there ought to be a continuum of infant clusters, from individual stars to massive protoglobular clusters. This appears to be the case. A continuum of infant clusters has begun to emerge, and the maximum mass of these clusters appears to correlate with the overall star formation rate of the host galaxies.

This correlation could be statistical (the more clusters are formed, the more likely it is that some of them will be massive), or it could be physical (the most massive clusters form in regions with the highest pressure, and high pressures also result in star formation). Theory might cause us to lean toward the latter; extremely high pressures appear to be required to form massive globular clusters (*8*, *9*).

We can now begin to understand where and how globular clusters formed by examining the formation of star clusters in the local universe. Important insights will come from investigating how the physical environment and properties of star formation scale between individual stars and extremely massive clusters. With the Expanded Very Large Array (EVLA) and Atacama Large Millimeter Array (ALMA) on the horizon, the next decade promises to address these questions with unprecedented sensitivity and resolution.

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PERSPECTIVES: CIRCADIAN RHYTHMS

A White Collar Protein Senses Blue Light

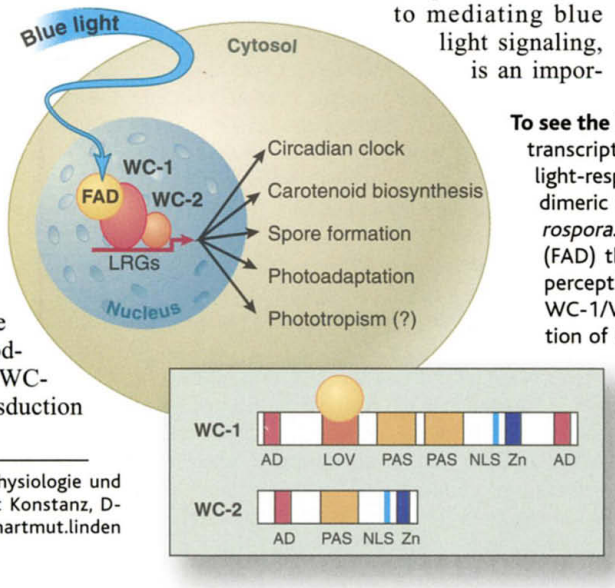
Hartmut Linden

The ability to perceive light is crucial for the survival of most organisms, enabling them to adapt to changing environmental conditions. Consequently, the capacity to sense and respond to light is widespread among bacteria, fungi, plants, and animals. In fungi, light regulates many developmental and physiological processes. For example, in the fungus *Neurospora crassa* blue light triggers the synthesis of photo-protective pigments (the carotenoids), spore formation, entrainment of the circadian clock, and growth toward a light source (phototropism) (*1*). *Neurospora* only perceives blue light, and seems to be "blind" to yellow, green, and red light. Exhaustive screening for light-insensitive *Neurospora* mutants only yielded two such mutants. These mutants are unable to synthesize carotenoids in response to light, and so have pigmented spores and white mycelia resulting in the moniker "*white collar* (*wc*)." Twenty years ago, it was proposed that the two *Neurospora* proteins encoded by the *wc* gene, WC-1 and WC-2, are involved in signal transduction in response to blue light and that one or both of these proteins might be the long-sought fungal blue-light photoreceptor (*2*). New studies reported by He *et al.* (*3*) and Froehlich *et al.* (*4*) on pages 840 and 815 of this issue, respectively, present convincing evidence that the WC-1 protein of *Neurospora* is indeed the blue-light photoreceptor of fungi.

Enhanced online at www.sciencemag.org/cgi/content/full/297/5582/777

The WC-1 and WC-2 proteins of *Neurospora* have features characteristic of transcription factors (see the figure) (*5*, *6*). They are found in the nucleus and bind to the promoters of light-regulated genes. The two proteins form heterodimers with each other, and dimerization is independent of the light conditions (*7*). The WC-1/WC-2 complex, in addition to mediating blue light signaling, is an important component of *Neurospora*'s circadian clock (*8*). Together with the circadian clock protein FREQUENCY (FRQ), the WC-1/WC-2 complex forms an autoregulatory feedback loop that is crucial for operation of the circadian oscillator, the molecular pacemaker that generates physiological rhythms (see the figure).

