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Effects of Environmental Factors on Seed Germination and Seedling Emergence of Common Teasel (Dipsacus fullonum).

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Abstract:	Common teasel is an invasive and widespread weed in Argentina. Germination experiments were carried out from 2011 to 2014 to determine the effect of various environmental factors on germination and emergence. Germination of recently dispersed seeds was 12% in darkness at constant temperature. In contrast, seed exposure to light and alternating temperatures enhanced germination to 95%. Requirement of light and alternating temperatures suggests that common teasel has physiological dormancy. Several experiments were carried out to determine whether: i) seed responses to light and alternating temperatures have a hormonal basis, ii) seed coats can suppress germination, iii) time and thermal conditions during seed storage reduces light and alternating temperatures requirements. Germination was reduced (≤ 13%) by a gibberellins synthesis inhibitor. In contrast, the presence of gibberellins and an abscisic acid synthesis inhibitor, increased germination to 95 and 38%, respectively. Results suggest that a higher ratio among gibberellins and abscisic acid (GA/ABA) lead to dormancy breakage. Germination was 100% when embryos were excised, suggesting that seed coats may suppress germination by mechanical restriction. Likewise, germination was enhanced by hydrogen peroxide (70%). This compound is known to increase GA/ABA ratio in agreement with a hormonal control of dormancy proposed for common teasel. An increment of storage time reduces light and alternating temperatures requirement, allowing seeds to germinate in darkness. Taking these results together confirms that common teasel has physiological dormancy. Seedling emergence was progressively reduced from 70 to 8% by increased burial depth from 0 to 3 cm. Information from these experiments may facilitate development of effective management for common teasel

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Running title: Common teasel germination

Effects of Environmental Factors on Seed Germination and Seedling Emergence of Common Teasel (*Dipsacus fullonum*).

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Common teasel is an invasive and widespread weed in Argentina. Germination experiments were carried out from 2011 to 2014 to determine the effect of various environmental factors on germination and emergence. Germination of recently dispersed seeds was 12% in darkness at constant temperature. In contrast, seed exposure to light and alternating temperatures enhanced germination to 95%. Requirement of light and alternating temperatures suggests that common teasel has physiological dormancy. Several experiments were carried out to determine whether: i) seed responses to light and

alternating temperatures have a hormonal basis, ii) seed coats can suppress germination, iii) time and thermal conditions during seed storage reduces light and alternating temperatures requirements. Germination was reduced (≤ 13%) by a gibberellins synthesis inhibitor. In contrast, the presence of gibberellins and an abscisic acid synthesis inhibitor, increased germination to 95 and 38%, respectively. Results suggest that a higher ratio among gibberellins and abscisic acid (GA/ABA) lead to dormancy breakage. Germination was 100% when embryos were excised, suggesting that seed coats may suppress germination by mechanical restriction. Likewise, germination was enhanced by hydrogen peroxide (70%). This compound is known to increase GA/ABA ratio in agreement with a hormonal control of dormancy proposed for common teasel. An increment of storage time reduces light and alternating temperatures requirement, allowing seeds to germinate in darkness. Taking these results together confirms that common teasel has physiological dormancy. Seedling emergence was progressively reduced from 70 to 8% by increased burial depth from 0 to 3 cm. Information from these experiments may facilitate development of effective management for common teasel.

Nomenclature: common teasel *Dipsacus fullonum* L. DIFU2; fluridone, gibberellins, trinexapac-ethyl.

Keywords: Alternating temperatures, depth of burial, GA/ABA ratio, physiological dormancy, germination, light.

Common teasel is a very aggressive, biennial, exotic species capable of colonizing natural and improved pastures, abandoned fields and roadsides. Common teasel plants are herbaceous 0.5 – 2 m tall, and have prickles on stem and leaves. Rosettes have large leaves that cover the soil around them, reducing resources available for establishment of other species (Rector et al 2006). Reproduction and dispersion is exclusively through seeds and each plant produces around 3000 seeds per year (Werner 1975). Due to this trait, knowledge of seed biology of this species is essential for developing sustainable management practices (Bhowmick 1997; Chauhan and Johnson 2008). Common teasel was observed in more than 40 states in USA and 4 Canadian provinces (Werner and Caswell 1977, Rector et al. 2006, USDA 2015). In Argentina, even though it was detected decades ago, colonized area has increased significantly in the past years, and it is considered as one of the most abundant weed species in grasslands and right of ways (Marzocca 1976; Busso et al. 2013). Nevertheless, there is no information available related to seed germination and seedling establishment of Argentinian seed populations.

Germination and emergence are important processes for weed establishment and persistence. Environmental factors, such as temperature and light, modulate these processes and influence weeds presence in the field. Werner (1975) and Beaton and Dudley (2007) reported that total germination of common teasel seeds exposed to light and/or alternating temperatures reached values higher than 90%. In contrast, there is no information regarding their germination under other environmental conditions, such as darkness. Batlla and Benech-Arnold (2010) pointed out that the stimulus of alternating temperatures and/or light are essential for germination of many species. These requirements are common for many species that have physiological dormancy (Finch-Savage and Footitt 2012). Requirement of one or both stimuli are gradually reduced as

seeds lose their dormancy under natural or artificial conditions (*e.g*. cold stratification or dry after-ripening) and, in many cases, when dormancy level is sufficiently low, seeds are able to germinate even at constant temperature in darkness (Benech-Arnold et al. 2000). To date germination requirements of Argentinian common teasel seeds are not known and some key questions are: I) is there a specific requirement of alternating temperatures and/or light to germinate II) is any of these stimuli, when present alone, enough for common teasel to germinate III) are common teasel seeds dormant at the time of being dispersed.

