HHS Public Access

Author manuscript

Environ Microbiol. Author manuscript; available in PMC 2017 January 01.

Published in final edited form as:

Environ Microbiol. 2016 January ; 18(1): 5–20. doi:10.1111/1462-2920.13055.

Seeing the world differently: variability in the photosensory mechanisms of two model fungi

Arko Dasgupta[1,±], **Kevin K. Fuller**[1,±], **Jay C. Dunlap**[1,*], and **Jennifer J. Loros**[2,*]

[1]Department of Genetics, Geisel School of Medicine at Dartmouth, Hanover, NH, USA

[2]Department of Biochemistry, Geisel School of Medicine at Dartmouth, Hanover, NH, USA

Summary

Light plays an important role for most organisms on this planet, serving either as a source of energy or information for the adaptation of biological processes to specific times of day. The fungal kingdom is estimated to contain well over a million species, possibly ten-fold more, and it is estimated that a majority of the fungi respond to light, eliciting changes in several physiological characteristics including pathogenesis, development and secondary metabolism. Two model organisms for photobiological studies have taken center-stage over the last few decades – *Neurospora crassa* and *Aspergillus nidulans*. In this review, we will first discuss our understanding of the light response in *N. crassa*, about which the most is known, and will then juxtapose *N. crassa* with *A. nidulans* which, as will be described below, provides an excellent template for understanding photosensory cross-talk. Finally, we will end with a commentary on the variability of the light response among other relevant fungi, and how our molecular understanding in the aforementioned model organisms still provides a strong base for dissecting light responses in such species.

Introduction

Light is a pervasive environmental signal that provides information to essentially all branches of life. In fungi, the presence of light may signal high temperature, the presence of genotoxic ultra-violet (UV) radiation, or the soil/air interface for optimal spore dispersal. In some fungi, light also serves to cue the organism's internal timekeeping system, termed the circadian clock, to anticipate daily environmental fluctuations (Idnurm *et al.*, 2010; Rodriguez-Romero *et al.*, 2010, Fuller *et al*., 2014; Fuller *et al*., 2014b). In this review, we will begin by discussing the molecular sensing apparatus of *Neurospora crassa*, the organism in which the light response is most thoroughly described and from which the first bone fide photoreceptors were cloned (Ballario *et al.*, 1996). We will also describe the photosensory mechanisms of another model fungus, *Aspergillus nidulans*, the photoresponse of which is dependent upon multiple photosensory inputs (Bayram *et al.*, 2010). A special

[*]Corresponding Authors: JC Dunlap or JJ Loros, Departments of Genetics and Biochemistry, Geisel School of Medicine at Dartmouth, 702 Remsen, HB 7400, Phone: 603-650-1108, jay.c.dunlap@dartmouth.edu or jennifer.loros@dartmouth.edu.
[±]These authors contributed equally to this work.

The authors declare no conflict of interest.

emphasis will be placed on the key differences between these two models and how both have provided important insights into fungal photosensing generally.

N. crassa: A model organism for studying photobiology

N. crassa, commonly observed as a red bread mold, has been a steadfast model organism in the dissection of several biological mechanisms (Davis & Perkins, 2002; Raju, 2009; Roche *et al.*, 2014). Besides the traditional features of a model genetic organism, a sequenced genome (Galagan *et al.*, 2003), an established core facility for knock-out or mutant strains, site-specific gene manipulation protocols (Colot *et al.*, 2006; Park *et al.*, 2011), development of CRISPR-mediated genome editing (Matsu-ura *et al*., 2015), optimized circadian reporters (Gooch *et al.*, 2008), microarray platforms (Lewis *et al.*, 2002; Chen *et al.*, 2009), and strains amenable for high-throughput proteomics (Baker *et al.*, 2009) have firmly established the value of this genetically facile organism as one thoroughly suited for the “omics” era.

An immense amount of research into the photobiology of *N. crassa* has been driven by the fact that this is a key model organism for the molecular dissection of circadian regulation, and light plays a dominant role as an entrainment cue in the circadian clock (Pittendrigh *et al.*, 1959; Baker *et al.*, 2012). Nevertheless, photobiology of the fungus has also been studied independently of circadian biology as exposure to blue light has several conspicuous effects on *N. crassa* physiology (Corrochano & Garre, 2010). Light exposure leads to the induction of carotene biosynthesis genes turning the dark-grown white mycelia to a bright orange color (Linden *et al.*, 1997), and induces branching in *N. crassa* cultures thereby making light-grown colonies appear more compact than dark-grown colonies (Lauter *et al.*, 1998; Ambra *et al.*, 2004). The development of protoperithecia (female sexual structures) is induced by blue light (Innocenti *et al.*, 1983), and the perithecial (fertilized perithecia) beaks are phototropic to blue light (Harding & Melles, 1983). Additionally, exposure to light is required for maximum asexual sporulation (Lauter *et al.*, 1997).

The blue light sensing apparatus in *N. crassa*

Fungi respond to specific wavelengths of light by utilizing wavelength-specific photoreceptor proteins, where the light-absorbing component is a chromophore that binds to specialized domains in the photoreceptors (Briggs & Spudich, 2005). Upon light absorption the chromophore undergoes a photochemical and structural change which is then transduced to biochemical and/or conformational changes in the photoreceptor protein (Nakasako *et al.*, 2004; Freddolino *et al.*, 2013; Hisatomi *et al.*, 2013; Conrad *et al.*, 2014; Vaidya *et al.*, 2011). The output from the light-induced changes in the photoreceptors comes in a couple of flavors – direct modulation of primary function, as in the case with photolyases (Carell *et al.*, 2001), or, the initiation of a signaling cascade that modulates downstream pathways, as in the case with White Collar-1 (WC-1) protein of *N. crassa* (Chen *et al.*, 2009).

N. crassa has been shown to respond to blue light and this response is mediated by the WC-1 and WC-2 proteins acting in a complex called the White-Collar-Complex (WCC) (Ballario *et al.*, 1996; Linden *et al.*, 1997; Ballario & Macino, 1997; Froehlich *et al.*, 2002). Inactivation of either *wc-1* or *wc-2* leads to defective carotenoid pigmentation of the mycelia, giving the strain a “white collar-like” appearance beneath the constitutively

pigmented conidia when grown on test-tube slants. Screening for strains defective in carotenoid synthesis only identified alleles of *wc-1* and *wc-2*; this suggested that the products of these two genes were either the primary effectors of the blue light response in *N. crassa* or were required to transduce the signal from the true photoreceptor. The finding that WC-1 and WC-2 comprised a transcription factor (Ballario *et al*., 1996; Linden *et al.,* 1997) suggested the latter, so it came as a surprise when WC-1 was also shown to be photoresponsive *in vitro* (Froehlich *et al.*, 2002). WC-1 is the photoreceptor component of the WCC, and it utilizes FAD (flavin-adenine dinucleotide) as a chromophore (Froehlich *et al.*, 2002; He *et al.*, 2002). The site of action of blue light is at the LOV (Light-Oxygen-Voltage) domain of WC-1 which covalently binds FAD at an active cysteine residue upon light exposure (Cheng *et al.*, 2003b). LOV domains are a specialized class of PAS (Per-ARNT-Sim) domains and are also found in bacteria and plants (Demarsy & Fankhauser, 2009; Herrou & Crosson, 2011). Additionally, WC-1 contains a Zn finger domain (GATA-like transcription factor domain), two PAS domains that modulate protein interactions, and a putative transcriptional activation domain (Ballario *et al.*, 1996; Lee *et al.*, 2003). The WC-2 protein dimerizes with WC-1 protein via a PAS domain (Ballario *et al*., 1998), and it also contains a Zn finger domain and putative transcriptional activation domains (Linden and Macino, 1997; Cheng *et al.*, 2002; Collett *et al.*, 2002). Thus, WCC functions as light-activated transcription factor.

Molecular changes in response to blue light: structural changes to DNA binding

In the absence of light, WCC exists as “dark” WCC comprised of a WC-1/2 heterodimer (Froehlich *et al.*, 2002; Cheng *et al.*, 2003b) which is able to bind to light response elements (LREs) in the promoters of light-responsive genes, although it appears to be unable to induce transcription *in vivo* after binding (Talora *et al.*, 1999; He & Liu, 2005; Smith *et al.*, 2010; Lewis *et al*., 2002). However, the “dark” WCC is proficient in gene expression from certain other promoter elements, e.g., the *c-box* in the *frq* promoter (Froehlich *et al.*, 2002; Baker *et al.*, 2012). Light activation is believed to induce structural changes in the WC-1 LOV domain that result in changes to the quaternary structure of the WCC: the “light” WCC is larger in size when compared to “dark” WCC, and contains the WC-1/2 heterodimer with additional light-activated WC-1 molecules (Froehlich *et al.*, 2002; Cheng *et al.*, 2003a). “Light” WCC transiently binds to the LREs (Early LREs, ELRE) of early light-responsive genes in a matter of a few minutes and the initiation of light-induced transcription is associated with transient phosphorylations of WC-1 that precedes removal of WC-1 from LREs followed by its degradation (Talora *et al.*, 1999; He & Liu, 2005; Schafmeier *et al.*, 2005; Froehlich *et al*., 2003).

Light response mediated through WCC at the DNA level

Chromatin modifications are involved in the induction of the expression of genes regulated by WCC in response to light. Light-induced acetylation of promoter histone H3-K14 is essential for the induction of these genes (Grimaldi *et al.*, 2006; Brenna *et al.*, 2012), and a strain expressing mutated H3 (*hH3K14q*), which cannot be acetylated at K14, phenocopies a *wc-1* mutant strain (Grimaldi *et al.*, 2006). This acetylation is carried out by histone acetyltransferase (HAT) NGF-1, and NGF-1 directly interacts with WC-1. Given that NGF-1 was found to interact with “dark” WC-1/2 heterodimer, a model has been proposed

wherein light activation of the WCC/NGF-1 complex leads to conformational changes in WCC (converting it to "light" WCC) which in turn leads to an enhancement of HAT activity of NGF-1 (Brenna *et al.*, 2012). More recently, H3K9me3 DNA methylation by methyltransferase DIM-5 and HP1 has been identified to be a component in the repression of light-induced gene expression (Ruesch *et al.*, 2014), and there is higher light-induced transcription of two genes, *frq* and *vvd*, in a *dim-5* mutant. WC-2 binding at the pLRE (the LRE of the *frq* promoter) is enhanced in the absence of H3K9me3 under light conditions (Ruesch *et al.*, 2014). Similar but less drastic results were found in a *set-1* mutant that lacks another histone lysine methyl transferase (Raduwan *et al.*, 2013). Thus, there are two levels of control of light-induced expression of genes at the level of chromatin. The chromatin at these light-response genes has to provide the right permissiveness for WCC binding, and after the initial binding event, additional modifiers (e.g. NGF-1) are required for the proper expression of the target genes. The aforementioned studies with NGF-1, DIM-5 and SET-1 have not been done on a genome-wide scale, so it is not yet known if the same mechanisms occur at the locus of every light-induced gene. There are major differences in the levels of induction of light-induced genes [*al-3* is induced to ~20 fold whereas *vvd* is induced to about ~200 fold (Chen *et al.*, 2009)] suggesting that the cis-acting LREs are not the only determinant of light-induced gene expression. Depending on the locus, ancillary chromatin modulators, transcription factors and transcriptional co-regulators are probably involved in determining expression levels (examples described below). A bromodomain-containing ATPase CATP, which removes histones (Cha *et al.*,2013), and the SWI/SNF complex are required for the action of WCC in the dark at the circadianly regulated *c-box* within the *frq* locus (Wang *et al.*, 2014); however, neither CATP nor SWI/SNF are required for light responses from this locus at least.

