



## Seed germination response of an invasive weed *Alternanthera ficoidea* to temperature and salinity stress

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### ABSTRACT

*Alternanthera ficoidea* (Sanguinaria) (Amaranthaceae) is an exotic weed that has widely spread in most of the terrestrial habitats including saline soils under different climatic conditions. Seed germination is an important stage in seedling establishment and maintaining health and vigour of every plant. Seed germination studies were carried out under temperature and salinity stress. Weed seeds showed adaptation to temperature as well as salinity stress in laboratory as well as field conditions. Such type of adaptation may be one of the reasons for rapid spread and establishment of this weed.

Worldwide biological invasion became a serious threat to native agriculture and natural ecosystems. Seed germination is an important development phase, playing a key role in seedling establishment and overall environmental adaptation in the life cycle of plant. High vigour seed is necessary to obtain fast and better establishment of seedling providing essential nutrients until it can photosynthesize independently. Frequency, amount of precipitation, seed germination and growth of seedlings during decrement of soil moisture and osmotic potentials, affects successful establishment of seedlings. Plants have a remarkable ability to cope up with extremely variable environmental stresses, including cold, drought, soils with changing salt and nutrient concentrations (Kreps *et al.* 2002). Drought and salinity are the two major environmental factors that reduce plant productivity. Temperature and salinity stress to invasive weeds were widely studied (Ebrahimi and Eslami 2011).

*A. ficoidea*, an exotic weed belongs to family Amaranthaceae, introduced from Tropical America is widely spread and established in most of terrestrial habitats all over India posing threat to biodiversity. Negative allelopathic effects on greengram and sorghum seed germination due to leaf leachates of *A. ficoidea* were previously reported (Patil and Kore 2017). It produces numerous small seeds and propagates rapidly vegetatively as well as by seeds and becomes a successful invader. Seed germination is one of the most important life-stages contributing

to the ability of a plant to become invasive. Acclimatisation to broad range of environmental conditions to germinate seeds rapidly is a crucial characteristic of invasive species. Investigation on effect of temperature and salinity stress on seed germination in *A. ficoidea* facilitates understanding invasive characteristics and adaptability of this weed to abiotic stress conditions studied.

Germination percentage alone does not reflect the speed or pattern of germination. Hence other germination indices, *viz.* rate, speed, percentage, vigour, establishment of the germinated seeds with addition to germination percentage made determination of germination potential of the seed. Lower the mean time for germination (MTG), faster the development of a population through seed germination. Coefficient of velocity (CVG), is an indication of rapidity of germination. Seedling length vigour index and seedling weight vigour index are the tests for vigour measurement.

In this study, eleven germination indices like germination percentage, radicle length, plumule length, seedling fresh weight, seedling dry weight, mean time for germination (MTG), mean daily germination (MDG), coefficient of velocity (CVG), seedling length vigour index (SLVI) seedling weight vigour index (SWVI) and daily germination speed (DGS) were studied. These parameters helped in comparison, interpretation and confirmation of results.

Experiments were carried out during 2015-2017 in triplicate. Thoroughly washed 10 seeds were kept in each sterilized Petriplate (9 cm diameter) lined with filter paper. For salinity stress, the filter papers in Petriplates were moistened with 10 ml solution of NaCl (10, 20, 30 and 40 m M concentrations). For temperature stress seeds were placed in air tight glass vials and were treated with high (60 °C) and low (4 °C) temperature, once for 24 h. before germination. These treated seeds then placed on germination paper moistened with D.W.

Untreated seeds were placed in Petriplates which lined with moistened filter papers by D.W. considered as control. Experiments were arranged in triplicates by using seed germination chamber at 26 °C with natural light and dark period. Finally, ten normal seedlings were selected randomly and average root and shoot length per seedling, and dry and fresh weight per 10 seedlings were recorded. In order to evaluate germination time and speed, Petri dishes were checked daily and number of germinated seeds were recorded. By daily counting of germinated seeds, seed germination and seedling vigour indices were calculated on 7<sup>th</sup> day as following:

