

## Factors Affecting Germination of Citronmelon (*Citrullus lanatus* var. *citroides*)

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Citron melon is a monoecious, hairy annual vine commonly found in citrus orchards, and cotton and peanut fields. Information is not available on the effect of various environmental factors on the germination of citron melon. Laboratory and greenhouse experiments were carried out in 2011 and 2012 to determine the effect of light, temperature, salinity, pH, simulated water stress, and depth of sowing on the germination of citron melon. Citron melon germination was affected by various environmental factors. Highest germination was observed at day/night temperatures of 25/20 to 30/25 C regardless of light conditions. At temperatures below 25 C and beyond 35 C, germination declined and was higher under dark condition than light. Germination decreased as osmotic potential became more negative (−0.3 MPa to −1.5 MPa) and salt concentration increased (50 to 350 mM). No germination was observed at > −0.9 MPa and ≥ 300 mM salt concentrations. However, germination was observed over a broad range of pH (3 to 9) and up to 10-cm sowing depths. Seeds sown at the surface did not germinate but maximum germination (88 to 96%) occurred at 2- to 4-cm depth. The results of this study suggest that citron melon can grow in a wide range of climatic conditions and therefore can persist in Florida because of favorable weather and environmental conditions.

**Nomenclature:** Citronmelon, *Citrullus lanatus* (Thunb.) Mats & Nakai var. *citroides* (L.H. Bailey) Mansf., CILAC.

**Key words:** Depth of sowing, emergence, light, osmotic potential, pH, salt concentration, temperature.

Citronmelon, also referred to as tsamma, is a member of the Cucurbitaceae family. A native of Africa, its spread was attributed to the escape from cultivation (Bryson and DeFelice 2009). It is widely distributed in the southern and eastern regions of the United States as well as in Southern California and Arizona (Bryson and DeFelice 2009). In Florida, citron melon is found in 23 of 67 counties (USDA 2012).

Citron melon is an annual, monoecious, and hairy vine weed. Seedlings of citron melon are very noticeable because of their large, thick, ovate, and shiny green cotyledons with very distinct venation (Hall et al. 1991). Leaves are alternate, rough, and have three to four pairs of rounded lobes. Flowers are solitary, with broad yellow petals. The fruit is a many-seeded hard berry that can be light green or variegated light and dark green in color. Seeds are ovoid, flattened with various colors ranging from white to dark reddish brown, tan, or blackish brown

to green. Mature plants possess tendrils on the side of deeply divided leaves (Bryson and DeFelice 2009; Hall et al. 1991).

Citron melon is a troublesome weed in many southern crops such as citrus (*Citrus* spp.) (Futch and Hall 2003), cotton (*Gossypium hirsutum* L.), grain sorghum [*Sorghum bicolor* L. (Moench)] (Smith and Cooley 1973), and peanut (*Arachis hypogaea* L.) (Grichar et al. 2001, 2002; Webster and MacDonald 2001). In cotton, citron melon interference reduced yield by 20 to 35% (Smith and Cooley 1973). Citron melon interferes during digging and inverting procedures in peanut production (Young et al. 1982), lengthening time for field drying of peanut vines and pods. This increases the chance of exposing peanut pods to rainfall events, thereby causing harvest losses (Grichar et al. 2001). It also serves as an alternate host of peanut rootknot nematode [*Meloidogyne arenaria* (Neal)] (Rich et al. 2009). In citrus, it interferes with grove operations such as herbicide application and harvesting and also competes with young citrus trees for nutrients and moisture.

Germination and emergence are important processes for weed establishment and persistence. Various environmental factors affect these processes and may determine weeds' presence in the field. Understanding the impact of environmental factors on the germination and seedling emergence of

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citron melon will aid in developing appropriate weed management strategies for its control. The objective of this study was to determine the effect of light, temperature, simulated water stress, salt stress, and pH on germination and effect of sowing depth on seedling emergence of citron melon.

## Materials and Methods

**Seed Sources and Preparation.** Fruits of citron melon were collected near the Citrus Research and Education Center in Lake Alfred, FL in 2011. Seeds were extracted from the fruit, washed, air dried, and then stored at 5 C until the initiation of all experiments. Before the initiation of the study, seeds were surface sterilized in 0.5% sodium hypochlorite solution for 10 min, and were rinsed with running tap water for 5 min.