Control of gene expression by WCC

The fraction of genes identified as light-induced has ranged widely (3%–31%) in various studies primarily due to differences in methodology (microarrays vs. RNA-seq), culture conditions, and fold-change cut-offs imposed (Lewis *et al.*, 2002; Dong *et al.*, 2008; Chen *et al.*, 2009; Wu *et al.*, 2014), with RNA-Seq, the most quantitative and sensitive technology (Wang *et al.*, 2009; Zhao *et al.*, 2014), revealing that nearly 31% of expressed genes show two-fold or more change when *N. crassa* is exposed to light (Wu *et al.*, 2014). Unique to this study was the finding that a subset of mRNAs was actually significantly down-regulated in response to light exposure (e.g. mRNAs specifying factors involved in rRNA metabolism), a phenomenon that had not been previously described in this fungus. Overall, exposure to light appears to induce a stress response, and an increase in cellular metabolism (Chen *et al.*, 2009; Wu *et al.*, 2014).

Although WCC is the primary conduit for light-induced gene expression, the timing of induction of these genes appears hierarchical (Linden *et al.*, 1997; Chen *et al.*, 2009; Chen *et al.*, 2010a; Wu *et al.*, 2014). A subset of genes, the early light-responders, show peaks in their mRNA levels between 15–30 minutes of light exposure, while the late light-responders take about 60–120 minutes to peak. Early light-responders include genes involved in stress-response (e.g. DNA repair/processing, photopigment synthesis), secondary metabolism and cellular signaling whereas the late light-responders are involved in carbohydrate

metabolism, oxidation of fatty acids and components of numerous detoxification systems (Chen *et al.*, 2009; Wu *et al.*, 2014). The repression of a subset of genes involved in ribosome biogenesis could be as a consequence of the stress faced by the organism upon light exposure (Wu *et al.*, 2014). The hierarchical control of gene expression appears to be in part dependent on the genes induced during the first light response. ChIP-Seq has recently identified ~400 direct targets of WCC in the presence of light, many of which are transcription factors that can potentially further activate downstream genes (Smith *et al.*, 2010). For example, a knockout for the GATA family transcription factor *sub-1*, which is directly induced by WCC binding to its LRE, shows defects in the induction of large number of late light-responsive genes (Chen *et al.*, 2009; Chen *et al.*, 2010a). Additionally, a certain subset of genes in a *sub-1* mutant have altered amplitude of induction which suggests that factors other than SUB-1 are required for activating these genes, and that SUB-1 takes up an ancillary role as a modulator of gene expression (Chen *et al.*, 2010a). This ancillary role was brought to the fore when it was found that for a subset of light-induced genes, although WCC was primarily essential for induction, SUB-1 facilitates chromatin remodeling by interacting with a transcription factor FF-7 (containing a putative O-acetyl transferase domain) in combination with WCC, thereby modulating the extent of induction of their common target genes (Sancar *et al.*, 2015). Interestingly, SUB-1 and FF-7 regulate the expression a much greater fraction of non-light induced genes, suggesting that this might be a general transcription regulatory module that is co-opted during the light response.

This requirement of additional factors for expression of WCC target genes has also been described in a circadian context, where the phase of binding of WCC to its target promoters does not appear to correlate in all cases with the phase of maximum expression of these genes (Hurley *et al.*, 2014). Additionally, strains where *N. crassa* homologs (*rcm-1* and *rcm-2*) of a well-defined yeast corepressor complex (Tup1/Ssn6) are mutated show a loss of repression of light-dependent gene expression, although this appears to be because of reduced expression of the photoadaptation protein VIVID (see below) (Olmedo *et al.*, 2010a; Ruger-Herreros *et al.*, 2014). It is not known if this repressor system works at a global level because high-throughput studies (RNA-Seq/microarray) have not been conducted using these mutants. *N. crassa* RCM-1/RCO-2 also forms a complex with CSP-1, a light induced TF and a homolog of another component the yeast Tup1/Ssn6 repressor complex, in bringing about repression of a certain subset of genes in phase specific manner in a circadian cycle (Lambreghts *et al*. 2009; Sancar *et al.*, 2011). Regardless of the fact that some of these studies have only been conducted in the context of certain gene families, it appears that general transcription regulatory modules are utilized in the modulation of the light response.

Other light receptors in *N. crassa*

Through whole genome sequencing of *N. crassa* it was determined that this organism encodes orthologs of putative photoreceptors identified in other organisms, namely, two phytochromes (PHY-1 and PHY-2), a cryptochrome (CRY), and two previously identified photoreceptors, the LOV domain protein, VIVID, (Heintzen et al., 2001) and an opsin protein, NOP-1 (Bieszke *et al.*, 1999; Galagan *et al.*, 2003). Of these, only VIVID (VVD) appears to have a major effect on the light response in *N. crassa.* It is important to re-iterate

that essentially all light responses cease to exist in strains lacking a functional WCC. There is, however, evidence that the other photoreceptors potentially modulate the light response in *N. crassa* in a gene family/developmental stage-specific manner (Froehlich *et al.*, 2010; Olmedo *et al*., 2010b).

VVD is an extensively studied blue-light photoreceptor in *N. crassa* and is one of the early light responsive genes (Heintzen *et al.*, 2001; Schwerdtfeger & Linden, 2003; Malzahn *et al.*, 2010; Chen *et al.*, 2010b; Hunt *et al.*, 2010). It contains a LOV domain that binds FAD, shows structural changes when exposed to light, and demonstrates a slow photocycle (Zoltowski *et al.*, 2007; Zoltowski & Crane, 2008; Lamb *et al.*, 2009; Vaidya *et al.*, 2011). VVD functions in photoadaptation, where it attenuates WCC-mediated light responses at a particular light intensity, yet allows the system to respond to the next higher light intensity (Heintzen *et al*. 2001; Schwerdtfeger & Linden, 2001; Schwerdtfeger & Linden, 2003). *vvd* mutants have an excessively bright orange coloration of the mycelia when grown in constant light which suggests hyper-carotenoid synthesis and hence higher WCC activity (Heintzen *et al.*, 2001; Schwerdtfeger & Linden, 2003; Chen *et al.*, 2010b). It localizes to the nucleus and physically interacts with the WCC through light-activated LOV domains and inhibits its transcriptional activity (Hunt *et al.*, 2010; Malzahn *et al.*, 2010; Chen *et al.*, 2010b). The VVD photocycle can be tuned over 4 orders of magnitude *in vitro* (Zoltowski *et al.*, 2009) and the native photocycle length is required to form a dynamic VVD-WCC pool *in vivo* important for photoadaptation (Dasgupta *et al.*, 2015). Thus, through analyses of VVD photocycle mutants in *N. crassa* the evolutionary significance for the conservation of a wide range of photocycle lengths among LOV domain proteins has been demonstrated.

N. crassa phytochrome (red light photoreceptors) orthologs, PHY-1 and PHY-2, do not have a defined function in the fungus and *phy-1* and *phy-2* mutants do not show significant global changes in the expression of light-responsive genes (Chen *et al.*, 2009). However, in a *phy-2* mutant there is greater accumulation of *con-10* mRNA suggesting that a small subset of genes might indeed be modulated by PHY-2 (Olmedo *et al.*, 2010b). PHY-2 shows typical photoreceptor properties of cofactor binding (biliverdin or phycocyanobilin) and an *in vitro* photocycle, while *phy-1* expression is circadianly regulated, thus further study of their function *in vivo* is warranted (Froehlich *et al.*, 2005).

CRY in *N. crassa* is a DASH (Cryptochrome-Drosophila, Arabidopsis, Synechocystis, Human)-type cryptochrome that binds chromophores, FAD and MTHF (5,10 methyltetrahydrofolate) but has no *in vivo* photolyase activity (Froehlich *et al.*, 2010). Like the phytochromes, knocking out *cry* appears not to cause changes in global gene expression in response to light (Chen *et al.*, 2009), but might alter the gene expression of a small subset of genes (Olmedo *et al.*, 2010b). Interestingly, both the transcript and protein of *cry* are strongly light-induced via the action of WCC, and CRY is capable of binding to DNA (Froehlich *et al.*, 2010). It is a bit of a surprise that cryptochrome is not an essential photoreceptor or clock component in *N. crassa* given the important role its orthologs play in photoreception and the clocks of some other eukaryotic organisms (Guo *et al.*, 1998; Somers *et al.*, 1998; Ma *et al.*, 2001; Matsumura *et al.*, 2014). The *N. crassa* opsin, NOP-1 also shows a photocycle *in vitro* and binds retinal (Bieszke *et al.*, 1999a; Bieszke *et al.*, 1999b) but the effect of inactivating *nop-1* appears, again, to be target specific and probably

development stage specific (Bieszke *et al.*, 2007; Chen *et al.*, 2009; Olmedo *et al.*, 2010b). *nop-1* expression is light-induced (Wu *et al.*, 2014) and *nop-1* mRNA accumulates during the later stages of conidiation (Bieszke *et al.*, 1999b). It has been proposed that NOP-1 regulates mRNA accumulation of a subset of genes regulated by light and conidiation (Bieszke *et al.*, 2007; Olmedo *et al.*, 2010b). It is important to note that more sensitive techniques such as RNA-Seq have not been applied to study the phytochrome, cryptochrome and opsin mutants in *N. crassa.* The functional landscape of these photoreceptors and the processes they control might change with the application of better, more sensitive approaches.

Other factors influencing the *Neurospora* light response

WCC was the founding example of the heterodimeric positive arm element in the conserved transcription-translation-based negative feedback loop of fungi and animals (Crosthwaite *et al*., 1997; Baker *et al.*, 2012). Briefly, WCC leads to activation of *frq* transcription, and FRQ (along with some partner proteins) subsequently inhibits WCC activity by physically interacting with it. Over the course of the circadian day FRQ is first stabilized and then inactivated through post-translational modifications, chiefly phosphorylation (Baker *et al*., 2009), leading to release and reactivation of the WCC (Larrondo *et al*., 2015). The phosphorylations also lead to FRQ degradation and the cycle repeats itself under constant conditions (Liu *et al*., 2000). Thus, the levels of FRQ protein oscillate and, depending on the phase of the oscillation, (or time in the circadian day) WCC's response to light is modulated (Merrow *et al.*, 2001; Heintzen *et al.*, 2001). The clock also modulates both early and late light responses in a FRQ-dependent manner (Chen *et al.*, 2009) and the absence of FRQ, which results in lower WC-1 levels (Lee *et al.*, 2000), causes a certain subset of genes to show defective light-induction (Merrow *et al.*, 2001; Tan *et al.*, 2004; Lee *et al*., 2003) whereas others maintain robust light-induction (Arpaia *et al.*, 1993; Arpaia *et al.*, 1995; Lee *et al*., 2003). Interestingly, inactivation of WCC by FRQ involves kinases that piggy-back on FRQ and are believed to subsequently phosphorylate and inactivate WCC (Baker *et al.*, 2012). This again suggests that there are locus-specific effects of tuning WC-1 levels/ activity in *N. crassa* when it comes to light-induced gene expression. VeA has been shown to inhibit red-light-mediated conidiation and promotion of fruiting body formation in *Aspergillus nidulans* (Mooney & Yager, 1990; Blumenstein *et al.*, 2005; Purschwitz *et al.*, 2008). Mutant phenotypes in *Aspergillus* can be rescued by *ve-1*, the *N. crassa* ortholog of *veA*, this despite the fact that there is no evidence of red-light responses in *N. crassa* (Froehlich *et al.*, 2005; Bayram *et al.*, 2008a; Chen *et al.*, 2009). It has been reported that the Δ*ve*-1 strain shows lower accumulation of carotenoids on prolonged light exposure (Olmedo *et al.*, 2010b). The mechanistic basis of this phenotype is unknown and it is possible that VE-1 in *N. crassa* affects the expression of a certain subset of genes similar to PHY-2 and NOP-1.

