

Differential photoperception by different phytochromes and their down-stream responses

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I. Diverse action spectra for phytochrome-dependent responses in plants:

Since early time, photomorphogenesis has been classified into 3 categories in terms of fluence required; i.e. very low fluence reaction (VLFR), low fluence red/far-red reversible reaction (LFR), and high irradiance reaction (HIR). Upon the action spectra for red/far-red reversible LFR (Borthwick et al 1952), phytochrome was discovered as the photoreceptor for LFR (Butler et al 1959). Phytochrome shows unique photoreversible spectral change between red light (R)-absorbing form, Pr, and far-red light (FR)-absorbing form, Pfr, speculating that Pr is biologically inactive while Pfr active. However, spectrophotometrically measured amount of Pfr *in vivo* was not matched with degree of biological responses in most cases of photomorphogenesis (Furuya 1993)¹.

Phytochrome gene family was discovered by Sharrock and Quail (1989), then the research in this field was focused to find which phytochrome regulates what photo-response(s). Action spectra for VLFR of seed germination (Shinomura et al 1996)² and Cab gene expression (Hamazato et al 1997) were determined using phytochromes A (PhyA)- and B (PhyB)-deficient mutants, finding that PhyA regulates VLFR while PhyB and other type II phytochromes control LFR. In the both cases, the phototransformation of Pr to Pfr induces the responses.

The recent studies on phytochrome-deficient mutants showed that FR-mediated HIR resulted from PhyA while R-mediated HIR from PhyB (Quail et al 1995), but the mode of photoperception was obscure. Shinomura et al (1999) succeeded to separate the elementary process of FR-HIR in *Arabidopsis* using intermittent irradiation with FR pulse light, and determined action spectra for this process at Okazaki large spectrograph. The results indicated that a) PhyA in Pr form synthesized newly in tissues and that in photoconverted Pfr form are not active in inducing HIR signal, but the signal is transmitted during phototransformation from Pfr to Pr, b) the FR pulse is required every few minutes for many hours to induce FR-HIR, suggesting that the signal is short-lived, and c) the effect of FR pulse is repeatedly photoreversible by R pulse if given subsequently after FR pulse. This mode of photoperception for FR-mediated HIR is quite different from that of VLFR by PhyA or LFR by PhyB, indicating that PhyA acts two essentially distinct roles in photoperception upon developmental stages of plants and environmental light condition (Table).

II. Down-stream responses:

Phytochromes regulate diverse processes at molecular, cellular and organ levels¹. In this lecture, I talk about our recent works on a) photoregulation of gene expression and b) photo-induced migration of cytosolic phytochromes to nucleus.

In the past two decades, the photoregulation of gene expression in higher plants was studied only on very limited number of genes in the genome. Hence we have recently developed a fluorescent differential display (FDD) technique³ to perform a large-scale screening for identification of a set of phytochrome-regulated genes in *Arabidopsis thaliana*. Seedlings of the wild-type and *phy*-mutans at different age were irradiated with diverse fluence of FR and R, and their total RNA extracts were applied to FDD analysis for finding any VLF-, LF- and HIR- specific gene expression. We have isolated and identified several genes, of which expression is regulated by PhyA. I will discuss the transcriptional hierarchy in down-stream responses of VLFR, LF and FR-mediated HIR.