Physiological dormancy is the most abundant form of dormancy in temperate species and is found in seeds of gymnosperms and all major angiosperm clades (Finch-Savage and Leubner-Metzger 2006). Some features of this type of dormancy are: embryos excised from seeds produced normal seedlings, gibberellins (GA) treatment can break dormancy and, depending on the species, dormancy can also be broken by scarification, after-ripening in dry storage, and cold stratification. Responses of common teasel seeds to these treatments will provide precise information about the existence and type of dormancy present in this species. As expressed above, light and alternating temperatures act as environmental stimulus to terminate dormancy. Once perceived by seeds, they regulate the expression of genes involved in hormones synthesis and degradation, and regulated the ratio between abscisic acid (ABA) and GA content (Footitt et al. 2011). Both ABA and GA are considered as key hormones in the induction of seed dormancy and promotion of seed germination respectively (Linkies and Leubner-Metzger 2012). In particular, the ratio between the contents of these two hormones are important in regulating induction, maintenance and termination of dormancy (Finkelstein et al. 2008). The termination of dormancy requires low ABA/GA ratios. In contrast, the maintenance

of dormancy is associated with high ABA/GA ratios (Finch-Savage and Leubner-Metzger 2006). The involvement of these hormones in light or alternating temperature responses has not been studied for common teasel.

The general aim of our research was to obtain a better understanding of seed germination and emergence responses of common teasel to different factors and environmental conditions. The particular objectives of this study were: i) to evaluate light and alternating temperatures requirements on germination, ii) to determine if seed responses to light and alternating temperatures are hormonally regulated, iii) to determine if common teasel has physiological dormancy and iv) to evaluate the influence of depth of burial on emergence.

Materials and Methods

Seed Source and Preparation.

Common teasel seeds were collected in January 2011 and 2012 at the School of Agriculture, Catholic University of Argentina (UCA) campus (S 34°34'; W 58°28'). Seed heads were collected from many randomly selected plants. Seeds were manually separated from each head and bulked. Seeds were then surface-sterilized by immersion in a 0.5% sodium hypochlorite solution for 10 min, then rinsed with distilled water several times, and then dried at room temperature. After that, dry seeds were kept at 6 C until the beginning of experiments.

General Procedure for Germination Studies

All experiments were conducted from 2011 to 2014 at the Seed Laboratory (School of Agriculture, UCA, Buenos Aires City, Argentina). All experiments were performed twice and consisted of four replicates of thirty sterilized seeds of common teasel. Seeds were placed on 9 cm Petri dishes lined with two sheets of filter paper (Double Ring # 102 Hangzhou Special Paper Industry Co., Ltd., China) in all experiments, unless stated otherwise. Filter papers were moistened with 8 ml of distilled water or corresponding test solution. All Petri dishes were placed in plastic bags (Ziploc® Johnson & Johnson, New Brunswick, NJ, USA) to prevent dehydration, and were kept for 21 days in germination chambers with day/night temperature of 20/10 C and 15 C for alternating and constant temperatures treatments, respectively. Temperatures used represent typical fall average high and low temperatures in our region. Light treatment was accomplished by seed exposure to six fluorescent Philips 40/15 40W lamps (Philips, Eindhoven, The Netherlands) to obtain a calculated proportion of Pfr of 0.87 (Casal et al. 1991), while dark treatment involved covering Petri dishes with two layers of aluminum foil. Germinated seeds for light treatments were counted daily until day 21, while germination for dark treatment was determined at day 21. Seeds with visible radicle protrusion were considered as germinated and were removed.

Effects of Alternating Temperatures, Light, Trinexapac ethyl, and Gibberellins on Seed Dormancy Breakage

To evaluate seed requirement of alternating temperatures and/or light for germination, and participation of GA on dormancy breakage, common teasel seeds were incubated at a combination of three factors: temperature (2 levels), light (2 levels) and media (2 levels). Temperature treatments were: alternating (20/10 C, 12 h each) and constant (15 C). Light treatments were: 12 h light/12 h darkness (light) and 0 h light (darkness). Media treatments were: distilled water (H_2O) and 250 µM trinexapac-ethyl (TE). Trinexapac-ethyl is a GA synthesis inhibitor (Rademacher 2000; Yamaguchi et al. 2008). Data was analyzed using a factorial design with three factors.

Effect of GA and Fluridone on Dormancy Breakage

Seeds were incubated in darkness at constant temperature (15 C). Gibberellins treatments were: 0, 50 and 250µM GA_3 (Sigma Chemical Company, ST Louis, MO, USA). The positive control was 15 C + light + 0 µM GA_3. To determine if primary dormancy is related to ABA present in the seed, seeds were incubated in darkness at 15 C in 10, 50 and 100 µM fluridone (1-methyl-3-phenyl-5-[3-trifluoromethyl-(phenyl)(-4-(1H)-pyridinone)]), (Phytotechnology Laboratories, Shawnee Mission, Kansas, USA). Fluridone is an ABA synthesis inhibitor (Yoshioka et al. 1998). Additionally, seeds were incubated in mixed solution 50 µM fluridone plus 250 µM GA_3. The pH of each solution was corrected to 6.5.

Effect of Seed Coat on Seed Germination

To determine if seed coat restricts seed germination, a surgical excision of embryos was carried out. Seed coats were cut at opposite end to the embryo with a scalpel, aided by tweezers and a magnification glass. Four replicates of 25 intact embryos were located in 9 cm Petri dishes on two filter paper discs moistened with 8 ml distilled water. Dishes were incubated for 9 d at 15 C in darkness and then germination was scored. Excised embryos germination was compared with that of whole seeds incubated for 21 d at 15 C in light (positive control) and in darkness (negative control).

