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Environmental Influences on Seed Germination and Seedling Emergence in Four *Echinochloa* Taxa

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Abstract: *Echinochloa* species are troublesome weeds that cause serious problems in paddy fields. Due to the fact that the genus *Echinochloa* comprises numerous species and subspecies, research on its seed germination and emergence ecology is still insufficient. In this study, the influence of varying temperatures; light, osmotic, and saline conditions; and depth of seed burial on the germination of *Echinochloa* seeds and the emergence of seedlings was determined through laboratory and pot tests: *E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis*, *E. crus-galli* var. *praticola*, and *E. caudata*. Seed germination of the constant temperatures in the four *Echinochloa* taxa was examined between 15 and 40 °C, and optimum germination occurred over the following temperature ranges: 25–35 °C for *E. crus-galli* var. *crus-galli*; 15–25 °C for *E. crus-galli* var. *mitis*; 15–40 °C for *E. crus-galli* var. *praticola*; and 15–35 °C for *E. caudata*. Fluctuating temperatures were conducive to seed germination in all four *Echinochloa* taxa. Except for *E. crus-galli* var. *crus-galli* and *E. crus-galli* var. *mitis* exposed to very acidic conditions (pH = 4), germination of seedlings from the four taxa of *Echinochloa* was not evidently affected by pH or light. Seed germination of the four *Echinochloa* taxa decreased as water stress decreased (<−0.2 MPa); however, it occurred across a wide spectrum of salt concentrations (0–320 mM NaCl). The seeds that were placed 0–0.5 cm below the surface had the highest rate of seedling emergence, which declined gradually as burial depth increased. This result demonstrates that deep tillage is an efficient management method for decreasing the seedling emergence of various weed species. This study's findings will enhance our comprehension of the conditions necessary for the germination and emergence of *Echinochloa* seeds, as well as furnish information that may assist in managing its growth.

Keywords: seed germination; seedling emergence; temperature; salinity and osmotic stress; burial depth

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1. Introduction

Echinochloa belongs to the Poaceae family, subfamily Panicoideae. Approximately 50 species of this genus are found in agroecosystems across various tropical and temperate habitats, particularly in rice paddies [1]. Rice serves as the primary food source for more than 65% of the population in China and holds an important role in the country's food security [2]. In China, direct-seeded rice is gaining popularity for its labor-saving benefits, whereas seedling-transplanted rice is utilized in regions with advanced mechanization. In rice-growing areas, *Echinochloa* species have historically been viewed as some of the most troublesome weeds [3], leading to considerable reductions in yield because of their competitive advantage over rice [4]. Llewellyn et al. [5] reported that the annual cost associated with the management and yield loss of *Echinochloa* species in Australia was estimated to be AU $14.6 million.

Echinochloa species exhibit a wide range of morphological, physiological, and biological variations [6]. In China, six *Echinochloa* species cause significant threats to rice cultivation: *E. crus-galli*, *E. glabrescens*, *E. oryzoides*, *E. colona*, *E. cruspavonis*, and *E. caudata* [7]. Furthermore, *Echinochloa crus-galli* comprises the varieties var. *crus-galli*, var. *zelayensis*, var. *mitis*, var. *austrojaponensis*, var. *breviseta*, and var. *praticola* [8]. The characteristics of the spikelets allow for identification of morphological differences between various *Echinochloa* species [9]. In addition, incorporating data from molecular markers, along with morphological characterization, is regarded as an effective approach for identifying *Echinochloa* species [10,11]. For weed management strategies to be successful and long-lasting, an understanding of weed biology is essential [12,13]. Damalas et al. [14] reported that the major *Echinochloa* species in northern Greece differ markedly in both structure and physiology, including factors such as seed germination, growth variability, herbicide susceptibility, and antioxidant enzyme activity. *Echinochloa* weed species' seed germination and seedling emergence might differ depending on environmental factors including soil moisture, light, and seed burial depth. It was shown [15] that *Echinochloa crus-galli* seeds germinated more readily than those of *Echinochloa colona* in the absence of light. Furthermore, *E. colona* seedlings emerged less frequently than those of *E. crus-galli* at similar planting depths. Another study highlighted that the rate of seedling emergence in *E. crus-galli* var. *oryzicola* was markedly higher than that of *E. crus-galli* var. *crus-galli* at every burial depth tested [16].

