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Effect of soaking periods and concentrations of priming agents in improving field emergence and fruit yield in bottle gourd

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Abstract
In the present investigation an attempt has been made to study the effects of soaking periods and concentrations of priming agents on field emergence and fruit yield of bottle gourd. Disodium hydrogen phosphate (Na_2HPO_4), potassium dihydrogen phosphate (KH_2PO_4), polyethylene glycol-6000 (PEG), gibberellic acid (GA3) were used in two different concentrations with 24 and 48 hours of soaking period and hydro priming was done with 24 and 48 hours of soaking period. One year old seeds were treated with these priming agents. Analysis of variance indicated significant differences among the treatments in respect of field emergence. With the increase in soaking period from 24 hours to 48 hours average field emergence increased by 13.25 %, 7.0 %, 5.75%, 0.25 % and 3.0 % in case of Na_2HPO_4, KH_2PO_4, GA_3, PEG-6000 and hydro priming. Effect of soaking period on speed of germination indicated that 48 hours of soaking period had better effect as compared to 24 hours of soaking period. PEG-6000 at lower concentration (0.5 MPa) with 48 hours of soaking period recorded significantly the highest fruit yield (161.19 q/ha).

Keywords: Bottle gourd, priming agent, soaking period, concentration, field emergence, fruit yield

Introduction
Food security has now become a challenging issue throughout the world. To meet this challenge scientists are focusing their effort in increasing the production of neglected underutilized crops like bottle gourd. Bottle gourd is one of the important vegetable crops of cucurbitaceae family and is considered as poor man's crop. A lot of information is known on the medicinal aspects of bottle gourd (Rahamn, 2003) [7]. In India bottle gourd consumption has gained popularity over last century. One of the important aspects in bottle gourd production that is often overlooked by many resource limited farmers is seed quality. Seed quality is considered as a major tool of a variety development in seed production and breeding process. There is limited and diffused information on seed quality of bottle gourd that determines the performance when the seed is either stored or sown. Seed priming is a technique that is followed to enhance seed quality notably with respect to rate and uniformity of germination (Taylor *et al*, 1998) [10], thereby improving seedling stand and enabling better crop establishment (Job *et al*, 2000) [4]. Various priming techniques have been followed to increase the speed and synchrony of seed germination (Bradford, 1986). Selection of priming technique for a particular crop is very important. In the present investigation an attempt has been made to study the effect of soaking periods and concentrations of different priming agents on field emergence and fruit yield of bottle gourd.

Materials and Methods
Seed Priming
Priming treatment was conducted at the Department of Seed Science and Technology, college of Agriculture, Orissa University of Agriculture and Technology, Bhubaneswar, India. One year old bottle gourd seeds (cv. Rajendra Chamtakar) used for the experiment were collected from All India Coordinated Research Project on Vegetable Crops, Bhubaneswar. Seed moisture content was determined by the high temperature oven method at **103 ^{0}C** for 17 hours (ISTA, 2003, ref: priming in lentil). Seeds were pretreated with thiram fungicide @ 3.0 g/kg to control possible fungal contamination during priming.

The priming agents used for the study were disodium hydrogen phosphate (Na_2HPO_4), potassium dihydrogen phosphate (KH_2PO_4), polyethylene glycol-6000 (PEG), gibberellic acid (GA3) and water. One year old seeds were treated with these priming agents with two different soaking periods (24 hours, 48 hours) and concentrations. The concentrations of disodium hydrogen phosphate and potassium dihydrogen phosphate were 10^{-1} Molar and10^{-3} Molar. Polyethylene glycol-6000 was used at concentrations of 0.5 and 1.5 MPa and GA3 was used at concentrations of 100 and 500 ppm. For hydro priming one year old seeds were immersed in distilled water at ambient temperature for 24 hours and 48 hours. All total there were 19 treatments including dry seeds as control. Details regarding the treatments (T1 to T19) is presented in Table 1. The treated seeds were surface dried and dried back to their original moisture content at room temperature.

Field experiment was conducted in a randomized block design with two replications. One hundred seeds were randomly taken from each treatment. Primed seeds were sown in the main field on 15.01.2015. The plot size was 2 x 4 m^2. Each plot contained four numbers of pits with a spacing of 1 x 2 m. Twenty five seeds were sown in each pit. Observations were recorded on field emergence % for five consecutive days. Observations on seedling root and shoot length were taken by uprooting randomly 10 seedlings at the time of final counting on field emergence % and then these seedlings were kept in hot air oven to take dry weight. Three seedlings were kept per pit. Normal package of practices were followed to raise the crop. Five plants were selected randomly to take observation on vine length (30, 45 and 60 days after sowing), number of leaves per plant (30, 45 and 60 days after sowing), node to 1st male flower, number of female flowers (at 60 DAS), number of fruit/plant, fruit length, fruit girth, average fruit weight (g) and finally fruit yield (kg) was taken on plot basis. Statistical analysis was done following SAS 9.3 version. Significant differences among the treatments were computed by using CD value (critical difference) at 5 % probability level. Correlation coefficient analysis was done to measure the degree of association between field emergence % and yield. Clustering of the treatments was performed using NTSYS PC software (Rohlf, 1993) [8].

