Boron-induced Bioelectric Field Change in Mung Bean Hypocotyl

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Takuma Tanada
Light and Plant Growth Laboratory, Agricultural Research Service, United States Department of Agriculture, Beltsville, Maryland 20705

ABSTRACT

Low concentrations of boron were found to affect the bioelectric field potentials of hypocotyls excised from 4-day-old mung bean (Phaseolus aureus cv. Oklahoma 612) seedlings grown in darkness and irradiated with red light. The significance of this effect is discussed in relation to the possible role of boron in some membrane function.

Over 50 years ago boron was found to be required in small amounts for normal plant growth (1, 13). Ever since this discovery, the basis for its physiological necessity has eluded the search of investigators. Several hypotheses have been suggested, and these have been reviewed by Gauch (4). Among these suggestions, two have received more attention than the others. One proposal implicates B as facilitating the movement of sugars into cells (5). The other suggests that B is involved in some manner in the growth and development of cells because actively growing cells are affected by its deficiency (12). In this connection, Shkolnik and Kositsyn (11) and Albert (2) have investigated the relationship of B to RNA metabolism.

During some measurements of the bioelectric field of mung bean hypocotyls with an electrometer, we found that the measured field potentials of hypocotyls from seedlings placed in low B water appeared to be different from those of seedlings placed in water containing some B. Because a change in the bioelectric field potential could be due to changes in the electric charges of cell surfaces or of the ionic fluxes through membranes, a B-induced potential change would implicate B with some membrane property. This study was undertaken to obtain supporting data for such a concept. The method used in this study is similar to that used by Jaffe (6) to demonstrate a phytochrome-mediated bioelectric potential change in mung bean roots.

MATERIALS AND METHODS

Mung bean seeds (Phaseolus aureus cv. Oklahoma 612), soaked in aerated deionized water for 8 hr, were germinated in darkness on cheesecloth over aerated deionized water at 25 C. Four days later, representative seedlings were selected for potential measurements. As depicted in Figure 1, a 2.5-cm hypocotyl section was excised 1.0 cm below the hook of a seedling in dim green light. Five hypocotyl sections were placed in a small glass cup with the apical end in 1 ml of solution containing 20 mM CaCl2, 10 mM MgSO4, and 0 to 1 μM H3BO3. The hypocotyl sections were then irradiated for 2 min with red light (1.3 μW cm−2 nm−1 at 660 nm). Green and red lights were isolated with interference filters. After exposure to red light, the hypocotyl sections were left in darkness for 10 min in the B solution. Their ends were then inserted in two capillary tubings (0.5 cm), containing about 20 μl of 10 mM KCl, which were connected with plastic tubing to Ag-AgCl glass electrodes containing 10 mM KCl (Fig. 1). The sensing electrode with the apical end of the hypocotyl was connected to a Keithley vibrating capacitor electrometer (Model 640). Some shielding was used to minimize electrical disturbances. Potential measurements were taken in darkness after the needle stopped drifting 1 to 2 min later. Some measurements were made without exposing the sections to red light. Whenever possible, the use of Pyrex glassware was avoided to prevent B contamination. The glass cup was cleaned for several minutes with 3 M H2SO4, and many washings of deionized water after each usage.

RESULTS AND DISCUSSION

Results of some measurements showing an effect of B on the bioelectric field potentials of mung bean hypocotyls from one lot of seedlings out of 10 lots are presented in Figure 2. Each point is the mean value for 10 seedlings. The results show that when the apical end of a hypocotyl exposed to red light is treated with dilute B solution, its potential is more positive up to a B concentration of about 0.4 μM. This positive response can be detected at B concentrations as low as 0.1 μM. Unreported data suggest that above 1 μM B the potentials decline to values which are about the same as those of the controls. Statistical analysis indicates that the B values are significantly different (P = 0.05) from that of the control up to 0.6 μM B. The finding that B appears to elicit a positive response over only a narrow concentration range—0.1 to 0.6 μM—could indicate its toxic properties at the higher concentrations.

As shown in Figure 3, hypocotyl sections not exposed to red light do not show any significant effect of B on the bioelectric field. These results and the lack of response to B at high B concentrations rule out the possibility that the B effect is due to an artifact such as an electrochemical effect on the electrode.

The significance of the electric field around plant tissues is still unresolved (10). However, the recent measurements of Jaffe (6) and of Newman and Briggs (8) of rapid phytochrome-induced changes in the bioelectric field suggest that the field is associated with some important constituent of plant cells.

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0.2

0.3

0.4

0.5

0.6

0.7

0.8

0.9

1.0

capillary tubing

plastic tubing

KCl

KCl

Ag - AgCl

Ag - AgCl

electrometer

interference filter

H2O

lamp

hypocotyl section

+ B

KCl

FIG. 1. Diagram of the experimental arrangement used to measure the bioelectric potentials of mung bean hypocotyls. Hypocotyl sections were excised in dim green light from 4-day-old seedlings, exposed to red light in B solutions, and placed as shown, between ends of two glass electrodes with 10 mM KCl.

FIG. 2. Effect of B concentrations on the bioelectric field potential of mung bean hypocotyls exposed to red light. Each point is the mean value of measurements from 10 seedlings. The length of the vertical lines represents the standard deviation of each mean.

which is involved in phytochrome-mediated responses. The discovery of Fondeville et al. (3) that leaflet movement of Mimosa is under phytochrome control and is due to K fluxes (9) implicates the plasmalemma as the constituent. If changes in the bioelectric field are caused by modifications of some

membrane components, the B effect could be due to its interaction with them to maintain structural integrity. These components, such as glycoproteins, would be rich in OH groups that can form complexes with B. Since the membrane is now believed to play more than a passive role in the growth and development of a cell (7), any impairment of its structure could result in abnormal growth. Hence, if B is a necessary component for membrane integrity, a deficiency of B would lead to inhibition of growth.

Since it is possible that the results reported could be due to a nonspecific effect of B, it would be desirable to show the necessity of B for some cellular processes such as ion uptake or phytochrome responses. With this in mind, attempts are being made to see whether B is required for phytochrome-mediated bioelectric field changes. This approach appears promising inasmuch as the B effect appears to be associated with some response of the hypocotyls to red light.

LITERATURE CITED