BRIEF COMMUNICATION

The effect of drought, temperature and irradiation on leaf rolling in *Ctenanthe setosa*

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Abstract

The effects of drought, temperature and irradiation on leaf rolling of *Ctenanthe setosa* were studied. Water deficit and air temperature were the most important, and have a synergistic effect on the degree and number of the rolled leaves. Irradiation also increased the rolling together with the water deficit but not alone. The stomata were open in rolled leaves.

Additional key words: water deficit, stomata.

Leaf rolling has been identified as one of the most familiar responses to water deficit in some plant species (*e.g.* Begg 1980, Blum 1988). Leaf rolling is induced by the loss of pressure potential in consequence of a loss of water from the bulliform cells in the upper epidermis of the leaf and reduces incident irradiation, leaf temperature and transpiration (*e.g.* O'Toole *et al.* 1979, Begg 1980, Ehleringer and Forseth 1980, Hsiao *et al.* 1984). It has been reported that the rolling increases drought resistance in cereal crops (Townley-Smith *et al.* 1979). Leaf rolling has been investigated especially in grasses (Clarke 1986, Ekanayake *et al.* 1993). However there is no available information about leaf rolling of *Ctenanthe*.

The first purpose of this study was to determine the interaction between temperature and irradiation on leaf rolling of *Ctenanthe setosa* in laboratory conditions. The second one was to determine the interaction between stomatal closure and leaf rolling.

Ctenanthe setosa (Rosc.) Eichler (*Marantaceae*) plants were vegetatively propagated and grown in plastic pots containing soil. In order to determine the effect of temperature on leaf rolling, plants were incubated in a growth chamber at 25, 30, 34, 36, 38, 40 and 42 °C, in a cycle of 12 h light (100 μmol(photon) m^{-2} s^{-1}) and 12 h darkness and 70 % relative humidity for 2 d. Plants were watered daily so that no water deficit appeared.

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To determine the effect of drought, plants were left to remain withholding water under a mild light and laboratory conditions during 56 d. To determine effect of irradiance, plants were incubated at 50, 100, 150, 200, 250, 300 μmol(photon) m^{-2} s^{-1}, 25 °C and 70 % relative humidity, for 2 to 12 h. Thereafter, the combined effects of these environmental factors were investigated. To estimate the effect of irradiation and drought, the plants were not irrigated for 4, 6, 8, 10 and 12 d, and were incubated at irradiance of 100, 150, 200, 250 and 300 μmol m^{-2} s^{-1} at 25 °C and 70 % relative humidity. Finally, the effects of drought and temperature were determined. For this purpose, plants were not irrigated for 2, 6, 10 and 12 d and held at temperature of 25, 30, 34, 36 °C, irradiance 250 μmol m^{-2} s^{-1}, and relative humidity 70 %.

At the end of these treatments, the number of rolled leaves was calculated and the degree of leaf rolling was estimated (Premachandra *et al.* 1993). The width of the mid-portion of leaves was measured and the degree of leaf rolling was calculated as the percentage reduction in leaf width by rolling.

The stomatal opening was determined by using surface sections of the leaves. For this purpose, unrolled, rolled and rolled-ABA sprayed (at 1mM concentration) leaves were detached at midday and fixed in FAA (formalin:acetic acid:alcohol) for 24 h and then stored in 70 % Et-OH. Hand cut leaf sections were stained with safranine-aniline blue and mounted in entellene.

There was not rolling in *Ctenanthe setosa* leaves up to 36 °C. The degree and number of rolled leaves increased from 38 to 42 °C (Table 1). At 42 °C, the degree and number of rolled leaves were found 53 and 78 %, respectively. When air temperature rose, the leaves rolled up more tightly and became needle-shaped.

Table 1. The effect of temperature on leaf rolling. The complete rolling reduced leaf surface by 61 - 65 %. At temperature of 25, 30, 34, 36 °C no rolling was observed. Means ± SE, n = 3.

Temperature [°C]	Rolled leaves [%]	Degree of rolling [%]
38	40 ± 1.2	32 ± 1.4
40	65 ± 2.3	40 ± 3.3
42	78 ± 1.2	53 ± 1.8

The leaves of *Ctenanthe* are arranged parallel to the soil surface in normal conditions. When the water content in the leaf tissues declines to the lethal level, the leaves roll up, forming two cylinders parallel to the central vein. The first rolling was observed at 28 d after withholding water. The number of rolled leaves was 75 % after 35 d. At 42 d all leaves were rolled (Table 2).

Table 2. The effect of drought on leaf rolling. Means + SE, n = 3.

