

SEED GERMINATION ECOLOGY OF *Lathyrus aphaca* L. AND *Vicia sativa* L. IN COMPARISON WITH *Triticum aestivum* L.

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ABSTRACT

*This study investigated the influence of various environmental factors and burial depths on seed germination and seedling growth of *Lathyrus aphaca* L. and *Vicia sativa* L. in comparison with wheat in order to suggest field modifications that would favor winter wheat (*Triticum aestivum* L.) germination and suppress weed germination. The highest germination percentage was recorded at 15°C for wheat and *L. aphaca* and at 20°C for *V. sativa*. Germination under light and dark conditions did not differ in *L. aphaca* and *V. sativa*, and were equivalent to wheat. Increase in drought stress level decreased germination and seedling growth and increased the days to 50% germination and mean germination time in all the three species. All salinity levels except 50% NaCl in wheat caused 100% germination inhibition. The seedling emergence decreased significantly ($P \leq 0.05$,) when seeding depth was increased beyond 3 cm in wheat and *L. aphaca*. Both the weed species showed greater emergence (100%) than wheat at 1, 2 and 3 cm depth. Emergence was slowest at 10 cm. These observations explore some of the factors contributing to success of these weed species in varied environments and provide useful information that could contribute to their control.*

Key words: Ecology, germination, *Lathyrus aphaca*, *Triticum aestivum*, *Vicia sativa*.

INTRODUCTION

Lathyrus aphaca L. and *Vicia sativa* L. are among the most important weed species affecting winter wheat production in rain-fed and irrigated conditions. Both are winter season annual broad leaved weeds growing from October-November to April. *Lathyrus aphaca* is the most problematic weed of wheat in rice-wheat cropping system in irrigated areas of Pakistan (Haleemi *et al.*, 1995); whereas, *V. sativa* is the most serious weed in rain-fed wheat (Dangwal *et al.*, 2010). Both

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weeds mature in early April before crop maturity and shed their seeds before harvest, thus adding to the soil weed seed bank and aggravating the problem in coming crop of winter season. The threshold density of *L. aphaca* in irrigated wheat has been reported 4.41-5.31 plants m⁻² whereas it is 2-3 plants m⁻² for *V. sativa* (Haleemi et al., 1995). Threshold density of *L. aphaca* and *V. sativa* in rain-fed wheat is 5.47 and 10.04 plants m⁻² (Naeem et al., 1995), respectively.

Germination is one of the most critical processes in determining the success of a particular weed under field conditions. Several environmental factors affect weed seed germination. The optimum temperature, light, pH, soil moisture and salinity conditions vary considerably, depending on species (Taylorson, 1987; Chachalis and Reddy, 2000; Khan et al., 2006; Khurshid et al., 2012). The burial depth of seed also affects seedling emergence (Koger et al., 2004). An understanding of germination and emergence behavior of *L. aphaca* and *V. sativa* in comparison with wheat would help to develop strategies that will suppress germination of these weeds. The objectives of this study were (i) to determine the effect of osmotic stress, salt stress, light and temperature on seed germination/emergence and seedling growth of wheat, *L. aphaca* and *V. sativa*. (ii) To find the optimum seeding depth that will give competitive advantage to wheat over weed species. (iii) To find the limiting depth to which the weeds cannot emerge.

MATERIALS AND METHODS

Drought stress

The experiment was conducted under normal (control) and stress conditions created with the use of polyethylene glycol 6000 (PEG-6000). Treatments were 0, 135, 180 and 225 g L⁻¹ PEG corresponding to 0, -0.4, -0.5 and -0.8 MPa. Solutions were prepared by dissolving the required amount of PEG in distilled water as described by Michel and Kaufman (1973). Ten wheat seeds were placed on two sheets of filter paper in 9 cm Petri plates and incubated at 25±3°C. The Petri dishes were covered to prevent the loss of moisture by evaporation. The 2 x 4 factorial experiment was arranged as a completely randomized design with ten replications. At the time of sowing, 4 mL of solution were added to each Petri dish. Moisture levels were assessed daily and additional solution was applied as necessary to compensate the losses due to evaporation. Germination percentage was recorded daily every 24 hours for 12 days. Seeds were considered germinated when the emerged radical reached 2 mm length. Time

taken to 50% germination of seedlings (T50) was calculated according to the following formulae of Coolbear *et al.* (1984) modified by Farooq *et al.* (2005). Fresh weight, root and shoot length of wheat seedlings were recorded at day 12. Plant dry weights were recorded after oven drying at 68°C for 24 hours to a constant weight. The same procedure was followed for *L. aphaca* and *V. sativa*.