Information on the conditions influencing the germination/emergence of *Echinochloa* species is very important for weed control measures and prediction of possible distribution areas of these weeds; however, minimal research has been conducted to date. Chauhan and Johnson [17] stated that *E. colona* seed germination was not affected by changing either day/night temperatures (25/15, 30/20, and 35/25 °C) or soil pH values between 4 and 9. However, salinity levels greater than 50 mM NaCl and osmotic potentials less than −0.2 MPa significantly reduced germination. Light exposure proved beneficial for germination, with the highest emergence following planting at 0.2 and 0 cm depths. The values of salinity and osmotic potential that reduced germination by 50% were determined to be 254–313 mM and −0.33 to −0.24 MPa, respectively [18].

Therefore, this study aimed to determine how various environmental conditions, such as light exposure, pH, saline stress, temperature, osmotic conditions, and planting depth, affected seed germination and the emergence of seedlings. It was conducted in China on four taxa of *Echinochloa*—three different *E. crus-galli* varieties *(E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis* and *E. crus-galli* var. *praticola*) and *E. caudata*—collected from rice crop fields.

2. Materials and Methods

2.1. Seeds

The seeds from four *Echinochloa* taxa (*E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis*, *E. crus-galli* var. *praticola,* and *E. caudata*, as shown in Figure S1) used in this experiment were obtained from several neighboring paddy fields in Fengxian district (30°53′59″ N, 121°23′16″ E), Shanghai City, in 2017. The seeds were cleansed and air-dried at ambient temperature after collection. They were later preserved in paper packets at 4 °C until they were used in further experiments.

2.2. Germination Test Protocol

In 2018, experimental studies were conducted in laboratory and greenhouse settings at the Eco-Environmental Protection Research Institute of the Shanghai Academy of Agricultural Sciences (30°57′15″ N, 121°28′31″ E). For the purpose of germination analysis, seeds were arranged in 9-cm Petri dishes (10 per dish) and overlaid with filter paper (2 layers of Whatman No. 1) saturated with either 5 mL of water from a distillery or appropriate test solutions. The

dishes were incubated at 30 °C with maintained temperature and with alternate 12-h light and dark cycles in a growth cabinet (MLR-352H, Sanyo Electric, Osaka, Japan). The fluorescent lamps generated a photosynthetic photon flux density of 150 μmol m^{-2} s^{-1}. To keep the paper saturated, more water was applied as required. Germination progress was noted down daily for 14 days, ending when no new seeds germinated. Germination was recognized by the appearance of a visible radicle (≥2 mm) [19]. The germination rates were calculated by evaluating the proportions of seeds showing successful germination. Each species included four randomly selected replicates (dishes). The experiment was conducted twice, maintaining consistency in the procedure each time. The first trial was conducted from 7 June to 21 July 2018. And the second trial was repeated from 15 August to 30 September 2018.

2.3. Temperature

The *Echinochloa* seeds were tested by placing dishes in a growth cabinet with both constant and variable temperatures. Seven constant (10, 15, 20, 25, 30, 35, and 40 °C) and five variable (10/15, 15/20, 20/25, 25/30, and 35/40 °C) temperature patterns were chosen. A 12-h light period was maintained, with the higher temperature in the fluctuating cycles coinciding with the light period. The remaining environmental factors were identical to those of the germination experiments.

2.4. Light

To assess light affecting germination of *Echinochloa* seeds, Petri dishes were placed under varying light conditions, including 24-h darkness, 24-h continuous light, and alternating light/dark cycles of 6/18, 12/12, and 18/6-h. The remaining environmental factors were identical to those of the germination experiments.

