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BLR-1 and BLR-2, key regulatory elements of photoconidiation and mycelial growth in *Trichoderma atroviride*

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In fungi, phototropism, the induction of carotenogenesis and reproductive structures, and resetting of the circadian rhythm are controlled by blue light. *Trichoderma atroviride*, a fungus used in biological control, sporulates in a synchronized manner following a brief pulse of blue light. Due to its apparent simplicity, this response was chosen for pursuing photoreceptor isolation. Two genes were cloned, blue-light regulators 1 and 2 (*blr-1* and *blr-2*), similar to the *Neurospora crassa* white-collar 1 and 2, respectively. The BLR-1 protein has all the characteristics of a blue-light photoreceptor, whereas the structure of the deduced BLR-2 protein suggests that it interacts with BLR-1 through PAS domains to form a complex. Disruption of the corresponding genes demonstrated that they are essential for blue-light-induced conidiation. *blr-1* and *blr-2* were also shown to be essential for the light-induced expression of the photolyase-encoding gene (*phr-1*). Mechanical injury of mycelia was found to trigger conidiation of *T. atroviride*, a response not described previously. This response was not altered in the mutants. A novel effect of both red and blue light on mycelial growth was found involving another light receptor, which is compensated by the BLR proteins.

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INTRODUCTION

Species of the common soil fungus *Trichoderma* are used for biocontrol of a variety of phytopathogenic fungi (Papavizas, 1985). *Trichoderma atroviride* is used as a photomorphogenetic model due to its ability to conidiate upon exposure to light. In total darkness, *T. atroviride* grows indefinitely as a mycelium provided that nutrients are not limiting. However, nutrient deprivation and light trigger the development of specialized asexual reproductive structures (conidia). A brief pulse of blue light (400–480 nm) given to a radially growing colony in a Petri dish induces synchronous sporulation. A ring of conidiophores bearing green conidia is produced at what had been the colony perimeter at the time of the light pulse (Gressel & Galun, 1967). The first event induced by light is a fast, first-order, photochemical reaction that does not require the presence of molecular oxygen and is independent of temperature. The photoinduction is 'remembered' while the culture is maintained in conditions that do not allow cellular growth (cold or absence of oxygen). When growth is resumed, under optimal conditions, the colony conidiates (Gressel *et al.*, 1975; Horwitz *et al.*, 1984a). According to the Bunsen–Roscoe law of reciprocity, a given quantity of photons could be delivered in pulses of different duration but the final response should be the same. A deviation from this rule indicates the participation of more than one photoreceptor or points to complexities of a single photoreceptor system such as photoreceptor recycling. For *T. atroviride* photoconidiation, reciprocity holds for pulses of blue light lasting from nanoseconds to minutes, indicating that photoconidiation is triggered by a single receptor system that is neither recycled to the photoreceptive form nor counted by enzymic processes during or immediately following irradiation (Horwitz *et al.*, 1990). Upon exposure to blue light, changes in membrane potential and in ATP levels, and a transient biphasic oscillation in intracellular cAMP levels, are observed (Gresik *et al.*, 1988). Exogenous cAMP promotes sporulation in the dark (Berrocal-Tito *et al.*, 2000; Nemcovic & Farkas, 1998), and a pulse of blue light results in the activation of adenylyl cyclase (Kolarova *et al.*, 1992).

A second light response in *T. atroviride* is the regulation of the expression of the photolyase gene *phr-1*. Blue light and

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The GenBank/EMBL/DDBJ accession numbers for the sequences reported in this paper are AY628431 for *blr-1* and AY628432 for *blr-2*.