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INFLUENCE OF ENVIRONMENTAL FACTORS ON SEED GERMINATION AND SEEDLING EMERGENCE OF YELLOWTOP (*Flaveria bidentis*)

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ABSTRACT

Laboratory experiments were conducted in Beijing from 2008-2010 to determine the effects of light, temperature and planting depth on Flaveria bidentis germination and emergence. F. bidentis had positively photoblastic seeds which required light in germination. Seed germination had a wide temperature range, 15°C to 40°C, with the optimum temperatures from 22.5°C to 35°C The threshold temperature for germination was 14.7°C and the effective accumulated temperature required to reach 90% germination was 40.4°C. Germination was increased by 12% to 100% when the temperature was from 12.5°C to 20°C. Seedling emergence was the highest on the soil surface, and no seedlings emerged from a soil depth of 1 cm or deeper. The information gained from the study could help predict its potential distribution area and facilitate the development of effective weed control strategies.

Key words: *F. bidentis*, light, temperature, burial depth.

INTRODUCTION

Yellowtop (*Flaveria bidentis* (L.) Kuntze), a member of the Asteraceae family, is an invasive weed native to North America (Powell, 1978), which was first observed in China in 2003 (Gao *et al*., 2004; Liu, 2005). At a surprising rate, the species has now spread to 84 counties of the middle south of Hebei province, five districts of Tianjin and three counties in Henan and Shangdong province. Invasion of diversified habitats in these areas, such as roadsides, transit points, construction waste grounds, wastelands, urban green spaces and ditch edges, has been attributed to its ecological adaptations, growth plasticity and competitiveness. It is worrying that *F. bidentis* has invaded farmlands adjacent to roads, and its rapid growth and high reproduction coefficient allow the weed to be a good competitor against crops. In addition, aqueous extracts of *F. bidentis* were documented to reduce germination of more than 20 plant speices, so the presence of the weed may therefore influence the growth of certain plants other than competition (Li *et al*., 2007). Consequently,

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invasion and colonization of *F. bidentis* in North China should be highly monitored for its widespread and distribution may pose a potential threat to ecosystem and agricultural production.

Seed germination is one of the most critical events for the success of any weed because it represents the first stage at which the weed can compete for an ecological niche (Forcella *et al*., 2000), and is mediated by various environmental variables such as temperature, light, pH, soil salinity, and moisture (Chachalis and Reddy, 2000; Chauhan *et al*., 2006). Acquiring the germination characteristics provides a biological basis for the spread and establishment of *F. bidentis* to allow better timing of weed control treatments and facilitate development of agronomic practices that discourage weed establishment in crops.

The present study was designed to examine the effects of constant and alternating temperature regimes, light and burial depth on seed germination and seedling emergence of *F. bidentis*. The information gained from the study could help understand current distribution, predict its potential spread area and facilitate the development of effective weed control programs.

MATERIAL AND METHODS

Seed collection

Experiments were conducted in 2008 and 2009 at Institute of Plant Protection, Chinese Academy of Agricultural Sciences. Seeds used in the experiments were collected in late October 2007 from wastelands in Handan City, Hebei Province, China. After air-drying, seeds were stored at room temperature (20 to 25^{o}C) until experiment initiation. The 10,000-seed weight of *F. bidentis* was 1.50 to 1.97 g.

General germination tests

F. bidentis germination was evaluated by placing 100 seeds evenly in a 7.5-cm- diameter petri dish containing two layers of Whatman No.1 filter paper, moistened with either 3 ml of deionized water or a treatment solution. Dishes were sealed with Parafilm and placed in a growth chamber set at 30^{o}C (temperature determined to be optimum for germination), except for the temperature experiment. The photoperiod was set at 12 h with fluorescent lamps used to produce a light intensity of 12000 Lux. A seed was characterized as germinated when the radicle was the same as seed length, while the cotyledon was equal to one half of seed length. Germination percentage was calculated as the total number of seeds germinated divided by the total number of seeds in each replication.