2.5. pH

Germination was assessed using pH 4, 6, 8, and 10 buffers, as described [20]. The pH 4 buffer contained 2 mM potassium hydrogen phthalate with 1 M hydrogen chloride (HCl), the pH 6 buffer was composed of 1 M sodium hydroxide (NaOH) and 2 mM MES, the pH 8 solution was made by combining 2 mM HEPES with 1 M NaOH, and the pH 10 solution combined 2 mM Tricine with 1 M NaOH. The control was deionized water (pH 6.86). The pH was measured using a pH meter (PB-21, Sartorius, Gottingen, Germany). The remaining environmental factors were identical to those of the germination experiments.

2.6. Salinity

Solutions with varying NaCl contents (0, 10, 20, 40, 80, 160, and 320 mM) were utilized for seed incubation. In accordance with the germination test protocol, other environmental conditions were kept the same.

2.7. Osmotic Potential

Solutions with 0, −0.1, −0.2, −0.4, −0.6, −0.8, and −1.0 MPa osmotic potentials were obtained by dissolving 0, 93.6, 132.4, 187.2, 229.2, 264.7, and 295.9 g of PEG 8000, respectively, in 1 L of distilled water [21]. The remaining environmental factors were identical to those of the germination test.

2.8. Burial Depth

Plastic containers (diameter, 12 cm; height, 21 cm) were used to plant thirty seedlings at depths of 0, 0.5, 1.0, 2.0, 4.0, 6.0, 8.0, 10.0, or 12.0 cm below the soil's surface. The soil (pH 6.9) contained 2.0% organic matter, and the soil texture was silt loam (35% sand, 40% silt, 25% clay). The experiments were performed in a growth cabinet with a 12-h light cycle, with the light and dark temperatures set to 30 °C and 25 °C, respectively. The containers

were watered as necessary to maintain moisture. The experiment terminated when no new seedlings emerged, and emergence was defined by the visible appearance of cotyledons.

2.9. Statistical Analysis

To guarantee the homogeneity of variances, the final germination percentages were arcsine square root transformed before the analysis. However, Bartlett's test revealed that the variance homogeneity did not benefit from the arcsine square transformation. The results of multiple experimental repeats were aggregated and analyzed using ANOVA, which revealed no significant interaction between trials and treatments ($p < 0.05$). Means were compared using Duncan's multiple-range test, with $p < 0.05$ considered significant. The duration required to achieve 80% germination (t_{80}) at various temperatures was determined using the following formula:

$$t_{80} = \left(H_p - L_p\right)^{-1} + L, \quad (1)$$

where L refers to day before achieving 80% germination, L_p represents the observed percentage germination on day L, and H_p denotes the observed percentage germination on the day when germination achieved or surpassed 80%.

The germination and emergence results at varying levels of salt concentration, osmotic potential, and burial depth were fitted to a three-parameter logistic model utilizing SigmaPlot (version 12.5). The fitted model is as follows:

$$G(\%) = G_{\max} / \left[1 + (x/x_{50})^{G_{\text{rate}}}\right], \quad (2)$$

where variable $G(\%)$ stands for the overall percentage germination or emergence at a specific value of salt content, osmotic potential, or soil depth (x). G_{max} is the highest recorded percentage germination or emergence, whereas x_{50} signifies the salt content, osmotic potential, or soil depth that reduced the maximum by 50%. The slope of the equation is represented by G_{rate}.