**General Procedure for Germination Studies.** All laboratory and greenhouse experiments were conducted in 2011 and 2012 at the Citrus Research and Education Center at Lake Alfred, FL. Ten sterilized seeds of citron melon were placed on 9-cm petri dishes lined with two sheets of filter paper (Whatman # 4 filter paper, International Ltd., Maidstone, U.K.) in all experiments unless stated otherwise. The filter paper was moistened initially with 10 ml of distilled water or test solution. All petri dishes were sealed with Parafilm (American National Company, Greenwich, CT 06836) to prevent desiccation and were kept for 7 d in a germination chamber with day/night temperature of 30/25 C and 12-h photoperiod except in the study on the effect of light and temperature. After 7 d, the number of seeds that germinated was counted. Seeds were considered germinated if at least 1 mm of the radicle was visible. All germination data were expressed as percentage germination. In all studies, treatments were replicated four times and were laid out in a randomized complete block with growth chambers serving as blocks. All experiments were repeated in time.

**Effect of Temperature and Light.** Seven day/night temperature and three light regimes were selected to determine the effect of temperature and light on germination of citron melon. The petri dishes were placed in corresponding growth cabinets maintained at day/night temperature of 10/5, 15/10, 20/15, 25/20, 30/25, 35/30, or 40/35 C and light regimes of either 12/12-h light and dark or complete light. Petri dishes were covered with two

layers of aluminum foil to create the completely dark condition.

**Effect of Simulated Water Stress.** Solutions with osmotic potential of 0.0, -0.3, -0.4, -0.6, -0.9, or -1.3 MPa were prepared by dissolving 0, 154, 191, 230, 297, or 350 g of polyethylene glycol (PEG 8000, Fischer Scientific, Fair Lawn, NJ 07410) in 1 L of deionized water (Michel 1983; Shaw et al. 1991).

**Effect of Salt Stress.** Solutions of 0, 50, 100, 150, 200, 250, 300, and 350 mM NaCl (Fischer Scientific, Fairlawn, NJ 07410) were prepared according to Michel (1983) and were used as germination media.

**Effect of pH.** Buffered pH solutions were prepared according to the method described by Shaw et al. (1987), using 0.1 M potassium hydrogen phthalate (Fischer Scientific) to make solutions with pH levels of 4, 5, and 6, and 25 mM sodium borate (Fischer Science Education, Hanover Park, IL 60133) to prepare solutions with pH levels of 7, 8, or 9. Distilled water was also used as germination medium for comparison.

**Effect of Depth of Sowing on Seedling Emergence.** Twenty five seeds were sown at depths of 0, 2, 4, 6, 8, or 10 cm below the soil surface in 18-cm-deep and 9-cm-diam Styrofoam® cups (Master Containers, Mulberry, FL) containing Florida Candler fine sand. The sand was sieved and moistened before seed sowing to facilitate easy wetting. The experiment was conducted in a greenhouse with day/night temperatures set at  $25 \pm 5/20 \pm 5$  C. Natural daylight was supplemented with sodium vapor lamps to provide 12 h of light. Germinated seedlings were counted every 7 and 14 d after planting. Seedlings were considered emerged when the two cotyledons could be visually discerned and were removed after weekly counts.

**Statistical Analysis.** Arcsine square transformation did not improve homogeneity of variance according to Bartlett's test; nontransformed percentage germination data were analyzed using the MIXED procedure in SAS version 9.3 (SAS Institute Inc., Cary, NC). All treatments were considered fixed effects, whereas experimental runs were considered random effects. Means were separated using Fisher's Protected LSD test at  $P = 0.05$ .

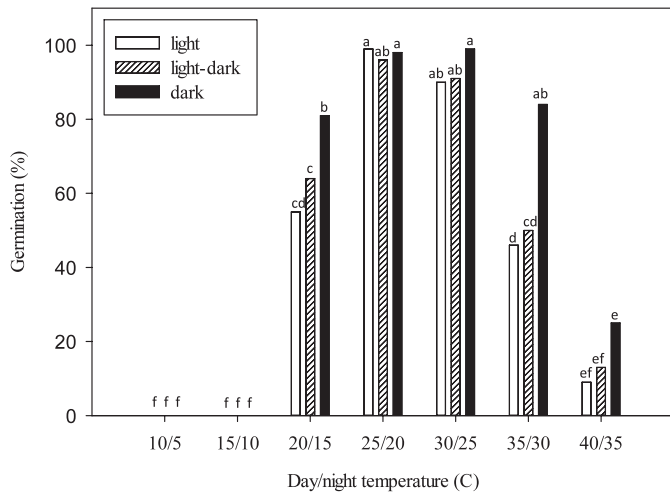


Figure 1. Effect of temperature and light on the germination of citronmelon. Bars with the same letters are not significantly different at  $\alpha = 0.05$ .

