ON THE PHOTOELECTROCHEMICAL EFFECT IN SOLID CHLOROPHYLL AND CHLOROPHYLL–PROTEIN FILMS

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Received 9 April 1973
Revised version received 1 June 1973

1. Introduction

The photogalvanic effect in solid films of chlorophyll and its analogues discovered by Evstigneev and Terenin [1, 2] is due to a combination of photochemical and semiconductive phenomena. Interest in the investigation of this effect [3–5] may be explained by the fact that the notions regarding the mechanism of photogeneration and subsequent separation of charges in condensed chlorophyll films in contact with donor or acceptor admixtures in an electrolyte can, with certain reservations, be applied to the primary photochemical processes of photosynthesis.

Since the high quantum efficiency of the primary photoprocesses of photosynthesis is, to a large extent, determined by the structural organization of chlorophyll and its environment, it seemed to be advisable to investigate the influence of the state of chlorophyll, and in particular its link with protein, on the photopotential generated in films of chlorophyll and other pigments of similar structure.

2. Materials and methods

Chlorophyll *a* extracted from nettle leaves by a familiar procedure [6] and a synthetic pigment, Mg-phthalocyanine whose molecular structure is similar to that of chlorophyll and which has been employed frequently in previous works for modelling chlorophyll reactions [7, 8], were used in the experiments. The purity of the pigments was checked spectroscopically.

Solid chlorophyll films deposited on a platinum (~ 0.5 cm^2) or quartz (~ 3 cm^2) plate were obtained by evaporation of a drop of an ether solution of the pigment; Mg-phthalocyanine films were obtained by vacuum sublimation [1].

Chlorophyll–protein films were prepared in the following way. An equal volume of acetone solution of chlorophyll *a* was added during continuous shaking to an aqueous solution of human serum albumin ($\sim 10^{-4}$ M) ("Reanal" reagent). A drop of the resulting mixture was deposited on a quartz or platinum plate; the plates were then placed in a exsiccator with $CaCl_2$. The films obtained after evaporation of the solvent were homogeneous and their immersion into a 90% ethanol solution for several hours did not reveal any extraction of the chlorophyll. This indicated that the pigment was comparatively strongly bound to the protein.

In the measurements the films were immersed in a 2 N KCl aqueous solution. A non-illuminated calomel or platinum electrode served as the reference electrode.

The photopotential was measured with a LPU-01 pH-meter with an input resistance of 10^{12} Ω. Absorption spectra were recorded with a Specord spectrophotometer.

Action spectra were measured with an apparatus which automatically equalized the energy or quantum flux in the 220–1000 nm spectral range.

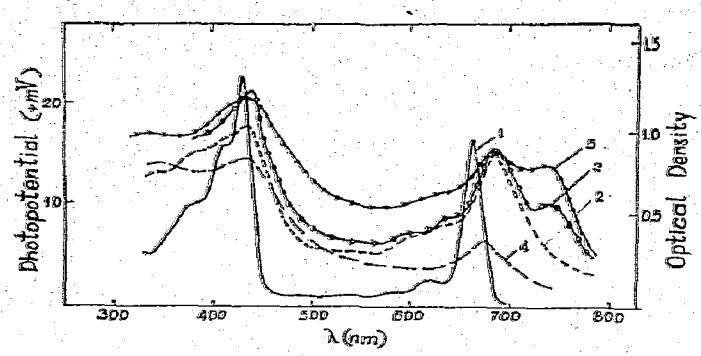

Fig. 1. Absorption spectra and photopotential action spectra of chlorophyll *a* films. 1) Absorption spectrum of chlorophyll *a* solution in ether. 2) Absorption spectrum of a "nonactivated" film on a quartz plate. 3) Absorption spectrum of an "activated" film. 4) Photopotential action spectrum of a "nonactivated" film. 5) Photopotential action spectrum of an "activated" film. The ordinates of curves 1, 2 and 3 are plotted along the right hand side axis, those for curves 4 and 5 – along the left axis.

3. Results and discussion

In a number of previous investigations it has been shown that the crystalline state of chlorophyll is characterized by the appearance of an absorption maximum in the 730–740 nm range; the maximum in the 670–680 nm range, on the other hand, can be ascribed to the amorphous state of the pigment [9–11].

The transition from the amorphous to crystalline state of chlorophyll may be caused by the interaction between the pigment and polar molecules (in our experiments this could be the result of exposure to water and ethanol vapour over a period of several hours). A consequence of such treatment is a significant increase of the photopotential.

The absorption and action spectra of the positive photopotential for the ordinary and "activated" (as described above) solid chlorophyll *a* films are shown in fig. 1. The curves show that the photopotential action spectra for the amorphous films (curve 4) and for those containing crystalline chlorophyll correlate with the respective absorption spectra (curves 2 and 3). This indicates the participation of the excited states of the pigment itself in generation of the photopotential.

