

## INFLUENCE OF TEMPERATURE AND OSMOTIC STRESS ON GERMINATION INDUCTION OF DIFFERENT CASTOR BEAN CULTIVARS

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### Abstract

Among the different factors temperature and moisture are considered very important for germination, crop maturity and productivity. Experiments were conducted in the laboratory to determine the effect of temperature and moisture stress on the speed and total germination of four Castor bean cultivars (DS-30, PR-7/1, PR-101 and Local) obtained from two locations viz., Attock and Chakwal. The seeds were conditioned to different moisture content ranging from 0 to -14 bars and temperature ranging from 0 to 35°C in hot air chamber/oven. The highest value indicated the fastest germination that was obtained from Attock location (3.992), in comparison with value (3.933) obtained from Chakwal when the Castor bean cultivars were incubated at 25°C. The maximum GRI was recorded at the osmotic pressure of 0 bar (control), whose value in case of Attock seed location was 3.857 and Chakwal seed location (4.788). The interaction effects of moisture, temperature and cultivars were found significant for GRI. The cultivars performed differently under different temperature and moisture conditions.

### Introduction

Castor bean plant is grown as an annual herb in temperate countries but grows to tree-like proportions on fertile ground in tropical climates. Castor bean is herbaceous when young but becomes woody with age. Castor is an important industrial oil crop of the world. Internationally, castor oil is used in more than 300 derivatives for use in soap, cosmetics, pharmaceutical, paints and varnishes/lubricant in high-speed jet engines. Pakistan also spends millions of rupees on the import of castor oil annually for use in automobile industry. It is being cultivated in about 43 countries of the world, whereas in Asia, it is being cultivated in 12 countries including Pakistan. However, the top producers are Brazil, India, China, Russia and Thailand (Kakakhel, 2008).

Different factors affect the seed germination of the crops. Crop productivity can be increased by increasing the germination rate which is possible by optimizing important parameters which are crucial for germination. The proportion of seeds of *Pisum sativum* germinated normally in standard germination tests declined progressively from 94 to 50% when the seeds were dried from 14.8 to 3.7% moisture content at ambient temperature (Ellis *et al.*, 1990; Ellis & Roberts, 1982). Imbibitional injury of seeds depends on several factors such as seed maturation, age, moisture content and temperature (Tully *et al.*, 1981; Powell & Matthews, 1979; Taylor & Prusinski, 1990).

The major aspects of seed germination include moisture content and viability (Ellis *et al.*, 1990), leachate conductivity (Powell & Matthews, 1981), soaking injury (Rowland & Gusta, 1977) and effect of temperature (Munro *et al.*, 2004). The germination of *Lotus*

*corniculatus* small seeds was reduced more than that of large seeds by temperatures of 25°C or higher. There were no significant differences between varieties or seed sizes in their over-all responses to moisture stress, but the times of some of the responses differed according to the variety or seed size (Woods & Macdonald, 1993). The present study was contemplated with the objective to optimize temperature and moisture conditions for high germination rate of *Ricinus communis*, so that it can be used in field to obtain maximum crop productivity.

## Materials and Methods

The seeds of Castor bean were collected from two locations; Attock and Chakwal. The reason for collecting seed from two locations is that seed produced in different environments behave differently in germination. The environmental conditions of both sites/locations regarding rainfall and freeze nights during the growth period of crop are given in Table 5. The collected seeds were conditioned to different moisture content ranging from 0 to 14 bars by treating with the temperature ranging from 0 to 35°C in hot air chamber/oven. Polyethylene glycol 20000 (PEG-20000), obtained from Merck-Schuchardt, Germany was used to create osmotic stress and chamber compartments for different temperature adjustments. To develop the moisture stress levels in the laboratory 16, 24, 36 and 48 grams of PEG-20000 were mixed in 100 ml of distilled water. This solution was stored in laboratory at room temperature for 12 hours. Different osmotic stress levels (0, -7 and -14 bars) were determined by using the Vapour Pressure Osmometer Model Vapro 5520. PEG-20000 with 24g and 36g per 100 ml of distilled water created osmotic potential of -7 and -14 bars respectively. Before sowing, the seeds were treated with NaOH and Captan 5% to disinfect any fungal contamination. Data used in analysis was the average of three replicates.

