Thylakoid and Grana Formation During the Development of Pea Chloroplasts, Illuminated by White, Red, and Blue Low Intensity Light

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Received January 21, 1986
Accepted May 20, 1986

Summary

We analyzed the formation of thylakoids and grana during the development of pea chloroplasts, illuminated by white, red and blue low intensity light. The total length of granal and intergranal thylakoids, and the length of granal thylakoids per unit area of plastid section were measured. Initially the greatest increase in length of granal thylakoids and the highest incidence of grana with large thylakoid content occurred in red light. On the other hand, with illumination times of over 12 hours blue light appeared to be more efficient in stimulating grana formation and thylakoid growth.

Keywords: Chloroplast development; Grana formation; Pisum sativum; Ultrastructure.

1. Introduction

Etiolated pea seedlings illuminated by white light develop into chloroplasts with thylakoid systems consisting of granal and intergranal thylakoids. Grana are also formed in high and medium intensity red light, and also after short treatment with high intensity red light (KLEIN et al. 1964, BERRY and SMITH 1971, TREFFRY 1973). According to BRADBEE et al. (1974) illumination treatments with red light; or light containing a red component, provide a rapid stimulus to thylakoid development, while blue and far-red light alone are inhibitory.

Data on the influence of red and blue light on grana formation are controversial. According to VOSKRESIENSKA (1979), VLASOVA et al. (1971) and VETTERMAN (1973) blue light is more effective in stimulating grana formation. The opposite conclusion was reached by BUSCHMANN et al. (1978), GUILLOT-SALOMON et al. (1978), LICHTENTHALER and BUSCHMANN (1978), KASEMIR (1979), TREMOLIÈRES et al. (1979). Blue light was also less efficient than white light in stimulating the formation of grana (DUYSEN et al. 1985).

The present work was undertaken in order to resolve this controversy. We therefore measured the total length of thylakoids, the length of granal thylakoids per unit area of plastid section and counted the number of grana with different numbers of thylakoids. This analysis was performed after illumination by white, red, and blue low intensity light.

This paper continues our studies on the effects of light quality on the development of pea chloroplasts (MOSTOWSKA 1985, 1986).

2. Material and Methods

Seeds of Pisum sativum L. var. Bördi sown in pots with sand were placed in darkness at 25 °C. Eight-day old, etiolated seedlings were illuminated by continuous white, blue or red light for 2, 4, 8, 12, 24, 48, and 72 hours. White light was provided by halogen lamps. Coloured light was obtained with the help of blue and red filters (BAUSH and LOMB) with maximal transmissions in the range 465–480 nm and 630–660 nm respectively (Fig. 1). Intensities were so chosen that the numbers of photons were comparable for each colour: 0.92 W/m² for red, 0.98 W/m² for white, and 1.25 W/m² for blue light.

Experiments were performed at least twice for each illumination time. At least 4 seedlings were illuminated per experiment. Thus the data are averages of at least 8 seedlings.

Samples for electron microscopy were always taken from the second leaf above the cotyledons. Pieces about 1 mm² in area were cut from...
the broadest part of this leaf near the main vein. Material was fixed in 2.5% glutaraldehyde in 0.05 M cacodylate buffer at pH 7.4 for 2 hours, washed in buffer and placed in cold 2% OsO₄ in the same buffer overnight. Manipulations were carried out in the dark with dim green light. Material dehydrated in a ethanol-propylene oxide series was embedded in epon (Luft 1961) and cut on an LKB ultramicrotome. Sections were stained with uranyl acetate and lead citrate (Reynolds 1963) and examined under a Tesla BS 500 electron microscope.

Fig. 1. Transmission diagram of the filters

After each illumination treatment 50–140 plastid cross sections were analyzed. Two or three areas of 1 μm² were chosen at random from each plastid cross section. Within such areas the length of granal thylakoids and the total length of granal and intergranal thylakoids were measured with the help of a curvometer (Brangeon 1973). The areas thus chosen provide a representative sample of plastid cross section area. The thylakoid length per 1 μm² is taken as a measure of stromal thylakoid content. The number of thylakoids per granum was also determined.

The data were analyzed by standard statistical methods.

3. Results

Pea etioplasts have a poorly developed thylakoid system (Fig. 2). During the first 2 hours of treatment by red, white, and blue light a rapid increase in the total length of thylakoids per 1 μm² area of plastid section took place (Fig. 2). After further illumination this increase was slower, and after 12 hours the increase in total thylakoid length began to depend on the light quality (Fig. 2). After 24 hours length was greater in blue light than in red light. After 24 hours the differences were even greater, and at the end of the experiment the greatest thylakoid length was attained in blue light and the lowest found with red light (Fig. 2).