Effect of Nitrogenous and Oxidant Treatments on Seed Germination

Four treatments, one rate of 0.2% potassium nitrate (KNO_3) and three rates of hydrogen peroxide (0.9, 4.5, and 8.9M), were tested on common teasel germination. All treatments were performed using 30 seeds per Petri dish and four replicates. Seeds were incubated at 15 C in darkness for 21 d. A positive control (15 C and light for untreated seeds) and a negative control (15 C and darkness for untreated seed) were also included.

Effect of Chemical and Mechanical Scarification on Seed Germination

Chemical scarification treatment was performed by seeds imbibition in concentrated sulphuric acid (H_2SO_4) by 0, 5, 15, 45, or 90 min. Seeds were washed with tap water and dried on absorbent paper. Mechanical scarification treatments consisted on: i) sanding the end opposite to the embryo by five times with a sandpaper (N°200), and ii) sanding entire seed surface by five times longitudinally on both sides. Sanded seeds were used to perform a germination test at 15 C in darkness.

Effect of Time and Temperature Storage on Seed Dormancy Relief

To determine the effect of time and temperature on a reduction in light or alternating temperatures requirement for germination, recently dispersed seeds were exposed to a combination of two treatments: i) cold stratification (*i.e*. hydrated seeds at 6 C) and ii) dry after-ripening (dry seeds at 15 C), and four durations: 0, 4, 8 and 12 weeks. After completion of the stratification treatments, seeds were incubated at 15 C in darkness for 21 d, when germination was recorded.

Effect of Seed Burial Depth on Seedling Emergence

The effect of seed burial depth on seedling emergence was studied in germination chambers set at 20/10 C (12 h light/12 h darkness). Eight seeds were placed on soil surface of 10 cm diameter plastic pots and covered with soil to 0, 0.5, 1, 1.5, 2, and 3 cm depth (five replicates per sowing depth). Soil used was sterilized topsoil and perlite mixed in a 2:1 (v/v) ratio. Containers were watered as needed to maintain optimum soil moisture for seed germination. Seedlings were considered emerged when cotyledons were visible. Seedling emergence was recorded 14 d after sowing.

Statistical Analysis

Data was subjected to Analysis of Variance (ANOVA) using a completely randomized design with four replicates for all experiments unless otherwise stated. Data presented are averages across two experimental runs because there was no significant

experiment-by-treatment interaction. Means were separated using Tukey's test at $P < 0.05$. Data were analyzed using Infostat (Di Rienzo et al. 2013).

RESULTS and DISCUSSION

Effects of Alternating Temperatures, Light and GA on Germination

There was a three-way interaction between temperature, light and incubation media ($P < 0.001$). Within H_2O treatments, combination of alternating temperature (20/10 C) in light (96 ± 2%) (Mean ± SE), and in darkness (100%), and constant temperature (15 C) in light (96 ± 2%), got the highest germination and there was no differences between them (Figure 1). The lowest germination in H_2O was for constant temperature (15 C) in darkness (13 ± 8%). Addition of TE to media reduced germination of all treatment combinations with, either or both, alternating temperature and light, which had the same low germination that scored at constant temperature in darkness (Figure 1). Also, low germination in all TE treatments was similar to that obtained in H_2O, at constant temperature and darkness (Figure 1). These results suggest that common teasel seed are dormant at time of dispersion, but it does not require both alternating temperatures and light to germinate. Either alternating temperatures or light are enough to trigger germination of common teasel. Total germination obtained in this study was in agreement with Werner (1975). Reduction in germination by TE suggests that GA have a role in seed dormancy breakage. Alternating temperatures or light requirement for germination of this population agrees with field conditions for common teasel germination in our region. Werner (1975) reported that common teasel germination is favored by open gaps, where

temperatures fluctuations are high and the incoming light, with a high R/FR ratio, satisfy both germination requirements. Therefore, those agronomic practices, such as seed burial or the presence of closed canopies, which prevent seeds to receive these signals, would reduce germination of recently dispersed seeds. Additionally, common teasel seed viability is short, not exceeding two years (Werner and Caswell 1977). Consequently, use of these practices together with prevention of a new seed rain would progressively reduce seed bank size.

Effect of Gibberellins and Fluridone on Seed Germination

Several authors reported that termination of dormancy by alternating temperature and light is related to an increase in GA/ABA ratio (Toyomasu et al. 1998, Sawada et al. 2008, Huarte et al. 2014). Low germination obtained in TE, which was similar to that scored at constant temperatures in darkness, suggested that dormancy breakage is mediated by GA. We wanted to evaluate if addition of exogenous GA and the inhibition of ABA synthesis, by means of fluridone, increase germination. While 50 μM GA, at constant temperature in darkness, did not increase germination, 250 μM GA increased germination to percentages similar to those of alternating temperatures and light positive control (Figure 2a). The presence of fluridone at constant temperatures (15 C) in darkness enhances germination in relation to negative control (*i.e.* 15 C + dark) ($p < 0,001$) (Figure 2b). The three fluridone concentrations tested were equally effective to increase germination (38 ± 8; 36 ± 10 and 25 ± 6% for 10, 50 and 100 μM respectively) when compared to negative control (i.e. 15 C + dark). However, germination was still 50% lower than the positive control (15 C + light) which had the greatest germination (75 ±

4%). Increment of GA/ABA ratio, by means of exogenous GA or blocking ABA synthesis, increases seed germination. Taken together, these results could suggest that common teasel dormancy breakage is under hormonal regulation. These results agree with those reported for other species with physiological dormancy, where a higher GA/ABA ratio trigger germination. A high GA/ABA ratio promote synthesis of cell wall remodeling proteins, which are responsible to increase embryo growth potential and/or seed coat weakening, leading to germination (Linkies and Leubner-Metzger 2012).

