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EFFECTS OF LIGHT STIMULI ON *DESMODIUM GYRANS* LATERAL LEAFLET MOVEMENT RHYTHMS

M. Idrish Miah*
Department of Physics, Chittagong University of Engineering and Technology, Chittagong, Chittagong-4349, Bangladesh.

Anders Johnsson
Biosystems and Biophysics Laboratory, Department of Physics, Norwegian University of Science and Technology, Trondheim, Norway.

ABSTRACT

Rhythmic up - down movements of the lateral leaflets in the plant Desmodium gyrans (L.F.) DC. have been studied. Leaflet movements were recorded with a video-computer system, where the digital video signals from the CCD camera were processed in an ATARI 1040 ST computer using special software. The average period of the lateral leaflet movements at 24^0C was estimated to be 3.5 min. It is proposed, however, that the rhythmic movements of the lateral leaflets might be due to the movement of water across the motor cells, i.e. shrinking and expansion of motor cells, and proton pumps, membrane potential changes and ion transports are responsible for water uptake and loss in motor cells. Leaflet movements in the presence of light stimuli were then studied. It was found that the leaflet always moved towards the direction of the light, whether it was applied from the abaxial or from the adaxial part of the pulvinus. A strong effect was found in both instances, namely the reduced amplitude of the leaflet movement and thus a shortened period. When the abaxial part was exposed to light, the leaflet oscillated in a lower position, but when the adaxial part was illuminated, it oscillated in an upper Detailed instructions for typing your article are given in the following. Since you have to send a camera-ready paper, you are personally responsible of the quality and appearance of your work. Please remember the following points in Particular: a) type upto 200 word abstract single column, b) use 9 pt Times New Roman font and single spaced lines for abstract, c) type the body of the paper in double columns, d) use 10 pt Times New Roman font and single spaced lines for body text, e) list of references in numerical order of appearance, f) supply good quality of figures and tables, g) the use of SI units is mandatory, h) no separate nomenclature, i) paper length should be limited to 10 pages, j) prepare the manuscripts in MS-WORD selecting US letter size paper (8.5″×11″), k) manuscripts should be submitted in DOS formatted 3.5″ FDD along with a hard copy or E-mail the manuscripts and post the hard copy.

Key words: Keywords: Desmodium gyrans; Lateral leaflet; Light stimuli; Motor cell; Rhythm; Plant physiology; Proton pump

1. INTRODUCTION

Desmodium gyrans has a terminal leaflet and one or two lateral leaflets. The terminal leaflet is ca. 3 to 7 cm long and the lateral leaflets are approximately 1 cm long. The terminal leaflets orient themselves horizontally or pointing upwards during the day (Fig. 1A) and hang down during the night (Fig. 1B). These up and down movements of the terminal leaflets occur with a periodicity approximating a day (*circadian; circa = approximately, dian = a day*), and the period is temperature compensated (i.e. the period does not change much with change in temperature). Whereas the lateral leaflets carry out up and down movements with a periodicity of 3-5 minutes (*ultradian*) and is dependent on temperature (Lewis et al., 1987 and Engelmann et al., 1998). The amplitude, phase and period of the lateral leaf movement rhythm in *Desmodium gyrans* could be

**Present Address*: Department of Computer Science and Engineering, Premier University, 1/A, O.R. Nizam Road, Prabartak Circle, Panchlaish, Chittagong – 4203, Bangladesh. Email: idrish_physics@yahoo.com

changed by a number of stimuli, such as, chemical as well as magnetic (Engelmann et al., 1998; Weber et al., 1992; Fostad et al., 1997 and Johnsson et al., 1993). In this work, we have used light stimuli to perturb the rhythmic leaf movements of *Desmodium gyrans* and to see whether visible light can influence the rhythm parameters in the same way as other stimuli.