Results and Discussion

Analysis of variance indicated significant differences among the treatments in respect of field emergence (Table 1). On the day of initial count (Fig.1) seeds treated with Na_2HPO_4 10^{-3} M for 48 hrs (T11) recorded the highest field emergence (57.0 %). During 2nd day, T12 (KH_2PO_4 10^{-1} M, 48 hrs) recorded the highest emergence (58.0 %) followed by T11 (57.0 %). During 3rd day, T1 recorded the highest emergence (65.0 %) followed by T10 (61.0 %). During 4th day, T1 recorded the highest emergence (65.0 %) followed by T10 (61.0 %). On the day of final count, T1 recorded the highest emergence (72.0 %) followed by T10 (71.0 %). On all these days of observation non primed seeds exhibited the lowest field emergence count.

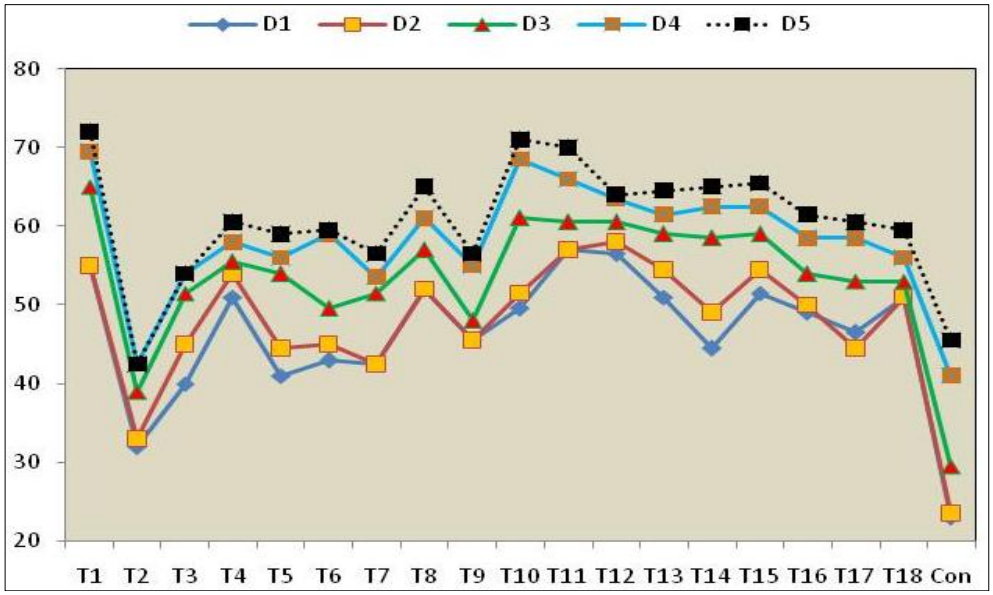

Fig 1: Field emergence count in bottle gourd at five consecutive days

Table 1: Effect of different priming agents on field emergence related traits

	Treatment	Field Emergence %	Root length (cm)	Shoot length (cm)	Seedling length (cm)	SL:RL ratio	Seedling dry wt. (mg/plant)	SV-I	SV-II
	24 hours soaking								
T1	Na_2HPO_4 10^{-1} M	72.00	5.00	14.01	19.01	2.89	0.20	1371.54	14.51
T2	Na_2HPO_4 10^{-3}M	42.50	5.03	14.45	19.47	2.98	0.21	836.77	9.05
T3	KH_2PO_4 10^{-1} M	54.00	5.45	15.25	20.70	2.88	0.25	1113.75	13.59
T4	KH_2PO_4 10^{-3}M	60.50	5.02	13.68	18.70	2.86	0.23	1137.64	14.27
T5	GA_3 100 ppm	59.00	5.89	14.92	20.80	2.59	0.25	1229.81	15.33
T6	GA_3 500 ppm	59.50	4.79	14.87	19.66	3.11	0.21	1169.94	12.31
T7	PEG 6000 (0.5 MPa)	56.50	5.44	14.08	19.52	2.61	0.21	1103.46	11.80
T8	PEG 6000 (1.5 Mpa)	65.00	5.22	14.36	19.58	2.82	0.20	1268.35	12.85
T9	Hydro priming	56.50	5.44	13.31	18.75	2.48	0.16	1066.33	9.24
	48 hours soaking								