Effect of Seed Coat on Germination

Germination of excised embryos incubated at 15 C in darkness was 100% not differing to that scored in the positive control (15 C + light) (Figure 3). These results, together with the enhancement of germination by means of GA (Figure 2a), suggest that physiological dormancy is the type of dormancy present in common teasel. Based on the hormonal response, germination impairment by seed coats seems to be mechanical. However, seed coats can also impede germination by other mechanisms (Nasreen et al. 2002). To solve this issue, we conducted additional experiments.

Effect of Scarification on Seed Germination

Sulphuric acid-treated seeds by 5 min and subsequently incubated in water at 15 C in darkness, obtained greater germination (18 ± 1%) to that of the negative control (0.4 ± 0.4%) and all other H_2SO_4 treatments (15, 45 and 90 min). However, efficiency of this treatment to enhance germination was not enough to get closer to positive control ($P <$

0.0001) (15 C + light) (Figure 4a). Increase in germination by H_2SO_4 treatment was smaller than that reported for other species (*e.g.* Aref et al. 2011; Aliero 2004; Barekart 2013; Muhammad and Amusa 2003), where the effect of H_2SO_4 scarification was suggested to be based on an increased seed permeability to water. Therefore, a reduced water uptake does not seem to be cause of germination inhibition by seed coats. Absence of germination observed in H_2SO_4 exposures longer than 5 min could be due to lesions in embryo vital parts (Aliero 2004). Mechanical scarification through sanding one end or both sides of the seed, only increased slightly germination of seeds incubated at 15 C in darkness in comparison to the untreated seed of the negative control (15 C + darkness) ($p < 0.0001$) (Figure 4b and 4c). Germination for seeds sanded on one end or both sides was 14 ± 6% and 9 ± 2%, respectively. Taken these results together it seems that seed coats do not inhibit seed germination by limiting water uptake and or gaseous exchange.

Effect of Nitrogenous and Oxidants Compounds on Seed Germination

Incubation in KNO_3 at 15 C in darkness increased germination to values intermediate of those scored of both controls ($p < 0.0001$) (Figure 4d). Germination in KNO_3 reach a 30 ± 4% while a 0.4 ± 0.4% and 75 ± 4% were scored for negative and positive control, respectively. Likewise, germination obtained in 0.89 M and 4.45 M H_2O_2 was 65 ± 7% and 70 ± 8%, respectively, and did not differ from the positive control (15 C + light) ($P < 0.0001$) (Figure 4e). Positive effect of H_2O_2 on seed germination has already been reported for many species (Schmidt 2000). Different mechanisms were proposed for this response. Liu *et al*. (2010) indicated that presence of H_2O_2, both endogenous and exogenous, increased ABA catabolism and GA biosynthesis. This role

is consistent with our results in which addition of GA and a reduction of ABA synthesis were effectives to promote of common teasel germination.

Effect of Seed Storage on Dormancy Relief

Germination of seeds treated with cold stratification was 43 to 58%, contrasting with no germination when seeds were not stratified ($P < 0.001$) (Figure 5a). In contrast, stratified seeds for 4 and 8 weeks reached a 43 ± 11% and 58 ± 3%, respectively. Extension of the stratification period to 12 weeks lead to a sharp reduction in germination (2 ± 2%) probably due to an induction into secondary dormancy. Our results suggest that seeds would be unable to germinate, in the absence of light and or alternating temperatures, during the fall next to dispersion. So buried seeds would only be partially able to germinate at the end of winter or early spring following dispersion. Seed storage at 15 C increased germination till 37 ± 8%, 7% ± 2 and 33 ± 7% for 0, 4, 8 and 12 w of storage, respectively (Figure 5b). These results partially agree with those of Bentivegna (2008), who reported an increase in seed germination with greater period of storage.

Effect of Seed Burial Depth on Seedling Emergence

Seed burial depth greatly affected seedling emergence of common teasel ($p < 0.0001$) (Figure 6). Seedling emergence was greatest when seeds were placed on the soil surface or buried shallowly (70 ± 5%, 42 ± 11, 42 ± 8 for 0, 0.5 and 1 cm depth). In contrast, seedling emergence at deeper depths decreased till 20 ± 5, 12 ± 4 and 7 ± 3 for 1.5, 2 and 3 cm, respectively. Decreased seedling emergence due to increased burial depth

has been reported for several weed species (Benvenuti et al 2001; Boyd and Van Acker 2009). Small-seeded species may have not enough energy reserves to support elongation from deeper depths. Also, a reduction in light amount with increasing burial depth has been suggested to reduce weed emergence from greater depths (Benech-Arnold et al. 2013). Results suggest that tillage operations leading to seed burial beyond maximum depth of emergence (*i.e.* 3 cm) would minimize seedling emergence.

Results of this study indicate that Argentinean common teasel population has physiological dormancy. Light and / or fluctuating temperatures were effective to trigger seed dormancy breakage. The presence of TE reduced germination suggesting that GA have a role in dormancy breakage in common teasel. Likewise, exogenous GA and Fluridone in dark constant temperatures-treated seeds, increased germination proposing that dormancy is under hormonal control. Dry after-ripening and cold stratification were effective to release seeds from this state. Excision of embryos from seeds induces fully germination at constant temperature in darkness. This result together with the increased germination after addition of GA confirms that the type of dormancy in common teasel is physiological dormancy. This result was supported by the effect of H_2O_2 on germination. Physiological effect of this compound was related to an increase of ABA catabolism and GA biosynthesis being consistent with the observed effect of GA addition and ABA reduction on common teasel germination.