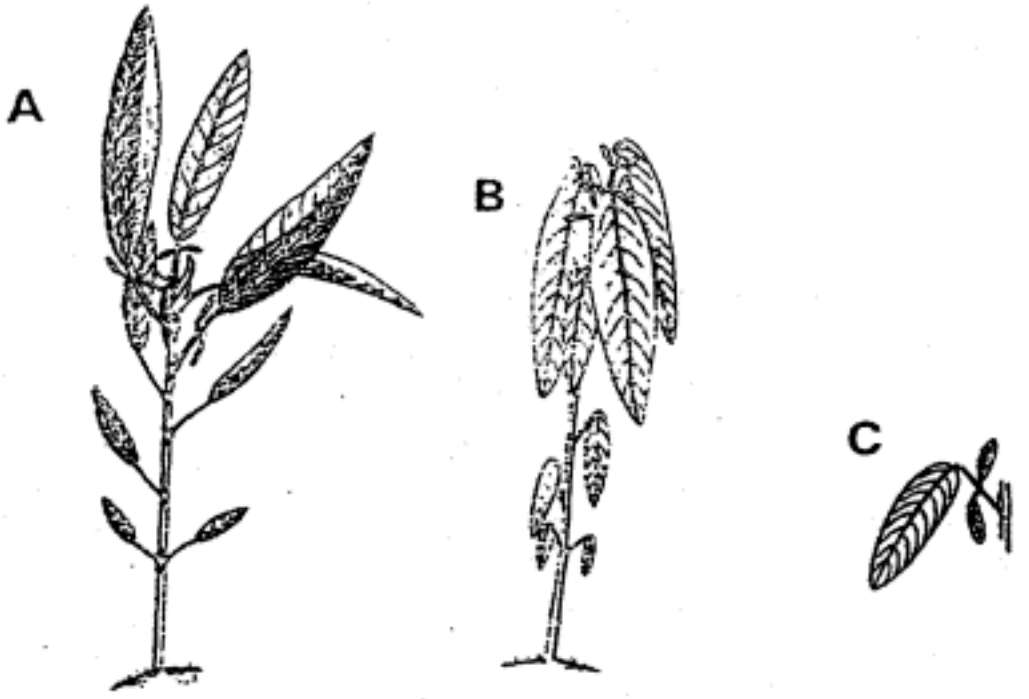

Fig.1. Plant *Desmodium gyrans*: A) in the day position and B) in the night position; C) a petiole with the terminal and two lateral leaflets.

2. MATERIALS AND METHODS

The plants, *Desmodium gyrans* (other name *Desmodium motirium* (Houtt.) Merril) were cultivated under 12:12 hour light/dark cycles in a greenhouse at 28^0C and in 65% humidity conditions. They were about 4 months old and had a height of approximately 70 cm when lateral leaflet were used. Leaflets displaying a regular oscillation were cut from the mother plants and the terminal leaflet removed. They were kept in distilled water in an acrylic glass holder inside an acrylic glass box to minimize temperature fluctuations. A small white styrofoam ball was attached to the tip of each leaflet, serving as an optical marker. A blue LED (light emitting diode) was clamped at a distance of 1 cm from the pulvinus. Leaflet movement was recorded by a video CCD camera (Fujitsu TCZ-250E), positioned in front of the box containing the leaflets. The video signal was digitized by a digitizer (VIDEO ST 1000, Berlin) and then processed in an ATARI 1040 ST computer using special software. The horizontal and vertical positions of the marker were determined by the program once every five seconds for ten hours and the data stored on disk. The period of the movement was estimated using a digital filter on the data and calculating the time between successive leaf position maxima.

3. RESULTS AND DISCUSSION

A typical example of the lateral leaflet movements recorded on video-computer system is shown in Fig. 2. This is the time-series of the sample (plant 1) out of ten. The period of the movements in absence of light stimuli was estimated and was found to be 3.5 min. at ambient temperature

(24^0C). Light was then directed onto the abaxial part as well as onto the adaxial part of the pulvinus (not shown). A strong effect was found in both instances, namely a reduced amplitude of the leaflet movement and a shortened period. When the abaxial part was illuminated, the leaflet oscillated in a lower position, but when the adaxial part was exposed to the light, it oscillated in an upper position. When light was switched off, the amplitude of the movements increased again, but it took some time to regain the same regularity of oscillations.

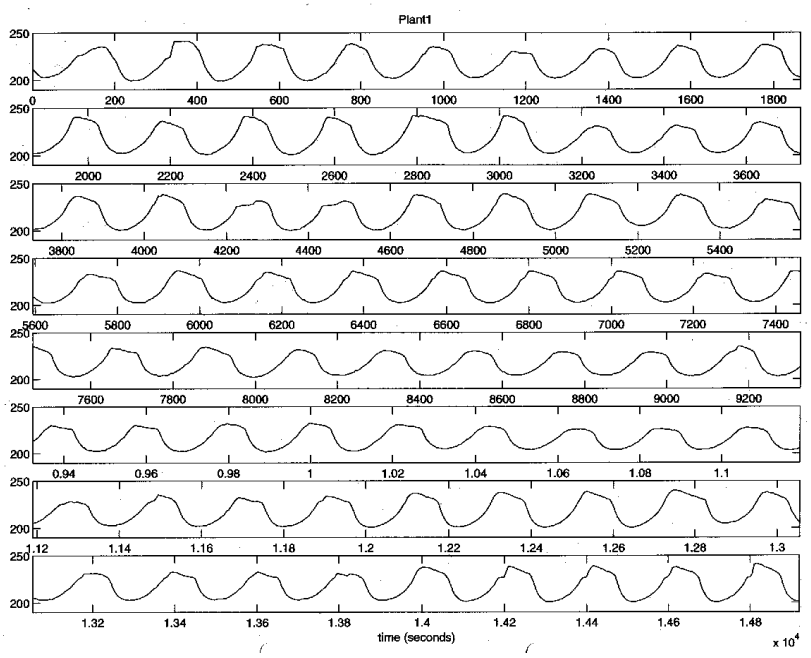

Fig. 2. The oscillatory movements of the lateral leaflets of *Desmodium gyrans* as a function of time.