T10	Na_2HPO_4 10^{-1} M	71.00	4.65	13.79	18.44	2.99	0.20	1310.02	14.00
T11	Na_2HPO_4 10^{-3} M	70.00	4.94	14.53	19.47	2.98	0.22	1363.38	15.21
T12	KH_2PO_4 10^{-1} M	64.00	5.31	15.18	20.49	2.87	0.24	1311.30	15.42
T13	KH_2PO_4 10^{-3} M	64.50	5.78	15.12	20.89	2.68	0.25	1347.68	16.47
T14	GA_3 100 ppm	65.00	4.81	14.52	19.32	3.03	0.25	1257.51	16.13
T15	GA_3 500 ppm	65.50	5.15	15.46	20.61	3.15	0.24	1348.92	15.60
T16	PEG 6000 0.5 Mpa	61.50	4.62	14.67	19.29	3.22	0.22	1187.12	13.44
T17	PEG 6000 1.5 Mpa	60.50	4.28	14.93	19.21	3.52	0.24	1130.84	14.07
T18	Water soaking 48 hours	59.50	4.53	13.86	18.39	3.06	0.20	1094.33	12.10
Con	Control (Dry seeds)	45.50	5.85	14.08	19.93	2.41	0.19	903.49	8.67
	CD	14.49	NS	NS	NS	NS	0.06	202.14	2.88
	CV	11.40	11.14	6.37	5.49	9.40	12.91	8.11	10.25

Speed of germination of seeds treated with different treatments is depicted in Fig. 2 indicated that seeds treated with Na_2HPO_4 10^{-1} M for 24 hours (T1) had the highest speed of germination (22.54) and the lowest was recorded in control (11.40). Effect of soaking period on speed of germination indicated that 48 hours of soaking period had better effect as compared to 24 hours of soaking period. The mean speed of germination across treatment was 18.44 for 24 hours of soaking period and 20.52 for 48 hours of soaking period (shown by solid straight line in Fig.2).

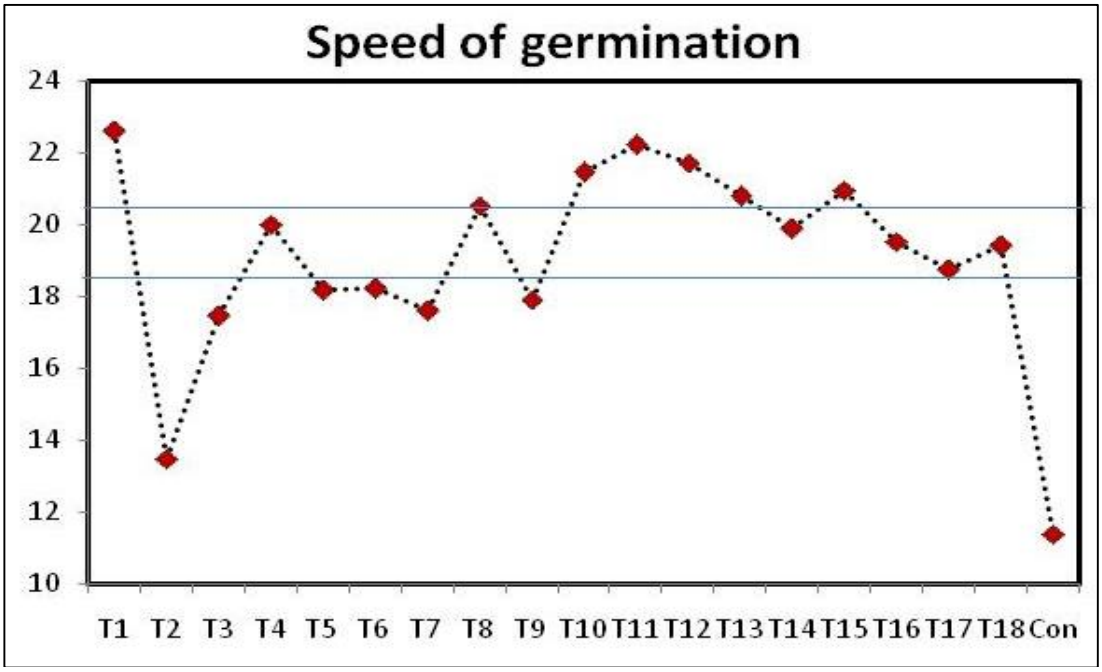

Fig 2: Speed of germination in bottle gourd seeds for different priming treatments

