SEED DORMANCY AND GERMI NATION BEHAVIOUR OF *ECHINOCHLOA COLONA*

Chun, J. C.* and K. Moody**

Echinochloa colona 種子의 休眠 및 發芽特性

全載哲* · 키이쓰 무디**

ABSTRACT

Seed dormancy and germination responses to light and gases were determined for *Echinochloa colona* (L.) Link. *E. colona* seeds did not require a period of after-ripening for breaking dormancy. Water movement occurred readily across the seed coat. Repeated cycles of hydration and dehydration reduced viability and thence germination. Water imbibition for 24 h increased seed moisture by 21%; seeds returned to their original weight after drying at room temperature for 13 h. Removal of seed-coats increased germination in the dark. Light stimulated germination. Germination at a daylight intensity of 51.9 Wm^{-2} or less was significantly reduced. Germination of seeds which were exposed to light for 1 h each day was significantly less than that of seeds exposed for longer than 2 h a day. Seeds subjected to blue light had delayed and decreased germination compared to seeds exposed to red light. Ethylene or carbon dioxide exogenenously added in the presence of light stimulated germination. The addition of the two gases together had a synergistic effect. In the dark, however, the two gases did not increase germination.

Key words: Germination, Dormancy, Light, Gas.

INTRODUCTION

Echinochloa colona (L.) Link is one of the worst weeds in the world.[3] This annual grass is commonly associated with rice (*Oryza sativa* L.) as well as various crops in tropical countries.

There is only a limited amount of information on the dormancy and germination behaviour of *E. colona*. Holm *et al.*[3] indicated that some strains have a short period of dormancy following harvest, but this dormancy disappeared in less than 8 weeks of dry storage. Ramakrishnan[5] reported that seeds of *E. colona* had a dormancy period of about two months and germinated best with continuous light.

The purpose of this study was to characterize the type of seed dormancy and to determine the effects of light and gases on germination behaviour.

* 全北大學校 農科大學

** 國際米作研究所

* Department of Agricultural Chemistry, Jeonbug National University, Jeonju 520, Korea.

** Department of Agronomy, The International Rice Research Institute, Los Banos, Laguna, Philippines.

Meterials and methods

The experiments were conducted at the International Rice Research Institute (IRRI), Los Banos, Laguna, Philippines. Mature seeds collected from *E. colona* plants growing in a maize (*Zea mays* L.) field at IRRI were air-dried in a greenhouse. They were germinated and grown to maturity to obtain uniform seeds for the experiments. Unless otherwise stated, 30 seeds were placed in a 250-ml Erlenmeyer flask on a piece of Whatman No. 1 filter paper and moistened with 3 ml of distilled water. The flasks were then sealed with a rubber stopper. Seeds were considered germinated when the shoot was 5 mm long. Since no change in germination occurred after 10 days after seeding (DAS), final germination percentage were determined after 10 days. A completely randomized design with four replications were used in all experiments. Duncan's multiple range test (DMRT) on percent germination was done after the data had been transformed using an arc sine transformation.

Length of drying of newly harvested seeds

To determine the effect of drying on dormancy, seeds were collected from the plants growing in the greenhouse as soon as the lowest spike had finished flowering and dried under sunlight. Four samples of one hundred seeds were selected at random and planted on the soil (sandy clay loam) surface in a plastic pot (9 x 9 cm) each day for 4 days beginning on the day after the seeds were harvested. The pots were sub-irrigated during the experimental period to prevent the seeds from being covered with soil. Accumulated percentage germination was determined each day for 7 consecutive days from 3 DAS.

Hydration-dehydration cycle

Ten days after harvesting forty samples of 100 air-dried seeds were alternately soaked in distilled water and dried for 24-h periods. Immediately after every hydration cycle, the water clinging to the seed coat was removed with a paper towel and the seeds were dried at room temperature (approximately 23°C). Four samples were chosen at random whenever one hydration-dehydration cycle was terminated, and these were planted on the soil surface in a plastic pot (9 x 9 cm) which was sub-irrigated. The number of germinated seeds was counted 10 DAS.

To determine the permeability of the seed coat to water during the hydration cycle, four samples of 100 seeds were soaked in distilled water in an incubator at 27°C after weighing. After 24 h of water imbibition, the seeds were dried by frequent stirring for 1 h at room temperature. The 100-seed weight was recorded every 2 h after the 1-h drying period until the weight returned to the initial weight.

Removal of seed coat

The effect of seed coat on dormancy was investigated by removing the seed coat carefully with a scalpel. Immediately after, the embryos were placed in flasks and moistened with 3 ml of distilled water. The flasks were kept in the dark covered with a black cloth in a steel box. Intact seeds and seeds with glumes removed were treated the same way.