Several kinases and phosphatases have been identified that alter WCC activity. Protein kinase A (PKA) and Protein Kinase C (PKC) inhibit light-dependent gene transcription in *N. crassa* (Franchi *et al.*, 2005; Huang *et al.*, 2007; Cha *et al.*, 2008). Of the two kinases, PKC has been shown to directly interact with WC-1 *in vivo* in a light-dependent manner (Franchi *et al.*, 2005). CK-1a and CK-II phosphorylate and inactivate WCC in a FRQ-dependent

(piggy-back) manner (He *et al.*, 2006), whereas PP2A and PP4 dephosphorylate and activate WCC (Schafmeier *et al.*, 2005; Cha *et al.*, 2007). Another kinase, GSK3, regulates WCC abundance by promoting its degradation (Tataroğlu *et al.*, 2012). It is difficult to elucidate the specific roles these kinases and phosphatases play in the *N. crassa* light response given that they have multiple targets and the final molecular phenotype might be a combination of several light response-independent molecular changes. Likewise, enzymes modulating response to ROS exposure have been implicated in the light response in *N. crassa*, but the effect is likely to be indirect (Yoshida & Hasunuma, 2004; Iigusa *et al.*, 2005; Wang *et al.*, 2007; Yoshida *et al.*, 2008). To conclude, the light sensing machinery in *N. crassa* can be summarized in Figure 1: On light activation, WCC binds to ELREs in the promoters of early light-responsive genes, and the activation of these genes requires chromatin modulators such as NGF-1 that de-repress the WCC target locus. Additionally, the methylation status of the loci appears to play a role in modulating the expression of the target genes. The VVD protein, a product of the early light-response, forms a dynamic interaction pool with WCC and brings about photoadaptation. Several early light-responsive genes are TFs that initiate the cascade of light signaling downstream of the initial light response. One such TF, SUB-1, has been found to co-regulate activation of late light-responsive genes in association with WCC. The other *N. crassa* light receptors and the VE-1 protein appear to play gene-family or developmental stage-specific roles. The clock protein FRQ, modulates the light response in circadian (time of day) manner and kinases and phosphatases, in general, inactivate or re-activate WCC respectively.

Aspergillus nidulans: a model for photosensory cross-talk

After *Aspergillus nidulans* came into play as a model some decades after *N. crassa*, the two fungi naturally settled into more parochial research roles. *N. crassa* remained a broader model for genetics and biochemistry, and effects of visible light were studied early on, driven by an active community studying mechanisms and rates of mutation. *A. nidulans* was initially used more narrowly for fungal parasexuality and recombination (Pontevervo *et al.*, 1953; Kafer, 1961). In the ensuing decades, work with *A. nidulans* has advanced the understanding of fungal developmental programming, primary and secondary metabolism, and nuclear duplication (Morris and Enos, 1992; Harris, 1997; Adams and Yu, 1998; Suelmann and Fischer, 2000; Osmani and Mirabito, 2004; Yu, 2006). The influence of light on these processes went largely unappreciated for several decades due to the isolation of the original “velvet” mutant (*veA1*) in 1965, so named because it undergoes conidiation without the production of aerial hyphae (Kafer, 1965; Mooney *et al.*, 1990). This isolate proved convenient for routine laboratory culture and became such a popular background strain for most developmental and metabolic studies that many researchers forgot it was in the genetic background. Not until 1990 was it understood that the *veA1* mutation actually abolishes the light-dependency of conidiation, although the mechanism remained unclear (Mooney *et al.*, 1990). With the breakthroughs in *N. crassa* paving the way, however, interest in *A. nidulans* photobiology has been rejuvenated in the last decade. The photo-responsive wild-type strains have been revisited and the contributions and mechanisms of light sensing pathways have been probed with genetics, genomics and biochemistry (Bayram *et al.*, 2010). Though the work in *A. nidulans* currently lacks the depth of that seen for *N. crassa*, it is clear that the

two models differ in several important ways, perhaps not surprisingly as *N. crassa* (in the Sordariomycetes) and *A. nidulans* (in the Eurotiomycetes) diverged over 300 million years ago, comparable to the evolutionary distance between humans and chickens or fish (Berbee & Taylor, 2010).

The *A. nidulans* light response is driven by multiple light qualities and photoreceptors

The initial description of the influence of light on conidiation in *A. nidulans* actually revealed a specific inducing effect at 680 nm (red light), with no effect seen below 500 nm (Mooney and Yager, 1990). A more recent investigation, however, showed that both red and blue light (450 nm) have equal inducing effects, but this approximates only one-third the level of induction observed in white light. Blue and red light treatment together fully restores conidiation, indicating that the two light qualities are sufficient and additive (Purschwitz *et al.*, 2008). The discord between the two mentioned investigations is currently unclear, but as will be discussed, bona fide blue light sensing in *A. nidulans* is now established at the molecular and biochemical level.

Blue and red light also act cooperatively/additively to repress two processes: the first is sexual development in the form of cleistothecium formation (Purschwitz *et al.*, 2008); the second is conidial germination (Röhrig *et al.*, 2013). In both cases the repression may serve as a possible protective mechanism, ensuring that sex or growth are not initiated in the presence of genotoxic and deleterious ultra-violet radiation (Fuller *et al.*, 2014). Indeed, the concept of light as a source of stress in *A. nidulans* is supported by the light-induction of various stress-resistance genes, including those involved in DNA repair (e.g. photolyase) and reactive oxygen species detoxification (e.g. superoxide dismutase) (Ruger-Herreros *et al.*, 2011).

Interestingly, an antagonistic interaction between red and blue light is seen with respect to their influence on secondary metabolism, namely the production of the mycotoxin sterigmatocystin (ST). ST is of particular interest as it is a precursor for various toxins of agricultural and medical importance, including aflatoxin and gliotoxin, respectively. Blue or white light treatments suppress ST production whereas red light treatment alone induces ST to levels beyond what is observed in the dark (Purschwitz *et al.*, 2008).

The light response machinery in *A. nidulans*

Analysis of the *A. nidulans* genome has revealed essentially the same suite of putative photoreceptors found in *N. crassa*. One exception includes the absence of a *nop-1* (rhodopsin) ortholog in *A. nidulans*, which is consistent with the fact that *A. nidulans* is not responsive to green light (536 nm), corresponding to the peak absorptive range for NOP-1 (Bayram *et al.*, 2010). *A. nidulans* also lacks an identifiable ortholog to the small LOV-domain protein VVD, the implications of which will be discussed later (Idnurm *et al.*, 2010). All putative photoreceptors that are present have been characterized, at a minimum, by gene deletion and will be briefly described next.

Blue Light Sensing—*A. nidulans* contains a single ortholog to both WC-1 and WC-2 of *N. crassa*, called LreA and LreB, respectively. Both proteins have the predicted domain

architecture of their *N. crassa* counterparts and their interaction to form a functional heterodimer has been inferred genetically and demonstrated through co-immunoprecipitation (Purschwitz *et al.*, 2008). In contrast to the essentiality of the WCC in the *N. crassa* light response, deletion of *lreA* and *lreB* is not sufficient to ablate the white light-induction of conidiation. To the contrary, the Δ*lreA/B* mutants display an increase in conidiation levels (beyond wild-type) in both the light and the dark. This suggests that the *A. nidulans* WCC somehow functions as a general repressor of conidiation which is relieved upon light activation. On the other hand, the *lreA/B* mutants display a general defect in both cleistothecium formation and ST production in both light and dark conditions, indicating a positive regulatory role by the WCC (Purschwitz *et al.*, 2008).

A. nidulans encodes a single cryptochrome ortholog, CryA, which displays absorption in the blue/near-UV range. Deletion of *cryA* leads to an increase in cleistothecium formation following induction with blue light (Bayram *et al.*, 2008b). This indicates that CryA plays a role in the light suppression of sexual development and, to this end, functions in opposition to the WCC.

Red light sensing—*A. nidulans* encodes a single phytochrome ortholog, called FphA, which covalently binds the tetrapyrole biliverdin and displays red and far-red absorption peaks (Brandt *et al.*, 2008). FphA also exhibits weak auto-phosphorylation and the kinase activity was recently shown to be essential for the red-light induction of gene expression (Brandt *et al.*, 2008; Hedtke *et al.*, 2015). Whole gene deletion of *fphA* leads to subtle repression of conidiation in both the light and the dark, as well as a general increase in cleistothecium development under both conditions (Blumenstein *et al.*, 2005). This suggests that the WCC and FphA of *A. nidulans* play opposing roles in regulating developmental programming in *A. nidulans*. Additionally, the *fphA* mutant displays a germination defect in the dark that reflects the wild-type germination levels seen in the light. This supports a model in which FphA serves as an activator of germination in the dark and this activity is repressed by light. Not only do the WCC and *cryA* mutants not display a germination defect, they still exhibit germination suppression by blue light (Röhrig *et al.*, 2013). This indicates that an additional, yet to be identified, blue light receptor senses blue light and impinges upon FphA.

In summary, *A. nidulans* can respond to both blue and red portions of the visible spectrum through at least three distinct photosensing modules: the WCC (i.e. LreA/LreB), phytochrome (FphA), and cryptochrome (CryA). This makes the photoresponse of *A. nidulans* inherently more complex than the blue light/WCC-centric model described for *N. crassa*. This then raises a fundamental question: how is the activity of distinct light sensors coordinated to generate a single physiological output? As will be discussed next, the genetic and functional interplay of these photoreceptors is beginning to come to light.

The mechanism of WCC signaling in *A. nidulans* is distinct from *N. crassa*

As described above, general photoreceptor functionality can be partially discerned from the genetic data. With respect to conidiation, for example, mutant analysis would suggest that the WCC represses conidiation genes in the dark, and this activity is released in the light. As

both WCC proteins contain DNA binding domains, one might expect this repression to be direct. Conversely, FphA is not thought to bind DNA yet serves as an activator of conidiation in both dark and light, but this activity is somehow partially blocked by the WCC in the dark (Purschwitz *et al.*, 2008). Interestingly, the WCC/FphA interaction is more than just genetic. Through a combination of bi-molecular fluorescence complementation and co-immunoprecipitation assays, it was shown that FphA binds to the WCC via a direct interaction with LreB. Within this complex, FphA also interacts directly with VeA, a central developmental regulator whose loss-of-function leads to the “velvet” phenotype described above (Purschwitz *et al.*, 2008; Purschwitz *et al.*, 2009). It is important to note that there is no evidence that VeA can directly sense light, rather it likely serves as a bridge between the photoperception machinery and developmental programming (Bayram *et al.*, 2008c).