**Final germination percentage (FGP):** (Scott *et al.* 1984)

FGP=Final number of seeds germinated in a seed lot × 100

**Mean time to germination (MTG):** Mean time to germination is an index of seed germination speed and velocity (Orchard 1977) and calculated by:  $MTG = \sum(n d) / \sum_n$

n : number of germinated seeds during seven days, d : number of day(s),

$\sum_n$ : total of germinated seeds

**Mean daily germination (MDG):** (Roberts 1981)

This is an index of daily germination speed and calculated by  $MDG = FGP/d$

FGP: final germination percent, d: test period

**Daily germination speed (DGS):** This index is converse of mean daily germination and calculated by  $DGS = 1/ MDG$

**Coefficient of velocity of germination (CVG):** (Jones and Sanders 1987)

This is another index of seed germination speed and velocity and calculated by:  $CVG = [(G_1 + G_2 + G_3 + \dots + G_n)] / [(1 \times G_1) + (2 \times G_2) + (3 \times G_3) + \dots + (n \times G_n)]$

G<sub>1</sub>-G<sub>n</sub>: number of germinated seeds from the first to the last day

**Daily Germination Speed (DGS):** Daily Germination speed calculated by this formula (ISTA 1999).  $DGS = (\text{number of normal seedlings/ Days to days to first count}) + \dots + (\text{number of normal seedlings/days to final count})$

**Seedling vigour index:** After measuring shoot and root length, fresh and dry weight of seedlings, weight and length vigour indices, were determined by using following formulae:

Seedling length vigour index (SLVI) = (mean shoot length + mean root length) × FGP.

Seedling weight vigour index (SWVI)/10seedlings = mean seedling weight × FGP

Most invasive plants primarily rely on seed dispersal and seedling recruitment for population establishment and persistence. Rapid spread of many invasive plants frequently correlated with germination and dormancy pattern. In nature, all living organisms including weeds frequently exposed to unfavourable environmental conditions that have adverse effects on their survival. Natural stress limit proliferation of weeds up to a certain extent.

Successful establishment of a plant species is dependent on adaptive mechanisms of seed germination and seedling growth. Seedling establishment is a critical stage in crop production and considerably depends on biochemical and physiological structures of seed. Seeds have the highest resistance to adverse environmental conditions during life cycle of a plant. They may be more sensitive to stresses than mature plants because of exposure to the dynamic environment close to the soil surface.

Salinity, drought stress biology and plant or seed responses at different levels have been discussed over two decades (Chauhan and Johnson 2009). Effect of salinity stress on germination of seeds of *A. ficoidea* are represented in **Table 1** and temperature stress in **Table 2**. Different indices were studied for clear interpretation and comparison of results.

### Salinity stress on seed germination

Reducing trend in germination per cent and seedling performance was observed. A considerable variation in all the germination attributes such as germination percentage and rate as well as seedling fresh and dry weights under salt stress was observed (**Table 1**).

CVG and MTG showed very minute effect. Seeds require higher amount of water uptake during the germination under salt stress and there is increase in osmotic pressure due to the accumulation of the soluble solutes around the seeds. This causes excessive uptake of ions which results in toxicity in plant (Jones 1986). Osmotic and toxic effects of the salts have been implicated in the inhibition of the germination (Machado Neto *et al.* 2004).

With increasing salinity level, germination per cent, seedling length and weight, MDG and SLVI were gradually decreased and are inversely proportional to salinity stress. This gradual declination was effect of reduction in ability to absorb water for germination, scarcity of nutrients for development of embryo as well as adverse effect of salinity on cell division and cell elongation.

The minimum SLVI and SWVI in all of the salinity levels were observed than that of control. Salt stress inhibited the seedling growth, but root length was more affected than shoot length. The reduction in growth observed in many plants subjected to salinity stress, is often correlated with salt-induced osmotic effect, nutrient deficiency or specific ion toxicity (Munns 2002). His work supports present results. Growth and survival of plants at elevated salinity levels depends on adaptation to stumpy water potentials and high sodium concentrations. DGS increased showed fluctuations as water imbibition and germination capacity of the seeds declined with increasing salt concentrations.

The maximum germination results are observed in control than stress conditions. The root and shoot lengths are the most important parameters for salt stress, as roots are in direct contact with soil and absorb water from soil and supply it to the rest of the plant. With increasing salinity level, the length of root and shoot of seedling was decreased. Interaction of salinity and root, shoot and seedling length was also showed a highly significant difference. Apparently ion active uptake by roots against concentration gradient

needs energy, so it may be one of the reasons for decrease in root length under salinity treatments

**Temperature stress to seed germination**

Temperature is an important factor for growth. Even though plants grow within very wide range of temperature, there may be difference in requirement of optimum temperature. More or less temperature than optimum value adversely affects seed germination. Seeds of *A. ficoidea* treated with temperature stress depicts (Table 2) inhibition of germination per cent, seedling length and weight, MTG, MDG, SLVI and SWVI. Gradual decrease in the above germination indices is more in hot stress than cold stress, possibly due to destruction of embryo and denaturation of enzymes required for germination .While due to cold stress, the enzyme activity may be trigger to increase germination per cent. More seedling fresh weight in hot stress could be due to growth of seedling than cold stress .