3. Results and Discussion

3.1. Influence of Temperature on Seed Germination

Under constant temperature conditions, the germination of seeds from all four *Echinochloa* taxa was zero at 10 °C (Table 1). Differences, however, were seen between the taxa in the temperature required for final germination higher than 80%: 25–35 °C for *E. crus-galli* var. *crus-galli* (ECc), 15–25 °C for *E. crus-galli* var. *mitis* (ECm), 15–40 °C for *E. crus-galli* var. *praticola* (ECp), and 15–35 °C for *E. caudata* (EC) (Table 1). At 40 °C, the germination rate of *E. crus-galli* var. *crus-galli* (ECc) was only 7.5%, whereas that of *E. crus-galli* var. *praticola* (ECp) was 91.2% (Table 1). Upon lowering the temperature to a steady 15 °C, the germination rate for *E. crus-galli* var. *crus-galli* (ECc) dropped to under 35.0%, whereas those of *E. crus-galli* var. *mitis* (ECm), *E. crus-galli* var. *praticola* (ECp), and *E. caudata* (EC) were >81.2% (Table 1). Understanding the cardinal temperatures essential for germination is crucial for forecasting the distribution of plant species because exposure to temperatures that exceed the optimal germination range can adversely affect seed germination [22]. Talaee et al. [23] observed that the optimal temperature for *Amaranthus blitoides* seed germination was 35 °C, whereas *Amaranthus hybridus* exhibited optimal germination within a consistent temperature range of 30–40 °C. In all *Echinochloa* taxa, the time to germination onset (T) and t_{80} decreased as the temperature increased from 15 to 35 °C, except for *E. crus-galli* var. *crus-galli* (ECc) at 35 °C and *E. crus-galli* var. *mitis* (ECm) at 25 °C (Table 1). Zhao et al. [24] reported that raising the temperature from 5 to 20 °C had minimal influence on *Alopecurus aequalis* seed germination rates; however, it caused a progressive shortening of the time taken for germination to initiate, along with the duration required to attain 90% germination (t_{90}).

Table 1. The rate of seed germination and days to achieve 80% germination (t_{80}) for four *Echinochloa* taxa (ECc, ECm, ECp, and EC) seeds after treatment at 13 temperatures.

Temperature (°C)	*T* (d)				Germination (%)				t_{80} (d)			
	ECc	ECm	ECp	EC	ECc	ECm	ECp	EC	ECc	ECm	ECp	EC
10	ND	ND	ND	ND	0 c	0 g	0 e	0 f	NE	NE	NE	NE
15	6	5	6	6	35.0 ± 7.5 b	97.5 ± 2.5 ab	81.2 ± 3.8 d	100 a	NE	6.5	10.2	8.3
20	4	3	3	4	40.0 ± 5.0 b	96.2 ± 3.8 ab	98.8 ± 1.2 ab	100 a	NE	4.0	6.2	5.0
25	2	2	2	2	96.2 ± 3.8 a	87.5 ± 2.5 bc	100 a	97.5 ± 2.5 a	3.5	4.3	3.5	3.5
30	2	2	2	2	93.8 ± 3.8 a	77.5 de	97.5 ± 2.5 abc	95.0 ± 5.0 ab	3.0	NE	3.5	3.0
35	2	1	1	2	93.8 ± 1.2 a	73.8 ± 1.2 de	97.5 abc	88.8 ± 1.2 ab	3.5	NE	2.5	3.0
40	2	2	1	2	7.5 ± 2.5 c	33.8 ± 6.2 f	91.2 ± 1.2 abc	55.0 ± 7.5 e	NE	NE	2.5	NE
10/15	7	6	7	7	93.8 ± 3.8 a	76.2 ± 3.8 de	87.5 ± 5.0 cd	73.8 ± 3.8 cd	8.5	NE	10.2	NE
15/20	3	3	3	3	92.5 ± 2.5 a	93.8 ± 3.8 ab	96.2 ± 1.2 abc	92.5 ± 7.5 ab	6.2	4.5	5.3	6.2
20/25	2	2	2	2	96.2 ± 1.2 a	98.8 ± 1.2 a	93.8 ± 1.2 abc	91.2 ± 3.8 ab	3.5	3.0	4.3	5.2
25/30	2	1	2	2	100 a	81.2 ± 1.2 cd	88.8 ± 6.2 bcd	87.5 ± 2.5 ab	3.5	4.2	3.5	3.5
30/35	2	1	1	1	90.0 ± 5.0 a	88.8 ± 1.2 abc	95.0 ± 5.0 abc	81.2 ± 3.8 bc	3.0	2.0	2.5	6.2
35/40	2	1	1	1	87.5 ± 2.5 a	67.5 ± 5.0 e	98.8 ± 1.2 ab	62.5 ± 5.0 de	4.3	NE	2.5	NE

The data (mean ± SE) were analyzed independently, and distinct letters indicate statistically significant differences ($p < 0.05$) within the same weed species. "*T*" refers to the time it takes for germination to begin, while "t_{80}" represents the days needed to reach 80% germination. ND means "not determined" due to the absence of germination, and NE stands for "not estimated" because the germination percentage never reached 80% in all replicates. The labels ECc, ECm, ECp, and EC refer to the species *E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis*, *E. crus-galli* var. *praticola*, and *E. caudata*, respectively.