The priming treatments did not show any significant differences in respect of root length, shoot length and shoot to root length ratio (Table 1). There were significant differences among the treatments in respect of seedling dry weight, seedling vigor index I & II. The lowest seedling dry weight was observed in T9 (0.16 mg). SV-I parameter of the treatments at 24 hours of soaking period ranged from 836.77 (T2) to 1371.54 (T1). Whereas it varied from 1094.33 to 1363.68 among the treatment soaked for 48 hours. SV-II values varied from 9.05 (T2) to 16.47 (T13). The mean SV- I value in case of 24 hours was 1144.18 and 48 hours was 1261.23 indicating that 48 hours of soaking had better effect than 24 hours. The mean SV-II value in case of 24 hours of soaking was 12.55 where as the value was higher (14.72) in case of 48 hours of soaking.

Vine length and number of leaves/plant at 30, 45, 60 DAS is presented in Table 2. At 30 DAS the highest vine length was recorded in T5 (48.70 cm) and the lowest in T15 (35.35 cm) i.e. GA3 at higher concentration with 24 hours of soaking period recorded the highest vine length where as GA3 at higher concentration with 48 hours of soaking period produced lowest vine length. This indicated that higher soaking period was not suitable for GA3. At 45 DAS, T6 produced the highest vine length of 179.10 cm follow by T14 (172.20 cm). At 60 DAS, T1 recorded the highest vine length (265.60 cm) followed by T5 (265.50). There were no significant differences among the treatment in respect of number of leaves/plant at 30 DAS and 60 DAS. At 45 DAS, T17 (PEG-6000, 1.5 MPa, 48 hrs.) recorded the highest number of leaves per plant (19.85) followed by T6 (18.75). Many authors reported the positive effect of GA3 on vegetative growth (Yogananda *et al.*, 2004; Kumar *et al.*, 2013; Saeedipour, 2013; Toklu, 2015) [12, 6, 9, 11]. Das *et al.* (2014) reported that treatment of bottlegourd seeds with GA_3 (500 ppm) for 24 hours gave significantly maximum germination, seedling vigour index-I and II and seedling dry weight than dry seeds.

Table 2: Effect of different priming agents on vine length and number of leaves

Treatment		Vine length (cm)			No. of leaves/plant		
		30 DAS	45 DAS	60 DAS	30 DAS	45 DAS	60 DAS
	24 hours soaking						
T1	Na_2HPO_4 10^{-1} M	38.15	134.30	210.10	6.10	13.45	23.80
T2	Na_2HPO_4 10^{-3}M	39.80	129.60	238.00	6.70	14.80	27.10
T3	KH_2PO_4 10^{-1} M	38.10	154.30	208.40	6.00	13.95	22.10
T4	KH_2PO_4 10^{-3}M	38.30	153.00	218.50	6.70	11.20	26.00
T5	GA_3 100 ppm	48.70	130.70	265.50	7.70	17.00	29.40
T6	GA_3 500 ppm	42.45	179.10	256.10	7.30	18.75	29.30
T7	PEG 6000 (0.5 MPa)	42.10	163.80	255.50	6.90	14.65	27.70
T8	PEG 6000 (1.5 Mpa)	39.25	168.60	221.90	6.00	16.90	25.70
T 9	Hydro priming	37.75	145.20	213.40	6.50	16.25	23.90
	48 hours soaking						
T10	Na_2HPO_4 10^{-1} M	40.85	124.60	212.40	6.20	15.15	26.20
T11	Na_2HPO_4 10^{-3} M	39.00	114.40	203.70	6.00	15.95	25.40
T12	KH_2PO_4 10^{-1} M	42.05	143.30	265.60	6.30	16.30	27.90
T13	KH_2PO_4 10^{-3} M	39.15	141.00	253.50	5.90	14.55	32.20
T14	GA_3 100 ppm	39.50	172.20	226.90	6.50	16.90	27.20
T15	GA_3 500 ppm	35.35	149.60	215.40	6.50	15.65	26.90
T16	PEG 6000 0.5 Mpa	35.80	140.70	242.50	6.00	13.85	30.90
T17	PEG 6000 1.5 Mpa	40.55	126.70	228.10	6.70	19.85	29.50
T18	Water soaking 48 hours	37.90	149.00	210.40	6.10	14.90	25.80
T19	Control (Dry seeds)	35.95	152.30	166.00	5.80	17.65	26.20
	CD	6.92	29.46	47.49	NS	2.29	NS
	CV	8.35	9.62	9.95	10.33	6.97	11.79

