value (88.35 %) though not significantly different from value of 86.51 obtained with *C. chinense* while *Capsicum annuum* recorded the least value. Highest values of germination index (5.94 days), seedling vigour index (1.72) were obtained with *C. frutescens* above other two species. Seedling emergence and mean emergence time had statistically similar values among the three species.

The main effect of species, priming media and duration on the seed quality of three *Capsicum* species is presented in Table 2. Of the three species studied, *C. frutescens* showed the best response to the priming treatments, while the other two species of *Capsicum* did not show any notable difference with regard to the seed quality characteristics studied. According to the data in Table 2, seed primed with distilled water (87.77%), 2% KCl (88.88%), 0.5% KH₂PO₄ (86.14%), 10% PEG (86.51%) and 20% PEG (86.59%) had the highest germination. Statistically similar germination index and seedling vigour index values were obtained after treatment with the seven priming media used for the experiment.

Seedling emergence value (88.22 %) was highest after treatment with distilled water. Though not significantly different from emergence values obtained from other priming solution except 2 % KCl and 4 % KCl which had lower emergence values of 84.70 and 84.92 % respectively. On the mean emergence time, all the solution except 4 % KCl had similar and higher mean emergence time values while 4 % KCl recorded the lowest mean emergence time of 4.69 days. On priming duration effect, 12 and 36 hrs duration had higher seed germination and germination index percentages while 24 hrs duration had the lowest. For seedling vigour index, 12 hrs and 24 hrs duration gave higher seedling vigour values of 1.71 and 1.64 respectively when compared with 36 hrs duration. Priming duration of 36 hrs gave the highest seedling emergence though not significantly different from value of 85.90 % obtained with 12 hrs priming duration. On mean emergence time, 12 hrs and 36 hrs durations had the highest value of 5.37 and 5.54 days while 24 hrs priming duration recorded the lowest mean emergence value (4.65).

Seedling emergence value (88.22%) was highest after treatment with distilled water, though not significantly different from treatment with other priming media except 2% KCl and 4% KCl which had lower emergence values of 84.70 and 84.92%, respectively. On the mean seedling emergence time, all the media except 4% KCl had similar and higher values while 4% KCl treatment recorded the lowest value of 4.69days. Priming the seeds for 12 and 24 h showed significantly higher seed germination, seed germination index, seedling vigour index. Moreover, priming for 24h also gave the highest seedling emergence (88.07%) and took the shortest time for seedlings to emerge (4.65 d).

Table 3 presents the effect of osmo priming duration on seed germination of three *Capsicum* species across priming media. The results showed that the seeds of *C. frutescens* and *C. chinense* recorded the highest seed germination initially (90.48 and 88.57%, respectively at 12h after priming and 90.10 and 87.62% after 24h). However, after 36 h of soaking, seed germination was the highest in *C. annuum*.

The effect of osmo priming duration on the mean seedling emergence time is presented in Table 4. The data revealed that when primed for 12 h, seeds of *C. frutescens* and *C. chinense* took comparatively longer time for seedling emergence (5.95 and 5.60d respectively) whereas when primed for 24h, *C. annuum* had the highest mean seedling emergence time (5.12 d). When soaked for 36 h, seeds of *C. frutescens* took significantly longer time for seedlings to emerge (5.95d).

Table 5 shows the effect of osmo priming solution on mean emergence time of three *Capsicum* species across priming duration treatments. From this result, seeds of *C. frutescens*, primed in a solution of 2% KCl had the highest mean emergence time (6.50 days) compared to the six other priming solution. Also seed primed in five other solutions except 4% KCl had statistically similar seed emergence values with seed of *C. frutescens* primed in distilled

water. Seeds of *C. chinense* primed in a solution of 4% KCl and 20% PEG were found with the highest mean emergence time of 5.14 and 5.30 days, respectively. Also, seeds of *C. chinense* primed in solutions of 0.5% KH₂PO₄, 1% KH₂PO₄ and 10% PEG were higher in mean emergence time above values of 4.40 days and 4.01 days recorded with distilled water and 2% KCl treatments respectively. *C. annuum* seeds primed in solution of 10 % PEG and distilled water recorded the higher mean emergence time of 6.80 and 6.10 days while all other solutions had mean emergence time below distilled water treatment. Also, seeds of *C. annuum* primed with 4 % KCl and 20% PEG had the earliest mean emergence time of 4.03 and 4.80 days.