Seeds of four Castor bean cultivars (DS-30, PR-7/1, PR-101 and Local) were sown in growth chamber on Whatman no. 2 filter paper in plastic Petri dishes with covers. Twenty seeds per Petri dish were sown and there were three dishes per replicate. A layer of filter papers in each Petri plate was moistened with the distilled water. Cracked, swollen seeds and abnormal germinants were excluded from the counts used in the analyses. During germination, distilled water was added as needed to maintain the filter papers at a visually constant moisture content (papers wet, but without surface water on the seeds or a water meniscus between paper and seed). GRI was calculated using the formula given below:

$$\text{GRI} = \frac{X_i}{Y_i} + \frac{X_{ii}}{Y_{ii}} + \frac{X_{iii}}{Y_{iii}} \text{ ----- } \frac{X_n}{Y_n}$$

GRI = Germination rate index

X = Seedlings germinated per day

Y = Days during which germination occurred

Every experiment was replicated three times at every combination of temperature and osmotic stress level. A computer based program MSTAT-C (Russell, 1986) was used for analysis using 5% significance level.

## Results and Discussion

Among the different factors temperature and moisture are considered very important for germination and later on for crop productivity. The cultivars were evaluated on the basis of germination rate index (GRI) under different temperature conditions in the

growth chamber. The highest value indicated the fastest germination that was obtained from seeds collected from Attock location (3.992), in comparison with value (3.933) obtained from Chakwal location when the Castor bean cultivars were incubated at 25°C. The lowest value indicated the slowest speed of germination that was observed when the Castor bean cultivars were incubated at 15°C (0.631) and 10°C (1.328) from Attock and Chakwal locations, respectively (Tables 1&2). The results were in agreement with the findings of Mwale (1994) who found significant variations among the cultivars in the way they responded to temperature.

Among the cultivars obtained from Attock location, DS-30 (2.831) was at the top for GRI that was followed by PR-101 (2.591). The minimum value was exhibited by the Local cultivar (Table 1) whereas, from Chakwal location, DS-30 (3.257) was at the top for GRI followed by Local (2.533). The minimum value was exhibited by the PR-101 (Table 2).

The interaction between temperature and cultivars was also significant. PR-7/1 cultivar gave the highest GRI (5.444) when incubated at 25°C, which differed significantly from the other cultivars (Table 1) and DS-30 cultivar gave the highest GRI (5.289) when incubated at same temperature (Table 2). The minimum GRI value was recorded in case of Local cultivar and PR-101 cultivar collected from Attock and Chakwal locations, respectively, that was statistically at par with rest of the cultivars at 10°C.

Moisture often limits germination and emergence in semi-arid regions. There was decrease in rate of germination due to low availability of moisture. The effect of different levels of PEG induced osmotic pressure (0, -7, -14 bars) was statistically significant for germination rate index (Table 3&4). The maximum GRI was recorded at the osmotic pressure of 0 bar (control), whose value in case of Attock location was 3.857 and Chakwal location (4.788). The GRI decreased when the PEG concentration was increased. At -7 bars, the GRI was 2.399 and 2.539 and the GRI was lowest i.e. 0.336 and 0.501 when osmotic pressure was -14 bars in both Attock and Chakwal locations, respectively. It was observed from the study that GRI value decrease when the osmotic stress increased (Table 3&4). Hegarty & Ross (1978) reported that difference in sensitivity to water stress between seed germination and growth of seedlings of several species is available. Atia *et al.*, (2006) and Khajeh-Hosseini *et al.*, (2003) also reported that seed germination in *Crithmum maritimum* (a perennial local oilseed halophyte) and *Glycine max* was more reduced in PEG-induced osmotic stress. The results are similar to findings of Smith *et al.*, (1989) and Livingston *et al.*, (1990) who reported that under osmotic stress, the germination was significantly reduced.

In this study, the interaction between the cultivars and the moisture levels was significant. The maximum value of GRI (4.511) was exhibited by the PR-7/1 cultivar at 0 bar but that was followed by PR-101 cultivar at the same moisture level from seeds of Attock location. Whereas, from seeds of Chakwal location, maximum value of GRI (5.633) was exhibited by the DS-30 cultivar at 0 bar followed by Local cultivar (5.239) at the same moisture level. At -7 bars osmotic stress, the highest value (3.572) was recorded for DS-30 cultivar and minimum (0.411) was recorded in case of Local cultivar from seeds of both sites. At -14 bars osmotic pressure, the maximum GRI values i.e. 0.556 (Attock location) and 1.00 (Chakwal location) were shown by DS-30 cultivar that was statistically at par with rest of the cultivars at this moisture level. The results presented in Table 3 and Table 4 showed that the GRI decreased when the moisture stress increased. Similar findings were noted by Ahmad *et al.*, (2009) in case of sunflower (*Helianthus annuus*) who reported that there was decrease in germination due to the increase in PEG-6000 concentration. Reports regarding germination reduction under osmotic stress were also articulated by Smith *et al.*, (1989); Livingston *et al.*, (1990).