Length of light exposure

Flasks were placed in either daylight in the greenhouse or in the dark for 10 days. In daylight, the flasks were exposed for varying periods - 1, 2, 3, 4, 5, and 10 days. Except for the 10-day light treatment, the flasks were transferred to darkness after exposure to light for the desired period. In the treatments involving 4- and 5-day light exposure, 25 ungerminated seeds out of 30 seeds originally placed were selected prior to being placed in the dark. To determine the stimulatory effect of light on germination, the flasks kept in darkness for 10 days were placed in light for another 6 days and the germination percentage were determined 16 DAS.

Light quantity and quality

Maximum daylight intensity as measured at 1300 hours daily was regulated not to exceed 0.6, 6.5, 51.9, and 311.2 Wm^{-2} during the experimental

period. A rectangular frame was covered with black cloth, and the flasks were placed in the frame. Light intensity inside the frame was measured by a light meter (Model Lambda).

To determine the effect of light exposure time on germination, the flasks were placed in sunlight between 1000 and 1500 h. The number of hours of exposure varied from 1 to 5 each day. Exposure to sunlight for 1, 2, 3, 4, and 5 h started from 1200, 1130, 1100, 1030, and 1000 h, respectively, each day for 10 days. This resulted in exposing all the treatments to a highest light intensity during a day.

Different coloured cellophanes were used to regulate light quality. Each flask was covered by one sheet of red, orange, yellow, green, or blue cellophane, and then placed in the greenhouse for 10 days. The wavelengths passing through each cellophane colour as determined by a spectrophotometer (Model Spectronic 20) were 608 nm with red cellophane, 580 with orange, 540 with yellow, 515 with green, and 487 with blue. Reductions in light intensity caused by the cellophane colours were 84.7, 41.0, 23.5, 70.6, and 84.7%, respectively.

Ethylene and carbon dioxide

Pure ethylene (C_2H_4) and carbon dioxide (CO_2) were used at a concentration (v/v) of 20 ppm and 1.0%, respectively. To supply the gases exogenously, C_2H_4 or CO_2 or both were injected into a flask through a flanged serum stopper installed in the centre of the rubber stopper in the flask. For removal of the gases endogenously evolved by imbibed seeds, 0.2 M $HgCl_2$ and saturated KOH were used as absorbents to trap C_2H_4 and CO_2, respectively. Two ml of each absorbent in a 20-ml bottle with a wick of filter paper was placed in each flask. The flasks were kept in either light or darkness for 10 days.

Results and discussion

Dormancy

Germination was affected by the length of drying (Fig. 1). Cumulative germination increased with increasing length of drying. A significant decrease

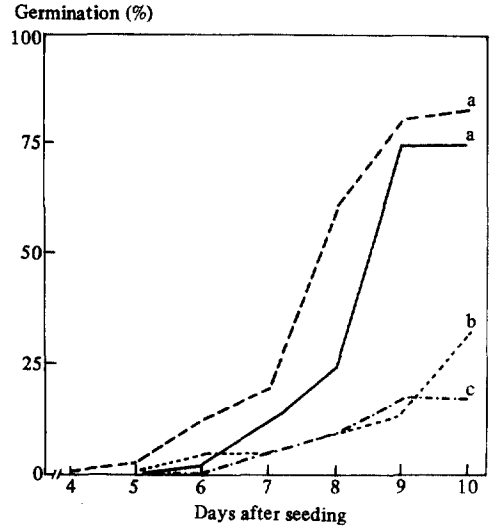

Fig. 1. Cumulative germination of newly harvested seeds of *Echinochloa colona*. Length of drying; 1 day (–·–·–), 2 days (·····), 3 days (———), and 4 days (– – – –). Means followed by a common letter 10 days after seeding are not significantly different at the 5% level by DMRT.

in germination was observed from seeds subjected to 1 or 2 days drying compared with seeds dried for 3 or 4 days. The low germination associated with a shorter drying period was probably due to incomplete conversion of assimilates into an available form for germination in the newly harvested seeds. During the experimental period, the conversion was completed, resulting in a continuous increase in cumulative germination.

E. colona seeds did not require a period of afterripening for breaking dormancy. These results disagree with the findings of Ramakrishnan[5] who reported that newly harvested seeds of *E. colona* had a dormancy period of about 2 months and did not readily germinate after harvest. In our experiment seeds were planted on the soil surface, whereas Remakrishnan[5] placed seeds in between moist filter paper, preventing exposure of the seeds to light.

