Enhancing Germination Capacity of Chloris barbata under in-vitro Conditions

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A B S T R A C T

Investigation was conducted at the laboratory at Tamil Nadu Agricultural University (TNAU), Coimbatore to enhance the germination capacity of Chloris barbata under in-vitro conditions with water and Potassium nitrate (KNO₃). The species performed poor for germination rate when placed in the petri plate at room temperature. To attain the germination rate and the uniformity of the germination an experiment was performed with six treatments viz. T₁ - Control (without KNO₃ treatment and soaking), T₂ - Soaking of seeds in water for 6 hrs, T₃ - Soaking of seeds in water for 12 hrs, T₄ - Seed treatment in 2% KNO₃ solution, T₅ - Seed treatment in 2% KNO₃ solution with 6 hrs soaking and T₆ - Seed treatment in 2% KNO₃ solution with 12 hrs soaking, all replicated four times. Germination behaviours of weed seeds were observed in all the treatments. The results showed that the germination percentage was significantly influenced by the seed treatment with KNO₃ and seed soaking. Seeds treated with 2% KNO₃ solution with 12 hrs soaking (T₆) recorded significantly higher seed germination (63%) compared to other treatments. Mean germination time of C. barbata reduced significantly when it was treated with 2% KNO₃ solution with soaking (6 & 12 hrs; T₅ & T₆, respectively). The maximum speed of germination, coefficient of velocity of germination, mean daily germination, peak value and germination value was recorded when the C. barbata seeds were treated in 2% KNO₃ solution with 12 hrs soaking.

Keywords
Chloris barbata, Water soaking, KNO₃, Germination, seed banks

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Introduction

The soil is a resting place for weed seeds, and they are the main source of weed population, which reproduces sexually, by seeds. Due to presence of the weed seed dormancy it’s very difficult to know the quantity of weed seeds in soil bank and this dormancy helps the seeds to thrive in soil over a period of time and cause threat to the crop production. To overcome these problems, the germination rate and uniformity of the weed seed germination has to be increased to attain reliable data from weed seed bank studies.

Chloris barbata, one of the annual or short-lived perennial species propagated through seeds, is an important weed in many tropical and subtropical regions. Under laboratory conditions C.barbata seeds were tested for germination and its response was very poor (< 10 %). Suhas and Joshi (2013) reported that
seeds of *C. barbata* showed little dormancy when germinated under non-saline conditions. Normally, to improve the seed germination it can be treated with water or any chemical to initiate the early germination process. Seed hydration with water encourages the germination by activating the enzymes and accelerates the starch and protein metabolism (Kikiuchi *et al.*, 2006). Sometimes, the presence of seed coat delays the speed of germination. Under such conditions, Potassium nitrate (KNO₃) stimulates the partial germination of seeds (Silva *et al.*, 2009). Despite some findings proved that the KNO₃ accelerates the seed germination in tomato (Lara *et al.*, 2014), *Calotropis persica* (Farajollahi *et al.*, 2014), Perennial ryegrass (Danneberger *et al.*, 1992), Shim *et al.*, 2008) reported that KNO₃ were used for breaking seed dormancy and promoting seed germination.

However, the duration of soaking also plays a major role in the germination behavior, which reduces the seeds natural defense against the germination and speeds up the process to initiate the germination. The study was conducted to find out the possible way to increase the germination percentage in Petridish to undertake the weed seed bank studies at normal conditions.

**Materials and Methods**

The present investigation was conducted at the laboratory in the Department of Agronomy, Agricultural College and Research Institute, Tamil Nadu Agricultural University (TNAU), Coimbatore, India, during 2019 to study the effect of water soaking and Potassium nitrate (KNO₃) on the germination of *C. barbata*. Seeds of *C. barbata* were collected from the mature plants of weeds in the field and the seeds were sun dried for three days and the seeds were kept in glass bottles at room temperature for the experiment. Twenty-five healthy seeds of *C. barbata* with uniform size were selected for all the treatments. Seeds were kept in Petri dish over filter paper (Whatman No. 1) and allowed to germinate by providing optimum moisture. After confirmation of poor germination, the seed lot was used for further studies. The experiment consists of six treatments with four replications viz. T₁ - Control (Without KNO₃ treatment and soaking), T₂ - Soaking of seeds in water for 6 hrs, T₃ - Soaking of seeds in water for 12 hrs, T₄ - Seeds treatment in 2% KNO₃ solution, T₅ - Seeds treatment in 2% KNO₃ solution with 6 hrs soaking and T₆ - Seeds treatment in 2% KNO₃ solution with 12 hrs soaking. Germination behaviours of weed seeds were observed in all the treatments. Germinated seeds number was counted (seed radical emergence which attained 2 mm were considered as the germinated seeds) on daily basis and observed up to 10 days after sowing, after that seedling root and shoot length were also observed. At the end following observations were recorded.

