Morphogenesis in *Candida albicans**

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Abstract

Candida albicans is termed a dimorphic fungus because it proliferates in either a yeast form or a hyphal form. The switch between these forms is the result of a complex interplay of external and internal factors and is coordinated in part by polarity-regulating proteins that are conserved among eukaryotic cells. However, yeast and hyphal cells are not the only morphological states of *C. albicans*. The opaque form required for mating, the pseudohyphal cell, and the chlamydospore represent distinct cell types that form in response to specific genetic or environmental conditions. In addition, hyperextended buds can form as a result of various cell cycle–related stresses. Recent studies are beginning to shed light on some of the molecular controls regulating the various morphogenetic forms of this fascinating human pathogen.

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INTRODUCTION

The fungi are a highly successful and diverse group of organisms. They exhibit two prominent modes of proliferation: the hyphal mode, in which continuous growth at a tip leads to an elongated tube wherein separate cells are generally delineated by septa, and the yeast growth mode, in which discrete cells elongate or bud-off daughter cells that typically disassociate from the mother cell. Most fungi are hyphal; even mushrooms are constructed from collections of filamentous hyphae. Understanding this mode of growth has been a long-standing scientific goal. The yeast form is less frequently encountered in nature but is found in economically important and scientifically well-investigated organisms such as *Saccharomyces cerevisiae* and thus is also a highly studied growth pattern.

Intriguingly, some fungi are not limited to a specific growth mode. Depending on the environmental conditions, these fungi are capable of growing in either the yeast or the hyphal pattern; such fungi are termed dimorphic. Among the best studied of these fungi is the human pathogen *Candida albicans*. Although dimorphic is a bit of a misnomer given the variety of cellular forms exhibited by this organism, specific conditions can be found in which essentially all the cells grow as yeast, and other conditions can be found in which most cells grow as hyphae, and the switch from one mode to the other in response to external signals is rapid. This ability to switch between such distinct modes of proliferation is intriguing, but it has also drawn interest because of its apparent relevance to the pathogenicity of the organism (120). In addition to this yeast-hyphal transition that defines *C. albicans* as a dimorphic fungus, a number of other naturally occurring morphological forms are characteristic of specific cellular functions (**Figure 1**). These distinct morphologies include the opaque form, characteristic of mating-competent cells (112); the chlamydospore, a currently enigmatic thick-walled cell formed typically under suboptimal growth conditions (44); and the pseudohyphal form, which often coexists with the yeast and hyphal forms in vegetative cultures and during infections (120).

In this article we provide an overview of developments in our understanding of the morphological variation exhibited by *C. albicans*. The general topic of *C. albicans* morphogenesis has been the subject of many recent reviews (21, 72, 80, 120, 130), so this manuscript focuses mainly on recent developments. There are also excellent reviews documenting our understanding of the regulation of morphogenesis in *Saccharomyces cerevisiae* (31, 58, 70) and of the growth regulation of obligate hyphal fungi such as *Aspergillus nidulans* and *Neurospora crassa* (94, 128). Ultimately our picture of *C. albicans* morphogenesis involves an understanding of both the processes and regulatory circuits unique to *Candida*, as well as of the general functions characteristic of both the budding and hyphal growth modes.