In case of node to 1st male flower (Table 3) it was observed that in all treatments 1st male flowers initiated on an average on 6th node irrespective of soaking periods and concentrations. In case of T15 1st male flower occurred at 7th node (6.90) where as in case of T10, 1st male flower occurred at 5th node (4.70). T2 recorded the highest number of female flowers (9.0) and T8 & T9 recorded the lowest number of female flowers (2.50). The highest number of female flowers was observed in T2 (9.0/plant). T16 recorded the highest number of fruits/plant (4.30) and T15 produced the lowest number of fruits/plant (1.30) but it had more positive effect on fruit length and girth as compared to others. The highest average fruit weight was observed in T5 (1216.50 g) and highest fruit yield was recorded in T16 (161.50 q/ha). Toklu (2015) [11] observed positive effect of GA3 and PEG-6000 seed priming treatments on yield component and grain yield of lentil.

Table 3: Effect of different priming agents on yield and yield attributing traits

Treatments		Node to 1st male flower	No. of female flowers (60 DAS)	Average no. of fruits/plant	fruit length (cm)	Fruit girth (cm)	Average fruit wt. (g)	Fruit yield/ plot (kg)	Fruit yield (q/ha)
	24 hours soaking								
T1	Na_2HPO_4 10^{-1} M	6.10	6.50	1.72	36.35	22.85	736.50	5.16	64.51
T2	Na_2HPO_4 10^{-3}M	6.10	9.00	2.18	44.15	20.80	730.00	6.54	81.75
T3	KH_2PO_4 10^{-1} M	5.50	4.50	2.54	30.45	21.15	1193.00	7.60	95.01
T4	KH_2PO_4 10^{-3}M	5.60	5.00	2.60	22.80	19.15	1157.00	7.81	97.57
T5	GA_3 100 ppm	5.40	6.50	3.60	37.15	20.90	1216.50	10.80	134.94
T6	GA_3 500 ppm	5.80	5.50	3.29	41.00	22.10	898.00	9.87	123.38
T7	PEG 6000 (0.5 MPa)	6.10	4.00	2.78	33.40	20.40	1124.00	8.34	104.19
T8	PEG 6000 (1.5 Mpa)	5.70	2.50	1.94	29.10	19.40	850.00	5.82	72.75
T9	Hydro priming	5.10	2.50	2.68	37.10	24.80	739.50	8.03	100.32
	48 hours soaking								
T10	Na_2HPO_4 10^{-1} M	4.70	4.50	2.49	29.00	20.10	663.50	7.47	88.38
T11	Na_2HPO_4 10^{-3} M	6.55	3.00	2.83	36.25	21.70	706.50	8.48	106.00
T12	KH_2PO_4 10^{-1} M	5.60	5.00	3.12	33.85	21.20	828.50	9.46	118.26
T13	KH_2PO_4 10^{-3} M	5.50	7.50	3.67	41.50	21.40	1110.00	10.99	137.38
T14	GA_3 100 ppm	5.90	6.00	1.83	36.85	23.75	998.50	5.47	68.38
T15	GA_3 500 ppm	6.90	7.00	1.36	43.50	24.50	677.50	4.09	51.07
T16	PEG 6000 0.5	5.00	8.50	4.30	36.70	21.35	1058.50	12.90	161.19

	Mpa								
T17	PEG 6000 1.5 Mpa	6.60	4.50	3.36	29.25	25.90	886.50	10.08	126.00
T18	Water soaking 48 hours	6.70	3.50	2.69	33.60	25.90	1160.00	8.05	100.63
T19	Control (Dry seeds)	6.30	3.00	1.62	35.50	24.25	783.00	4.84	60.50
	CD	1.13	1.52	0.93	10.74	5.07	355.47	1.70	20.96
	CV	9.21	13.99	16.70	14.22	10.65	18.36	10.14	10.02

Comparing effect of soaking periods and concentrations of priming agents on field emergence and yield

Comparing the effect of soaking periods of priming agents on field emergence it was observed that 48 hours of soaking period had increased effect on the field emergence as compared to 24 hours (Table 4).With the increase in soaking period from 24 hours to 48 hours average field emergence increased by 13.25 %, 7.0 %, 5.75%, 0.25 % and 3.0 % in case of Na_2HPO_4, KH_2PO_4, GA_3, PEG-6000 and hydro priming. Na_2HPO_4 at high concentration (10^{-1}M) showed positive effect on field emergence whereas KH_2PO_4 at high concentration (10^{-1}M) had negative effect on field emergence as compared to low concentration (10^{-3}M). GA_3 did not produce any differential effect on field emergence at both high (500 ppm) and low (100 ppm) concentrations. PEG-6000 at high concentration (1.5MPa) showed positive effect on field emergence (Table 5). At higher concentration field emergence increased by 27.10 %, -5.60 %, 0.40 % and 6.36 % in case of Na_2HPO_4, KH_2PO_4, GA_3 and PEG-6000 as compared to lower concentration.