Seed viability

Repeated cycles of water imbibition and drying resulted in reduced germination (Fig. 2). Seed viability was about 15% after 10 hydration-dehydra-

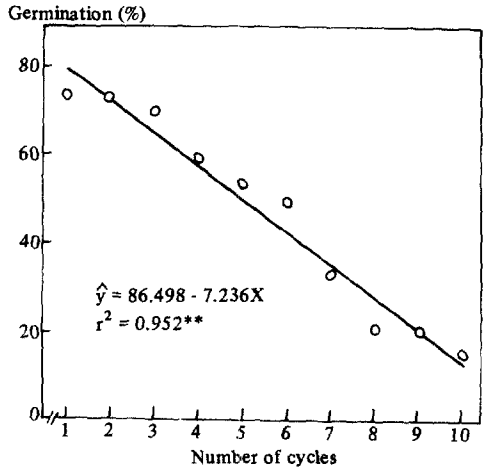

Fig. 2. Seed viability of *Echinochloa colona* as affected by the number of hydration-dehydration cycles. ** = Significant at P = 0.01 level.

tion cycles, whereas more than 70% germination was obtained from seeds subjected to only one hydration-dehydration cycle. A negative linear relationship was found between seed viability and the number of hydration-dehydration cycles. Hydration may stimulate induction of germination, but the subsequent reactions such as enzyme activity are probably inhibited by the following dehydration. Increasing the number of dehydrations would block the continuous activity of hydrolytic enzymes which respond to added moisture.

The seed coat of *E. colona* did not inhibit germination by preventing water imbibition. Water movement occurred readily across the seed coat. After the seeds were soaked in water for 24 h and then dried for 1 h, the 100-seed weight was 129.1 mg, compared to 106.7 mg before imbibition. The seeds gained 21 % of additional seed moisture and returned to their original weight after 13 h of drying at room temperature.

Germination of the embryo in the dark

When the seed coat of *E. colona* was removed, the germination percentage in the dark markedly increased. Compared with intact seeds, however, removal of glumes gave no increase in germination

Table 1. Percent germination in darkness as affected by removal of seed coats.[a]

Treatment	Germination (%)
Intact seeds	7.3 b
Seeds with glumes removed	7.8 b
Seeds with coats removed	83.9 a

[a]Means followed by a common letter are not significantly different at the 5% level by DMRT.

(Table 1). Hence, dormancy is not a condition of the embryo itself but is imposed by the surrounding structure. Therefore, the release of dormancy in *E. colona* seed in the dark could be completed by removing the seed coat. This indicates that a possible cause of the dormancy may be due to the presence of a germination inhibitor in the seed coat.

Stimulatory effect of light

Light had a stimulatory effect on germination of *E. colona* seeds (Table 2). The highest germination percentage occurred with seeds that were kept in light for 10 days; there was significantly less germination with less exposure to light. Exposure to light after the dark treatment brought about a significant increase in germination. The increase was due exclusively to the presence of light. Therefore, *E. colona* seeds need light for germination as has been reported by Ramakrishnan[5] and Holm *et al.*[3]

Table 2. Stimulatory effects of light on the germination of *Echinochloa colona* seeds.

Treatment (day, condition)*	Germination** (%)
10 L	54.6 a
10 D	2.5 de
10 D fb 6 L	49.2 b
1 L fb 10 D	1.7 e
2 L fb 10 D	4.2 de
3 L fb 10 D	5.8 d
4 L fb 10 D***	5.9 de
5 L fb 10 D***	12.5 c

*L = exposed to light, D = placed in darkness, fb = followed by.

** Means followed by a common letter are not significantly different at the 5% level by DMRT.

*** Ungerminated seeds were selected.

Although there was no significant difference in percentage germination between the light exposure treatments up to 4 days, germination of seeds exposed to light for 5 days before being placed in darkness increased significantly above all the other timings (Table 2). There seems to be a threshold level of light required for the germination process.

Harper[2] classified seed dormancy into three types: innate, induced, and enforced. Enforced dormancy is attributed to the absence of necessary conditions for germination. Based on te results obtained in the above experiments, the dormancy type of *E. colona* seed could be interpreted as enforced.

Light quality and quantity

Germination of *E. colona* seeds varied with the levels of maximum daylight intensity (Fig. 3). There was a significant reduction in percentage germination at 51.9 Wm^{-2} or less, compared to that at 311.2 Wm^{-2}. Less germination was observed at the lower daylight intensity levels.

