

PHOTOSYNTHETIC ACTION SPECTRA OF THE ENERGY STORAGE
IN BUNDLE SHEATH CELLS OF ZEA MAYS

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Photosynthetic action spectra ($F = \frac{\phi \cdot r}{\lambda} \alpha$), (Carpentier, R., Larue, B. and Leblanc, R. (1984) Arch. Biochem. Biophys. 228, 534-543.), from 400 to 750 nm were studied in bundle sheath cells of maize. Photosynthetic action spectra in the presence of 10 mM ascorbate or 4 mM ribose-5-phosphate were increased and shifted through all the spectra. After the addition of 10 μ M DCMU photosynthetic action spectra were remarkably diminished. On the basis of these results we suggest that the role of PSII in BS chloroplasts will be to prevent the overoxidation of PSI. It appears that in addition to PSII some endogen electron donor may regulate the PSI activity in bundle sheath cells.

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Thermal dissipation of absorbed light energy in photosynthesis can be measured with photoacoustic spectroscopy (1). A part of absorbed light energy not lost in thermal dissipation can be stored as chemical intermediates. Determination of such a type of energy storage is defined through the comparison of the acoustic signal from a photosynthetically active sample with that of an equivalent photochemically inactive reference. The difference between those two acoustic signals represents photochemically stored energy, usually referred to photochemical loss (2)

ABBREVIATIONS: BS, Bundle sheath; DCMU, 3-(3',4'-dichlorophenyl)-1,1-dimethylurea; HEPES, N-2-hydroxyethyl piperazine-N'-2 ethane-sulfonic acid; PSI, photosystem I; PSII, photosystem II; R₅P, ribose-5-phosphate; Tricine, N-[2-hydroxy-1,1-bis (hydroxy-methyl) ethyl] glycine.

To provide a photochemically inactive equivalent reference it is necessary to illuminate the reference sample with a strong light intensity (actinic beam) which saturates photochemistry (3,4).

Recently photoacoustic spectroscopy was used to monitor energy storage in bundle sheath (BS) cells of maize. It was demonstrated that during photochemical activity of BS cells some endogen electron donor participates in the reduction of PSI and in the energy storage process (5). It is accepted that BS cells chloroplasts have very low PSII activity and that major photochemical reactions are occurring at PSI (6, 7). It is postulated that the role of PSI in BS chloroplasts is to provide cyclic electron transport via plastoquinone and cytochrome f and consequently to induce ATP formation required for the carbon fixation (8). However, the role of PSII activity in BS cell chloroplasts is still unclear and contradictory (9). We report here photosynthetic action spectra of BS cells using photoacoustic spectroscopy.

MATERIALS AND METHODS

Maize (*Zea mays*) BS cells were isolated by a combined mechanical maceration-filtration technique (8, 10). The photoacoustic spectrometer was previously described (5). In the photoacoustic measurements, the amount of 10 mg chl of BS cells was resuspended in 3 ml of resuspending medium (with or without additives), then the sample was layered on a nitrocellulose filter. The photoacoustic spectra were normalized to the carbon black reference. Actinic light intensity was 30 mW/cm² and modulated light was to an average of 1 mW/cm². Chlorophyll concentration was determined by the Arnon method (11).

RESULTS AND DISCUSSION

Fig. 1a shows that the photoacoustic signal of BS cells was increased upon addition of nonmodulated background actinic light and returned to the initial level after removal of actinic light beam. Actinic light induces in BS chloroplasts the closing of photosystems electron traps and consequently the higher portion of modulated absorbed light energy is converted into heat, and this resulted in the enhancement of photoacoustic signal (12, 14). The difference between the photoacoustic signals with and without actinic light was used as the parameter to analyse

the energy storage process in BS cells chloroplasts. The parameter for spectrophotocoustic detection of energy storage is defined as the relative photochemical loss (or energy storage) expressed by the equation:

$$\phi_r' = \frac{Q_m - Q_c}{Q_m} \cdot 100 \quad (12, 13).$$

Q_m represents the maximum acoustic signal induced by the background actinic light. Parameter Q_c represents the control signal obtained by the modulated light. The photosynthetic effect (F) was expressed as $F = \frac{\phi_r' \alpha}{\lambda}$, where α is the optical absorption coefficient of the sample and λ represents the wavelength of modulated light (5). Light modulation frequency was 75 Hz as described earlier (5). Since we monitored photochemical loss which occurs in 1 ms according to modulated light pulse (5), we supposed that plastoquinone can correspond to the involved intermediary in the energy storage process (15).

Fig. 1b shows photosynthetic action spectra of BS cells in the pre-

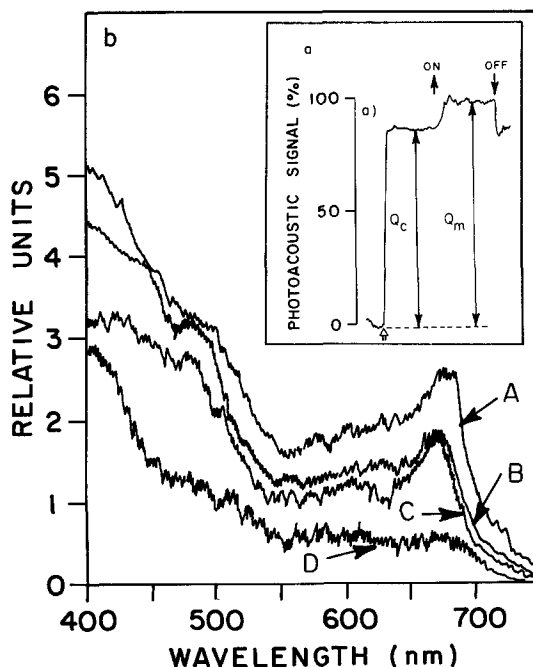


FIGURE 1. a. Photoacoustic signal from BS cells. Short dark arrows indicate the onset and terminating of the actinic beam, open arrow indicates the onset of the modulated light of 700 nm (Details in the text).
b. Photosynthetic action spectra of BS cells incubated with 10 mM ascorbate (A), 4 mM R_5P (B), no additives (C) and with 10 μM DCMU (D). Ordinate represents relative units of photosynthetic effect (F). (Details in the text).

sence of additives such as ascorbate, R_5P , DCMU and with no additives (control sample). We found that photosynthetic effect was increased at all wavelengths in the presence of ascorbate and R_5P compared to control sample. We noticed that photosynthetic action spectra, upon addition of ascorbate or R_5P , are shifted to longer wavelength through all spectra. However the presence of DCMU diminishes the value of the spectra by 40% compared to the control sample. Ascorbic acid as an electron donor initiates electron flow via PSI in BS chloroplasts and therefore photosynthetic effect is increased. Earlier it was confirmed that the addition of R_5P to chloroplasts induces enzymatically its conversion to ribulose 1,5 biphosphate and stimulates the cyclic electron flow via plastoquinone (16). Increased photosynthetic action spectra in the presence of R_5P supports this interpretation. In the presence of DCMU some existing PSII activity was inhibited (17) and this affected PSI electron transport in BS chloroplasts.

We concluded that PSI activity in BS cell chloroplasts is partially supported by PSII activity. We presume that the role of PSII in BS chloroplast is to prevent the overoxidized state of electron carries associated with PSII activity. In addition to PSII some endogen electron donor may support the PSI activity in BS cells.

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