**Final germination percentage (FGP)** (Orchard, 1977)

\[
FGP = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds}} \times 100
\]

**Speed of germination** (Maguire, 1962)

\[
\text{Speed of Germination} = \frac{\text{No. of seeds germinated}}{\text{Day of first count}} + \ldots + \frac{\text{Number of seeds germinated}}{\text{Day of final count}}
\]

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

\[
\text{MDG} = \frac{\text{Final germination percentage}}{\text{Total number of days}}
\]

**Mean Germination Time (MGT)** (Orchard, 1977)

\[
\text{MGT} = \frac{\sum (\text{nd})}{\sum n}
\]
Where, n - Number of seeds germinated; d - Number of days; \(\sum n\) - Total number of seeds germinated

Daily germination speed (DGS) Reshma and Basavaraj (2019)

\[
DGS = \frac{1}{MDG}
\]

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

\[
CVG = \frac{Ni + N2 + \ldots Ni}{100 \times N1Ti + N2T2 + \ldots NiTi}
\]

Where, \(Ni\) is the number of seeds germinated each day; \(Ti\) is the number of days from sowing corresponding to \(N\)

Peak value (Cazabator, 1962)

\[
\text{Peak value} = \frac{\text{Highest seed germinated}}{\text{Number of days}}
\]

Germination value (Cazabator, 1962)

\[
GV = PV \times MDG
\]

Time to 50 percent germination (Farooq et al., 2005)

\[
T_{50} = Ti + \left( \frac{N}{2} - \frac{Ni}{Nj - Ni} \right) \times 100
\]

Where, \(T_{50}\) is the median germination time, \(N\) is the final number of seed germinated and \(Ni\) and \(Nj\) are the total number of seeds germinated in the adjacent counts at time \(Ti\) and \(Tj\) respectively, when \(Ni < N/2 < Nj\).

Results and Discussion

Germination percentage

The germination percentage was significantly influenced by the seeds treatment with KNO\(_3\) and seed soaking. Non treated seeds (\(T_1\)) recorded poor germination (<10%) and it was on par with the water soaking for 6 hrs. When soaking time was increased to 12 hrs (\(T_3\)) it recorded significantly higher germination percentage (25 %) (\(T_3\)). Seeds treated with 2 per cent KNO\(_3\) solution had significantly positive effect on germination of \(C.\ barbata\) and also the germination percentage increases with increased time of soaking. Seeds treated with 2 per cent KNO\(_3\) solution with 12 hrs (\(T_6\)) soaking recorded significantly higher seed germination (63 %) compared to other treatments and it was followed by the treatment in which seeds treated with 2 per cent KNO\(_3\) solution with 6hrs (\(T_5\)) soaking (51 %). The KNO\(_3\) treated seeds showed higher percentage of germination and it is in accordance with the results of Zavariyan et al., (2015). It may be due to the presence of nitrate (NO\(_3^-\)) in KNO\(_3\) provided exogenously which acts as a signal molecule that favours the germination of \(C.\ barbata\) by involving in the gibberellins pathway (Alboresi et al., 2005) (Fig.1a-1b and Table 1).

Mean germination time

\(C.\ barbata\) shows significant response to the seeds treatment with respect to mean germination time. Mean germination time of \(C.\ barbata\) reduced significantly when it treated in 2% KNO\(_3\) solution with soaking (6 & 12 hrs) (\(T_5\) & \(T_6\)). The treatment \(T_4\) recorded higher mean germination and it may be due to the lesser time for the imbibition of KNO\(_3\) solution, since the MGT depends on the imbibition process in which priming activates the internal metabolic activities required for further germination process (Basra et al., 2005).

Soaking of seeds in water for 12 hrs (\(T_3\)) recorded higher mean germination time when compared to seeds soaked in water for 6hrs (\(T_2\)), it may be due to increase in germination percentage at lower rate of germination.


**Speed of germination**

The maximum speed of germination was recorded when the *C. barbata* seeds treated in 2% KNO$_3$ solution with 12 hrs soaking (3.40) and as soaking time decreases the germination speed also get decreased significantly. Seed of *C. barbata* without treatment of 2% KNO$_3$ solution recorded significantly lower speed of germination compared to the seeds treated with the 2% KNO$_3$ solution. Farooq *et al.*, 2006 recorded that the K$^+$ improves the cell water status and also act as the cofactor in activities of various enzymes most of which are active when reserve mobilization and radical protrusion are in progress. Time of soaking which plays a role in the ease of imbibition process leads to readily available food during germination thus complete the process of germination in the shorter time (Kant *et al.*, 2006; Kaur *et al.*, 2005) and it is accordance with the present study (Table 2).