Effect of light

Seed germination was studied under 12/12, 0/24, and 24/0 h light/dark regimes at 30^{o}C. Petri dishes assigned to the dark treatment were wrapped in two layers of aluminum foil to ensure no light penetration; other treatments were left uncovered to allow continuous light exposure (12000 Lux light intensity). Germinated seeds were counted after 5 d of incubation.

Effect of temperature

F. bidentis seeds were placed in petri dishes and incubated under constant (12.5, 15, 17.5, 20, 22.5, 25, 27.5, 30, 35, 40, 45°C) or fluctuating temperatures (15/10, 20/5, 20/10, 20/15, 25/5, 25/10, 25/15, 30/10, 30/15, 30/20, 35/10, 35/15°C day/night). Photoperiod was set at 12 h to coincide at high temperature. Seed germination was assessed every 12 h by counting and removing germinated seeds. The seed were kept hydrated by adding distilled water as needed to avoid a moisture effect.

Data about time to onset of germination and final germination percentages were used to compute the days required to reach 90% germination as overall germination(t90) (Ramon *et al*., 2004) and germination rates (V). The intercept (C, germination threshold temperature) and slope (K, effective accumulated temperature required to reach 90% germination) were calculated by the method of least squares (Cai *et al*., 2001) based on the effective accumulated temperature principle. Equations are as follows:

$$t_{90} = (H\,p - L\,p)^{-1} + L$$
$$V = 1 / t_{90}$$
$$K = (n\Sigma VT - \Sigma V\Sigma T) / [n\Sigma V^2 - (\Sigma V)^2]$$
$$C = (\Sigma V^2\Sigma T - \Sigma V\Sigma VT) / [n\Sigma V^2 - (\Sigma V)^2]$$

where L is the last day before 90% germination was reached. Lp is the observed germination percentage on day L, Hp is the observed germination percentage on the day when germination reached or exceeded 90%, T is the tested temperatures, and n is the value of temperature.

Effect of seed burial depth

Twenty seeds were planted in soil passed through a 2-mm sieve in 15-cm-diameter by 20-cm-deep plastic pots at depths of 0, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1,2,3,4 and 5 cm in each of four replications. For all treatments, the uppermost layer of soil was leveled and then pressured with the same force before and after seed placement to standardize depths. Pots were irrigated from the bottom as needed to maintain adequate soil moisture. Seedling emergence was counted every 5 d for 25 d.

Statistical analyses

All experiments were conducted in a randomized complete-block design, and each experiment was repeated at least twice. Effect of temperature on seed germination was replicated six times, and the other experiments were four replicates. Significant differences among treatments were identified with the use of Duncan's multiple-range test (P＝0.05). Regression analysis was used to determine the effect of salinity on germination. Data m*et al*l normality conditions; therefore, data transformation was not required. Means were separated using standard error of mean. All statistical analyses were performed using SPSS.