Additionally, regression analysis was used whenever applicable. Percent germination values at different osmotic potential or salt concentration were fitted to a functional two-parameter exponential decay model using SigmaPlot version 12.3 (Systat Software Inc., San Jose, CA 95110). The model fitted was:

$$G(\%) = G_{\max} \times \exp(-G_{\text{rate}}x)$$

where  $G$  represents germination at osmotic potential or salt concentration  $x$ ,  $G_{\max}$  is the maximum germination, and  $G_{\text{rate}}$  is the slope.

## Results and Discussion

**Effect of Temperature and Light.** Citron melon germination varied by temperature and light (P-value = 0.0367). Germination occurred at day/night temperatures ranging from 20/15 to 40/35 C at all light regimes (Figure 1). Maximum germination ranged from 90 to 99% at day/night temperatures of 25/20 to 30/25 C regardless of light regime and 35/30 C under dark conditions. Citron melon was sensitive to low temperature as there was no germination at day/night temperatures between 10/5 and 15/10 C regardless of light regimes. At day/night temperatures  $\leq 20/15$  and  $\geq 35/30$  C, light appeared to affect germination more than temperature as higher germination was observed under dark than under full light and alternating dark and light conditions. Similarly, Thanos and Mitrakos (1992) reported that germination of commercial watermelon cv. Sugar Baby [*Citrullus lanatus* (Thunb.) Matsum. & Nakai var.

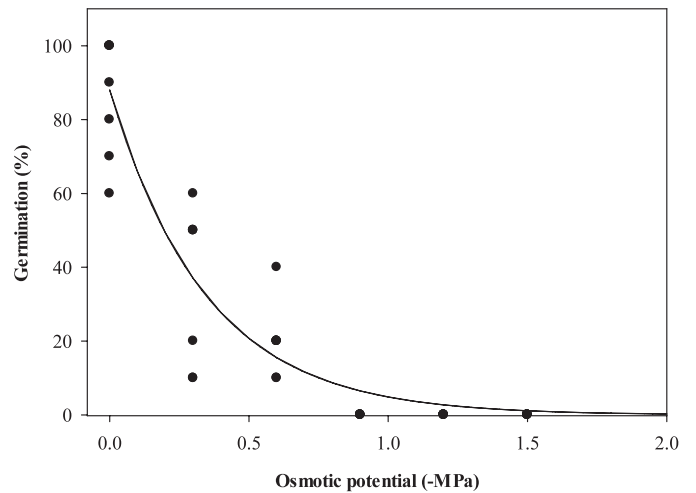


Figure 2. Effect of simulated water stress on the germination of citronmelon after incubating at 30/25C (day/night) with a 12-h photoperiod after 1 wk. For Equation 1, parameter estimates were  $G_{\max} = 87.97$ ,  $G_{\text{rate}} = 2.90$ , and  $R^2 = 0.89$ .

*lanatus*] was optimum at 20 to 40 C in the dark and exposure to continuous white incandescent light inhibited germination. Other weed species belonging to the same family exhibited similar germination behavior with respect to temperature effects. Germination of burcucumber (*Sicyos angulatus* L.) seeds occurred over a wide range of temperatures but was reduced at temperatures below 20 C and above 30 C (Mann et al. 1981). Similarly, buffalo gourd (*Cucurbita foetidissima* Kunth) germination occurred in the dark at constant temperatures ranging from 15 to 37 C, with optimum germination at 25 C (Horak and Sweat 1994).

**Effect of Simulated Water Stress.** Germination of citron melon was affected by simulated water stress under laboratory conditions (P-value < 0.0001). Germination was greatest (88%) when there was no simulated water stress and decreased as osmotic potential became more negative (Figure 2). Germination declined to 37 and 20% at osmotic potential of  $-0.3$  and  $-0.6$  MPa, respectively. There was no germination at  $> -0.9$  MPa. Similar results were obtained by Tingle and Chandler (2003) in smell-melon (*Cucumis melo* var. *dudaim* Naud.). These results suggest that citron melon can germinate in a wide range of soil moisture conditions but can be adversely affected by drought. Dry conditions can inhibit germination, thereby limiting citron melon occurrence under field conditions. However, continued presence of citron melon in citrus groves can be expected since irrigation is applied in most citrus groves, especially during summer months in Florida.