Compared to that at constant temperatures, seed germination of all four *Echinochloa* taxa was >80% during exposure to changing temperatures, with the exception of *E. crus-galli* var. *mitis* (ECm) and *E. caudata* (EC) at the least (10/15 °C) and greatest (35/40 °C) varying temperatures (Table 1). In comparison with maintaining the temperature constant at 15 °C and 40 °C, the largest increases in *E. crus-galli* var. *crus-galli* (ECc) germination rates were at the changing temperatures of 10/15 °C and 35/40 °C. These findings indicate that fluctuating temperatures may be more conducive to seed germination in the four *Echinochloa* taxa, likely because such temperature fluctuations can mimic the natural diurnal temperature variations occurring in their habitats [25]. These results are consistent with previous research, which indicated that fluctuating temperatures were effective in promoting the germination of *E. crus-galli* seed [26]. Wu et al. [27] found that fluctuating temperatures enhanced *Polypogon fugax* seed germination. Toole [28] theorized that fluctuating temperatures facilitate a proper balance and interaction of hormones, enzymes, substrates, and, possibly, preexistent active phytochrome in weed seeds.

The findings of the current study demonstrate that all the analyzed *Echinochloa* taxa exhibited germination capabilities over a broad temperature range. This adaptability suggests that these taxa are well-positioned to take advantage of the various climatic conditions found throughout the different rice cultivation phases in China, including early, middle, and late rice-cropping systems [29]. Such wide temperature tolerance enhances their potential to thrive and proliferate during these critical agricultural periods, thereby posing significant challenges for rice production management.

3.2. Influence of Light on Seed Germination

Seed germination was not influenced by photoperiod in any of *Echinochloa* taxa (Figure 1). Similar trends in seed germination between light and dark conditions have been observed in species of weed, including *Aegilops tauschii* [30] and *A. aequalis* [24], where no significant differences were noted in germination between light/dark regimes. It has been found [31] that although *E. crus-galli* and *E. colona* germination did not need light, germination was higher than in complete darkness. In some seeds, a sufficient amount of Pfr can be synthesized during seed development or upon imbibition, allowing them to germinate without additional light [32].

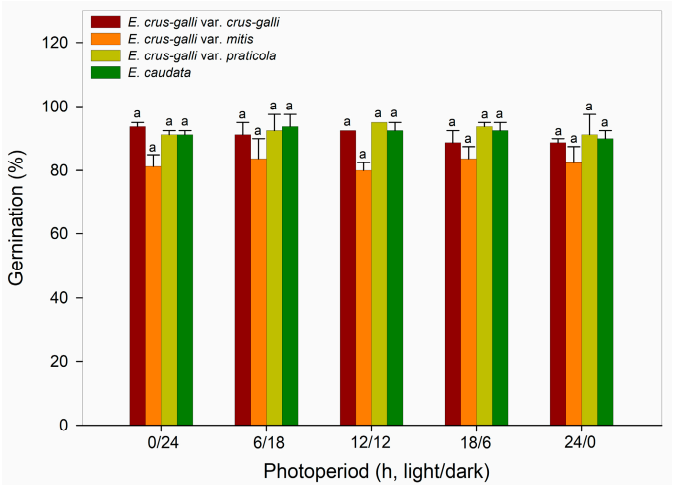

Figure 1. Impact of light on seed germination in four *Echinochloa* taxa. Data are mean values ± SE. Different letters indicate significant differences ($p < 0.05$) within the same weed species using Duncan's multiple-range test.