Salt stress

Sodium chloride solutions of 50, 100 and 150% were prepared. Distilled water was used as a control. Petri dishes each having ten *T. aestivum* seeds were incubated at 20°C in germinator for a period of 12 days. In the stress and normal treatments, 4 mL of salt solution and distilled water were added to each Petri dish, respectively at the time of sowing. The level of water and salt solution was assessed daily and applied time to time as per requirement to compensate the losses due to evaporation. Germination percentage, root/shoot length and their dry weights were recorded at the completion of experiment (after 12 days). Same procedure was followed for *L. aphaca* and *V. sativa*. Ranges of NaCl and osmotic potential were selected keeping in view the level of salinity and water stresses occurring in Pakistan's soils.

Temperature

Ten seeds of *T. aestivum* were sown on wet filter paper in Petri dishes. Germination was determined in germinators under constant temperatures of 15°C and 20°C. Seed germination in each Petri dish was recorded everyday for 10 days after seeding.

Light

To study seed germination in the dark, a set of four Petri dishes each having ten seeds of *T. aestivum* on a wet filter paper was wrapped in a layer of aluminum foil to ensure no light penetration. Another set of Petri dishes was left uncovered to ensure continuous light exposure. Petri dishes were placed in an incubator at 20°C. Seed germination in each Petri dish was recorded for 10 days after seeding.

Sowing depth

Ten seeds of *T. aestivum* were planted in sand in 25 cm-diameter plastic pots at depths of 1, 2, 3, 5 and 10cm. Each treatment was replicated four times following completely randomized design in factorial arrangement. Pots were watered as needed to maintain adequate moisture throughout the study. Seedling emergence was recorded daily and removed for a period of 30 days. Emergence was defined as the appearance of the shoot in wheat and two cotyledons in *L. aphaca* and *V. sativa*. Data on number of seedlings emerged, days to seedling initiation and total number of days taken to seedling

emergence was recorded. Same procedure was followed for *L. aphaca* and *V. sativa*.

Statistical analysis

The data collected was analyzed statistically using Fisher's analysis of variances technique and treatment means showing F-values significant were separated using least significant difference (LSD) at 0.05 probability level (Steel et al., 1997).

RESULTS AND DISCUSSION

Drought stress

The comparison of treatment means given in Table-1 shows that statistically maximum germination (93%) and CUG (1.19) was attained in *T. aestivum*, which were equivalent to *L. aphaca* and *V. sativa*, respectively under control treatments. Germination was decreased from 97 to 57%, 91 to 8% and 89 to 17% in wheat, *L. aphaca* and *V. sativa*, respectively as water stress increased from 0 to -0.8 MPa. A very similar reduction in the germination of *Mimosa invisa* was recorded by Chauhan and Johnson (2008) with identical water stress treatment. Rao et al. (2008) also reported a sharp decrease in germination of American slough grass (*Beckmannia syzigachne*) as osmotic potential reduced from 0 to -0.7 MPa. However, results also demonstrated that in any test specie germination was not inhibited completely up to osmotic potential of -0.8 MPa. In contrast to these species, germination in small flowered mallow was completely inhibited at an osmotic potential of -0.6 MPa (Chauhan et al., 2006b). Maximum mean germination time (5.07 days) and days to 50% germination (3.54 days) was recorded in *L. aphaca* at -0.5 MPa drought stress level. Minimum mean germination time (1.96 days) and days to 50% germination (1.47 days) was recorded in -0.8 MPa treatment for *V. sativa*. Comparison of test species showed that *L. aphaca* is most sensitive to drought stress followed by wheat as it was exhibited by the delay in germination.