The mechanistic interaction of the WCC, phytochrome and VeA has been partially dissected in a recent report in which the authors focused on a single light-regulated locus, *ccgA* (Hedtke *et al.*, 2015). The function of the *ccgA* gene (ortholog to the clock-controlled gene, *ccg-1*, of *N. crassa*) is unknown, but the gene is rapidly induced by red light and, to a lesser extent, blue light. Deletion of *fphA* completely abolishes the white light-induction of *ccgA*, whereas *lreA/B* deletion leads to an attenuated induction. Interestingly, VeA and LreA bind to the promoter of *ccgA* in the dark and LreA is released from the promoter following illumination with red or blue light (Hedtke *et al.*, 2015). This is in agreement with the conidiation data, in which LreA would serve as a repressor in the dark. The binding of VeA to the *ccgA* promoter is lost in the Δ*fphA* mutant and the binding of LreA is lost in the Δ*veA* mutant. It is currently unclear how red light alone can cause dissociation of LreA from the promoter, but it may involve a direct phosphorylation of the protein by FphA as the two proteins can physically interact.

Furthermore, the *ccgA* locus undergoes histone H3 acetylation upon light induction and this acetylation is lost in the Δ*fphA* or Δ*lreA* backgrounds (Hedtke *et al.*, 2015). Protein interaction assays demonstrated that LreA and FphA can interact with both histone acetylases and deacetylases to activate and inactivate transcription, respectively. Taken together, the following model is proposed and summarized in Figure 2: In the dark, VeA and LreA bind to the *ccgA* promoter and recruit both the histone deacetylase HdaA as well as the histone acetylase GcnE. LreA represses GcnE activity in order to maintain a de-acetylated (repressed) *ccgA* locus. In the light, LreA dissociates from the promoter and VeA and FphA promote histone acetylation by activating GcnE, thus promoting *ccgA* expression (e.g. see Hedtke *et al.*, 2015).

It will be interesting to see how WCC and FphA activity varies across loci, particularly with respect to genes that are down-regulated by light, e.g. genes involved in sexual development. For example, does LreA actively promote sexual developmental genes in the dark, or does it simply block the expression of a repressor? Chip-Seq experiments with LreA and VeA antibodies across a light-induction time-course will undoubtedly reveal insight into photoreceptor activity at a genome-wide level.

A. nidulans lacks a VIVID ortholog, but still displays photoadaptation

As described above, the term photoadaptation refers to the repression of light-regulated genes after the initial induction. In *N. crassa*, the photoadaption response is dependent upon VVD, which interacts directly with the WCC to suppress its activity. An ortholog to VVD is not found in the *A. nidulans* genome, yet photoadaptation-like kinetics have been observed for a number of genes, including *ccgA* and a central regulator of conidiation, *brlA* (Ruger-Herreros *et al.*, 2011; Olmedo *et al.*, 2013). The question, therefore, is how might photoadaptation occur in *A. nidulans*? Given that FphA is known to interact directly with the WCC, it is intriguing to speculate that light-activated FphA could take the place of light-activated VVD. In this scenario, FphA could directly phosphorylate the WCC and promote its re-association with target DNA. Alternatively, photoadaptation may have more to do with FphA, rather than the WCC. The conversion of the light-activated form (Pfr) to the dark form (Pr), either naturally or through an interaction with another protein (e.g. VeA), could promote closing of the chromatin and repress conidiation. An understanding of how photoadaptation occurs in *A. nidulans* would likely be applicable to a variety of fungal species as VVD is only found in restricted fungal lineages, including the Sordariomycetes, Leotiomycetes and Dothidiomycetes (Lombardi and Brody, 2005; Olmedo *et al.*, 2013; Salichos and Antonis, 2010).

Concluding Remarks

Taken together, two distinct models for photobiology have emerged: 1) The *N. crassa* system in which the WCC represents the central photosensory module that functions by actively promoting gene expression in the light, and its repressor, the photoreceptor VVD, and; 2) The *Aspergillus* model in which multiple photoreceptors (WCC, FphA, CryA) and ancillary proteins (VeA) interact physically and genetically to control gene expression. The difference between these two models does not simply reflect a disparity in components, as orthologs to both *fphA*, *cryA* and *veA* are all found in *N. crassa*; rather, it is the circuitry of the components that is fundamentally different between the two organisms. Whereas work with *N. crassa* has led the way in genome-wide and biochemical studies, *A. nidulans* has served as a model for photosensory cross-talk.

Is there a model for fungal photobiology?

The study of fungal light perception undoubtedly provides insights into signal transduction mechanisms in general, e.g. temporal spacing of gene induction via a hierarchical transcription factor cascade. However, understanding photobiology may have merit in its own right, as the importance of light on agriculturally, industrially and even medically important fungi becomes appreciated. For example, light enhances plant lesion size during infection with *Colletotrichum acutatum* and *Cercospora zeae-maydis*, but represses infection with *Magnaporthe oryzae* (Kim *et al.*, 2011a; Kim *et al.*, 2011b; Yu *et al.*, 2013). It is difficult to dissect the influence of light on the pathogen versus the host in these experiments, but at least in the case of *M. oryzae*, the light effect is lost upon deletion of *wc-1*. Moreover, *wc-1* deletion attenuates virulence in both *Botrytis cinerea* (Canessa *et al.*, 2013) and *C. zeae-maydis* (Kim *et al.*, 2011a) and, incredibly, in two distantly related human

pathogens, *Cryptococcus neoformans* (Idnurm and Heitman, 2005) and *Fusarium oxysporum* (Ruiz-Roldan *et al*., 2008).

Ideally, the work performed in *N. crassa* and *A. nidulans* should be predictive of the photosensing pathways in other organisms. For example, does the WCC generally serve as a repressor in the dark for organisms that respond to blue and red light? Does VVD, when present, always play a role in photoadaptation? We will briefly discuss recent data from two fungi, *Fusarium fujikuroi* and *Aspergillus fumigatus*, that provide some insight into these questions.

F. fujikuroi is an ascomycete whose production of plant developmental hormones (e.g. gibberellins) has made it an organism of interest in the agricultural and brewing industries (Tudzynski, 2005). Similar to *N. crassa*, *F. fujikuroi* accumulates large amounts of carotenoids in response to white and blue light; unlike *N. crassa*, however, this induction persists upon deletion of the *white collar-1* ortholog, *wcoA* (Estrada and Avalos, 2008; Castrillo and Avalos, 2015). Interestingly, the Δ*wcoA* mutant displays reduced carotenoids in the dark, but increased levels of additional pigments (e.g. bikaverins) under low nitrogen conditions (Estrada and Avalos, 2008). These data suggest that WCC of *F. fujikuroi* has regulatory functions in the dark.

Although red light does not lead to an overt increase in carotenoids in *F. fujikuroi*, it was shown to induce the expression of carotenoid biosynthetic genes to approximately 10% of the induction levels seen with blue or white light (Castrillo and Avalos, 2015). The role of the phytochrome ortholog is yet to be investigated, but the red light-induction was dependent upon the presence of *wcoA*. This suggests a direct transmission of light signals from phytochrome to WCC, potentially through a direct biophysical interaction as seen in *A. nidulans*. Furthermore, deletion of the cryptochrome ortholog, *cryD*, in *F. fujikuroi* leads to reduced light-induction of carotenoid genes. While this in some ways reflects the signaling role of CryA seen in *A. nidulans*, it is the first report of a cryptochrome regulating carotenoid biosynthesis (Castrillo and Avalos, 2015).

F. fujikuroi harbors an ortholog to *N. crassa's* VVD, VvdA; however, deletion of *vvdA* leads to a reduced accumulation of carotenoids in light, rather than a hyper-accumulation (i.e. photoadaptation defect) seen in *N. crassa* (Castrillo and Avalos, 2014). When analyzed further, the carotenoid response is biphasic, showing an initial burst in carotenogenesis, followed by a secondary and slower induction. Though the Δ*vvdA* mutant accumulates less pigment during the second phase, the rate and peak of the first phase is actually higher than wild-type. This suggests that VvdA may inhibit WcoA activity early in the light response, in partial agreement with its function in *N. crassa* (Castrillo and Avalos, 2015). A direct interaction between the two proteins has yet to be experimentally determined.

Finally, *F. fujikuroi* harbors a rhodopsin ortholog, *carO*. CarO is distributed primarily at the plasma membrane, where it serves as a green light-activated proton pump (Garcia-Martinez J. *et al*., 2015). Unlike NOP-1 of *N. crassa*, which does not affect light-dependent processes in that organism, loss of CarO function leads to enhanced conidial germination rates in light (Garcia-Martinez J. *et al*., 2015). This suggests that CarO plays a role in repressing conidial

germination, comparable to the role of the phytochrome in *A. nidulans*. Of note, light does not appear to inhibit conidial germination in *N. crassa* (K. Fuller, unpublished data) and *A. nidulans* does not harbor an opsin ortholog. Therefore, influence of light on germination is conserved between *A. nidulans* and *F. fujikuroi*, but the photoreceptor involved in regulating the process is differs between the two organisms.

Taken together, the light response of *F. fujikuroi* contains features that are characteristic to both *N. crassa* (e.g. VVD regulation of carotenogenesis through WCC) and *A. nidulans* (WCC activity in dark; responsiveness to red light), and is thus informed, at least in part, by both models. Further features of the *F. fujikuroi* light response appear distinct from either model, however, such as the positive roles in photocarotenogenesis by the VIVID and cryptochrome orthologs, or the regulation of germination by the opsin.

Aspergillus fumigatus is an important pathogen of immunocompromised patients. The photoresponse of *A. fumigatus* strain Af293 was shown to be distinct from *A. nidulans* in that light does not induce asexual sporulation. Rather, the major overt light response of Af293 is the accumulation of melanin, which occurs in both a blue light and *lreA*-dependent manner (Fuller *et al.*, 2013). The *lreA* mutants do not hyper-accumulate melanin in the dark, suggesting that LreA plays only a light-inducing role, as opposed to a dark-repressive role as it does in *A. nidulans*. Af293 mimics *A. nidulans* with respect to germination in that both red light and blue light inhibit the process. Moreover, the effect of *fphA* (phytochrome) deletion in both species with respect to germination is conserved (Fuller *et al.*, 2013).

Curiously, it was found recently that several strains of *A. fumigatus*, including the common lab strain H237 (Hensel *et al.*, 1998; Lopes Bezerra and Filler, 2004; Fortwendel *et al.*, 2008), do in fact induce conidiation in response to light (K. Fuller, unpublished data). However, there are important distinctions between the photoconidiation response in these strains versus *A. nidulans*: 1) the *A. fumigatus* induction is in response to blue light only, not red light; 2) deletion of *lreA* completely abolishes conidial induction in *A. fumigatus*; and 3) the *A. fumigatus* Δ*lreA* mutants do not hyper-conidiate in the dark (K. Fuller, unpublished data). Taken together, the story emerging from *A. fumigatus* is similar to that of *F. fujikuroi* in that the light response contains features of both *N. crassa* (WCC drives overt phenotypes in light) and *A. nidulans* (germination positively regulated by FphA).