Data in Table 6 shows the effect of osmo priming solution and priming duration on seed germination percentage of *Capsicum* species. With 12 hrs duration, 2% KCl (92.00 %), distilled water (89.11 %), 4% KCl (89.11 %), 10% PEG (87.11 %), and 20% PEG (88.44%) primed seeds had higher germination percentages over other priming treatments whereas seeds primed with 0.5% KH₂PO₄ and 1% KH₂PO₄ solution had lower germination percentages. At 24 hrs duration, seeds primed in solutions of distilled water (89.60 %), 2% KCl (90 %), 0.5% KH₂PO₄ (87.80 %), 1% KH₂PO₄ (88.22 %), and 10% PEG (88.22 %) recorded higher germination percentages above two other priming solution which had similar values. Also at 36 hrs duration, seeds primed in 4% KCl had the highest germination percentage (86 %) while the other treatments were not significantly different from one another.

DISCUSSION

The beneficial effects of osmo priming techniques have been observed in many vegetables species like in pepper, egg plant, tomato, among others. The result obtained from the study indicated that differential responses in seed germination and germination index occurred among the three pepper species after osmo priming. This implies that if higher seed germination and lower days to germination (germination index) are the objectives of seed improvement, superior *Capsicum* species can be selected. The result revealed that significant differences were observed among osmo priming treatment in respect to seed germination and seedling emergence, indicating the possibility of selection of best osmo priming condition for higher germination and seedling emergence. The result also revealed that differences in osmo priming duration (hours) were responsible for the differences observed in percentage seed germination and mean emergence time of the three *Capsicum* species examined. The differences observed in seed germination and mean emergence time among the three *Capsicum* species were due to differences in the effectiveness of the three priming durations. Variations in mean emergence time among the seven osmo priming solution were influenced by the differences in osmo priming durations. Shun *et al.* (2009) observed that efficacy of priming treatments on seed germination of sesame (*Sesamum indicum*) was found to depend on priming agent, concentration and duration of priming.

Data on main effect revealed that seeds of *Capsicum frutescens* had the germination and seedling vigour level but with highest number of days to germination as revealed by germination index. On osmo priming solution effect, 2 % KCl solution had higher seed germination which was slightly different from other treatments including distilled water. Lowest mean emergence time (4.69 days) was obtained with seeds primed in solution of 4 % KCl while other priming solution had mean emergence time above 5 days. In terms of priming duration effect, seeds primed in any of the priming solution solution for 12 and 24 hrs had higher germination, germination index, seedling vigour level and seedling emergence whereas seeds primed for 24 hrs recorded reduce mean emergence time (4.65 days).

On the response of three *Capsicum* species to osmo priming duration, seeds of *C. frutescens* and *C. chinense* primed in any of the solution for 12 hrs had distinct higher germination of 90.48 and 88.75 % than *C. annuum*. At 24 hrs priming duration, seeds of *C. frutescens* had highest germination whereas *C. annuum* seeds primed for 36 hrs recorded highest germination(84.50 %). Different responses in seed germination due to differences in effectiveness of priming duration were reported by Reis *et al.* (2012) in egg plant, Ajala *et al.* (2008) in NERICA rice and Asgharipour *et al.* (2011) in tomato. In respect to mean germination time, seeds of *C. annuum* primed for 12 hrs in any of the osmo priming solutions had lower days to germination (4.60 days) whereas seeds of other two capsicum species had lower mean emergence time above 5 days in the three species.