The WC-1 protein contains a chromophore-binding motif—similar to those identified in the blue-light photoreceptors of ferns and higher plants—called the light, oxygen, and voltage (LOV) domain (*9*). In their experiments, He *et al.* (*3*) removed the LOV domain from WC-1 and then expressed this protein in a WC-1–deficient strain of *Neurospora*. They found that light induction of the circadian clock gene *frq* and of another light-regulated gene was abolished in this mutant, suggesting that the LOV domain of WC-1 is important for light entrainment of the fungal circadian clock. Despite this defect in light regulation, the circadian clock could still be entrained according to temperature. He *et al.* also identified a flavin chromophore, flavin adenine dinucleotide (FAD), associated with the purified WC-1/WC-2 complex. The authors concluded that FAD is bound



To see the light, all you need is LOV. Light perception by the transcription factor WC-1 in the fungus *Neurospora*. The light-responsive proteins WC-1 and WC-2 form a heterodimeric complex that is localized to the nucleus of *Neurospora*. Blue light is perceived by a flavin chromophore (FAD) that is bound to the LOV domain of WC-1. Light perception may lead to a conformational change in the WC-1/WC-2 complex resulting in transcriptional activation of light-regulated genes (LRGs). These genes regulate physiological responses to light such as carotenoid biosynthesis, spore formation, and phototropism. The WC proteins have a dimerization (PAS) domain, a putative transcriptional activation (AD) domain, a zinc-finger DNA binding domain (Zn), and a nuclear localization signal (NLS). Only WC-1 contains the LOV domain.

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to the LOV domain of WC-1 and that WC-1 is the fungal blue-light photoreceptor. Interestingly, blue-light photoreceptors in plants carry a chromophore-binding motif that associates with another flavin molecule, flavin mononucleotide (FMN).

In complementary work, Froehlich *et al.* (*4*) arrived at the same conclusion via a different route. They identified two light-responsive DNA fragments in the promoter of the circadian clock gene *frq*. In electrophoretic mobility shift assays, they subsequently showed that both WC-1 and WC-2 were able to bind to these DNA fragments. They observed that one type of WC-1/WC-2 complex was present primarily in the dark, whereas the other prevailed under light conditions. Light intensity and wavelength requirements were similar for the formation of the light-specific WC-1/WC-2 complex in vitro and for the regulation of the circadian clock in vivo, supporting the notion that the light-sensitive WC-1/WC-2 complex contained the blue-light photoreceptor. WC-1, WC-2, and the chromophore FAD were sufficient for the formation of the light-specific complex, but only WC-1 seemed to be capable of binding FAD. Indeed, addition of FAD conferred light sensitivity on the WC-1/WC-2 dark complex. Corroborating the He *et al.* results, Froehlich and colleagues found that FAD but not FMN was required for light perception by *Neurospora*. Like He and colleagues, these authors also concluded that WC-1 is the blue-light photoreceptor responsible for light entrainment of the circadian clock in fungi.

Mutation of conserved amino acids in the WC-1 LOV domain results in a variety of defects in *Neurospora*'s response to light (*10*). These findings indicate that WC-1 is the blue-light photoreceptor of the fungal circadian clock, and is also a central switch that, with help from WC-2 and FAD, directs transcription of light-regulated genes in response to light. However, given that some *wc-1*–deficient *Neurospora* mutants can still respond to light, WC-1 may not be the only blue-light photoreceptor in fungi (*11*). Fungal proteins with the potential to be responsive to blue light include rhodopsin, proteins containing LOV domains, and proteins that resemble the photoreceptors of higher plants.