To conclude, it appears that remarkable photobiological variability exists between species, both in terms of the phenotypic output and signaling mechanisms. Indeed, even isolates within the same species may demonstrate fundamentally different light outputs, as has been observed in *A. fumigatus*. In spite of this heterogeneity, however, key features of the *N. crassa* and *A. nidulans* models are likely applicable across all fungi. For example, whether or not the photoreceptor serves as an inducer (e.g. WCC in *N. crassa*) or repressor (e.g. WCC at *ccgA* locus in *A. nidulans*) of transcription, the mechanism likely involves the interaction with one or more chromatin remodeling enzymes. Thus, the discord that exists between the two model organisms is less important than the unifying principles. This is an exciting time for fungal photobiology as studies move beyond the laboratory standards and into fungi of practical and environmental importance. Time will tell what novel

photosensory mechanisms have evolved in these species and to what extent understanding such mechanisms will be applicable in the world beyond the bench.

Acknowledgments

This work was supported by RO1 grants from the National Institute of General Medical Sciences of the National Institutes of Health to J.J.L. (GM08336) and J.C.D (GM034985).

References

Adams TH, Yu J. Coordinate control of secondary metabolite production and asexual sporulation in *Aspergillus nidulans*. Curr Opin Microbiol. 1998; 1:674–677. [PubMed: 10066549]

Ambra R, Grimaldi B, Zamboni S, Filetici P, Macino G, Ballario P. Photomorphogenesis in the hypogeous fungus *Tuber borchii*: isolation and characterization of Tbwc-1, the homologue of the blue-light photoreceptor of *Neurospora crassa*. Fungal Genet Biol. 2004; 41:688–697. [PubMed: 15275664]

Arpaia G, Loros JJ, Dunlap JC, Morelli G, Macino G. Light induction of the clock controlled gene ccg-1 is not transduced through the circadian clock in *Neurospora crassa*. Mol Gen Genet. 1995; 247:157–163. [PubMed: 7753024]

Arpaia G, Loros JJ, Dunlap JC, Morelli G, Macino G. The interplay of light and the circadian clock. Independent dual regulation of clock-controlled gene ccg-2(eas). Plant Physiol. 1993; 102:1299–1305. [PubMed: 8278550]

Baker CL, Kettenbach AN, Loros JJ, Gerber SA, Dunlap JC. Quantitative proteomics reveals a dynamic interactome and phase-specific phosphorylation in the *Neurospora* circadian clock. Mol Cell. 2009; 34:354–363. [PubMed: 19450533]

Baker CL, Loros JJ, Dunlap JC. The circadian clock of *Neurospora crassa*. FEMS Microbiol Rev. 2012; 36:95–110. [PubMed: 21707668]

Ballario P, Macino G. White collar proteins: PASsing the light signal in *Neurospora crassa*. Trends Microbiol. 1997; 5:458–462. [PubMed: 9402704]

Ballario P, Talora C, Galli D, Linden H, Macino G. Roles in dimerization and blue light photoresponse of the PAS and LOV domains of *Neurospora crassa* white collar proteins. Mol Microbiol. 1998; 29:719–729. [PubMed: 9723912]

Ballario P, Vittorioso P, Magrelli A, Talora C, Cabibbo A, Macino G. White collar-1, a central regulator of blue light responses in *Neurospora*, is a zinc finger protein. EMBO J. 1996; 15:1650–1657. [PubMed: 8612589]

Berbee ML, Taylor JW. Dating the molecular clock in fungi- How close are we? Fungal Biol Rev. 2010; 24:1–16.

Bayram Ö, Braus GH, Fischer R, Rodriguez-Romero J. Spotlight on *Aspergillus nidulans* photosensory systems. Fungal Genet Biol. 2010; 47:900–908. [PubMed: 20573560]

Bayram Ö, Krappmann S, Seiler S, Vogt N, Braus GH. *Neurospora crassa ve-1* affects asexual conidiation. Fungal Genet Biol. 2008a; 45:127–138. [PubMed: 17631397]

Bayram Ö, Biesemann C, Krappmann S, Galland P, Braus GH. More than a repair enzyme: *Aspergillus nidulans* photolyase-like CryA Is a regulator of sexual development. Mol Biol Cell. 2008b; 19:3254–3262. [PubMed: 18495868]

Bayram Ö, Krappmann S, Ni M, Bok JW, Helmstaedt K, Valerius O, et al. VelB/VeA/LaeA complex coordinates light signal with fungal development and secondary metabolism. Science. 2008c; 320:1504–1506. [PubMed: 18556559]

Bieszke JA, Spudich EN, Scott KL, Borkovich KA, Spudich JL. A eukaryotic protein, NOP-1, binds retinal to form an archaeal rhodopsin-like photochemically reactive pigment. Biochemistry. 1999a; 38:14138–14145. [PubMed: 10571987]

Bieszke JA, Braun EL, Bean LE, Kang S, Natvig DO, Borkovich KA. The *nop-1* gene of *Neurospora crassa* encodes a seven transmembrane helix retinal-binding protein homologous to archaeal rhodopsins. Proc Natl Acad Sci U S A. 1999b; 96:8034–8039. [PubMed: 10393943]

Bieszke JA, Li L, Borkovich KA. The fungal opsin gene *nop-1* is negatively-regulated by a component of the blue light sensing pathway and influences conidiation-specific gene expression in *Neurospora crassa*. Curr Genet. 2007; 52:149–57. [PubMed: 17676324]

Blumenstein A, Vienken K, Tasler R, Purschwitz J, Veith D, Frankenberg-Dinkel N, Fischer R. The *Aspergillus nidulans* phytochrome FphA represses sexual development in red light. Curr Biol. 2005; 15:1833–1838. [PubMed: 16243030]

Brandt S, von Stetten D, Günther M, Hildebrandt P, Frankenberg-Dinkel N. The fungal phytochrome FphA from *Aspergillus nidulans*. J Biol Chem. 2008; 283:34605–34614. [PubMed: 18931394]

Brenna A, Grimaldi B, Filetici P, Ballario P. Physical association of the WC-1 photoreceptor and the histone acetyltransferase NGF-1 is required for blue light signal transduction in *Neurospora crassa*. Mol Biol Cell. 2012; 23:3863–3872. [PubMed: 22875992]

Briggs, W.; Spudich, J. Handbook of photosensory receptors. Weinheim: WILEY-VCH Verlag GmbH and Co. KGaA; 2005.

Canessa P, Schumacher J, Hevia M, Tudzynski P, Larrondo L. Assessing the effects of light on differentiation and virulence of the plant pathogen *Botrytis cinerea*: characterization of the White Collar Complex. PLoS One. 2013; 8:e84223. [PubMed: 24391918]

Carell T, Burgdorf LT, Kundu LM, Cichon M. The mechanism of action of DNA photolyases. Curr Opin Chem Biol. 2001; 5:491–498. [PubMed: 11578921]

Castrillo M, Avalos J. Light-mediated participation of the VIVID-like protein of *Fusarium fujikuroi* VvdA in pigmentation and development. Fungal Genet Biol. 2014; 71:9–20. [PubMed: 25154020]

Castrillo M, Avalos J. The flavoproteins CryD and VvdA cooperate with the white collar protein WcoA in the control of photocarotenogenesis in *Fusarium fujikuroi*. PLoS One. 2015; 10:e0119785. [PubMed: 25774802]

Cha J, Chang S, Huang G, Cheng P, Liu Y. Control of WHITE COLLAR localization by phosphorylation is a critical step in the circadian negative feedback process. EMBO J. 2008; 27:3246–55. [PubMed: 19020516]

Cha J, Huang G, Guo J, Liu Y. Posttranslational control of the *Neurospora* circadian clock. Cold Spring Harb Symp Quant Biol. 2007; 72:185–191. [PubMed: 18419276]

Cha J, Zhou M, Liu Y. CATP is a critical component of the *Neurospora* circadian clock by regulating the nucleosome occupancy rhythm at the frequency locus. EMBO Rep. 2013; 14:923–930. [PubMed: 23958634]

Chen C, Dunlap JC, Loros JJ. *Neurospora* illuminates fungal photoreception. Fungal Genet Biol. 2010a; 47:922–929. [PubMed: 20637887]

Chen C, DeMay BS, Gladfelter AS, Dunlap JC, Loros JJ. Physical interaction between VIVID and white collar complex regulates photoadaptation in *Neurospora*. Proc Natl Acad Sci U S A. 2010b; 107:16715–16720. [PubMed: 20733070]

Chen C, Ringelberg CS, Gross RH, Dunlap JC, Loros JJ. Genome-wide analysis of light-inducible responses reveals hierarchical light signaling in *Neurospora*. EMBO J. 2009; 28:1029–42. [PubMed: 19262566]

Cheng P, Yang Y, Gardner KH, Liu Y. PAS domain-mediated WC-1/WC-2 interaction is essential for maintaining the steady-state level of WC-1 and the function of both proteins in circadian clock and light responses of *Neurospora*. Mol Cell Biol. 2002; 22:517–524. [PubMed: 11756547]

Cheng P, Yang Y, Wang L, He Q, Liu Y. WHITE COLLAR-1, a multifunctional *Neurospora* protein involved in the circadian feedback loops, light sensing, and transcription repression of *wc-2*. J Biol Chem. 2003a; 278:3801–3808. [PubMed: 12454012]

Cheng P, He Q, Yang Y, Wang L, Liu Y. Functional conservation of light, oxygen, or voltage domains in light sensing. Proc Natl Acad Sci U S A. 2003b; 100:5938–5943. [PubMed: 12719523]

Collett MA, Garceau N, Dunlap JC, Loros JJ. Light and clock expression of the Neurospora clock gene *frequency* is differentially driven by but dependent on WHITE COLLAR-2. Genetics. 2002; 160:149–58. [PubMed: 11805052]

Colot HV, Park G, Turner GE, Ringelberg C, et al. A high-throughput gene knockout procedure for *Neurospora* reveals functions for multiple transcription factors. Proc Natl Acad Sci U S A. 2006; 103:10352. [PubMed: 16801547]

Conrad KS, Manahan CC, Crane BR. Photochemistry of flavoprotein light sensors. Nature Chem Biol. 2014; 10:801–9. [PubMed: 25229449]

Corrochano LM, Garre V. Photobiology in the Zygomycota: Multiple photoreceptor genes for complex responses to light. Fungal Genet Biol. 2010; 47:893–899. [PubMed: 20466063]

Crosthwaite SK, Dunlap JC, Loros JJ. *Neurospora wc*-1 and *wc*-2: transcription, photoresponses, and the origins of circadian rhythmicity. Science. 1997; 276:763–769. [PubMed: 9115195]

Dasgupta A, Chen C, Lee C, Gladfelter AS, Dunlap JC, Loros JJ. Biological significance of photoreceptor photocycle length: VIVID photocycle governs the dynamic VIVID-White Collar Complex pool mediating photo-adaptation and response to changes in light intensity. PLoS Genet. 2015; 15:e1005215. [PubMed: 25978382]

Davis RH, Perkins DD. *Neurospora*: a model of model microbes. Nature Rev Genet. 2002; 3:397–403. [PubMed: 11988765]

Demarsy E, Fankhauser C. Higher plants use LOV to perceive blue light. Curr Opin Plant Biol. 2009; 12:69–74. [PubMed: 18930433]

Dong W, Tang X, Yu Y, Nilsen R, Kim R, Griffith J, et al. Systems biology of the clock in *Neurospora crassa*. PLoS One. 2008; 3:n/a.