Period of drought [d]	Rolled leaves [%]	Degree of rolling [%]
35	75 ± 2.0	27 ± 1.5
42	100 ± 0.4	51 ± 4.9
49	100 ± 0.3	71 ± 2.1
56	100 ± 0.1	85 ± 2.5

The degree of rolling also shows a gradual increase from 35 d after withholding irrigation water to the 56 d. When the plants were grown at relative humidity of 70 %, temperature 25 °C and irradiance 150, 200, 250 and 300 μmol m^{-2} s^{-1} the leaves remained unrolled.

Table 3. Dual effect of irradiance and drought on leaf rolling. Means ± SE. $n = 3$.

Period after withholding water [d]	Irradiance [μmol m^{-2} s^{-1}]	Rolled leaves [%]	Degree of rolling [d]
8	250	16 ± 1.2	23 ± 1.4
8	300	20 ± 1.4	28 ± 0.1
10	200	25 ± 2.2	26 ± 2.0
10	250	37 ± 1.6	36 ± 0.3
10	300	50 ± 3.3	47 ± 1.2
12	200	37 ± 2.2	27 ± 2.2
12	250	50 ± 1.2	44 ± 1.8
12	300	62 ± 1.2	52 ± 1.2

At the day 8 after withholding water, no rolling was observed in the leaves at irradiance of 200 μmol m^{-2} s^{-1}, but the number of rolled leaves was 16 and 20 % at irradiances of 250 and 300 μmol m^{-2} s^{-1}, respectively. The degree and number of rolled leaves continued to increase for 10 and 12 d after withholding water (Table 3).

The degree and number of rolling were enhanced in a parallel with increasing temperature and drought (Table 4).

Table 4. The effect of temperature and drought on leaf rolling. Means ± SE, $n = 3$.

Period after withholding water [d]	Temperature [°C]	Rolled leaves [%]	Degree of rolling [%]
2	25	-	-
2	30	25 ± 2.8	12 ± 2.4
2	34	25 ± 3.1	12 ± 2.0
2	36	25 ± 0.3	12 ± 1.6
6	25	-	-
6	30	34 ± 2.5	23 ± 1.7
6	34	34 ± 1.4	34 ± 2.3
6	36	34 ± 2.2	34 ± 3.1
10	25	36 ± 2.4	36 ± 1.6
10	30	44 ± 1.5	48 ± 4.2
10	34	44 ± 2.1	48 ± 2.6
10	36	44 ± 0.4	48 ± 0.8
12	25	50 ± 2.2	42 ± 1.3
12	30	56 ± 2.9	61 ± 1.8
12	34	56 ± 1.4	65 ± 0.6
12	36	56 ± 1.1	65 ± 1.2

It has been observed that the stomata of the rolled and unrolled leaves in *Ctenanthe setosa* were opened in the light. The stomata of rolled leaves treated with abscisic acid

(ABA) were closed and the degree of rolling decreased in these leaves.

The present study has shown that water deficit and air temperature are two important environmental factors causing rolling of leaves in *Ctenanthe setosa*. Omarova *et al.* (1995) noted a leaf rolling in rice at high temperature. Irradiation also increased the rolling together with water deficit but not alone.

In the stressed plants, all leaves of the plants rolled at 35 d after water was withheld but this period decreased to 10 d when the plants were grown under irradiance of 200 μmol m^{-2} s^{-1}. Similar results were observed in maize (Premachandra *et al.* 1993) and rice grown in the field (Turner *et al.* 1986). The degree of leaf rolling increased at midday in water-stressed plants. Leaf rolling in rice is alone used in selection of drought resistant cultivars (Loresto *et al.* 1976, O'Toole and Garrity 1984).

Besides rolling up, the leaves of plants can also change their position with respect to the direction of solar radiation during the day with the aim to minimize the radiation intercepted by leaf tissue. On the other hand, Turner *et al.* 1986 noted an increase in leaf temperature associated with leaf rolling in rice, possibly as a result of reduced transpiration. This is not surprising, since leaf rolling is known to reduce transpiration (O'Toole *et al.* 1979) and canopy temperature is a function of the rate of leaf transpiration (Jackson 1982).

Adaxial stomata are closed in rolled leaves of *Andropogon gerardii* and *Spartina pectinata* (Heckathorn and Delucia 1991), but remain partially opened in rolled leaves of rice (O'Toole and Cruz 1980). In the present study, we observed that the stomata were opened in both rolled and unrolled leaves. To explain whether the leaf rolling is a mechanism to prevent stomatal closure or not, we treated the plants with ABA and observed partial opening in rolled leaves. Leaf rolling can alter leaf microclimate (Matthews *et al.* 1990) in response to the stresses rather than closing the stomata, water loss can still be regulated while photosynthesis and growth continue.

In conclusion, we found that the effective environmental factors on leaf rolling of *Ctenanthe setosa* were similar to the plants which have rolling leaves. A cylinder leaf shape contributes to its cooling during the hottest part of the day so that stomata may remain open and growth continue without associated high rates of water loss.

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