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Bioelectrochemistry 63 (2004) 91–94
Bioelectrochemistry
www.elsevier.com/locate/bioelechem

Bioelectrochemical signaling in green plants induced by photosensory systems

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Received 23 June 2003; received in revised form 3 September 2003; accepted 8 September 2003

Abstract

Plants generate various types of intracellular and intercellular electrical events in response to environmental stress. The generation of electrophysiological responses induced by blue and red photosensory systems was observed in soybean plants. A phototropic response is a sequence of the following four processes: reception of a directional light signal, signal transduction, transformation of the signal into a physiological response, and the production of a directional growth response. It was found that the irradiation of soybean plants at 450 ± 50, 670, and 730 nm induces action potentials with duration times and amplitudes of approximately 0.3 ms and 60 mV respectively.

Keywords: Bioelectrochemistry; Phototropism; Plant electrophysiology; Action potential; Soybean plant

1. Introduction

Action potentials in higher plants could be the information carriers in intracellular and intercellular communication during environmental changes [1–8]. Plants are exposed to a diverse array of continuously varying perturbations, including variations of irradiation. The purpose of this paper is to describe the long-distance electrical communication in soybean plants induced by the irradiation of blue and red photosensory systems.

Plants generate different types of extracellular electrical events in connection to environmental stress [1–8]. A potential pathway for transmission of these electrical signals could be the phloem sieve–tube system, because it represents a continuum of plasma membranes [8]. Phloem is an electrical conductor of bioelectrochemical impulses over long distances [2]. Phloem consists of two types of conducting cells, the characteristic type known as sieve–tube elements and another type known as companion cells. Sieve–tube elements are elongated cells made up of end walls that are perforated by numerous minute pores through which dissolved materials are conducted. Such sieve–tube elements are connected in a vertical series known as sieve tubes.

Plants respond to light ranging from ultraviolet to far-red using specific photoreceptors. Natural radiation simultaneously activates more than one photoreceptor in higher plants. These receptors initiate distinct signaling pathways leading to wavelength-specific light responses. The three classes of plant photoreceptors that have been identified at the molecular level are phototropins, cryptochromes, and phytochromes [9–15].

2. Materials and methods

All electrochemical measurements were conducted at constant temperature inside a Faraday cage mounted on a vibration-stabilized table (Fig. 1). An IBM-compatible microcomputer with multi I/O plug-in data acquisition board KPCI-3107 (Keithley MetraByte) was interfaced through a multiplexed screw terminal accessory board (Keithley) with 0.1 mm nonpolarizable reversible Ag/AgCl electrodes and used to record digital data. This multifunctional KPCI-3107 data acquisition board provides high resolution and a wide gain range. It features continuous, high-speed, gap-free data

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1567-5394/$ - see front matter
doi:10.1016/j.bioelechem.2003.09.025

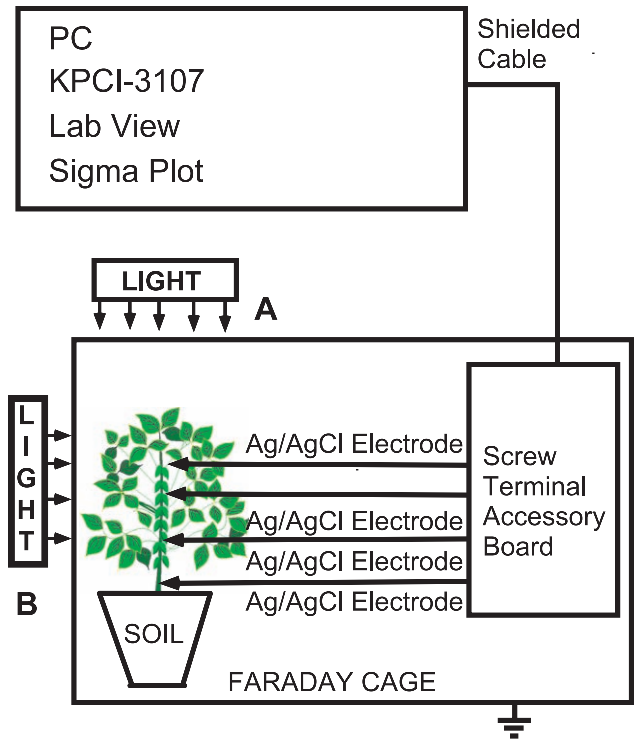

Fig. 1. Experimental setup for measuring electrical signals in green plants.

acquisitioning capacity. Any single channel can be sampled at any gain up to 100 kSamples/s. Measuring signals were recorded as ASCII files using LabView (National Instruments) software.