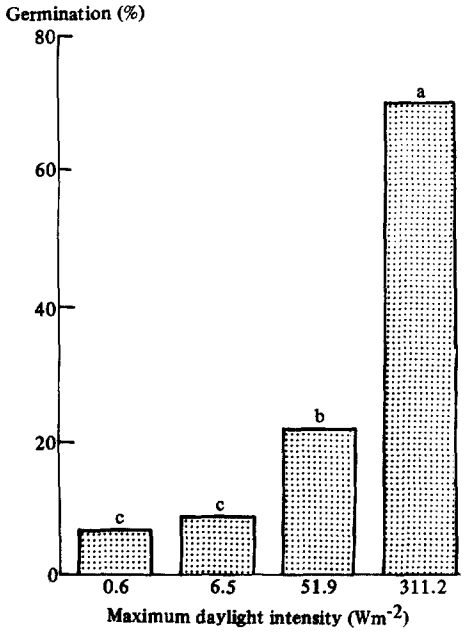

Fig. 3. Germination of *Echinochloa colona* at different light intensities. Means followed by a common letter are not significantly different at the 5% level by DMRT.

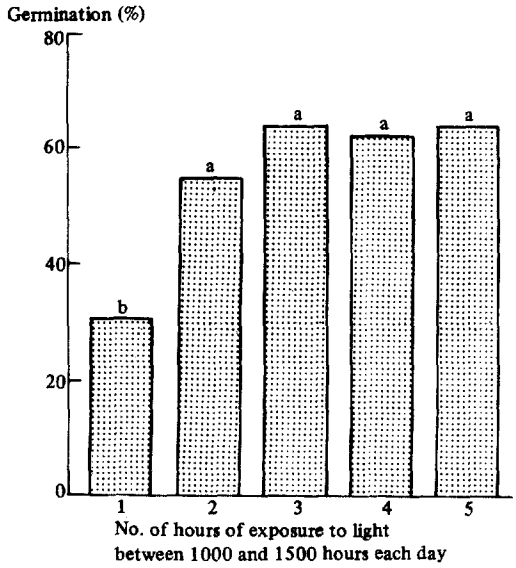

Fig. 4. Germination of *Echinochloa colona* as affected by different light exposure times. Means followed by a common letter are not significantly different at the 5% level by DMRT.

Different light exposure times also affected the germination of *E. colona* (Fig. 4). Compared to the other exposure times, 1-h exposure to light gave a significant decrease in seed germination. Germination was not significantly affected when the seeds were exposed to light for 2 h or more each day. Once a certain light requirement was satisfied, additional illumination did not increase germination.

Percentage germination was reduced by exposure to different colours of light compared to full sunlight (Fig. 5). This decrease was attributed to both decrease in light intensity and change in light quality imposed by the cellophane colours used. Germination with red, orange, or yellow light was significantly higher than that with green or blue light. Seeds subjected to green or blue light, showed delayed germination compared to seeds exposed to red, orange, or yellow light.

Different responses of the seeds to various light colours resulted from both a pigment system involved during the germination period and difference in light intensity given by the cellophane

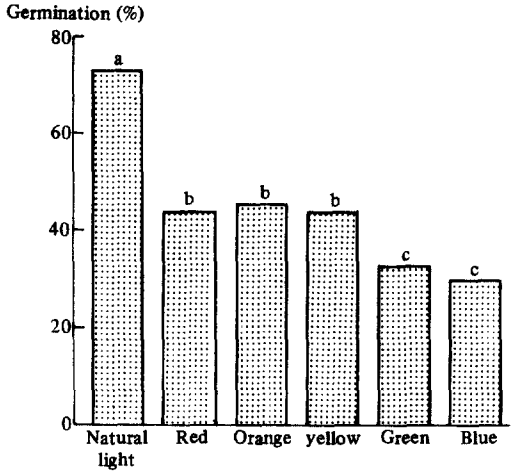

Fig. 5. Effect of different colours of light on the germination of *Echinochloa colona*. Means followed by a common letter are not significantly different at the 5% level by DMRT.

colours. The reduction in light intensity caused by red or blue cellophane was similar so that the increase in germination with red light compared with blue light was due to a pigment system involved. Higher germination with orange or yellow light compared to that with green or blue light was attributed primarily to less reduction in light intensity. Pagaspas[4] noted that germination of *E. colona* seed was promoted with red light, but inhibited with far-red light.

Ethylene and carbon dioxide

C_2H_4 or CO_2 exogenously added in the presence of light produced a stimulatory effect on germination. The greatest stimulatory effect was obtained from the addition of C_2H_4 + CO_2 or of C_2H_4 (Fig. 6). Using either $+C_2H_4$-CO_2 or $-C_2H_4$+CO_2 did not stimulate germination. The addition of the two gases together had a synergistic effect. Germination when the gases were added was higher than when either gas or both gases were removed during the experimental period.