Several osmo priming agents have been recommended for seed priming and positive effects have been reported (Fanani and Novembre, 2007, Lerna *et al* 2008; Adebisi *et al.* 2011(a and b); Ajala *et al.* 2007; Reis *et al.* 2012). The study showed that 1% KH₂PO₄ solution reduced the mean emergence time in *C. frutescens* while 2 % KCl increased mean emergence time in the same species. Conversely, 2 % KCl and distilled water solutions was found to reduce the mean emergence time in *C. chinense* whereas 20 % PEG recorded highest mean emergence time (5.30 days). Seeds of *C. annuum* primed in 4 % KCl solutions had the lowest mean emergence time whereas seeds primed in distilled water and 10 and 20 % PEG solutions resulted in higher mean emergence time. Asgharipour *et al.* (2011) observed that priming increased tomato seed emergence, emergence rate and plumule length whereas priming with PEG negatively affected the rate and growth of emerged seedlings. In terms of seed germination, priming for 12 hrs in 2 % KCl, 4 % KCl, 10 % PEG and 20 % PEG as well as distilled water increased seed germination, irrespective of capsicum species. Priming duration for 24 hrs in all the priming solution except 4 % KCl and 20 % PEG had higher and similar seed germination whereas priming in 4 % KCl for 36 hrs gave the highest seed germination above other priming agents including distilled water.

CONCLUSION

Efficacy of osmo priming treatments is dependent on capsicum species, priming agent and concentration duration if priming for enhancement of seed germination of capsicum species. Osmo priming improves the seed germination, seedling emergence and does not affect seedling vigour level and germination index of the capsicum species. Priming in distilled water, 2 % KCl, 4 % KCl and 10 and 20 % PEG from 12 hrs is efficient to improve seed germination in capsicum species.

Acknowledgement

The material support by the Department of Plant Breeding and Seed Technology (PBST), Federal University of Agriculture, Abeokuta (FUNAAB), Ogun State, Nigeria is highly appreciated. The authors acknowledged the intellectual contributions by academic staff of Department of PBST, FUNAAB during the defense of the proposal and final report of the experiments.

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Table 1: Summary of analysis of variance showing mean sum of squares of seed quality characters evaluated in three *Capsicum* species

Source of Variation	Degree of freedom	Germination percentage	Seedling Vigour Index	Seedling Emergence	Mean Emergence Time	Germination Index	Seedling Fresh weight	Seedling Dry Weight
Rep	2	17.72ns	0.01ns	68.85ns	0.58ns	2.48ns	5.29ns	1.72ns
Species(S)	2	94.16**	0.10ns	156.93ns	1.87ns	6.58**	9.58ns	1.02ns
Priming solution (P)	6	82.17ns	0.02ns	98.46*	2.15ns	1.96ns	0.00ns	3.53ns
Duration (D)	2	149.59**	0.09ns	170.20ns	13.97**	4.46*	7.83ns	4.32ns
P x D	12	46.82ns	0.04ns	92.73ns	4.92*	1.63ns	0.00ns	1.97ns
S x D	4	84.32**	0.05ns	52.83ns	7.29*	2.19ns	8.23ns	3.34ns
S x P	12	14.50ns	0.03ns	84.24ns	1.43ns	1.12ns	0.00ns	2.57ns
S x P x D	24	27.37ns	0.33ns	56.01ns	1.54ns	0.94ns	8.66ns	1.55ns
Error	124	30.73	0.32	66.55	1.87	1.35	7.80	2.24

** - significant at 1% probability * - significant at 5% probability level ns - not significant

Table 2: Main effect of species, priming treatment and priming duration on seed quality of three *Capsicum* species