Ag/AgCl electrodes were prepared from a Teflon-coated silver wire (A-M Systems) according to Ksenzhek and Volkov [3]. Because both Ag/AgCl electrodes were identical, we differentiated between the two by naming them reference and working electrodes. The reference electrode (−) was usually inserted into the stem or root and the working electrode (+) into the stem or a leaf. Following insertion of the electrodes, the plants were allowed to rest until a stable potential difference was obtained between the electrodes. Insertion them into the plants induced action potentials across the stem and slowed fluctuations in the streaming potential. After approximately 1–2 h, the streaming potential was stabilized.

Plants were irradiated from direction A or B (Fig. 1) at different wavelengths using narrow band pass interference filters from GS Edmund Scientific (Barrington, NJ) with a central wavelength tolerance of ± 2 nm.

The soybean seedlings used were approximately 3 weeks old [Glycine max (L.) Merrill, cultivar Hutchenson]. Each plant usually had five to six well-developed leaves. Surface sterilization of soybean seeds was performed before planting with 30% ethanol and 1% bleach. Plants were grown in clay pots with 0.5 l sterilized potting soil in a plant growth chamber (Environmental) at 22 °C. Plants were watered daily and exposed to a 12:12-h light/dark photoperiod. Humidity averaged about 45–50%.

3. Results and discussion

To maintain homeostasis, plants constantly make internal adjustments in response to changes in the external world. The coordination of these internal processes and their balance with the environment are connected to the excitability of plant cells.

Plant cells, and the cells of many other biological organisms, generate an electrical potential that may result in the firing of an electrical current. Electrical impulses can arise spontaneously or result from stimulation. Once initiated, they can propagate to adjacent excitable cells. This change in transmembrane potential creates a wave of depolarization, or action potential, which affects the adjoining resting membrane. Thus, when the phloem is stimulated at any point, the action potential is propagated over the entire length of the cell membrane and along the phloem, with a constant voltage. Once initiated, the action potential has a stereotyped form and essentially fixed amplitude called an “all or none” response [16–26]. The propagation of each impulse is followed by an absolute refractory period during which, the fiber cannot transmit a second impulse.

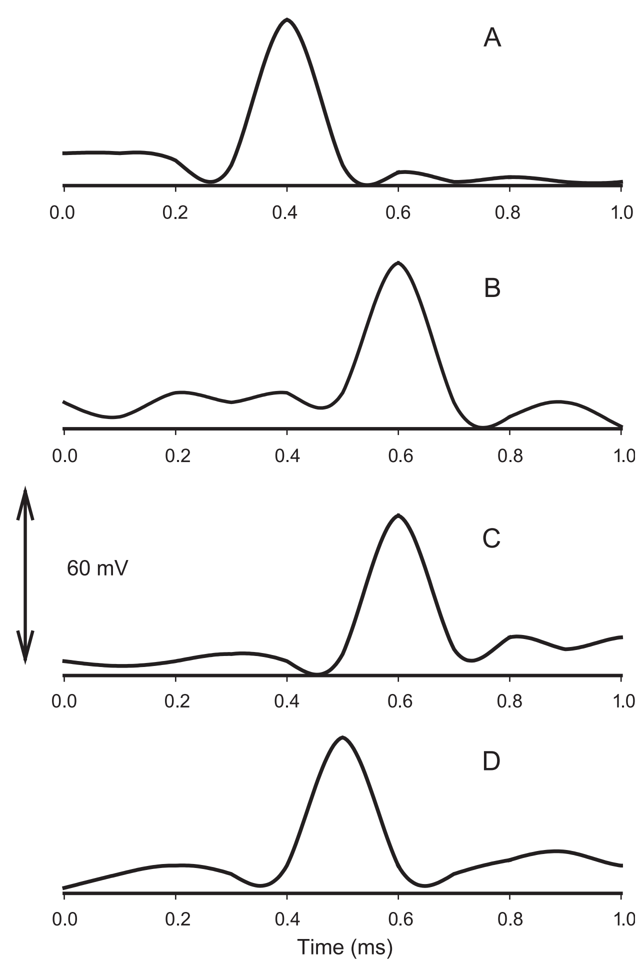

Fig. 2. Action potentials in soybean induced by irradiation at 450 nm (A), 470 nm (B), 671 nm (C), and 730 nm (D) in the direction B as shown on Fig. 1. Irradiance was 8.5 μE/m^{2}s.

