



Alleviating effect of extremely low frequency pulsed electric field on drought damage of maize seedling roots



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ABSTRACT

To investigate the effect of extremely low frequency pulsed electric field (ELF-PEF) on drought resistance of maize seedlings, drought stress was formed by PEG-600 solutions with osmotic potential of -001 MPa. Under such condition, ELF-PEF with electric field intensity 200 kV/m, pulse width 80 ms and frequency 1 Hz was applied to maize seedlings. The changes of dry weight of root, superoxide dismutase (SOD), malondialdehyde (MDA) and ultra-weak photon emission were recorded.

It was found that the dry weight of root increased, activity of SOD was greater; on the other hand, content of MOD decreased. The results indicated that the treatment of ELF-PEF promoted the growth of root. In addition, the ability of drought resistance and removal of free radicals were both enhanced. The damage of drought on root cells was alleviated. The ultra-weak photon emission was also analyzed, both spontaneous and induced biophoton emission increased, which showed that treatment by ELF-PEF improved the ability of self-organization and respiration metabolism of root cells.

1. Introduction

Root is important metabolic organ which absorbs water and nutrients to supply energy to the plant. Besides, it fixes the plant to keep it stable. Yield and growth of plant are closely related to the root. Root was affected at first when drought stress applied, throughout the period, drought causes damage to the root, thus root is closely related to the drought resistance of plant [28,32–34].

Previous studies showed that drought resistance of plant is enhanced by applying electrostatic field, which is beneficial to germination of seeds by stimulating the generation of physiological active substance and enzymes [2–4,15,32–34]. It was also proved that the activity of protective enzymes in germinating seeds and seedling such as SOD were promoted by electrostatic field, while the increases of malondialdehyde (MDA) content and acid phosphoesterase activity are limited. Those results indicated that electric field treatment applied to developing seeds could reduce cell membrane sensitivity to drought stress and process of peroxidation and phospholipid de-esterification was alleviated. Thus stability of cell membrane under drought stress was maintained, which increases the drought resistance [12,25,36]. However, there are few research on root system compared with cell membrane.

Current was induced by pulsed electric field inside the cell, induced current and motion of dipole will change the rate of chemical reaction

inside the cell which causes change of mechanism of chemical bonds between molecules. Through making change of structure of protein, it affects the biosystem. More important, reversible electroporation of cell membrane occurs, when ELE-PEF was applied which changes the permeability of cells [37]. Electrical potential coupling resonance takes place due to similarity between frequencies of pulsed electric field and potential fluctuation of cell membrane [40], as a result electric field of cell membrane was influenced significantly, such fluctuation plays essential role in key physiology process of plant, for instance, respiration metabolism, photosynthesis, water absorption and variation of stomatal conductance [6,19]. Based on these, in this paper, maize seedlings were used as material, osmotic stress was formed by PEG-600 solution, low frequency pulsed electric field (ELF-PEF) was applied to investigate its effect on root cells. It was found that damage of drought on root was alleviated effectively which showed the application potential of ELF-PEF to enhance the drought resistance of plants.