**Table 1. GRI of Attock seed source of four castor bean cultivars as influenced by different temperatures.**

Cultivars Temperature	Cultivars				
	DS-30	PR-7/1	PR-101	Local	Means
10 °C	1.178 fghi	0.800 hij	0.889 ghij	0.000 j	0.7170 C
15 °C	0.000 j	1.011 ghi	1.089 fghi	0.4220 ij	0.6310 C
20 °C	2.811 de	1.000 ghi	1.767 fg	0.0000 j	1.394 C
25 °C	4.544 ab	5.444 a	3.967 b	2.011 ef	3.992 A
30 °C	3.933 bc	3.789 bc	4.122 b	1.711 fgh	3.389 AB
35 °C	4.522 ab	2.000 ef	3.000 cd	1.200 fghi	3.061 B
Means	2.831 A	2.476 A	2.591 A	0.8910 B	---

Any two means not sharing a letter common in a row or column differ significantly at 5% probability level i.e.,  $\alpha = 0.05$

**Table 2. GRI of Chakwal seed source of four castor bean cultivars as influenced by different temperatures.**

Cultivars Temperature	Cultivars				
	DS-30	PR-7/1	PR-101	Local	Means
10 °C	1.611 jkl	1.467 klm	0.8110 n	1.422 klmn	1.328 B
15 °C	2.700 fgh	1.122 lmn	1.378 klmn	0.9560 mn	1.539 B
20 °C	2.456 ghi	1.844 ijk	1.578 jklm	2.156 hij	2.008 B
25 °C	5.289 a	3.400 de	2.644 fgh	4.400 b	3.933 A
30 °C	3.433 cde	3.500 cde	3.789 bcd	3.256 def	3.494 A
35 °C	4.056 bc	3.233 def	3.111 ef	3.011 efg	3.353 A
Means	3.257 A	2.428 B	2.219 B	2.533 B	---

Any two means not sharing a letter common in a row or column differ significantly at 5% probability level i.e.,  $\alpha = 0.05$

**Table 3. GRI of Attock seed source of four castor bean cultivars as influenced by different moisture levels.**

Cultivars Moisture level	Cultivars				
	DS-30	PR-7/1	PR-101	Local	Means
0 bar	4.367 a	4.511 a	4.422 a	2.128 d	3.857 A
-7 bar	3.572 ab	2.500 cd	3.111 bc	0.4110 e	2.399 B
-14 bar	0.5560 e	0.4170 e	0.2390 e	0.1330 e	0.336 C
Means	2.831 A	2.476 A	2.591 A	0.8910 B	---

Any two means not sharing a letter common in a row or column differ significantly at 5% probability level i.e.,  $\alpha = 0.05$

**Table 4. GRI of Chakwal seed source of four Castor bean cultivars as influenced by different moisture levels.**

Cultivars Moisture level	Cultivars				
	DS-30	PR-7/1	PR-101	Local	Means
0 bar	5.633 A	4.144 B	4.133 B	5.239 A	4.788 A
-7 bar	3.139 C	2.528 CD	2.522 CD	1.967 D	2.539 B
-14 bar	1.000 E	0.6110 EF	0.000 F	0.394 EF	0.501 C
Means	3.257 A	2.428 B	2.219 B	2.533 B	---

Any two means not sharing a letter common in a row or column differ significantly at 5% probability level i.e.,  $\alpha = 0.05$