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PERSPECTIVES: VIROLOGY

CMV Makes a Timely Exit

Veronica Sanchez and Deborah H. Spector

A virus faces many obstacles during its life cycle. As an obligate parasite, it must exploit the machinery of the host cell to ensure its own replication. This is particularly true for large DNA viruses, such as the herpesviruses, that replicate their genomes in the nucleus of the host cell but require both the nucleus and cytoplasm for assembly of mature viral particles (see the figure, next page). Thus, the subviral particles of herpesviruses must exit the nucleus after replication of their DNA, a task made more difficult by the physical barrier of the nuclear envelope. On page 854 of this issue, Muranyi *et al.* (*1*) reveal the tricks that cytomegalovirus (CMV), a β-herpesvirus, plays in order to dissolve the nuclear lamina of the host cell nuclear envelope and gain entry to the cytoplasm.

The herpesvirus virion has a complex structure: The large DNA core is packed into an icosahedral capsid that is surrounded by an amorphous layer called the tegument, which includes at least 15 distinct proteins. The tegument is enclosed in a cell-derived lipid membrane envelope containing virus-encoded glycoproteins that are important for attachment of the virus particle to the host cell (see the figure, next page). Successful assembly of virions requires that the virus overcome the barriers that impede (i) encapsidation of the newly replicated DNA in the host cell nucleus, and (ii) the subsequent envelopment and release of viral progeny from the cytoplasm.

The nuclear envelope provides a major obstacle to movement of herpesvirus virions into the cytoplasm. This structure consists of two concentric lipid bilayers, termed the inner and outer nuclear membranes, which are connected to each other at the nuclear pores (see the figure, next page). The outer nuclear membrane is contiguous with the endoplasmic reticulum, and membrane proteins diffuse freely between the two structures. The inner nuclear membrane has a distinct set of transmembrane proteins, and current models postulate that these proteins are held in place by the nuclear scaffold. Underlying the inner nuclear membrane are intermediate filament-like proteins called lamins, which form a network about 50 nm thick that stabilizes the nuclear envelope [for a review, see (*2*)].

The Muranyi *et al.* study suggests a provocative strategy by which CMV circumvents the physical barrier of the nuclear envelope (*1*). The authors propose that CMV is able to modify and destabilize the nuclear lamina by recruiting a host cell protein kinase C to the inner nuclear membrane. In uninfected cells, solubilization of the nuclear lamina is a prerequisite for dissolution of the nuclear membrane at mitosis. This solubilization is accomplished by phosphorylation of the lamins by protein kinase C or by the Cdk1–cyclin B complex. Accumulation of protein kinase C at the nuclear envelope in CMV-infected cells enables the virus to disrupt the nuclear lamina, allowing virion progeny access to the inner nuclear membrane and initiating the first stage of envelopment.

Herpesviruses replicate their DNA in nuclear domains called viral replication centers. The newly replicated DNA is inserted into preformed capsids in the nucleus [for a review, see (*3*)]. Because the subviral particles are too large to pass through the nuclear pore, they must cross the nuclear lamin meshwork to reach the inner nuclear membrane. The particles acquire some of their tegument proteins and a primary envelope by budding from the inner nuclear membrane, resulting in enveloped particles within the perinuclear space. The next step is still unclear, but most of the available data imply that there is de-envelopment and loss of the primary envelope by fusion of perinuclear particles with the outer nuclear membrane, followed by release of DNA-containing capsids into the cytoplasm. These naked nucleocapsids are ushered to the Golgi complex in the cytoplasm, where they acquire additional tegument proteins and their mature envelope. They then bud into Golgi-derived vesicles that travel to the plasma membrane, where the virions are released from the host cell by exocytosis. (The alternative and less popular model is that the perinuclear virions leave the cell through the secretory pathway.)

Genetic and biochemical studies of herpes simplex virus (HSV) and pseudorabies virus have set the stage for the Muranyi *et*

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