3.3. Influence of pH on Seed Germination

Compared with deionized water (pH 6.86), seed germination rates of the four *Echinochloa* taxa remained unaffected across a pH spectrum of 6 to 10, with rates exceeding 80% in each population (Figure 2). Likewise, numerous other weed species, such as *E. colona* [17], *Lolium perenne* [33], and *A. tauschii* [30], are capable of germinating over an extensive pH range. When exposed to a pH 4 solution, seed germination of *E. crus-galli* var. *crus-galli* and *E. crus-galli* var. *mitis* was significantly inhibited, and that of *E. crus-galli* var. *mitis* was only 23.8% (Figure 2). Wang et al. [34] also found that an alkaline environment was more appropriate for germination of *Alopecurus japoni* seeds. In contrast, *Phalaris minor*, *P. brachystachys*, and *P. paradoxa* seeds did not germinate at pH 9–11 [35]. The pH of most soils in China ranges from 5–8 [36]. Therefore, it is possible that soil pH does not restrict *Echinochloa* seed germination.

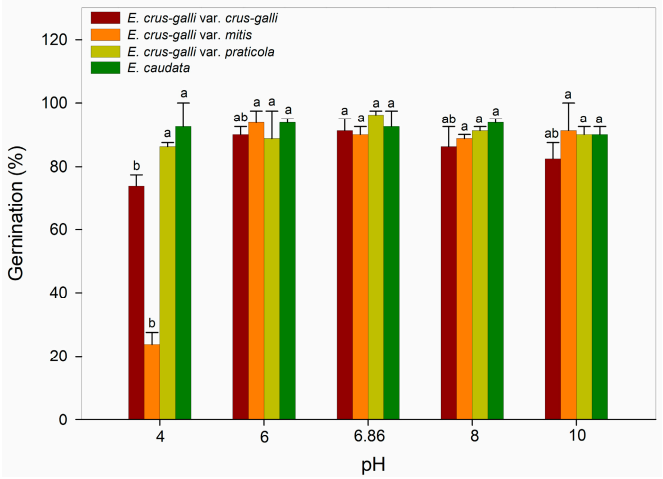

Figure 2. Impact of pH on seed germination in four *Echinochloa* taxa. Data are mean values ± SE. Different letters indicate significant differences ($p < 0.05$) within the same weed species using Duncan's multiple-range test.

3.4. Influence of Salt Stress on Seed Germination

Seed germination in the four *Echinochloa* taxa decreased with increasing NaCl concentrations (Figure 3). With x_{50} values of 241.6 mM and 225.1 mM, *E. crus-galli* var. *crus-galli* and *E. crus-galli* var. *mitis* showed similar tolerance to NaCl levels that reduced maximum germination by 50%, respectively. However, under extreme NaCl stress (320 mM), *E. crus-galli* var. *praticola* and *E. caudata* exhibited significantly higher germination rates than *E. crus-galli* var. *crus-galli* and *E. crus-galli* var. *mitis* (Figure 3). Similar weed species, including *E. glabrescens* [18], *E. colona* [17], and *Oryza sativa f. spontanea* [31], can also undergo germination in strongly saline environments. As we all know, soil salinity is becoming a primary abiotic constraint against the production of crops globally. Zhang et al. [37] found that salt stress markedly reduced rice yields and quality when salinity levels exceeded 17.1 mM NaCl. In future research, based on the high salt-tolerance characteristics of the four *Echinochloa* taxa, we could identify the relevant genes responsible for salt tolerance and utilize these genes to develop salt-tolerant rice varieties. In addition, the current study's findings indicate that controlling the spread is challenging, four *Echinochloa* taxa weeds in China and other countries through soil salinization.

