



Temperature, pH and light effect on germination and growth behavior of grassy weeds of direct-seeded rice

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ABSTRACT

Environmental factors have significant implications on the biology of weeds, hence the study of biology of major weeds in a crop, could prove an ecological and economical viable tool for their management. *Echinochloa glabrescens*, *Leptochloa chinensis*, *Eragrostis japonica* and *Dactyloctenium aegyptium* are the major weeds of direct-seeded rice (DSR) and many other *Kharif* crops. The effect of temperature, pH and light was studied on the biology of these four weed species under laboratory conditions during the *Kharif* seasons of 2012 and 2013. Temperature regimes of 15, 20, 25, 30, 35, 40 and 45°C; pH 5.0, 7.0, 9.0 and 11.0 and light period of 0, 3, 6, 12, 24 and 48 hours were evaluated for their effects on germination, shoot and root growth. Conducive temperature for germination of all four-weed species was 35°C except *Dactyloctenium aegyptium*, which has maximum germination at 30°C. Similar to germination, maximum shoot and root length was recorded at 35°C in all the weed species except *E. japonica*, for which 30°C was the optimum. Seed germination was observed over a broad range of pH of all weed species; however, it was highest at pH 7.0. *Echinochloa glabrescens* was most sensitive to a given pH range among all the weed species. Light periods didn't alter the process of germination, shoot and root growth. Manipulation of these factors at field level could be helpful in reducing the weed pressure in DSR by preventing their germination.

INTRODUCTION

Weed biology is an important, but neglected tool of integrated weed management compared to chemical weed management. Studying weed biology will help to develop a robust and sustainable weed management program (Kumari *et al.* 2010). But, there are concerns about laboratory experimental results that can be translated into field weed management programme (Acker 2009). Many factors like seeding depth, temperature, soil moisture, soil pH have influence on germination of weed species and their subsequent growth behavior that will affect crop-weed competition (Singh and Punia 2008, Chauhan and Johnson 2010, Kumari *et al.* 2010). Alteration of above factors by agronomic management could reduce the weed crop competition. Direct-seeded rice (DSR) has more weed competition compared to transplanted rice, because of the absence of a size differential between the crop and weeds and the suppressive effect of standing water on weed growth at crop establishment (Chauhan and Johnson 2010). Weed control is major limitation for the success of DSR (Rao *et al.* 2007). When optimum weed control was achieved in

DSR, there was non-significant yield difference compared to transplanted crop (Sipaseuth *et al.* 2000, Chauhan *et al.* 2015). Reducing the weed-crop competition by integration of weed biology could reduce the dependency on chemicals for weed management, hence harnessing high yields in DSR.

Popular tool fascinating the farmers, chemical weed management is also facing problems of resistance, residue for next crop, leaching into ground water, *etc.* Singh and Punia (2008) argued that understanding the biology and ecology of weeds could be helpful for effective weed management in herbicide era where there is a rapid rate of evolution of resistant weed biotypes. According to Zimdhal (1991), unlike entomology and pathology, weed science evolved as 'how to control' weeds and focuses on mainly chemical weed control, whereas, weed biology stress on 'cause.' Advance information on weed biology and ecology is a key to improve weed management programme and determining the biology of the species determines the strengths and weakness in its growth cycle and allow for the development of better weed control strategies. So, integration of multifaceted characterization of weed

biology of DSR weeds can result in designing an efficient weed management system for this crop.

MATERIALS AND METHODS

Experiments were conducted at CCS Haryana Agricultural University during 2012 and 2013 under laboratory conditions. Effect of temperature, pH and light at different levels on germination and growth were studied. In all studies, each treatment included four replications (four Petri dishes) and data of two experiments was used for analysis. Seed of four weed species (*Echinochloa glabrescens* (Tall Barnyard grass), *Leptochloa chinensis* (Chinese sprangletop), *Eragrostis japonica* (Love grass) and *Dactyloctenium aegyptium* (Crowfoot grass) were used in the study. Seed of these weeds were collected in 2011 and 2012 from the plants growing in DSR fields. Harvested seeds were bulked, cleaned and sieved to remove any extraneous plant or floral material and then stored in weed science laboratory of department of Agronomy at 15°C temperature. These seeds were kept for approximately 6-7 months. The seeds collected in 2011 were used for sowing in June of 2012 and those collected in 2012 were used for 2013 studies. Seeds were treated with 0.1% sodium hypochlorite immediately before each experiment for 30 minutes and washed 3-4 times with distilled water so as to ensure disease free seeds. Before starting the experiment, seeds of each weed species were tested for viability with 1% tetrazolium chloride solution. A germination test was carried out for each species, at room temperature using Whatman filter paper No. 1 in Petri dishes in both the seasons.