Results presented here proposed that, as for other weed species that require light or fluctuating temperatures to germinate, defoliation patterns selected for pasture management would prevent the fulfillment of both germination requirements by common teasel seeds. For instance, by maintaining some residual leaf area after mowing to reduce both the thermal fluctuations and the light reaching the soil surface. Once common teasel

seeds loss its dormancy, and consequently its germination requirements, the presence of a close canopy will reduce possibility of common teasel seedling establishment due to its small seedling size. Also, seedling emergence was optimal at shallow burial depths, indicating that farming practices that bury seeds beyond the maximum zone of emergence would minimize seedling emergence. Inferences thought from obtained results should therefore be tested in field experiments.

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Figure legend

Figure 1. Effect of temperature, light and incubation media on germination of common teasel seeds. TE is a gibberellins synthesis inhibitor (see Materials and Methods). T bars indicate the SE. Different letters at the top of the bars indicate significant differences according Tukey's Test (⟨ = 0.05).

Figure 2. (A) Germination of common teasel seeds exposed to constant 15 C + dark + different GA_3 concentrations (black columns); fluctuating temperatures (20/10 C) + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). (B) Germination of common teasel seeds exposed to constant 15 C + dark + different Fluridone concentrations (black columns); fluctuating temperatures (20/10 C) + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). T bars indicate the SE. Different letters at the top of each bar indicate significant differences according Tukey's Test ⟨ = 0.05).

Figure 3. Germination of common teasel seeds and isolated embryos exposed to 15 C + dark (black columns) and constant 15 C + light (white column). T bars indicate the SE. Different letters at the top of each bar indicate significant differences according Tukey's Test (P = 0.05).

Figure 4. (A) Germination of common teasel seeds exposed to constant 15 C + dark + different H_2SO_4 exposure times (black columns); constant 15 C + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). (B) Germination of single and double side scarified common teasel seeds exposed to constant 15 C + dark + H_2O (black

columns); constant 15 C + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). (C) Germination of both sides scarified common teasel seeds exposed to constant 15 C + dark + H_2O (black columns); constant 15 C + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). (D) Germination of common teasel seeds exposed to constant 15 C + dark + KNO_3 (black columns); constant 15 C + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). (E) Germination of common teasel seeds exposed to constant 15 C + dark + different H_2O_2 concentrations (black columns); constant 15 C + dark + H_2O (black column); and constant 15 C + light + H_2O (white column). T bars indicate the SE. Different letters at the top of each bar indicate significant differences according Tukey's Test (p = 0.05).

Figure 5. (A) Germination of common teasel seeds exposed to different cold stratification (6 C) weeks durations + constant 15 C + dark + H_2O (black columns) (B). Germination of common teasel seeds exposed to different dry after ripening (20 C) weeks durations + constant 15 C + dark + H_2O (black columns). T bars indicate the SE.

Figure 6. Depth of emergence of common teasel seedlings. Data points are the means.T bars indicate the SE.