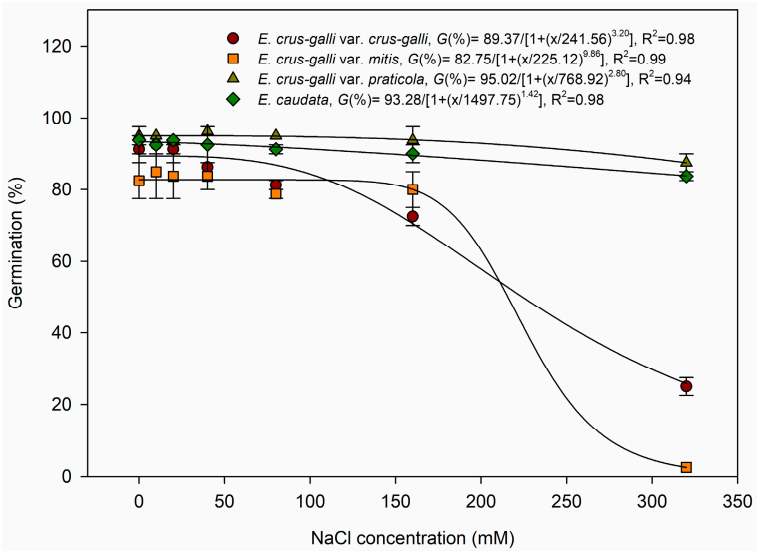

Figure 3. Impact of NaCl levels on seed germination in four *Echinochloa* taxa. Data are mean values ± SE.

3.5. Influence of Osmotic Stress on Seed Germination

Reduced osmotic potentials (from −0.2 to −1.0 Mpa) led to a gradual decline in the germination of the four *Echinochloa* taxa (Figure 4). Osmotic potentials that induced 50% suppression of maximal germination (x_{50}) in *E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis*, *E. crus-galli* var. *praticola*, and *E. caudata* were −0.53, −0.48, −0.32, and −0.67 MPa, respectively (Figure 4). Germination was 13.8% in *E. caudata* after reducing the water potential to −0.8 MPa, and was <1.3% in *E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis*, and *E. crus-galli* var. *praticola* (Figure 4). Similar germination responses were observed in *E. colona* [17] and *E. glabrescens* [18]. Osmotic potentials of −0.8 Mpa lowered the germination rates to 1% and 4%, respectively. These findings demonstrate that moist environments, such as paddy fields, are preferred by *Echinochloa* for germinating seeds.

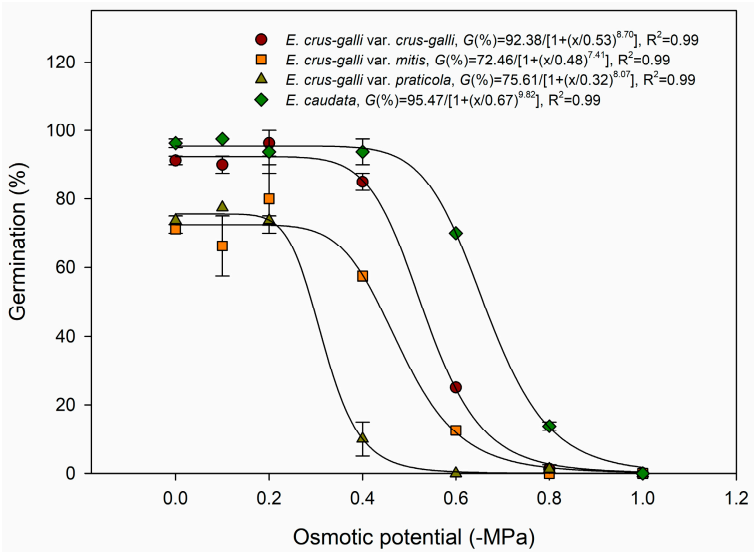

Figure 4. Impact of osmotic potentials on seed germination in four *Echinochloa* taxa. Data are mean values ± SE.

3.6. Influence of Burial Depth on Seedling Emergence

The depth of soil had a marked impact on seedling emergence in all four *Echinochloa* taxa (Figure 5). Based on the model's predictions, the depths needed to lower the maximum