Estrada A, Avalos J. The White Collar protein WcoA of *Fusarium fujikuroi* is not essential for photocarotenogenesis, but is involved in the regulation of secondary metabolism and conidiation. Fungal Genet Biol. 2008; 45:705–715. [PubMed: 18203635]

Fortwendel JR, Fuller KK, Stephens TJ, Bacon WC, Askew DS, Rhodes JC. *Aspergillus fumigatus* RasA regulates asexual development and cell wall integrity. Eukaryot Cell. 2008; 7:1530–1539. [PubMed: 18606827]

Franchi L, Fulci V, Macino G. Protein kinase C modulates light responses in *Neurospora* by regulating the blue light photoreceptor WC-1. Mol Microbiol. 2005; 56:334–345. [PubMed: 15813728]

Freddolino PL, Gardner KH, Schulten K. Signaling mechanisms of LOV domains: new insights from molecular dynamics studies. Photochemical Photobiol Sci. 2013; 12:1158–1170.

Froehlich AC, Chen C, Belden WJ, Madeti C, Roenneberg T, Merrow M, et al. Genetic and molecular characterization of a cryptochrome from the filamentous fungus *Neurospora crassa*. Eukaryot Cell. 2010; 9:738–750. [PubMed: 20305004]

Froehlich AC, Liu Y, Loros JJ, Dunlap JC. White collar-1, a circadian blue light photoreceptor, binding to the *frequency* promoter. Science. 2002; 297:815–9. [PubMed: 12098706]

Froehlich AC, Noh B, Vierstra RD, Loros J, Dunlap JC. Genetic and molecular analysis of phytochromes from the filamentous fungus *Neurospora crassa*. Eukaryot Cell. 2005; 4:2140–2152. [PubMed: 16339731]

Froehlich AC, Loros JJ, Dunlap JC. Rhythmic binding of a WHITE COLLAR-containing complex to the frequency promoter is inhibited by FREQUENCY. Proc Natl Acad Sci U S A. 2003; 100:5914–5919. [PubMed: 12714686]

Fuller, KK.; Hurley, JM.; Loros, JJ.; Dunlap, JC. Photobiology and circadian clocks in *Neurospora*. In: Nowrousian, M., editor. The Mycota. 2. Vol. XIII. Springer-Verlag; Berlin: 2014a.

Fuller KK, Loros J, Dunlap J. Fungal photobiology: light as a signal for stress, space and time. Curr Genet. 2014b10.1007/s00294-014-0451-0

Fuller KK, Ringelberg CS, Loros JJ, Dunlap JC. The fungal pathogen *Aspergillus fumigatus* regulates growth, metabolism, and stress resistance in response to light. mBio. 2013:4.

Galagan JE, Calvo SE, Borkovich KA, Selker EU, Read ND, et al. The genome sequence of the filamentous fungus *Neurospora crassa*. Nature. 2003; 422:859–68. [PubMed: 12712197]

García-Martínez J, Brunk M, Avalos J, Terpitz U. The CarO rhodopsin of the fungus *Fusarium fujikuroi* is a light-driven proton pump that retards spore germination. Sci Rep. 2015; 5:7798. [PubMed: 25589426]

Gooch VD, Mehra A, Larrondo LF, Fox J, Touroutoutoudis M, Loros JJ, Dunlap JC. Fully codon-optimized luciferase uncovers novel temperature characteristics of the *Neurospora* clock. Eukaryot Cell. 2008; 7:28–37. [PubMed: 17766461]

Grimaldi B, Coiro P, Filetici P, Berge E, Dobosy JR, Freitag M, et al. The *Neurospora crassa* White Collar-1 dependent blue light response requires acetylation of histone H3 lysine 14 by NGF-1. Mol Biol Cell. 2006; 17:4576–4583. [PubMed: 16914525]

Guo H, Yang H, Mockler TC, Lin C. Regulation of flowering time by Arabidopsis photoreceptors. Science. 1998; 279:1360–3. [PubMed: 9478898]

Harding RW, Melles S. Genetic analysis of phototropism of *Neurospora crassa* perithecial beaks using White Collar and albino mutants. Plant Physiol. 1983; 72:996–1000. [PubMed: 16663152]

Harris SD. The duplication cycle in *Aspergillus nidulans*. Fungal Genet Biol. 1997; 22:1–12. [PubMed: 9344627]

He Q, Cheng P, Yang Y, Wang L, Gardner KH, Liu Y. White collar-1, a DNA binding transcription factor and a light sensor. Science. 2002; 297:840–3. [PubMed: 12098705]

He Q, Cha J, He Q, Lee H, Yang Y, Liu Y. CKI and CKII mediate the FREQUENCY-dependent phosphorylation of the WHITE COLLAR complex to close the *Neurospora* circadian negative feedback loop. Genes Dev. 2006; 20:2552–2565. [PubMed: 16980584]

He Q, Liu Y. Molecular mechanism of light responses in *Neurospora*: from light-induced transcription to photoadaptation. Genes Dev. 2005; 19:2888–2899. [PubMed: 16287715]

Hedtke M, Rauscher S, Röhrig J, Rodriguez J, Yu Z, Fischer R. Light-dependent gene activation in *Aspergillus nidulans* is strictly dependent on phytochrome and involves the interplay of phytochrome and White-collar-regulated histone H3 acetylation. Mol Microbiol. 2015 In Press.

Heintzen C, Loros JJ, Dunlap JC. The PAS protein VIVID defines a clock-associated feedback loop that represses light input, modulates gating, and regulates clock resetting. Cell. 2001; 104:453–464. [PubMed: 11239402]

Hensel M, Arst HJ, Aufauvre-Brown A, Holden DW. The role of the *Aspergillus fumigatus areA* gene in invasive pulmonary aspergillosis. Mol Gen Genet. 1998; 37:553–557. [PubMed: 9669338]

Herrou J, Crosson S. Function, structure and mechanism of bacterial photosensory LOV proteins. Nat Rev Microbiol. 2011; 9:713–23. [PubMed: 21822294]

Hisatomi O, Takeuchi K, Zikihara K, Ookubo Y, Nakatani Y, Takahashi F, et al. Blue light-induced conformational changes in a light-regulated transcription factor, aureochrome-1. Plant Cell Physiol. 2013; 54:93–106. [PubMed: 23220692]

Huang G, Chen S, Li S, Cha J, Long C, Li L, et al. Protein kinase A and casein kinases mediate sequential phosphorylation events in the circadian negative feedback loop. Genes Dev. 2007; 21:3283–3295. [PubMed: 18079175]

Hunt SM, Thompson S, Elvin M, Heintzen C. VIVID interacts with the WHITE COLLAR complex and FREQUENCY-interacting RNA helicase to alter light and clock responses in *Neurospora*. Proc Natl Acad Sci U S A. 2010; 107:16709–16714. [PubMed: 20807745]

Hurley JM, Dasgupta A, Emerson JM, Zhou X, Ringelberg CS, Knabe N, et al. Analysis of clock-regulated genes in *Neurospora* reveals widespread posttranscriptional control of metabolic potential. Proc Natl Acad Sci U S A. 2014; 111:16995–17002. [PubMed: 25362047]

Iigusa H, Yoshida Y, Hasunuma K. Oxygen and hydrogen peroxide enhance light induced carotenoid synthesis in *Neurospora crassa*. FEBS Lett. 2005; 579:4012–4016. [PubMed: 16004990]

Idnurm A, Verma S, Corrochano LM. A glimpse into the basis of vision in the kingdom Mycota. Fungal Genet Biol. 2010; 47:881–892. [PubMed: 20451644]

Idnurm A, Heitman J. Light controls growth and development via a conserved pathway in the fungal kingdom. PLoS Biol. 2005; 3:e95. [PubMed: 15760278]

Innocenti FD, Pohl U, Russo VE. Photoinduction of protoperithecia in *Neurospora crassa* by blue light. Photochem Photobiol. 1983; 37:49–51. [PubMed: 6220417]

Kafer E. The origin of translocations in *Aspergillus nidulans*. Genetics. 1965; 52:217–232. [PubMed: 5857597]

Kafer E. The processes of spontaneous recombination in vegetative nuclei of *Aspergillus nidulans*. Genetics. 1961; 46:1583–1609.

Kim H, Ridenour J, Dunkle L, Bluhm B. Regulation of stomatal tropism and infection by light in *Cercospora zeae-maydis*: evidence for coordinated host/pathogen responses to photoperiod. PLoS Pathog. 2011a; 7:e1002113. [PubMed: 21829344]

Kim S, Singh P, Park J, Park S, Friedman A, Zheng T, et al. Genetic and molecular characterization of a blue light photoreceptor MGWC-1 in *Magnaporthe oryzae*. Fungal Genet Biol. 2011b; 48:400–407. [PubMed: 21241815]

Lamb JS, Zoltowski BD, Pabit SA, Li L, Crane BR, Pollack L. Illuminating solution responses of a LOV domain protein with photocoupled small-angle X-ray scattering. J Mol Biol. 2009; 393:909–919. [PubMed: 19712683]

Lambreghts R, Shi M, Belden WJ, Decaprio D, Park D, Henn MR, Galagan JE, Bastürkmen M, Birren BW, Sachs MS, Dunlap JC, Loros JJ. A high-density single nucleotide polymorphism map for *Neurospora crassa*. Genetics. 2009; 181:167–781.

Larrondo LF, Olivares-Yañez C, Baker CL, Loros JJ, Dunlap JC. Circadian rhythms. Decoupling circadian clock protein turnover from circadian period determination. Science. 2015; 347:1257277. [PubMed: 25635104]

Lauter FR, Marchfelder U, Russo VE, Yamashiro CT, Yatzkan E, Yarden O. Photoregulation of *cot*-1, a kinase-encoding gene involved in hyphal growth in *Neurospora crassa*. Fungal Genet Biol. 1998; 23:300–310.

Lauter F, Yamashiro CT, Yanofsky C. Light stimulation of conidiation in *Neurospora crassa*: Studies with the wild-type strain and mutants *wc*-1, *wc*-2 and *acon*-2. J Photochem Photobiol B: Biol. 1997; 37:203–211.

Lee K, Loros JJ, Dunlap JC. Interconnected feedback loops in the *Neurospora* circadian system. Science. 2000; 289:107–10. [PubMed: 10884222]

Lee K, Loros JJ, Dunlap JC. Roles for WHITE COLLAR-1 in circadian and general photoperception in *Neurospora crassa*. Genetics. 2003; 163:103–114. [PubMed: 12586700]

Lewis ZA, Correa A, Schwerdtfeger C, Link KL, Xie X, Gomer RH, et al. Overexpression of White Collar-1 (WC-1) activates circadian clock-associated genes, but is not sufficient to induce most light-regulated gene expression in *Neurospora crassa*. Mol Microbiol. 2002; 45:917–931. [PubMed: 12180913]

Linden H, Ballario P, Macino G. Blue light regulation in *Neurospora crassa*. Fungal Genet Biol. 1997; 22:141–150. [PubMed: 9454641]

Linden H, Macino G. White collar 2, a partner in blue-light signal transduction, controlling expression of light-regulated genes in *Neurospora crassa*. EMBO J. 1997; 98:109.