The higher photopotentials in films containing crystalline forms of chlorophyll signify that in these forms the conditions are more favourable for efficient photogeneration and separation of charges.

The most probable mechanism of production of the photopotential is the following: the excited states created by light (possibly excitons) migrate in the film and split up into electrons and holes at the dissociation centers. Under aerobic conditions such centers at the film–electrolyte interface may be identified as adsorbed electron acceptor (oxygen) molecules. Inside the film they may be microcrystal imperfections, imbedded impurity polar molecules, etc.

However, the fact that removal of dissolved oxygen from the electrolyte results in an appreciable decrease of the photopotential and frequently even in its disappearance, apparently signifies that it is precisely the surface dissociation centers which play the most essential role. This is also confirmed by the fact that the effect of oxygen removal is practically observed only for non-treated pigment films. If the pigment film is exposed to an ethanol solution of benzoquinone for several hours (one of the "activating" procedures) then besides a general increase of the photopotential, the removal of oxygen ceases to significantly affect the photopotential.

Experiments on the temperature dependence of the photopotential of Mg-phthalocyanine films, which mechanically and thermally are more stable compared to chlorophyll films, confirm the important role of the adsorbed acceptor. An increase of the electrolyte temperature from 10°C to 50°C results in a decrease of the photopotential by more than four times and this can be ascribed to a decrease of the concentration of adsorbed oxygen on the pigment film surface with increase of temperature.

Artificial chlorophyll–protein films may be regarded as a rough model of the state of affairs prevailing *in vivo* [12, 13]. In our experiments illumination of chlorophyll *a*–albumin films deposited on a platinum electrode in 90% ethanol (fig. 2A) induced a pronounced increase of the positive potential. The stationary photopotential of such films may reach 150–200 mV which considerably exceeds the values for non-activated chlorophyll *a* films under identical experimental conditions.

Removal of dissolved air oxygen leads to an almost complete disappearance of the photopotential which may appear again after subsequent admission of oxygen (fig. 2A). Thus interaction with the electron acceptor (oxygen) is also of great importance for photopotential generation in chlorophyll–protein films.

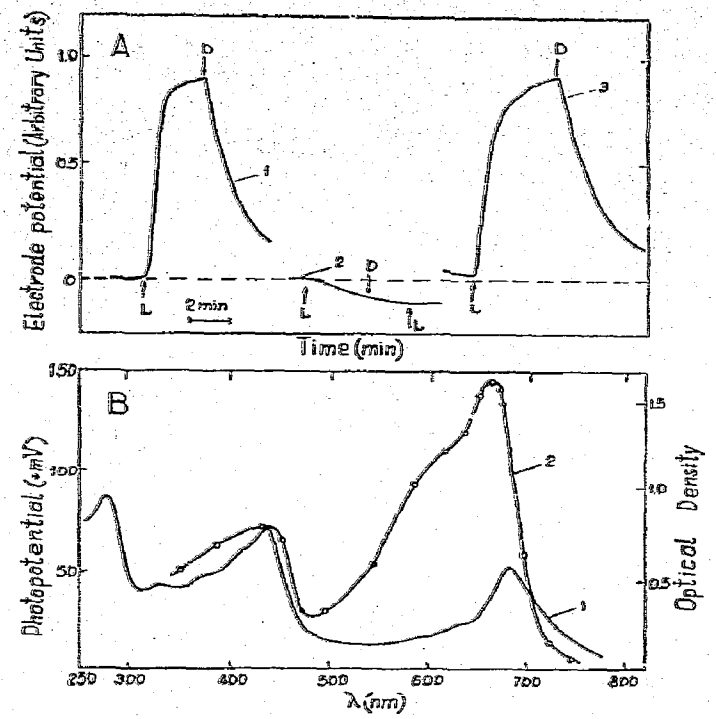

Fig. 2. A) Variation of the electrode potential on illumination of chlorophyll–protein films and after switching off of the light. 1) In the presence of air. 2) After evacuation of air. 3) After admission of air. L = Light on; D = light off. B) 1 = Absorption spectrum of chlorophyll–protein film. 2 = Photopotential action spectrum for chlorophyll–protein film. The ordinates of curve 1 are plotted along right hand side axis, those of curve 2 – along the left axis.

Both major absorption bands are manifest in the photopotential action spectrum (fig. 2B). As can be seen from the figure, the action spectrum markedly differs from the absorption spectra of either chlorophyll *a* or chlorophyll *a*–protein films. The peaks are broader and their ratio is different.

Further investigations are required for elucidation of the nature of the chlorophyll–protein complex.

In conclusion it may be stated that an increase of the degree of crystallization of chlorophyll films and the interaction between chlorophyll and protein may appreciably enhance the efficiency of photogeneration and separation of charges in the films and hence the magnitude of the photopotential.

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