emergence by 50% (x_{50}) were 8.4 cm for *E. crus-galli* var. *crus-galli*, 9.3 cm for *E. crus-galli* var. *mitis*, 2.3 cm for *E. crus-galli* var. *praticola*, and 8.0 cm for *E. caudata* (Figure 5). Studies have shown that burial depth influences seedling emergence differently across various weed species. Chauhan et al. [38] reported an absence of emergence from *Leptochloa chinensis* seeds located ≥0.5 cm below the soil surface. In contrast, *Aegilops cylindrica* seeds could emerge from soil depths of 10.2 cm [39]. The absence of light, restrictions on gas diffusion within the soil, and insufficient nutrient levels in weed seeds often hinder seedling emergence from deep soil [35,40,41]. Yan and Chen [32] reported that seeds buried underground can determine their distance from the ground by detecting the quality and intensity of light signals. Although we found that seed germination in the four *Echinochloa* taxa was not dependent on light in this study, the higher proportion of albinotic seedlings in dark condition indicated that light played an important role in the regular growth of seedlings. This finding is similar to previously reported that the budding seedlings could not reach the surface to commence photosynthesis before the energy reserves contained within the seeds were depleted when seeds were planted too deeply in the soil [42]. Thus, deep tillage may be a useful option for weed management.

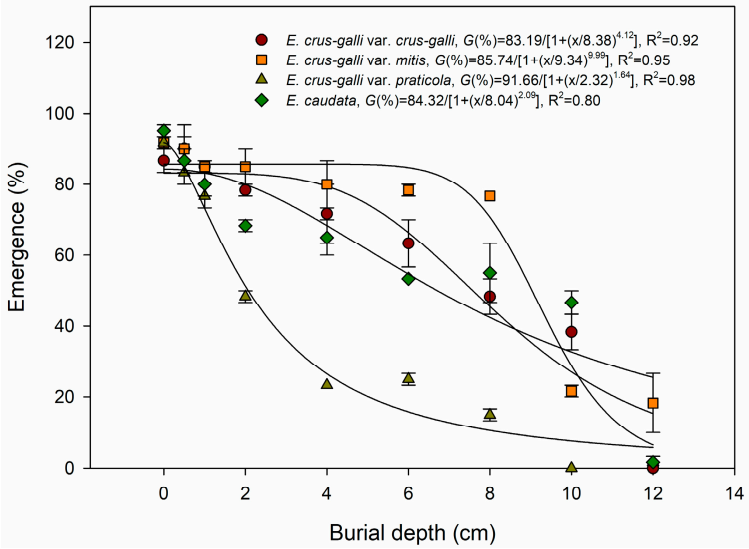

Figure 5. Influence of burial depth on seed germination in four *Echinochloa* taxa. Data are mean values ± SE.

4. Conclusions

In conclusion, the study highlights that various environmental factors affecting the germination of four *Echinochloa* taxa. The seeds of the four species were found to be able to germinate at a variety of constant temperatures, with maximum germination observed within specific temperature limits: 25–35 °C for *E. crus-galli* var. *crus-galli*, 15–25 °C for *E. crus-galli* var. *mitis*, 15–40 °C for *E. crus-galli* var. *praticola*, and 15–35 °C for *E. caudata*. Our findings indicated that fluctuations in temperature enhanced the germination of seeds in all four *Echinochloa* taxa used in the present study. Light and pH had no marked influence on germination in the four *Echinochloa* taxa, except for *E. crus-galli* var. *crus-galli* and *E. crus-galli* var. *mitis*, exposed to highly acidic conditions. Seed germination of the four *Echinochloa* taxa decreased as water stress decreased. However, it has occurred across a wide spectrum of salt concentrations. Seeds of the four *Echinochloa* taxa were vulnerable to low water potential, and their distribution was restricted to moist soils. Seedling emergence of the four *Echinochloa* taxa was greatest from seeds on soil surfaces, with emergence reducing with increasing soil depth, indicating the effectiveness of deep tillage for controlling these

weed species. The results of this research will offer valuable insights into the false seedbed method, which decreases the weed seed reservoir in the upper layer of soil and consequently lessens the competition posed by annual weeds in the following crop. Meanwhile, the future studies could focus on the mechanistic analysis of why different *Echinochloa* taxa respond variably to environmental factors. Potential limitation of this study is the relatively small number of seeds used for testing each factor, but it still provides important data and we hope that this study can provide a reference for future research.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/agronomy15020401/s1: Figure S1: Photos of four *Echinochloa* taxa: *E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *mitis*, *E. crus-galli* var. *praticola*, and *E. caudata*.

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