Plants species, drought stress and their interaction have significant effect on root length. The comparison of treatment means (Table-2) shows that under controlled conditions maximum root length (3.43, 2.42 and 2.19 cm) and shoot length (4.77, 11.26 and 10.85 cm) was recorded in *T. aestivum*, *L. aphaca* and *V. sativa* with statistically highest value in wheat for root length and for shoot length in *L. aphaca*. Minimum root length (0.87, 0.05 and 0.05 cm) and shoot length (0.04, 0.0 and 0.04 cm) was recorded in *T. aestivum*, *L. aphaca* and *V. sativa*, respectively at -0.8 MPa. *Triticum aestivum*, *L. aphaca*

and *V. sativa* root and shoot dry weight was influenced by the effects of osmotic potential. The comparison of treatment means given in Table 2 shows that under controlled conditions maximum root dry weight (2.14, 0.34 and 0.57 mg) and shoot dry weight (12.35, 8.73 and 11.12 mg) was recorded in *T. aestivum*, *L. aphaca* and *V. sativa* when there was no drought stress. Minimum root dry weight (1.34, 0.007 and 0.017 mg) and shoot dry weight (0.12, 0.0 and 0.02 mg) was recorded in *T. aestivum*, *L. aphaca* and *V. sativa*, respectively at -0.8 MPa. *Lathyrus aphaca* and *V. sativa* showed statistically minimum value as compared to wheat.

Table-1. Drought stress effects on germination of *Triticum aestivum*, *Lathyrus aphaca* and *Vicia sativa*.

Plant spp.	Drought level	Germination %	Coefficient of uniformity of germination (CUG)	Days to 50% germination (T ₅₀)	Mean germination time (Days)
<i>Triticum aestivum</i>	Control	93a	1.19a	1.89de	3.57def
	-0.4 MPa	86bc	0.99b	2.42c	3.91cd
	-0.5 MPa	80d	0.92b	2.84b	4.68b
	-0.8 MPa	57f	0.79c	2.98b	4.48b
<i>Lathyrus aphaca</i>	Control	91a	1.00b	2.09d	3.80cde
	-0.4 MPa	83cd	0.99b	2.93b	4.46b
	-0.5 MPa	64e	0.98b	3.54a	5.07a
	-0.8 MPa	8h	0.50d	2.50c	3.40f
<i>Vicia sativa</i>	Control	89ab	0.99b	1.75e	3.53ef
	-0.4 MPa	82cd	0.99b	2.41c	4.02c
	-0.5 MPa	62e	0.99b	2.96b	4.52b
	-0.8 MPa	17g	0.52d	1.47f	1.96g
LSD 5%		0.4539	0.0864	0.2763	0.3544

Table-2. Drought stress effects on seedling growth of *Triticum aestivum*, *Lathyrus aphaca* and *Vicia sativa*.

Plant spp.	Drought level	Root length (cm)	Shoot length (cm)	Dry root weight (mg)	Dry shoot weight (mg)
<i>Triticum aestivum</i>	Control	2.42b	4.77c	2.14a	12.35a
	-0.4 MPa	2.21c	0.84de	1.98b	1.78d
	-0.5 MPa	1.52d	0.18fg	1.81c	0.42e
	-0.8 MPa	0.87fg	0.04g	1.34d	0.12e
<i>Lathyrus aphaca</i>	Control	2.19c	11.26a	0.34f	8.73c
	-0.4 MPa	1.10e	1.12d	0.24fg	0.30e
	-0.5 MPa	1.08e	0.71e	0.25fg	0.10e
	-0.8 MPa	0.05h	0g	0.007h	0e
<i>Vicia sativa</i>	Control	3.43a	10.85b	0.57e	11.12b
	-0.4 MPa	0.96ef	0.80de	0.11gh	0.27e
	-0.5 MPa	0.75g	0.49ef	0.08h	0.14e
	-0.8 MPa	0.05h	0.04g	0.017h	0.02e
LSD 5%		0.186	0.364	0.145	0.478

Table-3. Salt stress effects on germination and growth of *Triticum aestivum*, *Lathyrus aphaca* and *Vicia sativa*.