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Figure 1

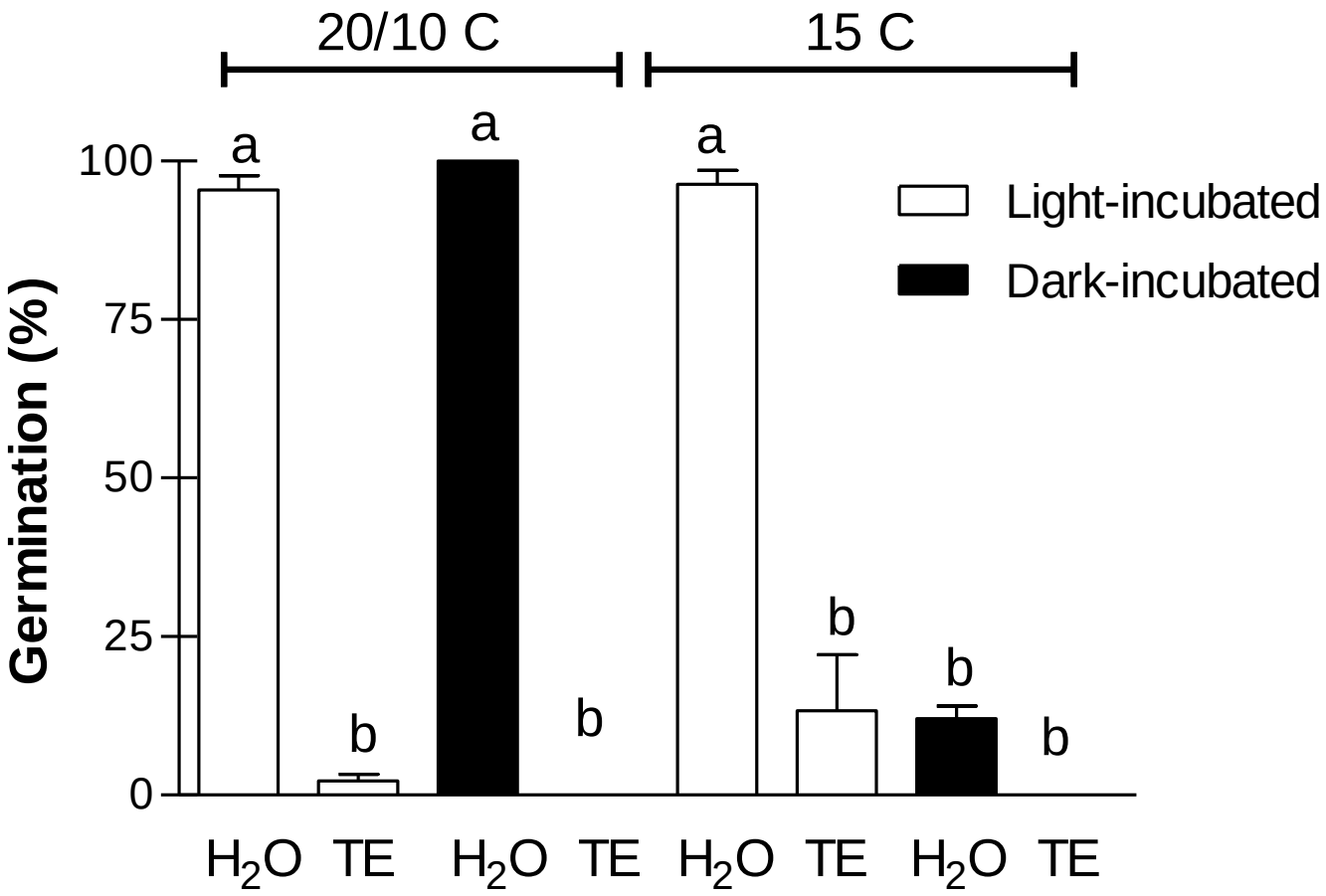

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Figure 2

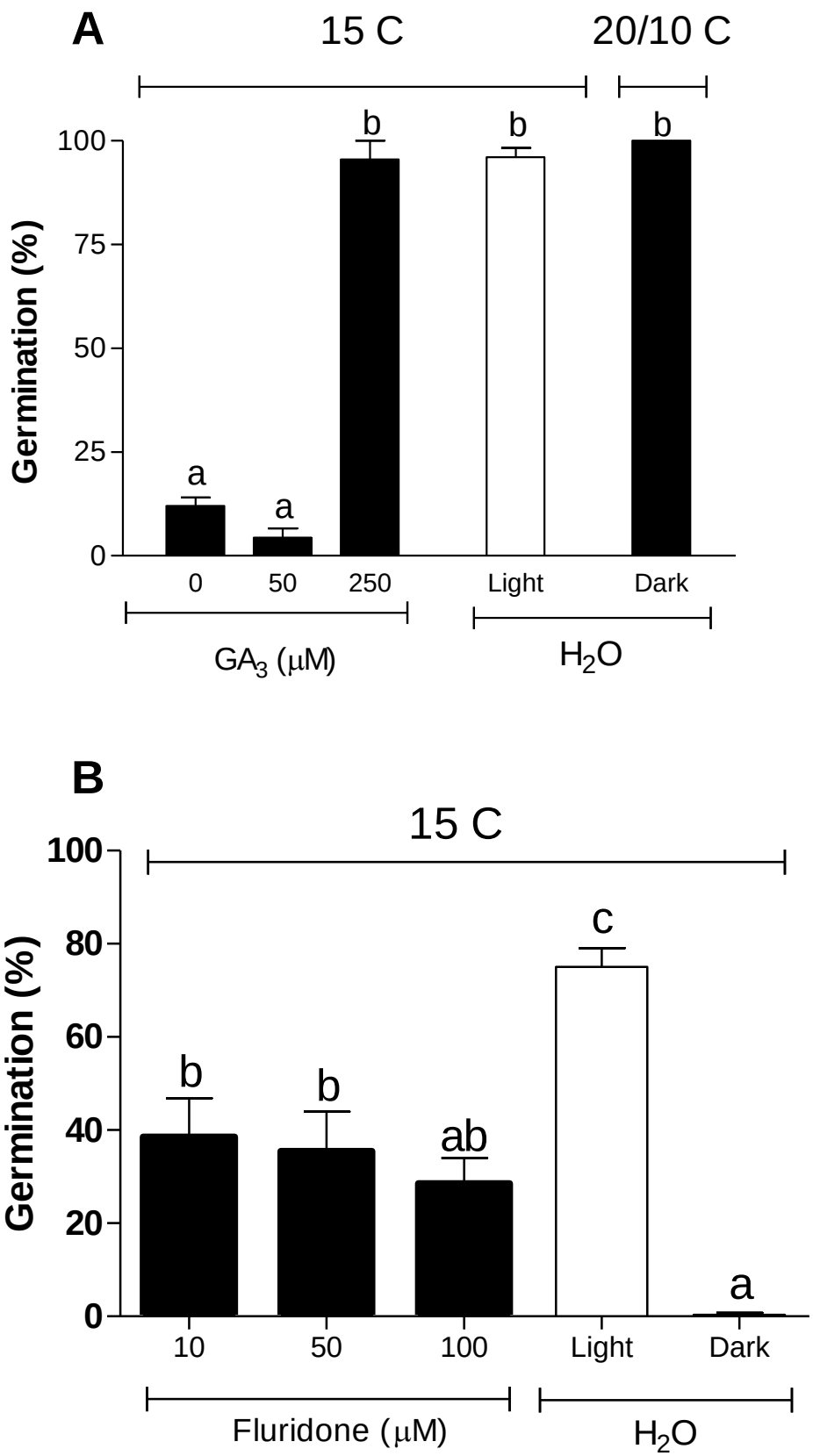