The high sensitivity of protoplasm and all cell organelles to any natural and or chemical effect is the basis of excitability. It is important to note that plants can be developed and maintained in a continuously varying environment only if all cells, tissues and organs function in concordance.

The first step in any plant photomorphogenic response is the perception of an incoming light signal. Prior to the actual experiment, the soybean plants were irradiated in direction A (Fig. 1) by white light in a Faraday cage for 2 days with a 12:12-h light/dark photoperiod.

Changing the direction of irradiation from A to B generated action potentials in the plant (Fig. 2). This effect depends on the wavelength of the irradiating light. Irradiation at wavelengths 400–500 and 650–730 nm induced fast action potentials in soybean plants that lasted for approximately 0.3 ms. Irradiation from direction B with wavelengths between 500 and 630 nm did not generate action potentials. The action spectrum of light-induced action potentials is shown in Fig. 3. Irradiating the plant with blue light induced movement of the top of the plants towards the direction of light. This is known as phototropism and is one of the best known plant tropic responses. A phototropic response is a sequence of the four of following processes: reception of the directional light signal, signal transduction, transformation of the signal to a physiological response, and the production of directional growth response. Irradiation between 500 and 600 nm did not induce phototropism.

Electrical signaling at 670 and 730 nm in the soybean plant (Figs. 2 and 3) is caused by phytochromes. Phytochromes provide photosynthetic organisms with the means to detect suboptimal light conditions that direct appropriate responses via changes in growth and development. Light perception by the highly conserved photosensory domain of phytochromes entails double bond photoisomerization of the covalently linked linear tetrapyrolle prosthetic group. This leads to a change in conformation and a change in the biochemical function of the associated protein moiety. The phytochrome is the best characterized plant informational photoreceptor.

The duration of an action potential does not depend on the location of the working electrode, or on the distance between the working and reference electrodes. Action potentials are essential to the expeditious character of response reactions in plants as they respond to external changes. These impulses transfer a signal about the changes of conditions in a conducting bundle from the root system to the point of growth and conversely. Solitary waves function as information carriers in soybean plants due to the impulses generated by changes in environmental conditions [23]. Action potentials are signals caused by the depolarization of the plasma membrane. Mechanical, physical, or chemical external irritants act not only at the place of occurrence, but the electrical excitation they create can also be transferred throughout the entire plant.

The speed of transfer depends on many factors, such as the intensity of irritation, temperature, chemical treatment or mechanical wounding, and is also influenced by previous excitations. The excitation reaction travels from the top of a stem to the roots and vice versa. The transfer of excitation has a complicated character, and is accompanied by an internal change in cells and tissues. The most rapid methods of long-distance communication between plant tissues and organs are bioelectrochemical or electrophysiological signals. The effectiveness of such long-distance communication is evident on account that plants can rapidly respond to external stimuli (e.g., changes in temperature or osmotic environment, plant pathogens, herbivory, insects, irradiation level, wounding, cutting, mechanical stimulation or water availability), and changes can be detected in distant parts of the plant soon after the injury is inflicted.

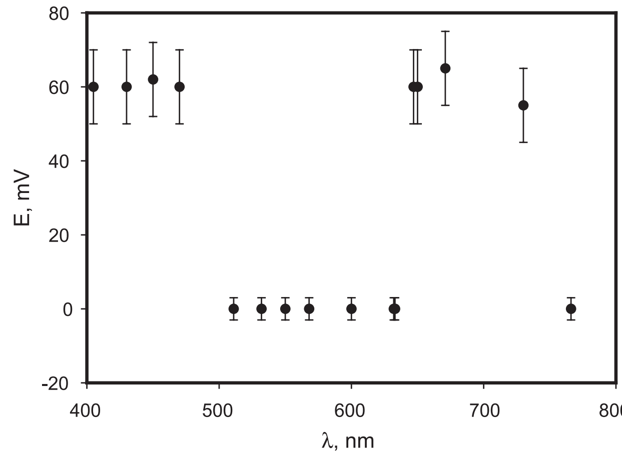

Fig. 3. Action spectrum: dependence of action potential amplitude on wavelength of irradiation.

The automatic measurements of the electrical potential difference can be effectively used in environmental plant bioelectrochemistry. The electrochemical measurements are also useful in the study of molecular mechanisms of ion transport as well as the influence of external stimuli on plants.

Acknowledgements

This work has been supported by the National Science Foundation grant HRD-0043162 and NASA grant NAG8-1888.

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