Table 4: Effect of soaking period on field emergence

Priming agent	Field emergence % (average of low and high concentrations)		% increase
	24 hours	48 hours	
Na_2HPO_4	57.25	70.50	13.25
KH_2PO_4	57.25	64.25	7.0
GA_3	59.50	65.25	5.75
PEG 6000	60.75	61.0	0.25
Hydro priming	56.50	59.50	3.0

Table 5: Effect of concentrations on field emergence

Priming agent	Field emergence % (average of 24 and 48 hrs of soaking period)		% increase
	low	high	
Na_2HPO_4	56.25	71.50	27.10
KH_2PO_4	62.50	59.00	-5.60
GA_3	62.25	62.50	0.40
PEG 6000	59.0	62.75	6.36
Hydro priming	-	-	-

Effect of priming treatment on fruit yield revealed that PEG-6000 at lower concentration with 48 hours of soaking period recorded significantly the highest fruit yield (161.19 q/ha). T13 (KH_2PO_4, 10^{-3}M, 48 hours) that ranked 2nd position in fruit yield (137.9/ha) was found to be at par with T4 (GA_3, 100 ppm, 134.09/ha), T5 (GA_3, 500 ppm, 123.0 q/ha), T12 (118.26 q/ha) and T17 (126.09 q/ha). Hydro priming at different soaking period did not exhibited any significant difference in yield but PEG, KH_2PO_4, Na_2HPO_4 resulted in significantly higher yield at 48 hours of soaking period as compared to 24 hours of soaking period irrespective of concentrations. GA3 both at lower & higher concentration with 24 hours of soaking period resulted in significantly higher yield than 48 hours of soaking period. These results indicated that 48 hours of soaking period has immense effect in increasing yield in case of PEG, KH_2PO_4 and Na_2HPO_4. Comparing the effect of lower & higher concentrations of priming treatments it is realized that all priming agents (except water) produced higher yield at lower concentration than higher concentration irrespective of their soaking period. From this study it may be concluded that lower concentration with higher soaking period has pronounced effect in increasing yield. Kaur *et al.* (2015) [5] reported that soaking of okra seeds for 24 hours with 5 % PEG gave higher yield as compared to hydro priming and non primed seeds. Ghassemi *et al.* (2010) [3] studied the effect of hydro priming duration on grain yield of pinto bean and suggested that hydro priming duration of 7 hours and 14 hours had improved grain yield.

Table 6: Effect of soaking periods and concentrations on fruit yield

	Yield (q/ha)			
	24 hours		48 hours	
	Low concentration	High Concentration	Low concentration	High Concentration
Na_2HPO_4	81.75	64.51	106.0	83.38
KH_2PO_4	97.57	95.01	137.38	118.26
GA_3	134.94	123.38	68.38	51.07
PEG 6000	104.19	72.75	161.19	126.0
Water soaking	100.32	100.32	100.63	100.63
Mean	91.15		106.37	

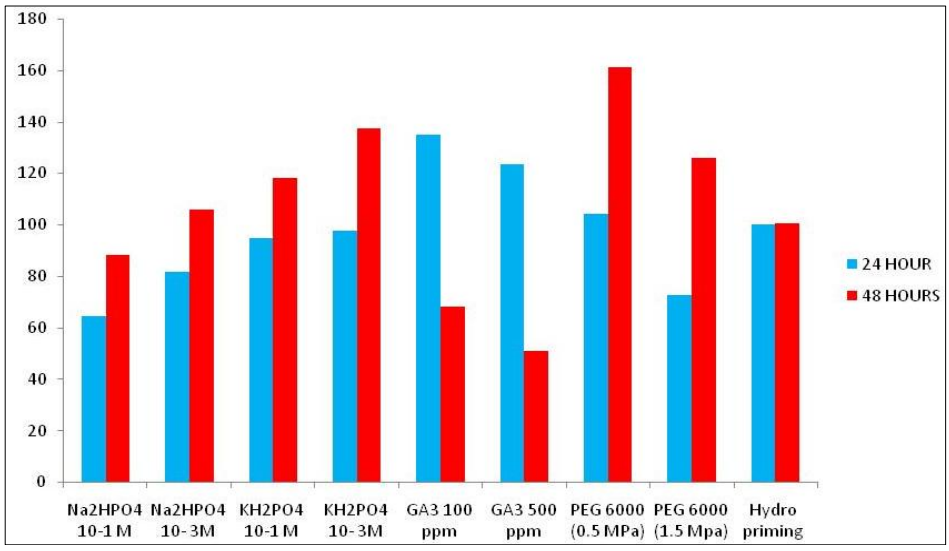

Fig 3: Comparing the effect of soaking period of priming treatments on fruit yield (q/ha)