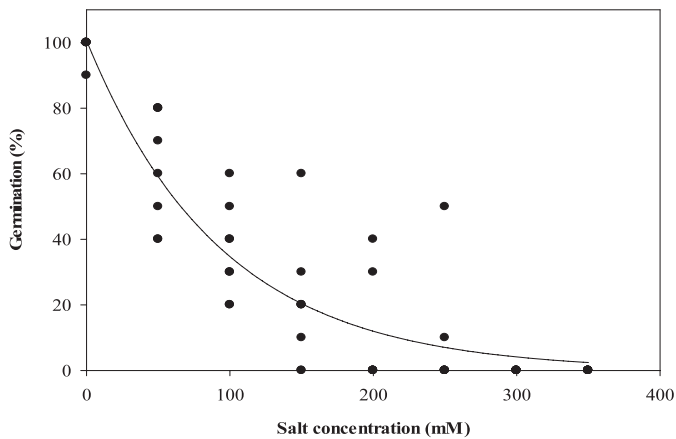


Figure 3. Effect of salt concentration on the germination of citronmelon after incubating at 30/25C (day/night) with a 12-h photoperiod after 1 wk. For Equation 1, parameter estimates were  $G_{\max} = 100.61$ ,  $G_{\text{rate}} = 0.01$ , and  $R^2 = 0.87$ .

**Effect of Salt Stress.** Germination of citron melon decreased as salt concentration increased and occurred over a wide range of salt concentrations (P-value < 0.0001). Highest germination (96%) was observed under no saline stress conditions (Figure 3). Germination was reduced to almost 50% and further declined to 13 to 21% when salt concentration was increased 100 to 150 mM. Germination was lowest (< 10%) at salt concentration  $\geq 200$  mM. A small percentage of seeds germinated at higher salt concentrations ( $\geq 200$  mM to < 300 mM), indicating some degree of tolerance to highly saline conditions. Germination was completely inhibited at salt concentration  $\geq 300$  mM. Other species known to tolerate salinity includes giant sensitiveplant (*Mimosa invisa* Mart ex Colla) (Chauhan and Johnson 2008) and tall morningglory [*Ipomoea purpurea* (L.) Roth] (Singh et al. 2012).

**Effect of pH.** Germination of citron melon was affected by pH (P-value < 0.0001) and occurred over a wide range of pH (pH 3 to 9) (Figure 4). At pH 3 to 5, germination of citron melon was lower (49 to 51%) compared with pH 6 to 7, where germination was 73 to 77%. At pH > 7 germination was minimal ( $\leq 5\%$ ). These results suggest that citron melon germination was favored by acidic to neutral pH. Similar to citron melon, several weed species commonly found in Florida such as common beggar's-ticks [*Bidens alba* (L.) D.C.] (Ramirez et al. 2012a), goatweed (*Scoparia dulcis* L.) (Jain and Singh 1989), and stranglervine [*Morrenia odorata* (Hook. & Arn.) Lindl.] (Singh and Achhireddy 1984) germinate over a wide range of pH. Other species such as silverleaf nightshade

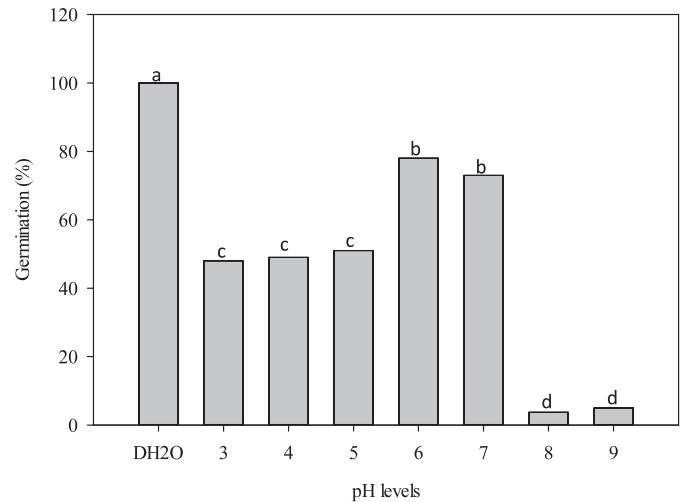


Figure 4. Germination of citronmelon under varying pH levels. Bars with the same letters are not significantly different at  $\alpha = 0.05$ . DH<sub>2</sub>O stands for distilled water.