When either of the two gases endogenously evolved was removed, germination percentage was not significantly different between treatments, regardless of the addition of the other gas. No significant difference in germination was obtained when CO_2 or C_2H_4 was added. This indicates that the

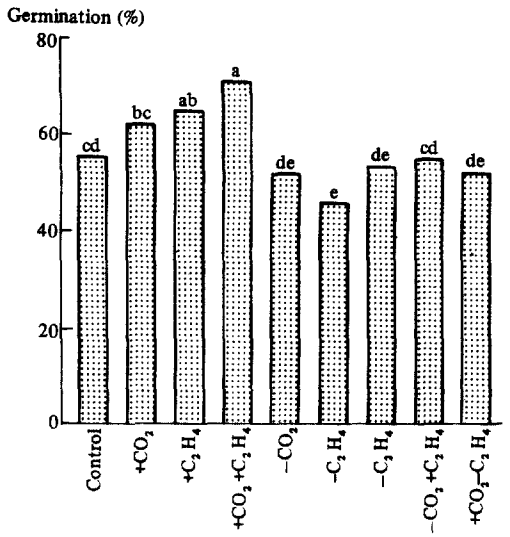

Fig. 6. Effect of ethylene and carbon dioxide on the germination of *Echinochloa colona*. Means followed by a common letter are not significantly different at the 5% level by DMRT.

two gases have relatively independent action on the germination of *E. colona* seeds, as was observed for *Trifolium subterraneum* L. by Esashi & Leopold.[1]

In darkness, however, germination was completely inhibited in all treatments (data not presented). C_2H_4 or CO_2 did not contribute to the regulation of dormancy when the seeds were kept in darkness.

As a whole, the data obtained indicate that dormancy in *E. colona* is regulated possibly by a germination inhibitor present in the seed coat. The coat-imposed dormancy can be released by removal of the seed coat, resulting in germination of the embryo in the dark. Neither the after-ripening process nor hardness of seed coat is responsible for imposing dormancy. Dormancy was overcome best by light treatment, but the response varied with light quantity and quality. This may be an effect of a threshold level of light required for counteracting the germination inhibitor and a pigment system involved during the germination process.

C_2H_4 and CO_2 are the principal gases evolved by seeds known to affect germination. The results with *E. colona* suggest that although addition of exogenous C_2H_4 or CO_2 contributes to stimulate

germination, response to light appears to be a prerequisite in overcoming dormancy.

Light is one of important environmental factors necessary for germination of *E. colona*. In rice area *E. colona* usually begins to germinate when rice seeds are planted with enough moisture. At the beginning of rice culture sufficient light reaches the ground floor to stimulate the germination of *E. colona* seeds. Although *E. colona* produces and shatters seeds much earlier than the rice plants, the mature seeds of *E. colona* fallen on the soil surface do not germinate immediately because of insufficient light transmitted after great portion of light as well as most effective light to germination has been absorbed by the rice canopy. Then, *E. colona* seeds may be thrown into dormancy until conditions become favorable. The dormancy is a means for optimizing growth and dispersal of *E. colona* in time.

摘　　要

E. colona 種子의 休眠性 및 光과 發芽關與氣體에 대한 發芽 反應性을 調査하였다. *E. colona* 는 休眠 覺醒을 위해 後熟期間을 要하지 않하였다. 種皮를 通한 水分 移動은 容易하였다. 水和 및 乾燥의 反復處理는 種子의 發芽力을 減少시켰다. 24時間의 種子 浸潤으로 水分含量은 21%까지 增加되었으나, 室溫에서 13時間 後에는 原狀態로 回復되었다. 種皮의 除去는 暗條件下에서 發芽率을 현저히 增加시켰으며, 光은 完全種子의 發芽를 促進시켰다. 日光度 51.9 Wm^{-2} 以下의 條件下에서의 發芽는 현저하게 減少되었다. 光照射 1日 1時間 處理는 1日 2時間 以上 處理에 비하여 현저하게 낮은 發芽率을 보였다. 靑色光 照射 種子는 赤色光 照射에 비하여 發芽가 遲延되었으며, 發芽率도 낮았다. 光存在下에 外生的으로 處理한 에칠렌 및 탄산가스는 發芽를 促進시켰으며, 이 두 氣體의 同時處理는 共力效果를 나타내었다. 그러나 暗條件下에서는 이 두 氣體의 作用力이 나타나지 않하였다.

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