Classification of the treatments

Classification of the treatments was done to study whether the 19 treatments were similar in their effect or not as four priming agents along with water was taken for the study with two different concentrations and soaking periods. Fig.4 shows classification of the treatments based on germination as well as growth and yield related traits. The number of clusters at 0 % distance coefficient was 19; at 12.5 % distance coefficient the total number of clusters were 15; at 25 % distance coefficient the cluster number was reduced to 9; at 50 % distance coefficient the 19 treatments were grouped into 3clusters; at 85 % distance coefficient all the treatments were appeared as a single unit. No treatments were found to be 95 % similar with each other in their effect. But T1(Na_2HPO_4, 10^{-1} M, 24 hrs) was 90 % similar to T11(Na_2HPO_4, 10^{-3} M, 48 hrs) and formed one cluster; T3(KH_2PO_4, 10^{-1} M, 24 hrs), T4 (KH_2PO_4, 10^{-3}M) & T18 (Hydro priming, 48 hrs) were 90 % similar to each other and formed another cluster. Here the farmers' community may prefer to go for hydro priming instead of KH_2PO_4 as seed soaking with water for 48 hours is cheaper and easier to follow.

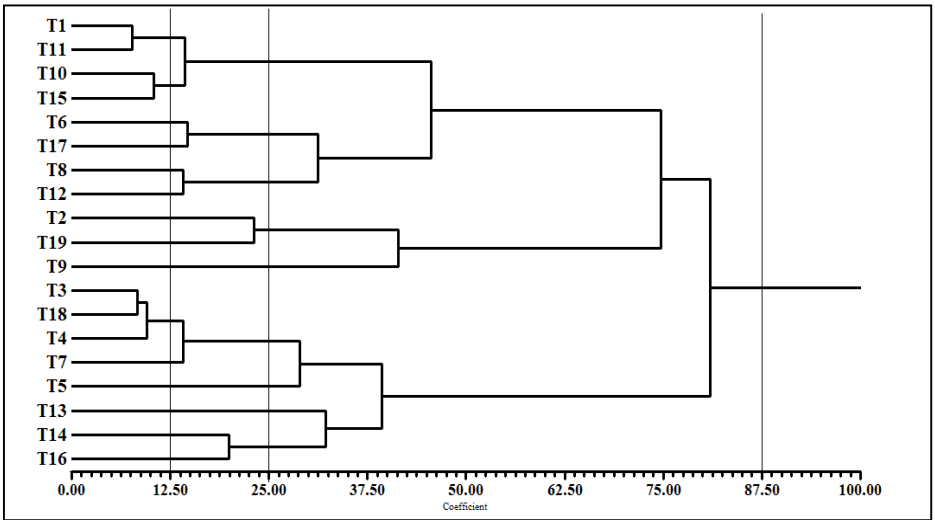

Fig 4: Classification of treatments based on numerical taxonomic approach

Association of fruit yield with FE %, SV-I and SV-II

Field emergence %, SV-I and SV-II are three important seed quality parameters. Therefore correlation coefficient study was made to envisage that which quality parameter could be used as an indicator for predicting yield. Field emergence % and SV-I showed very weak positive correlations (0.025 and 0.079) with yield where as SV-II showed high but non significant positive correlation (0.204). It was observed that there was no linear relationship between FE % and yield (Fig. 5); SV-I and yield (Fig. 6), SV-II and yield (Fig. 7) as the coefficient of determination (R^2 value) was very low. Therefore quadratic equation was considered which still did not show high R^2 value. In the present study efficacy of different priming treatments was judged on the basis of yield *per se* directly.