Effect of temperature

To determine the effect of temperature on germination of the above mentioned weed species, twenty seed of each weed were placed uniformly between two layers of filter papers (Whatman No. 1) of 90 mm in Petri dishes of 100 mm diameter (Borosil glass). They were moistened with distilled water and then incubated at 15, 20, 25, 30, 35, 40 and 45°C ± 1.5°C in seed germinator. The filter paper was kept moist throughout the period by regular application of deionized water. Constant temperatures were maintained in the incubator without any diurnal fluctuations. Germination, shoot and root length were recorded at 1, 2 and 3 weeks after sowing (WAS).

Effect of pH

Buffered solutions of pH 5.0 was prepared by using citric acid and pH of 7.0, 9.0 and 11.0 by using potassium hydroxide pellets. Twenty seeds of each weed species were placed on filter paper in Petri dishes. A 10 ml pH solution was added to each Petri

dish and these were watered as and when required with freshly prepared pH solutions. Germination, shoot and root length was recorded at 1, 2 and 3 WAS.

Effect of light

To evaluate the effect of light on germination, twenty seeds were placed in Petri dishes with 10 ml deionized water applied/Petri dish and kept under six regimes of light periods (0, 3, 6, 12, 24 and 48 hour). After a given light hour, the Petri dishes were immediately wrapped with double layer aluminum foil to ensure no light penetration. In case of 0 h light, Petri dishes were covered with aluminum foil immediately after moisture application. Wrapped Petri dishes were kept undisturbed for seven days and then were unwrapped to observe germination and then maintained under natural day and light conditions. Germination, shoot and root length was recorded at 1, 2 and 3 WAS.

Statistical analysis

All the experiments were conducted in completely randomized block design. Treatments of each experiment were replicated four times, and each experiment was conducted twice and experimental data were analyzed using software SPSS version 7.5. Arcsine transformation was used wherever needed. Formula used for arcsine transformation in SPSS was= $\text{ARSIN}[\text{SQRT}(\text{germination}/100)] \times 90/1.571$

RESULTS AND DISCUSSION

Effect of temperature

Optimum temperature recorded for germination of *Eragrostis japonica* was 30 to 35°C and it was the lowest at 15°C (**Figure 1**), whereas shoot and root length were recorded highest at 30°C (**Table 1**). Some of the weed species may germinate at a higher temperature, but may not grow at that high temperature, as the optimum temperature for the germination of small flower morning glory (*Jacquemontia tamnifolia*) was 35 to 40°C, but optimum growth was between 25 to 35°C (Shaw *et al.* 1987). This can be attributed to high photosynthesis rate at these temperature regimes as Kebede *et al.* (1989) recorded carbon exchange rate of *Eragrostis tenella* was the highest between 36 and 42°C. Singh and Singh (2009) reported that Germination of *Sida spinosa* (prickly sida) and *Desmodium tortuosum* (Florida beggar weed) was highest between 25 to 40°C and *Senna obtusifolia* (sicklepod) from 20 to 40°C, but maximum germination of Feather Lovegrass [*Eragrostis tenella* (L.)] occurred at 30/20°C alternating temperature (Chauhan 2013). Seed germination,