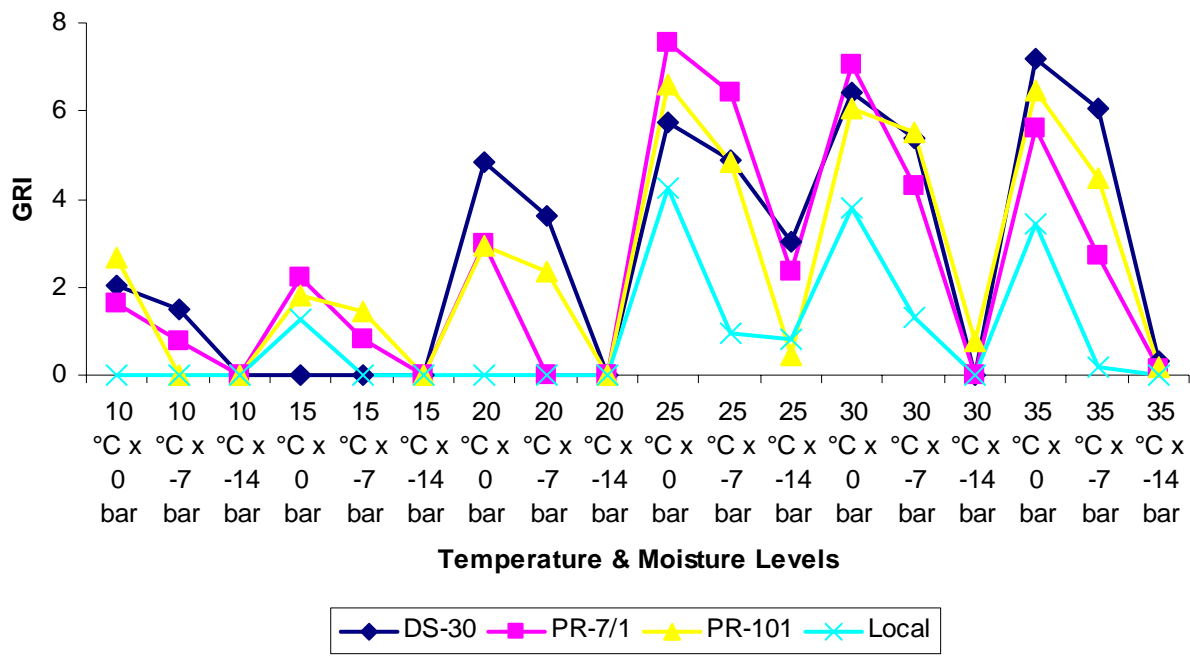


Fig. 1. Interaction between temperature, moisture and seed source (Attock) of four Castor bean cultivars.