(*Solanum elaeagnifolium* Cav.) had significantly higher germination at pH 10 (Stanton et al. 2012).

**Effect of Depth of Sowing on Seedling Emergence.** Citron melon emergence was affected by sowing depth (P-value < 0.0001). Higher seedling emergence (88 to 96%) was observed when seeds were sown at 2- to 6-cm depth, whereas there was no emergence when seeds were sown at the surface (Figure 5). At lower depths (8 to 10 cm) emergence was reduced to < 65%. Similarly, Smith and Crowley (1973) reported that citronmelon emergence from 2.5 to 6 cm was observed under greenhouse conditions, with highest emergence occurring at depths closer to the surface. There was minimal emergence (20%) at 30 cm and citronmelon failed to emerge from the 35-cm depth (Smith and Crowley 1973). Other weed species from the Cucurbitaceae family exhibit the same emergence characteristics. Horak and Sweat (1994) reported that surface-sown seeds of buffalo gourd did not germinate, but emergence was greatest (> 50%) at 2- to 8-cm sowing depth. Smellmelon emergence was 62 to 97% at depths of 1 to 6 cm, whereas at 9- to 15-cm depth germination was < 25% (Tingle and Chandler 2003). In contrast, other weed species such as common beggar's-tick and horseweed [*Conyza canadensis* (L.) Cronq.], germination was highest when seeds were sown at the surface (Nandula et al. 2006; Ramirez et al. 2012a). Citronmelon's ability to emerge from deeper depths of sowing could be attributed to its large seed (6-mm diam). Large seeds often have more seed reserve, thus allowing them to emerge from greater depths (Baskin and Baskin 1998).

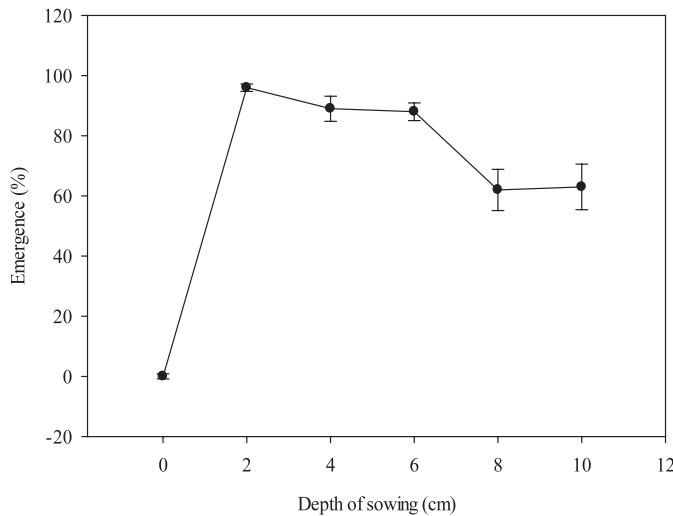


Figure 5. Germination of citronmelon at various sowing depths 7 d after sowing. Horizontal bars are standard error of the mean.

The results of this study indicate that citronmelon germination is affected by various environmental factors. Citronmelon germinates best at day/night temperatures of 25/20 to 30/25 C and under dark condition at day/night temperatures lower than 20/15 and higher than 35/30 C. Germination decreased as water stress and salt concentration increased but occurred over a broad range of osmotic potential and salt concentration, indicating some degree of tolerance to dry and saline conditions. Acidic to neutral conditions favored germination more than alkali pH. Seeds of citronmelon did not germinate when sown on the soil surface; however, burying seeds at 2 cm or deeper favored emergence. Therefore, burying seeds through cultivation will not be effective in controlling citronmelon. These results suggest that citronmelon can grow well and persist in Florida because of favorable weather and environmental conditions. The average maximum temperatures for spring and summer months in Florida ranged from 24 to 34 C and 28 to 35 C, respectively, conducive for germination of citronmelon (FAWN 2013). Citrus-producing areas in Florida receive an average of 20 to 25 cm of precipitation during summer months (USDA-NASS 2013), adequate enough to support growth of citronmelon that might emerge in spring. Furthermore, Florida soil is slightly acidic, with pH of 5 to 6 (Shober et al 2008), which also favors citronmelon germination. Under these conditions, citronmelon control is imperative. Application of PRE and POST herbicides is a good option for citronmelon control as it is highly susceptible to several herbicides (Ramirez et al.

2012b). Overall, this information can be used in modeling the germination and emergence dynamics of citronmelon and developing strategies for effective management of this weed not only in citrus but in other agricultural production areas in Florida.

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