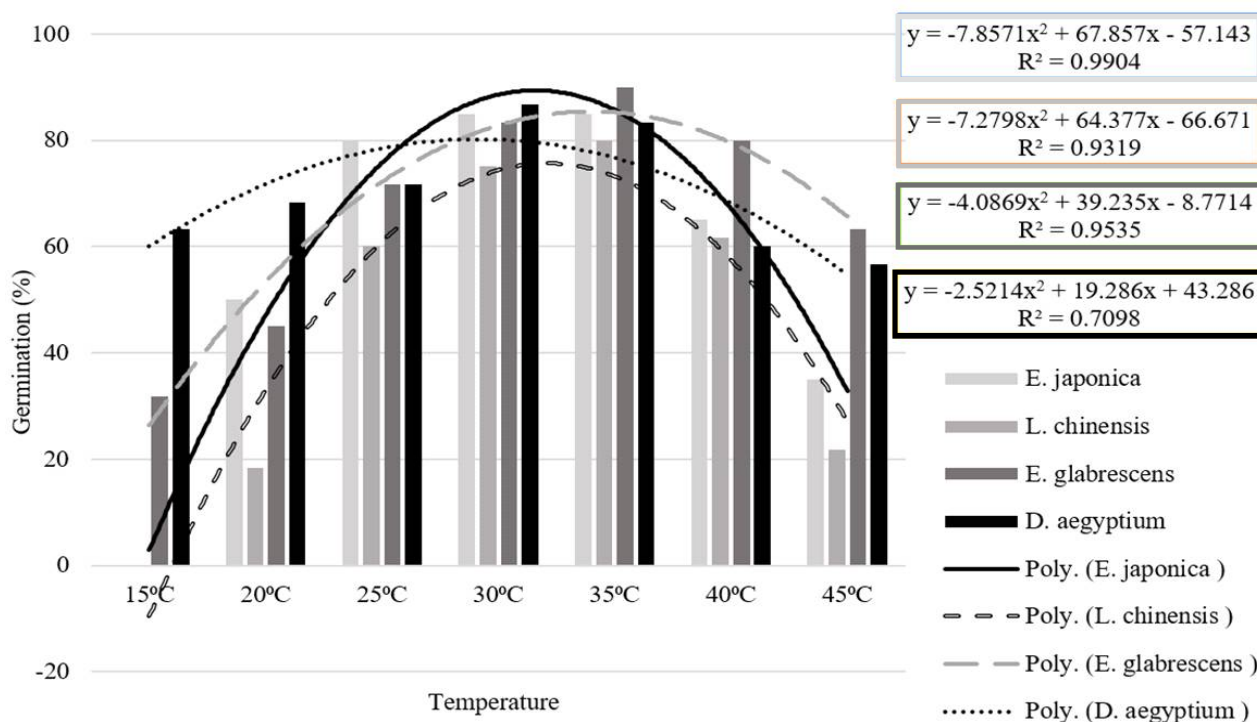


Figure 1. Effect of temperature regimes on germination (%) of different weed species (3 WAS)

Table 1. Effect of temperature regimes on shoot length and root length (cm) of different weed species (3 WAS)

Temperature	Shoot length (cm)				Root length (cm)			
	<i>E. japonica</i>	<i>L. chinensis</i>	<i>E. glabrescens</i>	<i>D. aegyptium</i>	<i>E. japonica</i>	<i>L. chinensis</i>	<i>E. glabrescens</i>	<i>D. aegyptium</i>
15°C	0.00 (0.10)	0.00 (0.10)	3.90 (1.98)	1.40 (1.18)	0.00 (0.10)	0.00 (0.10)	1.17 (1.08)	2.43 (1.56)
20°C	0.97 (0.98)	0.60 (0.78)	4.27 (2.06)	1.87 (1.37)	0.47 (0.68)	0.53 (0.71)	1.57 (1.25)	2.57 (1.60)
25°C	1.23 (1.11)	1.47 (1.21)	5.60 (2.37)	2.17 (1.47)	0.67 (0.79)	0.63 (0.82)	2.30 (1.52)	4.40 (2.10)
30°C	2.17 (1.47)	1.77 (1.33)	7.37 (2.71)	2.40 (1.55)	1.04 (1.02)	0.97 (0.98)	4.40 (2.10)	5.20 (2.28)
35°C	1.73 (1.32)	2.10 (1.45)	7.77 (2.78)	2.57 (1.60)	1.03 (1.02)	0.97 (0.98)	4.67 (2.16)	5.37 (2.32)
40°C	0.90 (0.94)	1.37 (1.17)	6.10 (2.47)	1.97 (1.40)	0.47 (0.68)	0.90 (0.95)	4.23 (2.06)	3.73 (1.93)
45°C	0.37 (0.60)	1.17 (1.08)	6.23 (2.50)	1.53 (1.24)	0.27 (0.51)	0.73 (0.86)	3.17 (1.78)	2.27 (1.50)
LSD (p=0.05)	0.18	0.12	0.10	0.12	0.19	0.14	0.13	0.17