Liu Y, Loros J, Dunlap JC. Phosphorylation of the *Neurospora* clock protein FREQUENCY determines its degradation rate and strongly influences the period length of the circadian clock. Proc Natl Acad Sci U S A. 2000; 97:234–237. [PubMed: 10618401]

Lombardi LM, Brody S. Circadian rhythms in *Neurospora crassa*: clock gene homologues in fungi. Fungal Genet Biol. 2005; 42:887–892. [PubMed: 16154782]

Lopes Bezerra L, Filler S. Interactions of *Aspergillus fumigatus* with endothelial cells: internalization, injury, and stimulation of tissue factor activity. Blood. 2004; 103:2143–2149. [PubMed: 14630829]

Ma L, Li J, Qu L, Hager J, et al. Light control of Arabidopsis development entails coordinated regulation of genome expression and cellular pathways. Plant Cell. 2001; 13:2589–607. [PubMed: 11752374]

Malzahn E, Ciprianidis S, Káldi K, Schafmeier T, Brunner M. Photoadaptation in *Neurospora* by competitive interaction of activating and inhibitory LOV domains. Cell. 2010; 142:762–772. [PubMed: 20813262]

Matsumura R, Tsuchiya Y, Tokuda I, Matsuo T, Sato M, Node K, et al. The mammalian circadian clock protein period counteracts cryptochrome in phosphorylation dynamics of circadian locomotor output cycles kaput (CLOCK). J Biol Chem. 2014; 289:32064–32072. [PubMed: 25271155]

Matsu-ura T, Baek M, Kwon J, Hong C. Efficient gene editing in *Neurospora crassa* with CRISPR technology. Fungal Biol Biotechnol. 2015:2.10.1186/s40694-015-0015-1

Merrow M, Franchi L, Dragovic Z, Gorl M, Johnson J, Brunner M, et al. Circadian regulation of the light input pathway in *Neurospora crassa*. EMBO J. 2001; 20:307–15. [PubMed: 11157738]

Mooney JL, Hassett DE, Yager LN. Genetic analysis of suppressors of the *veA1* mutation in *Aspergillus nidulans*. Genetics. 1990; 126:869–874. [PubMed: 2076818]

Mooney JL, Yager LN. Light is required for conidiation in *Aspergillus nidulans*. Genes Dev. 1990; 4:1473–1482. [PubMed: 2253875]

Morris NR, Enos AP. Mitotic gold in a mold: *Aspergillus* genetics and the biology of mitosis. Trends Genet. 1992; 8:32–33. [PubMed: 1369734]

Nakasako M, Iwata T, Matsuoka D, Tokutomi S. Light-induced structural changes of LOV domain-containing polypeptides from Arabidopsis phototropin 1 and 2 studied by small angle X-ray scattering. Biochemistry. 2004; 43:14881–14890. [PubMed: 15554695]

Olmedo M, Navarro-Sampedro L, Ruger-Herreros C, Kim S, Jeong B, Lee B, Corrochano LM. A role in the regulation of transcription by light for RCO-1 and RCM-1, the *Neurospora* homologs of the yeast Tup1-Ssn6 repressor. Fungal Genet Biol. 2010a; 47:939–952. [PubMed: 20709620]

Olmedo M, Ruger-Herreros C, Luque EM, Corrochano LM. A complex photoreceptor system mediates the regulation by light of the conidiation genes *con*-10 and *con*-6 in *Neurospora crass*a. Fungal Genet Biol. 2010b; 47:352–363. [PubMed: 19932184]

Olmedo M, Ruger-Herreros C, Luque EM, Corrochano LM. Regulation of transcription by light in *Neurospora crassa*: A model for fungal photobiology? Fungal Biol Rev. 2013; 27:10–18.

Osmani SA, Mirabito PM. The early impact of genetics on our understanding of cell cycle regulation in *Aspergillus nidulans*. Fungal Genet Biol. 2004; 41:401–410. [PubMed: 14998523]

Park G, Colot HV, Collopy PD, Krystofova S, Crew C, Ringelberg C, et al. High throughput production of gene replacement mutants in *Neurospora crassa*. Methods Mol Biol. 2011; 722:179–189. [PubMed: 21590421]

Pittendrigh CS, Bruce VG, Rosensweig NS, Rubin ML. Growth patterns in *Neurospora*: a biological clock in *Neurospora*. Nature. 1959; 184:169.

Pontevervo G, Roper J, Hemmons L, MacDonald K, Bufton A. The genetics of *Aspergillus nidulans*. Adv Genet. 1953; 5:141–238. [PubMed: 13040135]

Purschwitz J, Müller S, Fischer R. Mapping the interaction sites of *Aspergillus nidulans* phytochrome FphA with the global regulator VeA and the White Collar protein LreB. Mol Genet Genomics. 2009; 281:35–42. [PubMed: 18936976]

Purschwitz J, Müller S, Kastner C, Schöser M, Haas H, Espeso EA, et al. Functional and physical interaction of blue- and red-light sensors in *Aspergillus nidulans*. Curr Biol. 2008; 18:255–259. [PubMed: 18291652]

Raduwan H, Isola AL, Belden WJ. Methylation of histone H3 on lysine 4 by the lysine methyltransferase SET1 protein is needed for normal clock gene expression. J Biol Chem. 2013; 288:8380–8390. [PubMed: 23319591]

Raju NB. Neurospora as a model fungus for studies in cytogenetics and sexual biology at Stanford. J Biosci. 2009; 34:139–59. [PubMed: 19430125]

Roche CM, Loros JJ, McCluskey K, Glass NL. *Neurospora crassa*: Looking back and looking forward at a model microbe. Am J Bot. 2014; 101:2022. [PubMed: 25480699]

Rodriguez-Romero J, Hedtke M, Kastner C, Müller S, Fischer R. Fungi, hidden in soil or up in the air: light makes a difference. Annu Rev Microbiol. 2010; 64:585–610. [PubMed: 20533875]

Röhrig J, Kastner C, Fischer R. Light inhibits spore germination through phytochrome in *Aspergillus nidulans*. Curr Genet. 2013; 59:55–62. [PubMed: 23385948]

Ruesch CE, Ramakrishnan M, Park J, Li N, Chong HS, Zaman R, et al. The histone H3 lysine 9 methyltransferase DIM-5 modifies chromatin at frequency and represses light-activated gene expression. G3. 2014; 5:93–101. [PubMed: 25429045]

Ruger-Herreros C, Gil-Sánchez MD, Sancar G, Brunner M, Corrochano LM. Alteration of light-dependent gene regulation by the absence of the RCO-1/RCM-1 repressor complex in the fungus *Neurospora crassa*. PLoS One. 2014; 9:e95069. [PubMed: 24747913]

Ruger-Herreros C, Rodríguez-Romero J, Fernández-Barranco R, Olmedo M, Fischer R, Corrochano LM, Canovas D. Regulation of conidiation by light in *Aspergillus nidulans*. Genetics. 2011; 188:809–822. [PubMed: 21624998]

Ruiz-Roldan MC, Garre V, Guarro J, Marine M, Roncero MIG. Role of the White Collar-1 photoreceptor in carotenogenesis, UV resistance, hydrophobicity, and virulence of *Fusarium oxysporum*. Eukaryot Cell. 2008; 7:1227–1230. [PubMed: 18503005]

Sancar C, Ha N, Yilmaz R, Tesorero R, Fisher T, Brunner M, Sancar G. Combinatorial control of light induced chromatin remodeling and gene activation in *Neurospora*. PLoS Genet. 2015; 11:e1005105. [PubMed: 25822411]

Sancar G, Sancar C, Brügger B, Ha N, Sachsenheimer T, Gin E, et al. A global circadian repressor controls antiphasic expression of metabolic genes in *Neurospora*. Mol Cell. 2011; 44:687–697. [PubMed: 22152473]

Schafmeier T, Haase A, Káldi K, Scholz J, Fuchs M, Brunner M. Transcriptional feedback of *Neurospora* circadian clock gene by phosphorylation-dependent inactivation of its transcription factor. Cell. 2005; 122:235–246. [PubMed: 16051148]

Schwerdtfeger C, Linden H. Blue light adaptation and desensitization of light signal transduction in *Neurospora crassa*. Mol Microbiol. 2001; 39:1080–1087. [PubMed: 11251826]

Schwerdtfeger C, Linden H. VIVID is a flavoprotein and serves as a fungal blue light photoreceptor for photoadaptation. EMBO J. 2003; 22:4846–55. [PubMed: 12970196]

Smith KM, Sancar G, Dekhang R, Sullivan CM, Li S, Tag AG, et al. Transcription factors in light and circadian clock signaling networks revealed by genome-wide mapping of direct targets for *Neurospora* White Collar Complex. Eukaryot Cell. 2010; 9:1549–1556. [PubMed: 20675579]

Somers DE, Devlin PF, Kay SA. Phytochromes and cryptochromes in the entrainment of the Arabidopsis circadian clock. Science. 1998; 282:1488–90. [PubMed: 9822379]

Salichos L, Rokas A. The diversity and evolution of circadian clock proteins in fungi. Mycologia. 2010; 102:269–278. [PubMed: 20361495]

Suelmann R, Fischer R. Nuclear migration in fungi – different motors at work. Res Microbiol. 2000; 151:247–254. [PubMed: 10875281]

Talora C, Franchi L, Linden H, Ballario P, Macino G. Role of a white collar-1-white collar-2 complex in blue-light signal transduction. EMBO J. 1999; 18:4961–8. [PubMed: 10487748]

Tan Y, Merrow M, Roenneberg T. Photoperiodism in *Neurospora crassa*. J Biol Rhythms. 2004; 19:135–143. [PubMed: 15038853]

Tataroğlu Ö, Lauinger L, Sancar G, Jakob K, Brunner M, Diernfellner ACR. Glycogen synthase kinase is a regulator of the circadian clock of *Neurospora crassa*. J Biol Chem. 2012; 287:36936–36943. [PubMed: 22955278]

Tudzynski B. Gibberellin biosynthesis in fungi: genes, enzymes, evolution, and impact on biotechnology. Appl Microbiol Biotechnol. 2005; 66:597–611. [PubMed: 15578178]

Vaidya AT, Chen C, Dunlap JC, Loros JJ, Crane BR. Structure of a light-activated LOV protein dimer that regulates transcription. Sci Signal. 2011; 4:ra50. [PubMed: 21868352]

Wang B, Kettenbach AN, Gerber SA, Loros JJ, Dunlap JC. *Neurospora* WC-1 recruits SWI/SNF to remodel *frequency* and initiate a circadian cycle. PLoS Genet. 2014; 10:e1004599. [PubMed: 25254987]

Wang N, Yoshida Y, Hasunuma K. Loss of Catalase-1 (Cat-1) results in decreased conidial viability enhanced by exposure to light in *Neurospora crassa*. Molecular Genet Genomics. 2007; 277:13–22.

Wang Z, Gerstein M, Snyder M. RNA-Seq: a revolutionary tool for transcriptomics. Nature Rev Genet. 2009; 10:57–63. [PubMed: 19015660]

Wu C, Yang F, Smith KM, Peterson M, Dekhang R, Zhang Y, et al. Genome-wide characterization of light-regulated genes in *Neurospora crassa*. G3. 2014; 4:1731–1745. [PubMed: 25053707]

Yoshida Y, Hasunuma K. Reactive oxygen species affect photomorphogenesis in *Neurospora crassa*. J Biol Chem. 2004; 279:6986–6993. [PubMed: 14625272]

Yoshida Y, Maeda T, Lee B, Hasunuma K. Conidiation rhythm and light entrainment in superoxide dismutase mutant in *Neurospora crassa*. Molecular Genet Genomics. 2008; 279:193–202.