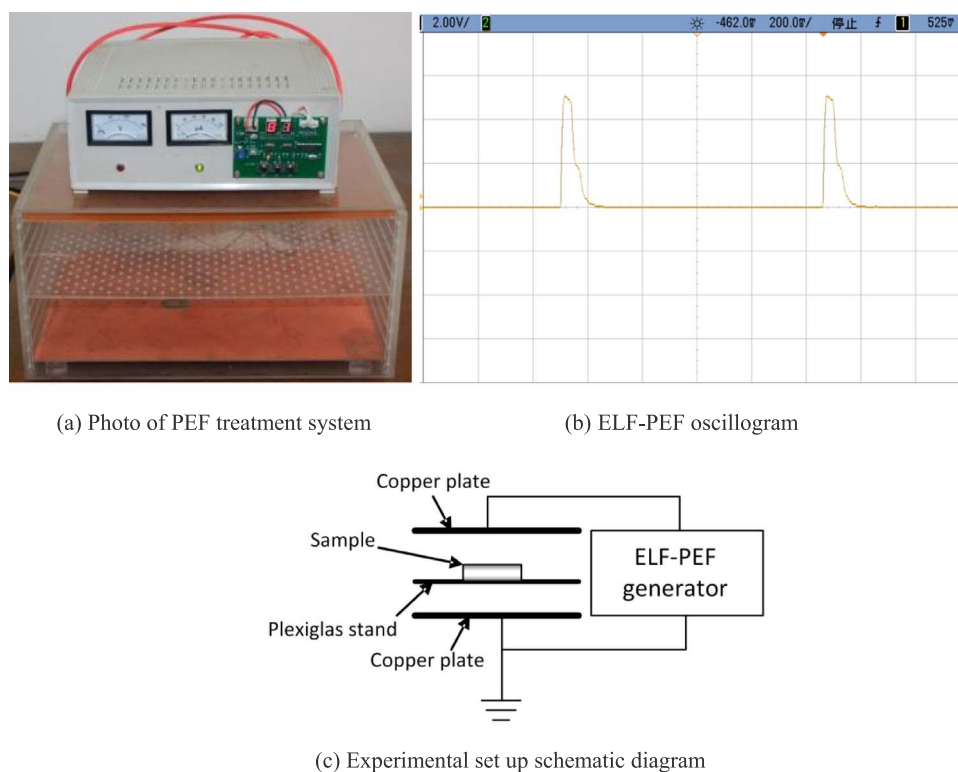
2. Materials and methods

2.1. Materials and cultivation

Maize seeds YanDan8 with similar outlook were selected as material, they were sterilized with 0.1% HgCl_2 solution after cleaned by distill water, then washed carefully again. Then seeds were

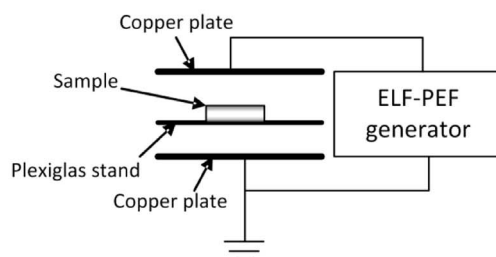
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(a) Photo of PEF treatment system

(b) ELF-PEF oscillogram



(c) Experimental set up schematic diagram

Fig. 1. Photo of ELF-PEF treatment system, ELF-PEF oscillogram and experimental set up schematic diagram.

submerged in water for 24 h, during the period, the temperature was kept as 24 °C then germinated in box with constant temperature 30 °C for two days.

2.2. Extremely low frequency pulsed electric field treatment

Self-made ELF-PEF system contains two copper plates, a perspex stand, and ELF-PEF generator [38]. Photo of the system is shown by Fig. 1a, as Fig. 1b shows the oscillogram of electric field with field intensity 200 kV/m vertically downwards, pulse width of 80 ms, frequency of 1 Hz. 400 seeds were chosen as sample, they were divided into treated group and control group, each group contains 200 seeds. Seeds of treated group were placed evenly in a culture dish, at the bottom of which there was wet filter paper. The culture dish then was positioned in the center of perspex stand. After that ELF-PEF was applied for 30 min on same period every day. The operation was repeated for 5 days.

2.3. Drought stress

According to previous study [8], nutrient solution of both group was removed, then PEG-600 solution with osmotic potential -0.1 MPa was added to form drought stress, measurements were done on different periods.

2.4. Measurement of dry weight of seedling roots

Five maize seedlings were chosen from both groups respectively, they were put in paper bag after their leaves were cut off, to avoid mixture, each paper was labeled with name to show which group the seeds came from. When the temperature of heat oven reached 100 °C, two bags were put inside for 10 min. Then temperature of oven was turned down to 80 °C, wait until there was no change on dry weight. Bags were turned out after their temperature dropped to room temperature. Their weights were measured with electronic balance. Then repeated 3 times and take the average value.

2.5. Determination of MDA content

MDA content was measured with thiobarbituric acid chromatometry [27]. The unit is $\mu\text{mol/g}$ (fresh weight). During the growth of maize seedlings, maize seedlings with uniform growth vigor were selected from the control group and the treated group respectively every 24 h. The roots were cut off 2 g from multiple plants of each group. Each of them was added with 5 ml precooled 5% trichloroacetic acid (TCA) and then was ground into homogenate, which was centrifuged at 4000 r/min for 10 min. 2 ml supernatant was obtained and added with 2 ml 0.67% thiobarbituric acid (TBA). After mixing, the solution was boiled for 15 min in 100 °C water bath, and was then centrifuged at 4000 r/min for 10 min after cooling to room temperature. The supernatant was obtained for the absorbance test at 450 nm, 532 nm and 600 nm. There were 3 repeats in each group. The determination of MDA content was conducted continuously for 5 days.

2.6. Determination of superoxide dismutase (SOD) activity

Activity of SOD in maize seedlings roots was measured with nitroblue tetrazolium (NBT) method [23,29]. One unit of SOD activity (U) means ability to prevent 50% deoxidizer of NBT. Unit of activity is U/g (fresh weight). The measurements were done 3 times for each group every day. The operation was repeated for successive 5 days, then average value was obtained.

2.7. Measurement of spontaneous biophoton emission

The instrument used to measure spontaneous biophoton emission is BPCL-T-Q ultra-weak biophoton emission analyzer manufactured by Institute of Biophysics of Chinese Academy of Sciences. It adopts CR131 photomultiplier (PMT) manufactured by Hamamatsu Photonics company as light sensor, whose quantum efficiency is 20–40% from 300 nm to 400 nm. Relevant measurement parameters were set in accordance with the method mentioned in references [8]. 5 maize seedlings were chosen for each group, 2 g of roots from seedlings were cut down,

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