shoot and root length in case of *Leptochloa chinensis* was the highest at 35°C, which was similar at 25°C, 30°C and 40°C at all observation stages. However, Aulakh *et al.* (2006) reported differences in germination of *L. chinensis* were non-significant due to temperature variations. At initial stage of experiment, *L. chinensis* did not germinate at 15 and 20°C, but at later stages emergence took place. Temperature and time (*i.e.* degree days) is a more comprehensive parameter for the study of temperate effect on germination (Ritchie and NeSmith 1991). Different weed species has different degree days requirement for germination, therefore, variation was recorded in germination of different weed species. Degree days requirement of *L. chinensis* could be more compared to other weed species, resulting into delay germination at lower temperature.

The maximum germination, shoot and root growth of *E. glabrescens* was observed at 35°C. The germination of *E. glabrescens* at 15°C in first week was very less, but it increased at 3 WAS. Germination, shoot and root growth of *E. glabrescens* at all temperature regimes suggest that this species could emerge throughout the year at low altitudes in tropical countries. Similar results were reported for *E. prostrata*, in which seeds germinated at all test temperatures (*i.e.*, 25/15, 30/20, and 35/25°C). The germination of *E. glabrescens* and *D. aegyptium* was faster and showed the earliest germination among all the four-weed species tested. So, it could establish itself very quickly under field conditions and found major species in rice fields. Maximum seed germination was reported at 30°C in *D. aegyptium* and minimum at 45°C, shoot and root length was also

high at 30°C. Similar to this study, Burke *et al.* (2003) found that *D. aegyptium* germinated over a range of 15-40°C with the optimum germination occurring at 30°C. So, *D. aegyptium* has a wide range of temperature for germination, this may be due to its wider adaptability from temperate to tropical climate. Whereas, Singh and Singh (2009) found that when data averaged over different weed species and temperatures, highest weed seed germination was recorded in the temperature regime of 25 to 35°C and maximum being at 30°C. Higher germination in the present study was observed in *E. glabrescens* among the four test species and minimum in *E. japonica*. There was no germination recorded in *E. japonica* and *L. chinensis* at 15°C.

Temperature changes may affect a number of processes during seed germination and growth including the membrane permeability, activity of membrane-bound protein and cytosol enzymes reported by Bewley and Black (1994). Seed germination includes two stages: water absorption and radicle emergence; may be the first stage is not related to temperature, but the second stage is temperature dependent. However, Horowitz and Taylorson (1983) found effect of temperature on imbibition of velvet leaf (*Abutilon theophrasti* Medic.). Exposure of seed to lower or higher temperature than optimum at the second stage cause inhibition of germination while few seed could germinate. Optimum temperature regime is important for embryo development before germination (Harris *et al.* 1998). Some researchers have pointed out that high humidity and temperature environments may readily cause seeds to age and to lose some vigor (Walters 2000).

Effect of pH

The present experimental results indicated that *E. japonica* showed germination at all pH ranges, with highest germination at pH 7.0 (**Figure 2**). Solution of pH 9.0 and 11.0 had similar effect on the germination of *E. japonica*. Similar to *E. japonica*, pH had an effect on the germination of *L. chinensis* and showed tolerance to change in pH. At 1 WAS, pH 5.0 and 7.0 had similar effect on germination. This trend was followed at all observation time. Also, increase in pH from 9.0 to 11.0 had non-significant effect on the germination at each observation stage. Moderate effect of pH was recorded by Altop *et al.* (2015) on germination of bearded spangled (*Leptochloa fusca*) as maximum germination (92%) occurred at a pH of 7 and the lowest germination (54%) at a pH of 10. Similar to this, spotted spurge (*Euphorbia maculate* L.) germination was not affected by the tested levels of pH as germination was

95% over a broad pH range from 4 to 10 (Agropur *et al.* 2015). In a study of 12 Florida weed species, Singh and Singh (2009) found no weed seed germination at pH 3, except *Cyperus esculentus* (yellow nutsedge). A pH range of 5 to 11 had no adverse effect on the germination, when data were averaged over 12 weed species. Germination of prickly sida was the highest at pH 9 and any increase or decrease in pH resulted in reduced germination. Yellow nutsedge tubers germinated 14% at pH 3 compared to 47% at pH 7.