Treatment	Germination percentage	Germination Index	Seedling Vigour Index	Seedling Emergence	Mean Emergence Time
(a) Species					
<i>Capsicum frutescens</i>	88.35a	5.94a	1.72a	88.03a	5.36a
<i>Capsicum Chinense</i>	86.51ab	5.53b	1.68b	84.97a	5.28a
<i>Capsicum annuum</i>	85.23b	5.30b	1.64b	85.84a	5.01a
Mean	86.70	5.60	1.70	86.30	5.22
(b) Osmo priming solution					
Distilled water	87.77ab	5.72a	1.76a	88.22a	5.40a
2% KCl	88.88a	5.99a	1.72a	84.70b	5.30a
4% KCl	85.25b	5.35a	1.70a	84.92b	4.69b
0.5% KH ₂ PO ₄	86.14ab	5.42a	1.76a	86.88ab	5.32a
1% KH ₂ PO ₄	85.70b	5.33a	1.64a	86.55ab	5.05a
10% PEG	86.51ab	5.94a	1.71a	86.37ab	5.53a
20% PEG	86.59ab	5.51a	1.73a	86.29ab	5.03a
Mean	86.70	5.61	1.71	86.30	5.93
(c) osmo priming duration					
12 hours	88.31a	5.78a	1.71a	85.90b	5.37a
24 hours	87.30a	5.69a	1.64a	88.07a	4.65b
36 hours	84.47b	5.28b	1.58b	84.85b	5.54a
Mean	86.70	5.60	1.64	86.27	5.17

Means followed by the same alphabet along the column are not significantly different from each other at 5% probability level according to Tukey HSD test

Table 3: Effect of osmo priming duration on seed germination of three cross *Capsicum* species across priming solution

Species	Seed germination (%)		
	12 h	24 h	36 h
<i>Capsicum frutescens</i>	90.48a	90.10a	84.47b
<i>Capsicum Chinense</i>	88.57a	87.62b	83.33b
<i>Capsicum annuum</i>	85.91b	84.20c	85.63a
Mean	88.32	87.31	84.50

Means followed by the same alphabet along the column are not significantly different from each other at 5% probability level according to Tukey HSD test

Table 4: Effect of osmo priming duration on mean seedling emergence time of three *Capsicum* species across priming solution

Species	Mean seedling emergence time (d)		
	12 h	24 h	36 h
<i>Capsicum frutescens</i>	5.95a	4.30b	5.95a
<i>Capsicum chinense</i>	5.60a	4.60b	5.44b
<i>Capsicum annuum</i>	4.60b	5.12a	5.33b
Mean	5.38	4.71	5.63

Means followed by the same alphabet along the column are not significantly different from each other at 5% probability level according to Tukey HSD test

Table 5: Effect of Osmo Priming Media on Mean Seedling Emergence Time of Three *Capsicum* spp across Priming Duration Treatments

Treatment	Mean Seedling Emergence Time (days)		
	<i>Capsicum frutescens</i>	<i>Capsicum chinense</i>	<i>Capsicum annuum</i>
Distilled water	5.80b	4.40c	6.10a
2% KCl	6.50a	4.01c	5.40b
4% KCl	4.90c	5.14a	4.03c
0.5% KH ₂ PO ₄	5.34b	4.62b	5.99b
1% KH ₂ PO ₄	4.94c	4.50b	5.74b
10% PEG	5.14b	4.70b	6.80a
20% PEG	5.02b	5.30a	6.80a
Mean	5.38	4.67	5.58

Means followed by the same alphabet along the column are not significantly different from each other at 5% probability level according to Tukey's HSD test.

Table 6: Effect of osmo priming solution and priming duration on seed germination percentage of *Capsicum* species across species

Treatment	Seed germination (%)		
	12h	24h	36h
Distilled water	89.11a	89.60a	84.70b
2% KCl	92.00a	90.00a	84.70b
4% KCl	89.11a	80.70c	88.00a
0.5% KH ₂ PO ₄	86.00b	87.80ab	84.70b
1% KH ₂ PO ₄	86.44b	88.22ab	82.44b
10% PEG	87.11ab	88.22ab	84.22b
20% PEG	88.44a	86.70b	84.70b
Mean	88.32	87.32	84.50

Means followed by the same alphabet along the column are not significantly different from each other at 5% probability level according to Tukey HSD test