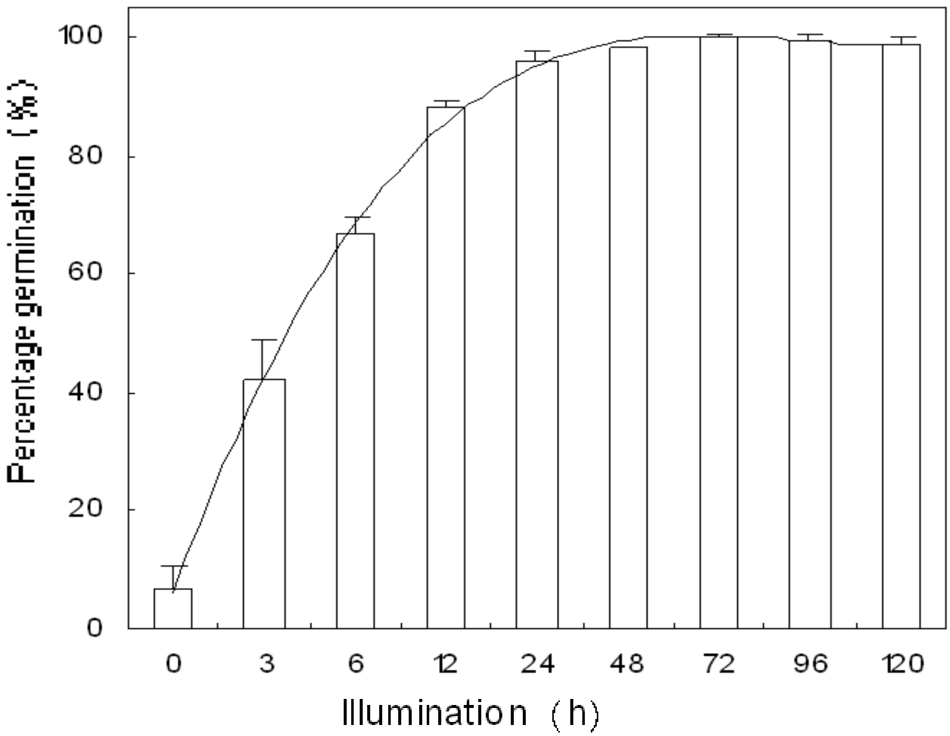

Figure 1. Germination of *F. bidentis* in illumination and darkness.

RESULTS AND DISCUSSION

Effect of light

Light had an impact on germination. There were no differences (P=0.05) in germination percentage between seed placed in a 12-h photoperiod and seed exposed to continuous light, which was significantly higher than the germination of complete darkness (Fig. 1).

It is reported that seed germination response to light is species-specific. Seeds of some species germinate equally in light and dark (Norsworthy and Oliveira 2005; Zhou *et al*. 2005; Wei *et al*.

2009); some require light to stimulate germination (Lu *et al*. 2006; Chauhan and Johnson 2008); others are favored by darkness (Chauhan *et al*. 2006). Our results demonstrated that seeds of *F. bidentis* were positively photoblastic and light stimulation was necessary for seed germination, whereas no or few seeds germinated in the dark.

Effect of temperature

Germination percentages differed significantly among constant temperature regimes (P=0.05) (Table-1). In general, germination increased from 2.7 to 99.3% between 15 and 22.5^oC; it was greater than 97% over the temperature range of 22.5^oC to 35^oC with no significant differences (P=0.05), and then dropped with increased temperature to 19% at 40^oC with obvious radical length reduction. No germination occurred at either 12.5^oC or 45^oC (data not shown).

Time to onset of germination gradually decreased as the temperature rose from 15^oC to 35^oC. For example, when exposed to constant temperature regimes of 22.5, 25, 27.5, 30 and 35^oC, seed germination began and final germination percentage was greater than 90% after 36 to 60 h of incubation and 2 to 6 d of incubation, respectively. Together, the optimum germination temperature was between 22.5 and 35^oC. The threshold temperature for germination was 14.7^oC and the effective accumulated temperature required to reach 90% germination was 40.4^oC.

The final germination percentages and germination rates of all the constant temperature treatments was compared with the germination of all the alternating temperature treatments (Table 1). In comparison, germination of alternating temperatures increased significantly (P=0.05) in response to mean temperatures of 12.5 to 20^oC; constant temperature treatments and alternating temperature treatments with 22.5^oC and 25^oC mean temperatures had no significant effect on seed germination (P=0.05); germination was greater than 94.0% at mean temperatures ranging from 15 to 25^oC, which indicated that *F. bidentis* seeds had a broad temperature range for germination.

Increasing the mean temperature from 12.5^oC to 25^oC reduced time to onset of germination and time to 90% germination (t_{90}). Temperature alternation not only stimulated *F*. *bidentis* germination but also reduced the mean temperature required to promote the same germination as the maximum germination shown by treatments with constant temperature. Increased amplitude of the diurnal temperature alternation increased percent germination as well as germination rate, and this was more evident at the lower temperature of 12.5^oC.

Table-1. *F. bidentis* seed germination for 30 d in alternating and constant temperatures (C).