Among all the weed species, *E. glabrescens* showed more pH sensitivity as there was no germination recorded at pH 11.0 during the whole experiment. Maximum germination took place at pH 7.0. There was a steep reduction in the germination at all three observation stages when pH was increased from 7.0 to 9.0. Similar to *E. glabrescens* in this study, *E. crus-galli* was also sensitive to pH change as reported by Sadeghloo *et al.* (2013) and recorded very less germination at pH 9. Although, there was highly variable difference in *D. aegyptium* germination at different pH levels initially, this variability was narrowed down with advancement of the experiment. It showed that higher and lower pH had slowed down the process of germination. Maximum germination occurred at pH 7.0 and this could be explained to favorable ion concentration at this pH. Similarly, germination of sheep sorrel (*Rumex acetosella* L.) seed was occurred in a wide range of buffered pH solutions but the highest germination occurred over a pH range of 6 to 7 (Yazid *et al.* 2013).

Maximum shoot and root length of all weed species was recorded at pH 7.0 (**Table 2**), as this pH may not have affected different enzymic activities and also absorption of nutrients could be more at this pH, compared to other levels. *E. japonica* was less sensitive to pH change as considerable shoot and root length was recorded at all pH levels, maximum being at 7.0. Effect of pH was lowered as the experiment advanced. Similar effect of pH 7.0 was observed in *L. chinensis* on shoot and root growth. However, pH 5.0 affected shoot and root length in similar way at all observation times. Similar to germination, *E. glabrescens* had no shoot and root growth at pH 11.0 during the whole experiment and proved most sensitive to higher pH. Like other weed species, favorable pH for shoot and root growth of *E. glabrescens* was 7.0. As the pH was lowered from 7.0 to 5.0 there was only 9% reduction in shoot length but when it was increased from 7.0 to 9.0, 31% reduction was recorded. In contrast to other species, *D. aegyptium* had less reduction in shoot and root length as pH was increased from 9.0 to 11.0. Hydrogen ion concentration produced its effect on