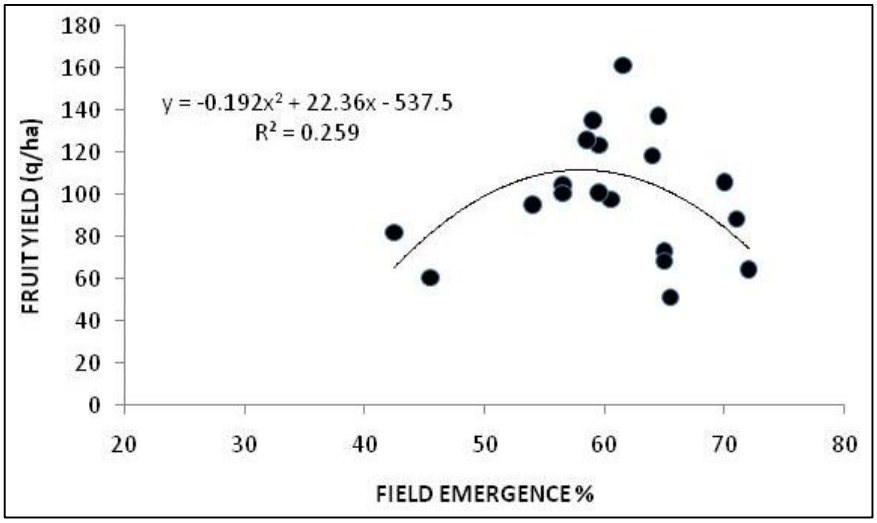

Fig 5: Relationship between fruit yield and field emergence

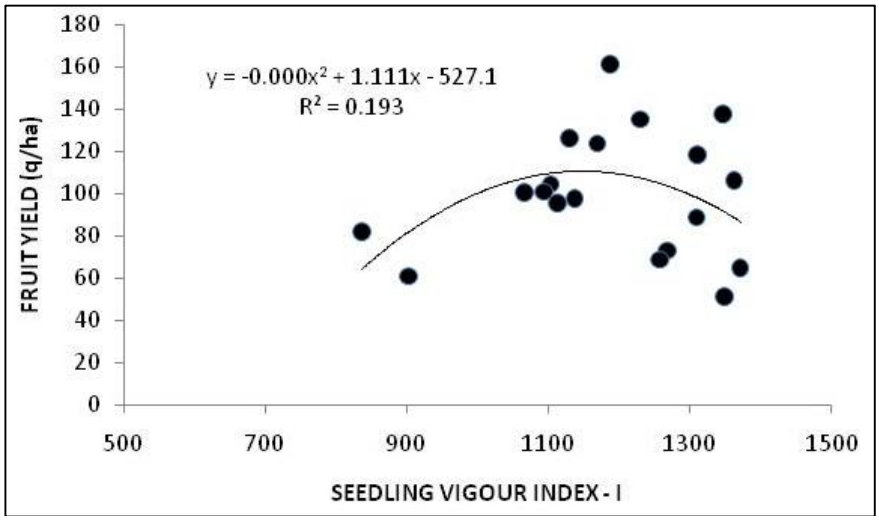

Fig 6: Relationship between fruit yield and SV-I

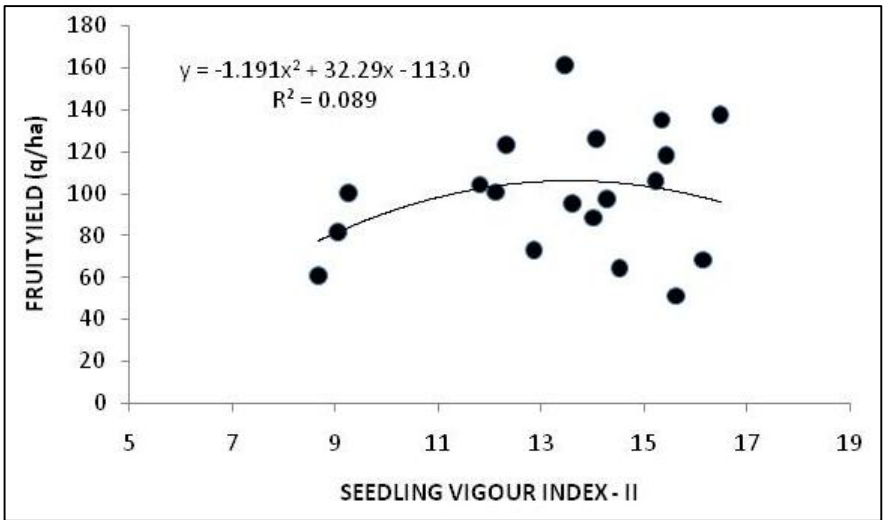

Fig 7: Relationship between fruit yield and SV-II

Conclusion

The priming agents with lower concentration recorded higher yield than higher concentration irrespective of soaking period. Comparison of soaking period revealed that 48 hours of soaking period gave better yield in all the priming agents except GA3 irrespective of the concentrations.

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