**Table 1** Effect of seeds treatment on the germination indices of *Chloris barbata*

| Treatments                                      | Mean germination time | Speed of germination | Mean daily germination |
|-------------------------------------------------|-----------------------|----------------------|------------------------|
| T$_1$ - Control (Without KNO$_3$ treatment and soaking) | 5.13                  | 0.36                 | 0.29                   |
| T$_2$ - Soaking of seeds in water for 6 hrs      | 5.15                  | 0.55                 | 0.46                   |
| T$_3$ - Soaking of seeds in water for 12 hrs     | 5.53                  | 1.18                 | 0.92                   |
| T$_4$ - Seeds treatment in 2% KNO$_3$ solution   | 5.98                  | 1.86                 | 1.54                   |
| T$_5$ - Seeds treatment in 2% KNO$_3$ solution with 6 hrs soaking | 4.75                  | 2.76                 | 1.90                   |
| T$_6$ - Seeds treatment in 2% KNO$_3$ solution with 12 hrs soaking | 4.76                  | 3.40                 | 2.25                   |
| S.Ed                                            | 0.28                  | 0.13                 | 0.12                   |
| C.D.(p=0.05)                                    | 0.59                  | 0.28                 | 0.26                   |

**Table 2** Effect of seeds treatment on the Time to 50 % germination, Peak value and Germination value

| Treatments                                      | Time to 50 % germination | Peak value | Germination value |
|-------------------------------------------------|--------------------------|------------|-------------------|
| T$_1$ - Control (Without KNO$_3$ treatment and soaking) | 4.38                     | 0.23       | 0.07              |
| T$_2$ - Soaking of seeds in water for 6 hrs      | 4.38                     | 0.32       | 0.15              |
| T$_3$ - Soaking of seeds in water for 12 hrs     | 5.13                     | 0.54       | 0.50              |
| T$_4$ - Seeds treatment in 2% KNO$_3$ solution   | 5.54                     | 0.74       | 1.14              |
| T$_5$ - Seeds treatment in 2% KNO$_3$ solution with 6 hrs soaking | 4.32                     | 1.44       | 2.76              |
| T$_6$ - Seeds treatment in 2% KNO$_3$ solution with 12 hrs soaking | 4.26                     | 1.75       | 3.96              |
| S.Ed                                            | 0.44                     | 0.08       | 0.27              |
| C.D.(p=0.05)                                    | 0.925                    | 0.18       | 0.57              |
Mean daily germination

The mean daily germination of *C. barbata* was lower (0.18) in the control and it was on par with the soaking of seeds in water for 6 hrs (0.28). When seeds soaked for 12hrs in water (T₃) and treated with the 2 % KNO₃ solution recorded significantly higher mean germination. Increasing in germination percentage and speed of germination due to the positive response of *C. barbata* seeds also significantly reflect on the mean daily germination.

Daily germination speed (DGS)

The higher daily germination speed (6.25) of *C. barbata* is recorded in the control treatment (T₁) and significantly low value of DGS is recorded in the treatment in which,
seeds of *C. barbata* is treated in 2 % KNO$_3$ solution with 12 hrs soaking (T$_6$) (0.64). Results shows that DGS decreased as treatments favours germination capacity of the seeds and its accordance with the findings of Reshma and Basavaraj (2019) reported DGS increased as water imbibition and germination capacity of the seeds declined with increasing salt concentration (Fig. 2).

**Coefficient of velocity of germination (CVG)**

Seeds treated in 2 % KNO$_3$ solution with 12 hrs soaking (T$_6$) recorded significantly higher coefficient of velocity of germination (11.88) and lower coefficient of velocity of germination were resulted in the non-treated seed (control) (0.16). Busso *et al.*, (2005) stated the coefficient of velocity of germination get increased when more number of seeds germinate in the lesser time and the value get decreased when less number of seeds took more days to germinate.

**Time to 50 percent germination**

Days to 50 % germination is significantly influenced by the seeds treatments. Seeds treated in 2 % KNO$_3$ solution with 12 hrs soaking (T$_6$) shows significantly lesser number of day for 50% germination when compared to seeds treated with KNO$_3$ without soaking. When soaking time decreases the T$_{50}$ tends to increase. When compare the treatments without KNO$_3$ higher day for 50 % germination recorded in higher time of soaking. Soaking the seeds in water for 12 hours without KNO$_3$ recorded a higher germination percentage when compared to T$_1$ & T$_2$, so it took longer days to germinate. It is in accordance with the findings of Farooq *et al.*, (2007) who reported that all the seed primed treatments reduced the time to 50 % emergence compared with control.

**Peak value and Germination value**

The peak value and germination value was observed maximum in seed treated in 2 % KNO$_3$ solution with 12 hrs soaking (1.75 and 2.77) and minimum in control(0.23 and 0.44). It might be due to the soaking duration, the value varied as the soaking time increases the peak value and germination value increased in the respective treatment (Cazabator, 1962).

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