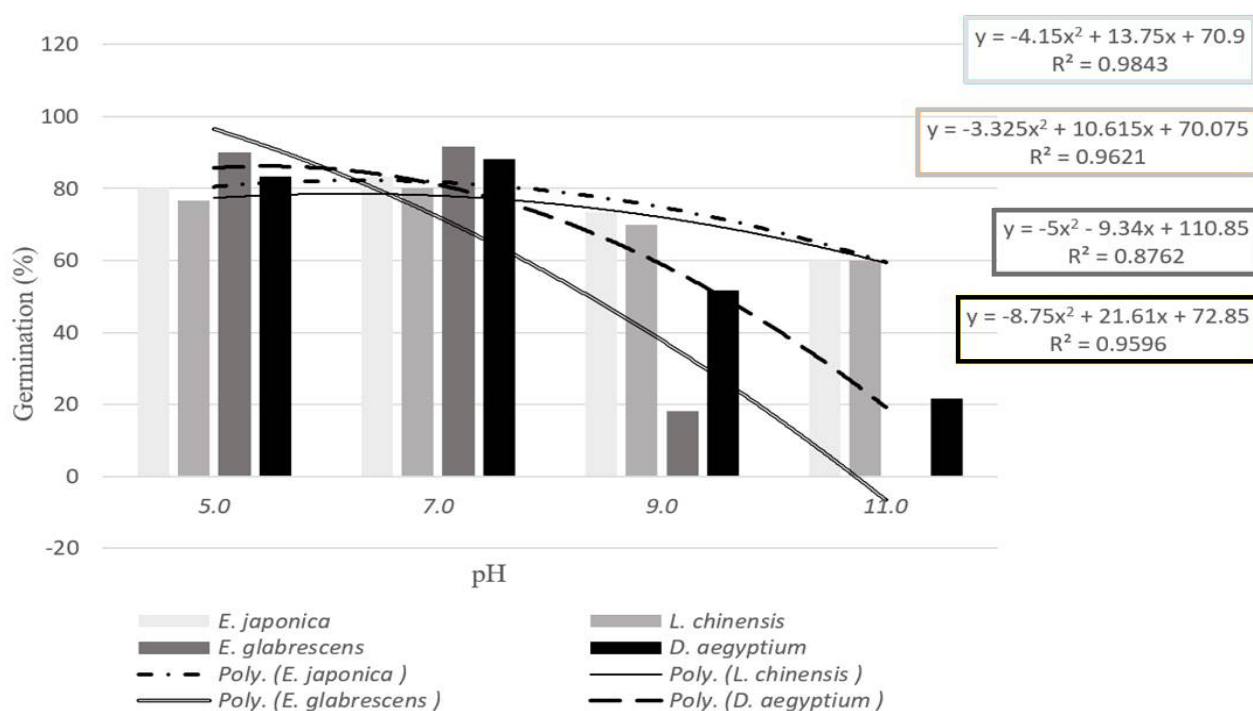


Figure 2. Effect of pH on germination (%) of different weed species (3 WAS)

Table 2. Effect of pH on shoot length and root length of different weed species (3 WAS)

pH	Shoot length (cm)				Root length (cm)			
	<i>E. japonica</i>	<i>L. chinensis</i>	<i>E. glabrescens</i>	<i>D. aegyptium</i>	<i>E. japonica</i>	<i>L. chinensis</i>	<i>E. glabrescens</i>	<i>D. aegyptium</i>
5.0	2.10	2.10	6.10 (2.47)	2.07 (1.44)	1.27	0.90	2.03 (1.43)	2.90 (1.70)
7.0	2.37	2.37	6.67 (2.58)	2.37 (1.54)	1.43	1.27	2.23 (1.50)	3.33 (1.83)
9.0	1.43	1.67	4.57 (2.14)	1.27 (1.13)	1.03	1.03	1.33 (1.15)	2.43 (1.56)
11.0	1.07	1.00	0.00 (0.10)	1.07 (1.04)	0.93	0.67	0.00 (0.10)	2.03 (1.42)
LSD (p=0.05)	0.13	0.17	0.12	0.12	0.18	0.19	0.17	0.20

root length of *D. aegyptium*, as there was 40% change in maximum and minimum at 3 WAS. But, Burke *et al.* (2003) recorded highest *D. aegyptium* growth at pH 4.0 compared to other higher pH. Similar to present experiment results, Buchanan *et al.*, (1975) recorded higher growth of *D. aegyptium* at pH 6.3 than 5.4.

Effect of light periods

Results indicated that light was not a prerequisite for germination of these weed species (Data not presented). These results are similar to sicklepod (*Senna obtusifolia*) germination, which was not responsive to light (Norsworthy and Oliveira 2006), while dissimilar to the germination of four weeds under study, *Celosia argentea* was stimulated by light for higher germination (Chauhan and Johnson 2007). Seed germination response to light varies from species to species. Seed of some species require light to germinate (Chauhan and Johnson 2008b, Chauhan and Johnson 2008c) and others can germinate equally in light and dark (Chauhan and Johnson 2008a). Higher germination under both conditions *i.e.* light

and dark shows that these weeds can germinate from deeper depths. So, weed species under the present study could germinate from deeper depths if other factors are favorable.

Singh and Singh (2009) found no germination inhibition for any of the 12 test species in dark, except *Richardia brasiliensis* (Brazil pusley). After 168 h, germination of Brazil pusley ranged from 2 to 10% with light exposure of 0 and 16 h, respectively, before placing them in dark. Under alternate light and darkness cycle of 12 h, germination of Brazil pusley increased to 59%. Other than Brazil pusley no other species exhibited the photoblastic effect.

For shoot growth, light is the prime requirement mainly for photosynthesis, even though in the beginning darkness for seven days had non-significant effect on shoot growth. It was observed that in absence of light, achlorophyllous growth occurred during first week. After 7 days when Petri dish were kept in normal day and night conditions, seedlings resumed normal growth. Maximum shoot length (7.43 cm) was recorded in *E. glabrescens* at 3

WAS. Therefore, exposing to light or dark for 7 days did not affect the growth of weeds significantly. These results are in conformity with that of Wang *et al.* (2009) on *Urena lobata* which was not light dependent and emerged from depths up to 9 cm. There was no effect of light/dark period on the root length when seed placed in dark or in light for initial one week, in all weed species. Therefore, it can be concluded that these weeds can emerge from deeper depths where light cannot reach.

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