Plant spp.	Salt stress level	Germination %	Root Length (cm)	Shoot Length (cm)	Root Dry Weight (mg)	Shoot Dry Weight (mg)
<i>Triticum aestivum</i>	Control	47.5 b	2.80 a	4.13 a	3.83 a	4.55 a
	50% Nacl	35.0 b	1.85 b	2.14 b	2.50 b	3.20 b
	100% Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
	150%Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
<i>Lathyrus aphaca</i>	control	37.5 b	0.61 cd	1.26 bc	0.00 c	0.00 c
	50% Nacl	12.5 c	0.41 cd	0.35 cd	0.00 c	0.00 c
	100% Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
<i>Vicia sativa</i>	150%Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
	control	80.0 a	0.94 c	4.23 a	0.00 c	0.00 c
	50% Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
	100% Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
	150%Nacl	0.00 c	0.00 d	0.00 d	0.00 c	0.00 c
LSD 5%		17.41	0.790	1.062	0.922	1.266

Table-4. Temperature effects on germination of *Triticum aestivum*, *Lathyrus aphaca* and *Vicia sativa*.

Plant spp.	Temperature	Germination %
<i>Triticum aestivum</i>	15°C	100.0 a
	20°C	97.5 ab
<i>Lathyrus aphaca</i>	15°C	92.5 abc
	20°C	87.5 bc
<i>Vicia sativa</i>	15°C	72.5 d
	20°C	85.0 c
LSD 5%		12.20

Table-5. Light effects on germination of *Triticum aestivum*, *Lathyrus aphaca* and *Vicia sativa*.

Plant spp.	Light	Germination %
<i>Triticum aestivum</i>	Light	95.0 a
	Dark	80.0 b
<i>Lathyrus aphaca</i>	Light	90.0 ab
	Dark	90.0 ab
<i>Vicia sativa</i>	Light	90.0 ab
	Dark	90.0 ab
LSD 5%		12.61

Root length is an important trait against drought stress in plant species. In general, species with longer roots have resistance against drought (Leishman and Westoby, 1994). The results of drought stress on germination suggest that wheat is more tolerant to drought stress as compared to *L. aphaca* and *V. sativa*. Restricted seedling growth in

both weed species, even at minimum drought stress level (-0.4 MPa) indicated that they are sensitive to low water potential. Similar decline in Sickle pod (*Senna obtusifolia*) root and shoot length was noted by Norsworthy and Oliveira (2006) when they exposed the seedlings to -1.0 MPa osmotic potential compared with absence of osmotic stress. Similar results have also been reported by Shaw *et al.* (1991) and Reddy and Singh (1992). It can be predicted that wheat may have competitive advantage over *L. aphaca* and *V. Sativa* under low moisture conditions because drought stress caused delayed emergence of these weeds as compared to wheat which give wheat a non competitive period to establish before the emergence of weeds. Increased level of drought stress caused severe decrease in root/shoot length and their dry weight which may be due to retardation of cell division and differentiation in absence of water needed for these activities. These results help to explain association between main events and flushes of these weeds in the field which occur during rainy season (Jan-Feb). Similar results have been shown by Jain and Singh (1989), and Baird and Dickens (1991).