Yu J. Heterotrimeric G protein signaling and RGSs in *Aspergillus nidulans*. J Microbiol. 2006; 44:145–154. [PubMed: 16728950]

Yu SM, Ramkumar G, Lee YH. Light quality influences the virulence and physiological responses of *Colletotrichum acutatum* causing anthracnose in pepper plants. J Appl Microbiol. 2013; 115:509–516. [PubMed: 23663215]

Zhao S, Fung-Leung W, Bittner A, Ngo K, Liu X. Comparison of RNA-Seq and microarray in transcriptome profiling of activated T cells. PLoS One. 2014; 9:n/a-e78644.

Zoltowski BD, Crane BR. Light activation of the LOV protein vivid generates a rapidly exchanging dimer. Biochemistry. 2008; 47:7012–7019. [PubMed: 18553928]

Zoltowski BD, Schwerdtfeger C, Widom J, Loros JJ, Bilwes AM, Dunlap JC, Crane BR. Conformational switching in the fungal light sensor Vivid. Science. 2007; 316:1054–1057. [PubMed: 17510367]

Zoltowski BD, Vaccaro B, Crane BR. Mechanism-based tuning of a LOV domain photoreceptor. Nature Chem Biol. 2009; 5:827–34. [PubMed: 19718042]

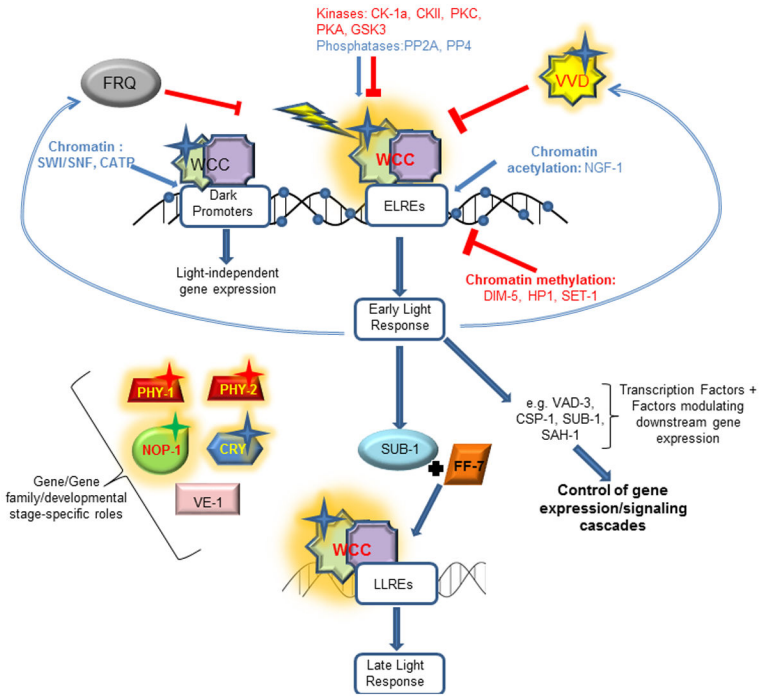

Figure 1. The *N. crassa* light response cascade

Light-activated WCC binds transiently to early light-responsive elements (ELREs) in the promoters of early light-responsive genes and induces the expression of several genes such as *frq*, *vvd*, *sub-1* and several transcription factors. The FRQ protein (via the circadian clock) modulates this initial light response in a circadian fashion, whereas VVD forms a dynamic pool with WCC activity to bring about photoadaptation. WCC activity and levels in response to light are also regulated by kinases and phosphatases. Chromatin state (histone acetylation and methylation) modulates light response at the level of target DNA, and histone acetylation (via NGF-1) is required for activation (gene expression), while methylation (via DIM-5, HP1 and SET-1) represses light responses. SUB-1, after its synthesis collaborates with FF7 and WCC to induce expression from late light response elements (LLREs) in the promoters of late light-responsive genes. Other photoreceptor proteins in *N. crassa* (PHY-1, PHY-2, NOP-1 and CRY), and proteins like VE-1 potentially modulate light responses in gene-family or developmental stage specific contexts. Finally, TFs induced after the first light response control gene expression cascades in a hierarchical manner. Components that respond to light are shown with halos and the proteins and chromophores are colored according to wavelength of light they absorb.

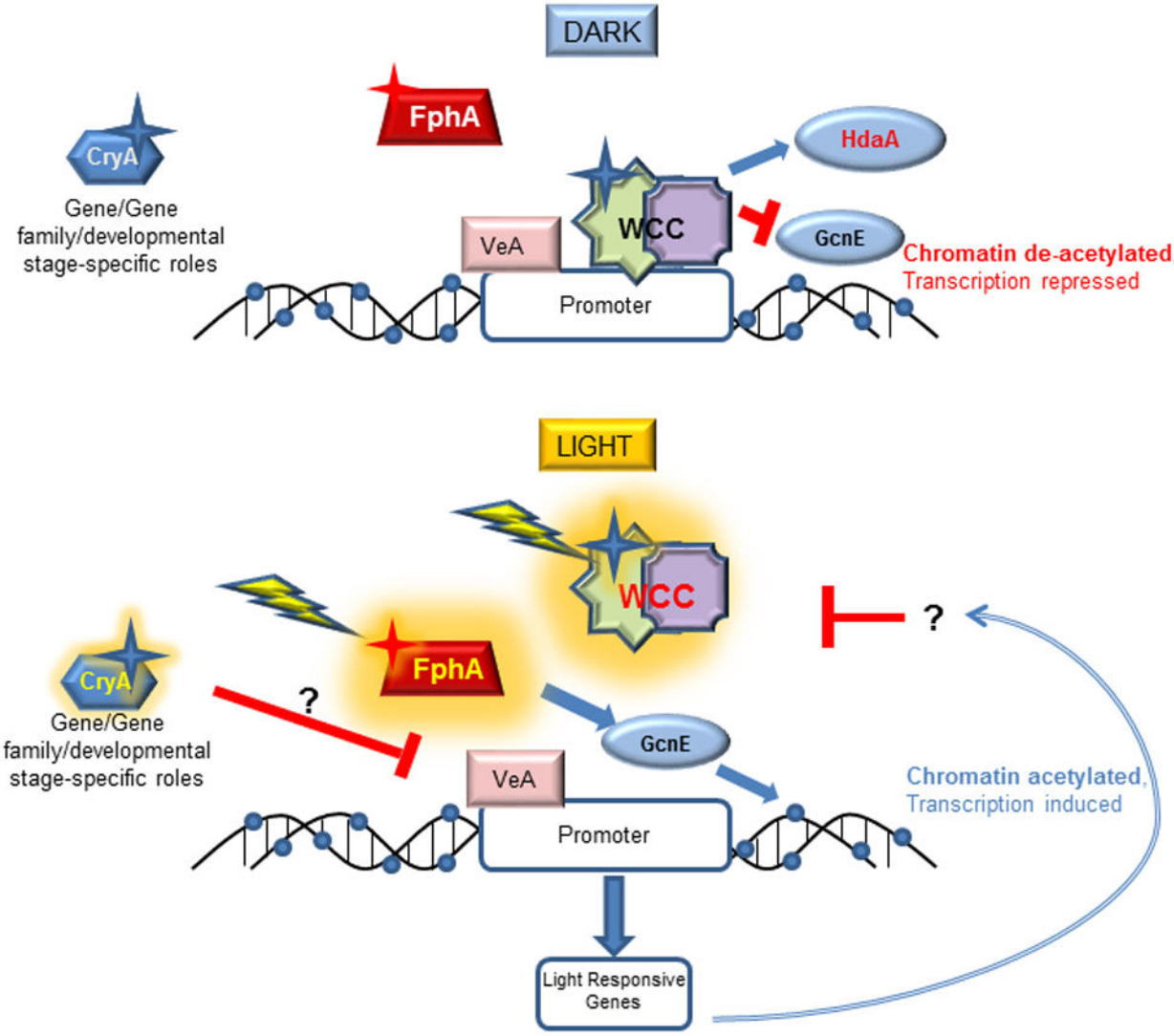

Figure 2. The *A. nidulans* light response cascade

Model is based on studies using the *ccgA* locus (Hedkte et al., 2015) *Dark*: The WCC complex (consisting of LreA/LreB) binds the target promoter and represses gene expression. WCC mediates its repression, at least in part, by promoting the activity of the histone deacetylase, HdaA. The WCC-DNA interaction is dependent upon VeA, which is also bound to the promoter. The phytochrome, FphA, does not bind DNA, but is required for VeA binding. *Light*: Either blue or red light leads to WCC dissociation from the DNA, which relieves its repressive function. FphA promotes the activity of the histone acetylase, GcnE, which opens the chromatin for gene induction. The role of cryptochrome, CryA, in *ccgA* expression has not been investigated. However, its role in repressing sexual developmental genes in blue light has been described and a postulated role in *ccgA* regulation is depicted.

Table 1
Functional summary of the major fungal photoreceptors

The protein nomenclature for the White Collar Complex, cryptochrome, VIVID, phytochrome and opsin orthologs are provided for *N. crassa*, *A. nidulans*, and *F. fujikuroi*. Included is a brief description of known protein functions or phenotypes of the corresponding mutants.

White Collar Complex		
Organism	**Protein**	**Notes**
N. crassa	WC-1/2	Promotes transcription at target loci; essential for all light-induced responses; essential for circadian gene expression.
A. nidulans	LreA/B	May promote or repress transcription of target loci; involved in both blue and red- light responses; not essential for all light-regulated processes; no reported clock in *A. nidulans*.
F. fujikuroi	WcoA/B	Regulates, but not essential for, the photo-induction of carotenoids; generally regulates conidiation; involved in blue and red light transcriptional response; no reported clock in *F. fujikuroi*.
VIVID-like		
N. crassa	VVD	Essential for photoadaptation via a direct interaction with the WCC.
A. nidulans	-	No known ortholog.
F. fujikuroi	VvdA	Regulates kinetics of carotenoid induction in response to light (positive and negative regulation); direct interaction with WcoA is unknown.
Cryptochrome		
N. crassa	CRY	Mutant displays no defect in the light response; may modulate WCC activity at specific loci.
A. nidulans	CryA	Represses sexual development in response to blue light; displays photolyase activity.
F. fujikuroi	CryD	Regulates, but not essential for, the light-induction of carotenoids.
Phytochrome		
N. crassa	PHY-1/2	No known red light response in *N. crassa*; mutant displays no overt phenotype in the light response; may modulate WCC activity at specific loci.
A. nidulans	FphA	Involved in both blue and red light responses; interacts directly with the WCC (LreA/B).
F. fujikuroi	Not-annotated	Uncharacterized; proposed role in red light-induction of carotenoids.
Opsin		
N. crassa	NOP-1	No known green light response in *N. crassa*; mutant displays no defect in the overt light response; may modulate WCC activity at specific loci.
A. nidulans	-	No known ortholog.
F. fujikuroi	CarO	green light-driven proton pump; represses spore germination in light.