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Figure 3

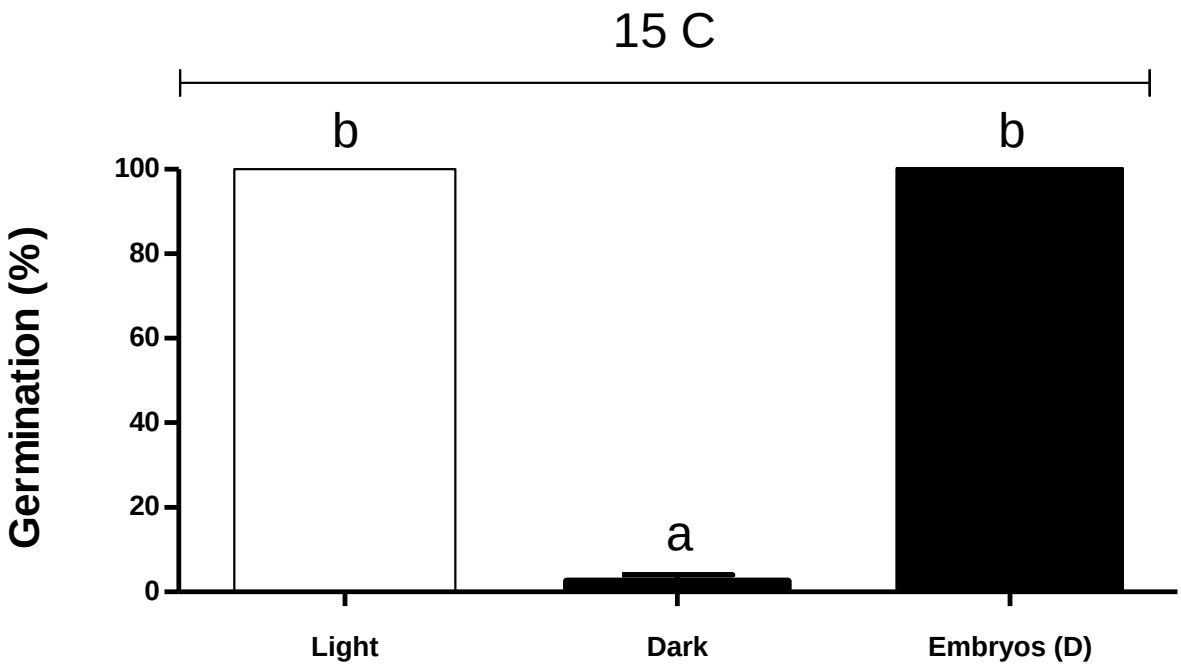

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Figure 4

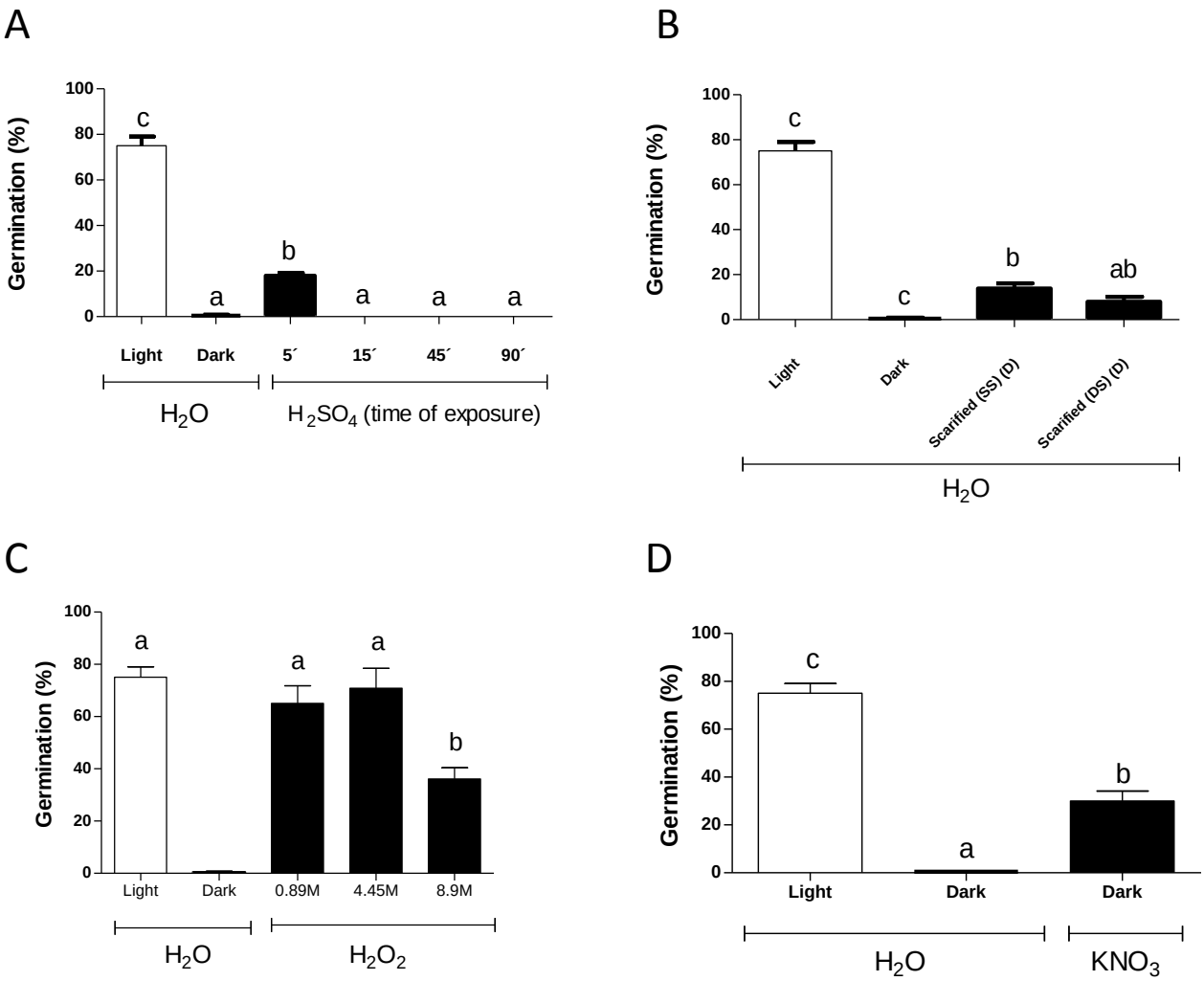

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Figure 5