Salt stress

Data regarding salinity stress suggested that *V. sativa* was most susceptible to salinity stress as no germination was observed at any salinity level investigated. Wheat is more tolerant to salinity as compared to weed species as 35 % wheat seed germination was recorded at 50 % NaCl level while *L. aphaca* showed 12.5% germination on the same salinity level. Root-shoot length and their respective dry weights were also affected significantly by salinity stress levels. At low salinity levels root and shoot elongations declined in *T. aestivum* and *L. aphaca* compared with the absence of salinity stress. No germination and ultimately no growth occurred in all test species when salinity stress was more than 50% NaCl. Root-shoot dry weights were not measurable in case of *V. sativa* and *L. aphaca* while in *T. aestivum* minimum root shoot weight was recorded at 50% NaCl level. *L. aphaca* and *T. aestivum* showed relatively more tolerance to salt stress at 50% NaCl level. But beyond this level both of these species showed zero germination and seedling growth. The significant difference in salinity tolerance between two weed species (*L. aphaca* and *V. sativa*) is in accordance with the findings of Chauhan and Johnson (2008) who noted that India crab grass and coat button germination was influenced to a greater extent than that of Southern crab grass and Siam weed, respectively by increasing salt concentration.

Similarly, Koger *et al.* (2004) and Chachalis and Reddy (2000) reported that crofton weed has greater salt tolerance than trumpet creeper (*Complis Redicons* L.) and Texas weed (*Coperonia*). Similar to *L. aphaca*, 18% of African mustard (a Brassicaceae weed species) seed germinated at a concentration of 160 mM NaCl (Chauhan *et al.*, 2006a). Our data suggested that *L. aphaca* could germinate in saline conditions and crop cultivation may be limited not only by salinity but also by competition from this species. Similar results were reported by Bhagirath and Johnson (2008) who found that goose grass could germinate and cause problem for crop cultivation in saline soils.

Temperature

Effect of temperature on germination of *T. aestivum*, *L. aphaca* and *V. sativa* was significant ($P < 5\%$). Statistically lowest germination (72.5%) was recorded at 15°C in case of *V. sativa* while the difference between two temperature regimes (15°C and 20°C) was non-significant in case of *T. aestivum* and *L. aphaca*. Temperature is an important factor in seed germination that plays a vital role in governing seasonability and range expansion. The maximum germination was recorded in *T. aestivum* at both temperature regimes tested. At 15°C the germination of *V. sativa* was inhibited indicating that this specie may not be a serious problem in relatively cooler wheat growing areas. Mattana *et al.* (2009) reported similar increase in germination of *Rhamnus perricifolia* shrub at warm temperatures (>20°C) as we noted in *V. sativa* which confirm that plant species vary in temperature adoptability range. Seeds of *V. sativa* germinated over a narrow range of temperatures which indicate that it has limited period of germination during a crop season. Some seeds germinate over a wide range whereas others require critical levels of relatively high temperatures (Labouriau, 1978).

Light

Wheat crop seed germination was significantly ($P \leq 0.05$) reduced when they were exposed to continuous darkness (80%) as compared to continuous light (95%) treatment while the effect of light and dark was non significant in case of *L. aphaca* and *V. sativa*. *Lathyrus aphaca* and *V. sativa* seeds germinated equally well (90%) under both conditions of light and darkness indicating that seeds of both species are not photoblastic. Similarly night shade seeds have been reported to germinate in light as well as in dark conditions (Jingkai *et al.*, 2005). These results are contradictory to the findings of Chejara *et al.* (2008) who reported a significant effect of light and dark treatments on the germination of *Hyparrhenia hirta*. Lower *T. aestivum*

seed germination in darkness suggested that wheat seeds are light sensitive. Similar results were reported by Anderson *et al.* (1997) and Ping *et al.* (2006).

Sowing depth

The comparison of treatment means given in Table 6 shows that emergence percentage of *V. sativa* (100%) was not affected by sowing depth. Likewise, in *L. aphaca* (100%) emergence percentage was not affected by sowing depth up to 3 cm but showed marked decline in emergence percentage with increase in sowing depth. Germination of *L. aphaca* was reduced to 5% and 2.5% with sowing depth of 5 and 10 cm, respectively. *Triticum aestivum* showed relatively less emergence (90%) at 1 cm depth than at 2 or 3 cm depth (95%) followed by 92.5% at both 5 and 10 cm sowing depths.

Table-6. Sowing depth effects on emergence of *Triticum aestivum*, *Lathyrus aphaca* and *Vicia sativa*.