Temperatures	Time to onset of germination (d)	Germination (%)	t_{90}(d)
12.5	/	0.0 f	NE
15/10	13	19.8±6.9 e	NE
20/5	6	93.0±2.1 b	29.11
15	7	2.7±1.1 f	NE
20/10	6	97.0±1.0 ab	26.15
25/5	4	94.0±1.4 ab	13.43
17.5	4	38.8±3.0 d	NE
20/15	4	98.0±0.5 ab	24.20
25/10	4	98.2±0.5 ab	6.11
20	3	85.5±1.9 c	NE
25/15	3	99.0±0.5 ab	5.06
30/10	2.5	97.2±0.7 ab	4.64
22.5	2.5	99.3±0.3 a	5.95
30/15	2.5	99.5±0.3 a	4.04
35/10	2.5	98.5±0.7 ab	3.65
25	2	99.7±0.2 a	3.57
30/20	2	99.2±0.3 ab	3.58
35/15	2.5	98.7±0.5 ab	3.02

Notes: "/" indicated no seed germinated.

Temperature is an important factor in seed germination. *F. bidentis* seed germination had a wide optimum temperature range and the germination threshold temperature was 14.7°C, partially explaining why the invasive weed may emerge throughout the season from the first ten days of April through mid-October provided soil moisture is favorable in North China. Consequently, it is vital to establish a season-long control system. The low but insignificant germination, particularly at the low temperatures (15°C), may be biologically significant. Even an individual plant, if uncontrolled, is capable of producing approximately 360, 000 seeds to the soil seed bank (Li *et al*., 2006).

The stimulation in germination with alternating temperatures had been also reported in other studies (Ramon *et al*., 2004; Steckel *et al*., 2004). Moreover, the ability of *F. bidentis* seeds to germinate quickly on account of temperature alternation would be of benefit for rapid seedling establishment in the field following summer rainfall events prior to soils becoming dry. So the greater response to warm fluctuating temperatures may suggest greater emergence of these species on bare ground, where the greatest diurnal fluctuations would be expected.

Effect of seed burial depth

Seedling emergence decreased dramatically with increasing planting depth. Emergence was at its maximum for seeds planted on the soil surface with the emergence rate of 96%; emergence fell sharply even with shallow burial (data not shown). No seedlings emerged from seed buried at 1 cm. These results agree with that of the light study, in which *F. bidentis* seeds had a light requirement for germination. Benvenuti and Macchia (1995) reported that light penetration fell below 0.01% at a depth of no more than 4 mm and that with increasing soil depth, light permeability was proportional to wavelength, leading to progressive decline in the red-far red ratio. The reason of no seedlings emerged from seeds placed at a depth of 1 cm was in part that very little light reached at that depth or the shortwave lights required for emergence were filtrated. Further research will need to focus on effect of light quality on seed germination. In addition, emergence from different soil depths has been found to be positively proportional to seed energy reserves (Benvenuti *et al*. 2001; Li *et al*. 2004; Zheng *et al*. 2006). The 10,000-seed weight of *F. bidentis* was only 1.50 to 1.97 g, so the species might not have enough energy to emerge from deeper soil layers. Furthermore, physical obstacles from soils and decreasing thermal fluctuation (Kegode *et al*. 1998) are responsible for depth inhibition. The observed preference for seed germination from the soil surface could result in greater abundance of *F. bidentis* under no-till farming systems. In these systems, the weed seed bank would be concentrated on or near the soil surface (Clements *et al.* 1996). In our study, seedling emergence on the soil surface was slightly lower than germination observed in petri dishes in the light. This difference could be due to lower soil-to-seed contact and water availability on the soil surface than on the filter paper (Ghorbani *et al*., 1999).

Inferences drawn from the results of this study should be limited to the weed population sampled because weed ecotypes often vary in germination requirements. *F*. *bidentis* seeds were positively photoblastic; thus, shallow tillage will help reduce the weed populations and seedling emergence was the greatest for seeds present on the soil surface. These results suggest that *F*. *bidentis* has the potential of becoming a problematic weed under no-till systems. *F*. *bidentis* was able to germinate over a broad range of temperatures, and the germination threshold temperature was low, which contribute to in part explain why the invasive weed may emerge throughout the season from the first ten days of April through mid-October provided soil moisture is favorable in North China.

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