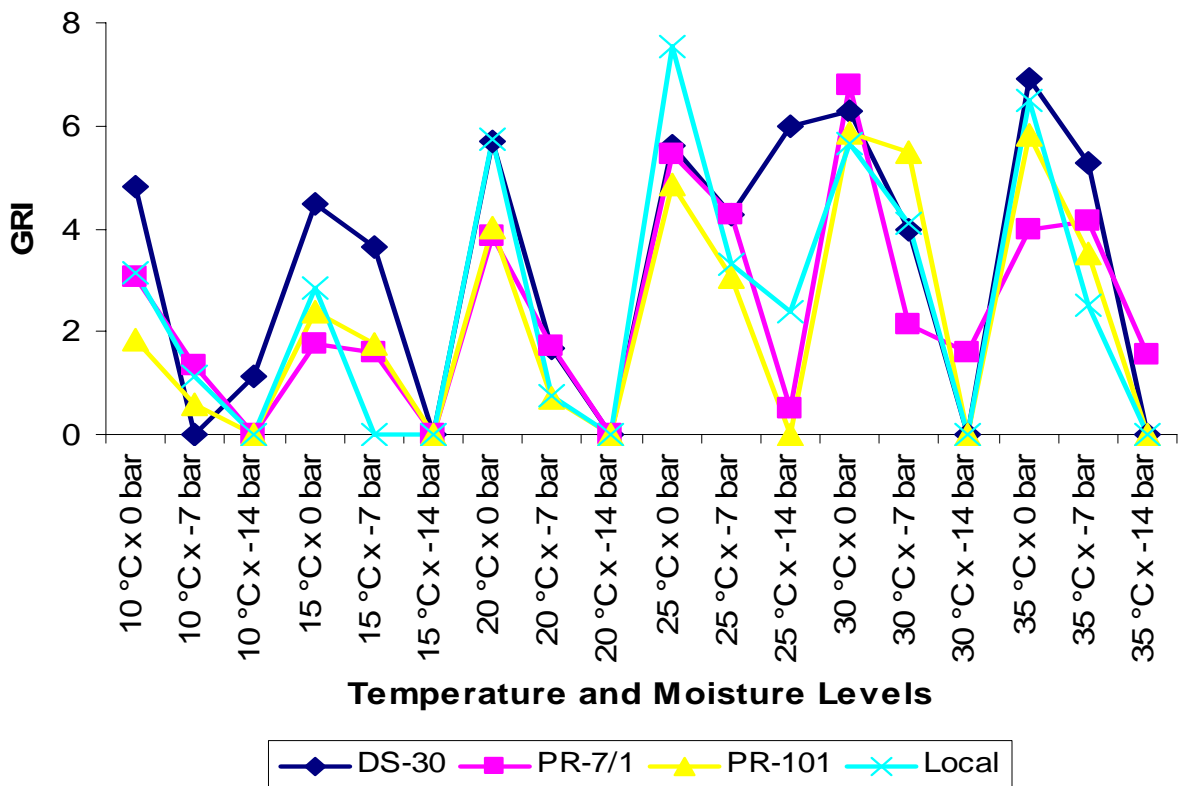


Fig. 2. Interaction between temperature, moisture and seed source (Chakwal) of four Castor cultivars.

The interaction affects of moisture, temperature and cultivars were found significant for GRI. The cultivars performed differently under different temperature and moisture conditions. There was very low or no germination at 10°C and 15°C at -14 bars osmotic pressure and all the cultivars were statistically similar (Figs. 1&2).

**Table 5. Summary of average rainfall and freeze nights at various locations during the growth period of crop in 2004-05 and 2005-06.**

Location	Rainfall				Freeze nights	Yield kg/ha
	Jul.	Aug.	Sep-Dec.	Total		
GRS Attock 2005-06	226	148	57	431	0	533
BARI, Chakwal 2005-06	167	93	102	362	29	288

In case of Attock location, maximum GRI value (7.533) was recorded for the PR-7/1 cultivar at 0 bars at 25°C. At-7 bars, the maximum GRI (6.433) was recorded in case of same cultivar at 25°C followed by the DS-30 cultivar (6.067) at 35°C. The minimum value was obtained at 10°C for -14 bars (Fig. 1). But for the Chakwal seed locations, the maximum GRI (6.9) was recorded for the DS-30 cultivar at 0 bars and 35°C. At-7 bars, the maximum GRI (5.5) was recorded in case of PR-101 cultivar at 30°C followed by the DS-30 cultivar (5.267) at 35°C. The minimum value obtained at 10°C for -14 bars (Fig. 2). These results suggested that when different cultivars were subjected to different levels of water stress and temperatures they performed differently. It has been concluded that variable temperature and moisture often are limiting factors during germination (Ashraf & Abu-shkara, 1978). Somers *et al.*, (1983) used a germination test on sunflower genotypes submitted to PEG water stress as a screening technique and concluded that it could be used as screening test. These variations in GRI found in different Castor bean cultivars could be attributed to genetic potential of cultivars to withstand differently at various moisture and temperature regime. These treatments may be used to predict tolerance or susceptibility of important genotype subjected to moderate water and temperature stress.

The reason for collecting seed from two locations was to see the effect of different environments on seed germination as the seed of various species vary in their degree of germination ability with respect to maternal environment. This variation is due to genetic make up as well as phenotypic environment. Phenotypic variation in seed due to a-biotic factors is caused by local conditions in which seed is matured. These conditions consist of micro-environments experienced during seed development as well as temperature, moisture and sun light (Gutterman, 1992).

The germination results elaborated in Table 1&2 shows that seed produced at Attock has more germination as compared to Chakwal. Maximum germination (5.444) was noted in PR-7/1 at 25°C whereas maximum GRI (5.289) at Chakwal was observed in DS-30 at the same temperature. The seed of cultivars PR-7/1 and PR-101 also showed more GRI values obtained at Attock than Chakwal. This is due to the reason that the conditions of seed produced at Attock were more conducive for seed production as compared to Chakwal. There was appropriate temperature and more moisture was available at Attock whereas the environment of Chakwal was harder (Table 5) and less moisture was available. So seed of Attock was healthier and vigorous. It was further noted that germination at 30°C is more in both cases as 27°C to 30°C is appropriate temperature for all seed germinations.

## Conclusion

Seed germination level is the indicator for further crop maturity, leading to high value production. It is concluded from the experimental study that optimum temperature and moisture availability are the important components which affect seed germination. So

selection of suitable time when proper temperature and moisture level is available may be considered on priority basis before cultivation of *Ricinus communis* L. to get better GRI and ultimately production. It is further concluded that seed should be produced in proper conditions as seed from harder environment is not good for further crop production.

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