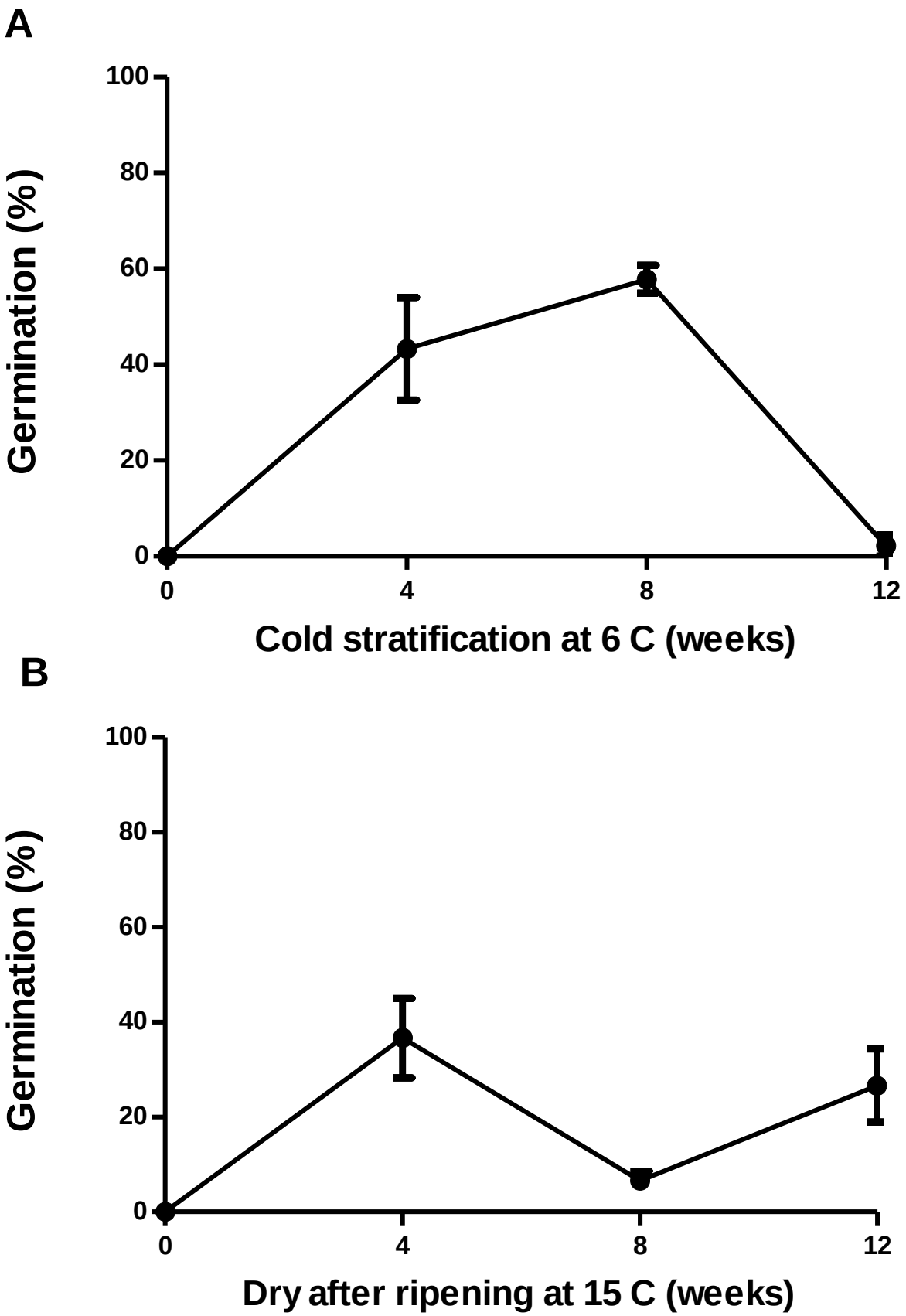

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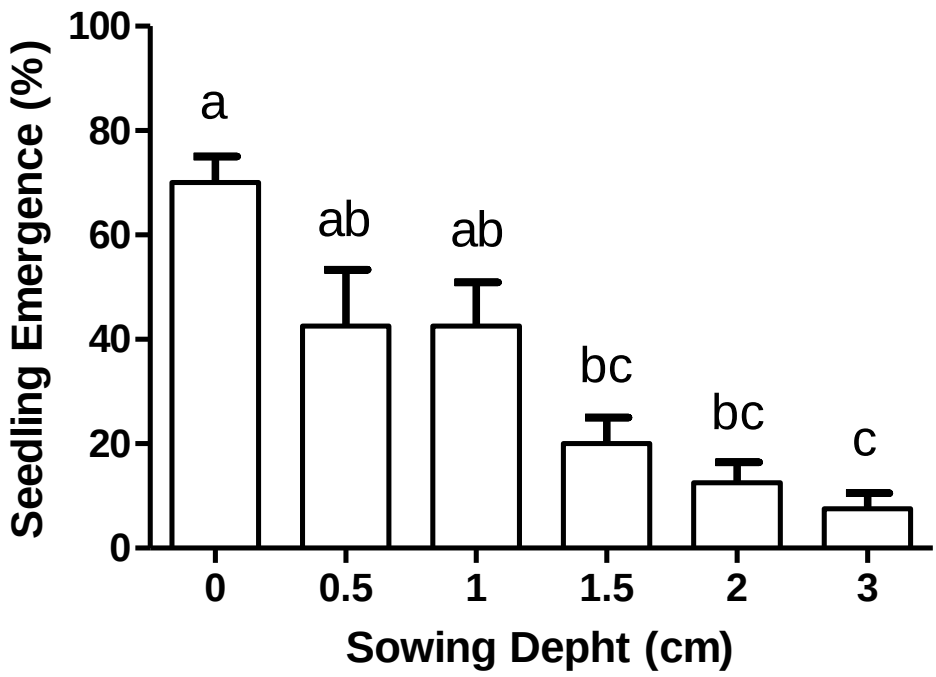
Figure 6



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