Seedling length and fresh weight is affected more in cold stress whereas hot stress showed unnoticeable effect, this might be due to some genetic factors as this weed acclimatizes to hot climatic conditions from its native region. Length of seedling was less at lower temperature as cell division and cell elongation process might have affected. Vigour index and speed of germination decreased as germination capacity affected. Seedling length and fresh weight affected more in cold stress whereas hot stress showed very minute effect might be due to some genetic reasons as this weed acclimatizes to hot climatic conditions from it’s native region.

**Table 1. Effect of salinity stress on seed germination**

Parameter Stress	FG %	Radicle length mm	Plumule length mm	Seedling f. w.mg/10 seedlings	Seedling d.w. mg /10 seedlings	MTG	CVG	MDG	DGS	SLVI	SWVI/10 seedlings
	Control	98.2±0.8	25±3	32±3	104±4	9 ±2	1.15±0.05	0.2±0.01	14.1±0.6	0.065±0.01	560±27
10 mM NaCl	84±2	20±2	25±3	62±6	6±3	0.9±0.04	0.2±0.01	12±0.7	0.08±0.01	227±11	5.2±0.3
20mM NaCl	73±3	20±1	22±2	53±7	5±2	1.02±0.04	0.19±0.1	10.4±0.5	0.095±0.01	175±10	3.9±0.2
30mM NaCl	71±3	15±3	18±2	52±5	4±2	0.6±0.05	0.22±0.1	10.1±0.5	0.098±0.01	138±12	3.7±0.2
40mM NaCl	65±2	15±2	17±2	51±2	2±2	0.68±0.03	0.24±0.1	9.28±0.4	0.107±0.01	120±13	3.3±0.2

**Table 2. Effect of temperature stress on seed germination**

Indices Stress	FG %	Radicle length mm	Plumule length mm	Seedling f.w. mg/10 seedlings	Seedling d.w.mg/10 seedlings	MTG	CVG	MDG	DGS	SLVI	SWVI/10 seedlings
	Control	98.2±0.8	25±3	32±3	104±4	9 ±2	1.15±0.05	0.2±0.1	14.1±0.6	0.065±0.01	560±27
Cold(4 °C)	98±0.2	23±2	28±1	95±5	6±3	0.87±0.02	0.21±0.1	14±0.2	0.071±0.01	500±22	9.31±0.5
Hot (60°C)	79.5±0.5	24±2	30±1	79.5±0.5	5±3	0.76±0.01	0.21±0.1	13.7±0.1	0.067±0.01	528±25	9.97±0.45

Length of seedling was less at lower temperature as cell division and cell elongation process might have affected. Vigour index and speed of germination decreased as germination capacity affected. DGS and CVG were inversely proportional to the MTG so it minutely increased from control to cold stress.

Seed germination decreases with increase in temperature showed signs of stagnation or decrease in seedling growth in important crops like maize, rice and sorghum reported by Lloh *et al.* (2014). According to Neelambari *et al.* (2018), germination percentage, root shoot ratio, fresh weight ratio decreased in some wheat varieties due to salinity and temperature stress.

*A. ficoidea* seed germination showed very minute effect of temperature stress as interpreted in **Table 2**. Negligible adverse effect of temperature stress shown by all germination indices studied proves its adaptability to temperature stress and potential to use as subsidiary food during scarcity.

Species with great plasticity and environmental tolerance may easily find suitable habitats in the introduced localities. Establishment of a plant species in high and low temperature area than optimum is related to germination response of seeds to temperature and early establishment usually decides population's survival to maturity.

Greater tolerance and adaptability exhibited by seeds of *A. ficoidea* to temperature and salinity stress. This may be one of the reason for invasiveness and rapid spread of this exotic weed in terrestrial habitats including saline soil as well as drastic temperatures. Different germination indices studied helped in comparison and confirmation of the results. This invasive weed may be used as an alternative to regular food during adverse conditions for removing scarcity.

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