Plant spp.	Planting depth	Emergence %	Days to emergence initiation	Days to complete emergence
<i>Triticum aestivum</i>	1cm	90d	9h	12i
	2cm	95b	7i	11j
	3cm	95b	7i	8k
	5cm	92.5c	11f	39c
	10cm	92.5c	11f	36d
<i>Lathyrus aphaca</i>	1cm	100a	10g	16g
	2cm	100a	9h	15h
	3cm	100a	13d	17f
	5cm	5e	28b	28e
	10cm	2.5f	29a	36d
<i>Vicia sativa</i>	1cm	100a	12e	17f
	2cm	100a	12e	17f
	3cm	100a	12e	17f
	5cm	100a	12e	46b
	10cm	100a	27c	68a
LSD 5%		0.498	0.320	0.300

The comparison of treatment means given in Table 3 shows that at 10 cm sowing depth, days to seedling initiation was maximum (11, 29 and 27 days) in *T. aestivum*, *L. aphaca* and *V. sativa*. Minimum number of days to initiate emergence was recorded at 2 cm depth in all plant species tested. Statistically highest number of days to initiate emergence and days to complete emergence was recorded in *V. sativa* at all sowing depths. The comparisons of treatment means given in Table 3 show that at 5 cm sowing depth total days to seedling emergence was maximum (39 days) in *T. aestivum* while in *L. aphaca*

and *V. sativa* it was recorded at 10 cm planting depth. Minimum values for it was recorded at 3 cm depth in *T. aestivum* while in *L. aphaca* and *V. sativa* it was recorded at 2cm depth.

Lathyrus aphaca seeds showed 100% emergence at 1, 2 and 3 cm sowing depth but emergence decreased at 5 and 10 cm sowing depth. On the other hand *V. sativa* showed 100% emergence at all sowing depths. Seeds of *T. aestivum* showed low percentage of emergence at 1 cm sowing depth that could be due to less moisture availability needed for emergence. Germination and emergence of plants is affected by many factors under field conditions. Benvenuti et al. (2001) stated that seed burial inhibited germination of *Rumex obtusifolius* in proportion to depth. According to Grundy et al. (2003), *Tripleurospermum inodorum* and *Veronica arvensis* having smaller seeds, showed a sharp decline in germination when their burial depth exceeded 1 cm. Usually, the level of oxygen is decreased with the greater soil depths, which probably resulted in dormancy in the seeds of *P. minor*. Similarly, Omami et al. (1999) and Mennan (2003) reported that seeds of *Amaranthus retroflexus* and *Galium aparine* had higher germination percentage on soil surface than those buried at different soil depths. All species showed more reduction in emergence except *V. sativa* at 10 cm depth which was most probably due to greater depth and inability of shoot to come out of soil. These results are supported by the findings of Hassan et al. (2003).

Minimum number of days required by wheat (at 1, 2 and 3 cm seeding depths) to seedling emergence initiation indicate that this species establish earlier. Maximum number of days to complete emergence taken by *L. aphaca* indicate that this weed continue to infest crops late in growing season. When weeds of Fabaceae (*L. aphaca* and *V. sativa*) and crop of Poaceae family (*T. aestivum*) were compared, it was noted that number of days required to initiation of seedling emergence and to complete seedling emergence was minimum in crop than weeds at all depths.

CONCLUSION

The observations of this study have shown that both *L. aphaca* and *V. sativa* possess potential to establish in ecologically diverse habitats. Our findings suggest that these weeds will dominate wheat crop if farmers will allow their seeds to remain on soil surface with out water stress. Further, it can be concluded that *L. aphaca* and *V. sativa* can be managed by burying their seeds below 10 cm depth by a tillage operation and subsequently using shallow tillage operation to

overcome bringing back the seeds on the soil surface. Sowing of *T. aestivum* at 5 cm depth gave it competitive edge in respect of earlier and maximum emergence over *L. aphaca*. Salinity is a constraint to *V. sativa* germination but *L. aphaca* could adapt to saline conditions that normally prevail in Asian sub-continent and create problem in harvesting higher crop fields.

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