It is believed that the rhythmic movements of the lateral leaflets occur due to the movement of water across the special cells, called motor cells, at the base of the lateral leaflets. The motor cells are surrounded by conducting vessels (supporting the leaflets with water, etc.) and supportive tissues. These cells, conducting vessels and the supporting tissues form an organ called a pulvinus. The pulvini attach the leaflets to the petioles. The motor cells extend and shrink in a coordinated way. If, for instance, the motor cells of the upper longitudinal rows shrink and those of the lower longitudinal rows expand, the leaflet would rise upward. A special arrangement of cellulose microfibrils in the cell walls allows lengthening and shortening of the motor cells, but prevents the cells from increasing in diameter. Thus, water uptake lengthens the motor cells while water loss shortens them (see Fig. 3). Due to such coordination, the events of lengthening and shortening of the longitudinal segments are performed.

The effects of electric currents and 27 MHz radio frequencies have been studied (Bose, 1913, Guhathakurta et al. 1962 and Johnsson,et al., 1993). They reported that these slimuli could directly affect the ion movement across the motor cells of the pulvinus. It was found that when direct current of strength 10-100 μA was applied for a few seconds to the tip of the lateral leaflets, the phase of the rhythm was delayed. Pulses of dc current were also found to stop the leaf movement rhythms in these plants if applied at an appropriate phase. Ellingsrud et al. (1993) perturbed the leaf movement rhythm of *Desmodium gyrans* using pulses of 27 MHz radio frequencies. It was observed that such pulses could change the amplitude, phase and period length of the rhythm. Radio frequencies were also found to stop the leaf movement. The results of experiments where electrical stimuli affected the ultradian leaf movement rhythm further support a model, which explains the leaf movement rhythm in terms of proton pumps, membrane potential changes and ion

transports. Electrophysiological studies suggest that proton pumps, membrane potential changes and ion transports are responsible for water uptake and loss in motor cells. The following events occur during the up and down movements of the lateral leaflets (Fig. 3):

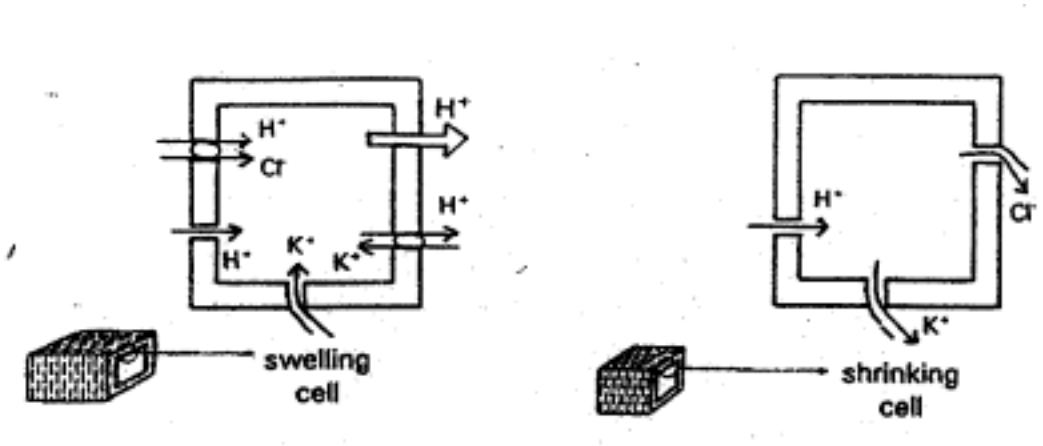

Fig. 3. A model for the membrane processes responsible for the cell volume changes leading to movement of the leaves. Notice that the diameter of the shrinking and swelling cell is the same while the length changes.

Proton pumps in the plasma membrane transport H^+ ions out of the cell. The cells become more negatively charged. This allows K^+ to enter the cell via K^+ channels. Cl^- is imported into the cell probably via Cl^-H^+ transport. The resulting increase in osmotically active donors (K^+ and Cl^-) leads to water uptake and expansion of the motor cells. When a certain pressure is reached, the cells depolarize. The trigger for this depolarization is yet unknown. It is speculated that pressure sensitive Ca^{2+} channels open when a certain pressure is reached and allow Ca^{2+} to pass into the cell. The less negative potential opens outwardly directed K^+ and Cl^- channels. Ion loss is followed by water loss and the cells shrink. According to this model, the ultradian rhythms of the lateral leaflets of *Desmodium* are the outcome of the processes involved in the shrinking (depolarization) and expansion (hyperpolarization) of the motor cells. Light apparently affected physiological parameters that were involved in the control of period. The reason is not yet completely understood.

In conclusion, the period of lateral leaflet movement in plant *Desmodium gyrans* at 24^0C was found to be 3.5 min. It is proposed that the ultradian rhythms of the lateral leaflets are the outcome of the processes involved in the shrinking and expanding of the motor cells. Illumination of the lateral leaflet pulvini of *Desmodium gyrans* not only reduced the amplitude, but also changed period length. Light might affect physiological parameters that are involved in the control of period. Thus light influences the oscillation itself. It was found that the leaflet always moved towards the direction of light, whether it was applied from the abaxial or from the adaxial part of the pulvinus.

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