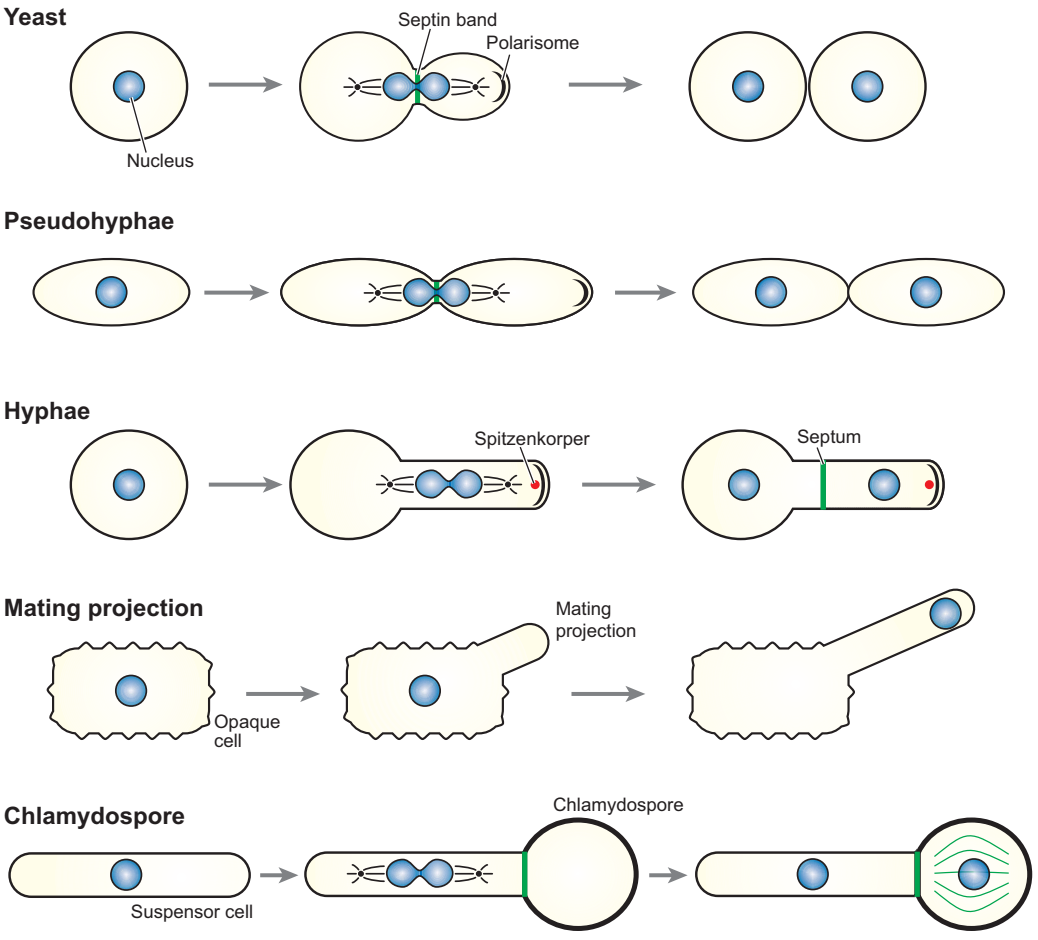

Figure 1

Distinct morphological forms of *C. albicans*. In yeast-form growth a blastospore buds off a new cell, resulting in two discrete cells. The separated spindle pole bodies elaborate spindles that separate the chromosomes across the mother-daughter junction defined by a septin band (*green*). Polarized growth is defined by a crescent-shaped polarisome at the tip of the growing bud. In pseudohyphal growth the nuclear division also crosses the mother-daughter junction defined by a septin band, and polarized growth is characterized by the polarisome. The cells themselves are more elongated than during yeast growth, and the cells remain attached after cytokinesis. Hyphal growth is defined by both a polarisome and a Spitzenkorper at the tip of the growing hyphae. The nucleus divides within the elongating germ tube across the region that defines the eventual septum and then one nucleus migrates back to the mother cell and the other moves farther into the elongating germ tube. Opaque-form cells are capable of responding to mating pheromone by elongating a mating projection, or shmoo. The nucleus migrates into the projection and, after fusion of the mating projection to a cell of the opposite mating type, undergoes a Kar3p-mediated fusion with the other nucleus. Chlamydospores are formed at the end of suspensor cells. They have a thicker cell wall and are larger than blastospores; the nucleus divides within the suspensor cell and then the daughter nucleus migrates into the chlamydospore across a septin structure. The final chlamydospore has an elaborate septin-derived substructure. See text for details.

THE YEAST-HYPHAL TRANSITION

Polarity Determinants

GTPase: guanosine triphosphate binding/hydrolyzing enzyme

Polarisome: complex of proteins involved in directing polarized growth

Germ tube: initial elongating structure that will become a hypha

The ability to switch rapidly from yeast-form growth to hyphal growth is a defining characteristic of *C. albicans* cells, and it is believed that each form of growth provides critical functions required for the pathogenic lifestyle (120). In both modes the cells exhibit polarized growth; the degree of polarity is more extreme in the hyphal state, because the alternation of polarized growth with isotropic growth that characterizes the yeast mode of proliferation is replaced by the essentially permanently polarized apical growth of the hyphal cell. In general, polarized growth is an important characteristic of cells, with intrinsic cues and extrinsic signals combining to determine both the shape of individual cells and their spatial organization in multicellular structures. Eukaryotic cells typically use small GTPases of the Rho superfamily, in particular members of the Cdc42/Rac group of enzymes, to regulate the overall process (47, 59); these GTPases appear implicated primarily in regulating actin polymerization and thus in controlling polarized growth and secretion through modulation of the actin cytoskeleton (63). Cdc42p is a central regulator of a complex of proteins required for proper polarized growth. This complex, initially identified in budding yeast and termed the polarisome (109), contains the formin Bni1p as well as the proteins Bud6p, Spa2p, and Pea2p (**Figure 1**).

Members of the polarisome and its GTPase regulator are also involved in polarized growth in *C. albicans*. Deletion (91) or regulated shut-off (122) of the *C. albicans CDC42* homolog blocks cell cycle progression under both yeast- and hyphal growth conditions and generates apolar growth, as did inactivation of the protein in *S. cerevisiae*. During hyphal growth the Cdc42p GTPase remains at the tip of the elongating hyphal tube; this localization is disrupted by latrunculin A treatment, suggesting it is dependent on the actin cytoskeleton (52), unlike the localization control during yeast-form growth. Regulation of the GTPase activity of Cdc42p plays an important role in controlling the formation of hyphae, as reduction in GTPase function through loss of the exchange factor Cdc24p blocks the ability to form hyphae and induce hyphae-specific transcripts (14), and an increase in the activated form by deletion of the GTPase-activating proteins Rga2p and Bem3p allows cells growing under normally pseudohyphae-inducing conditions to form hyphal-like germ tubes (36).

The polarisome is also implicated in polarized growth in *C. albicans*, as deletion of the homologs of *BNI1* (30, 88), *SPA2* (135), and *BUD6* (113) results in similar phenotypes. Yeast-growth cells exhibit random budding and delocalized surface growth, leading to round cells with large bud necks; in hyphal growth conditions these mutants make abnormally thick hyphae. Although cells can still polarize in the absence of polarisome factors, polar growth cannot be maintained. Unlike *S. cerevisiae*, but like higher cells, *C. albicans* also has a homolog of the Rac GTPase. This protein, termed Rac1p, plays a role distinct from that of Cdc42p, and is not needed for budding and proper actin organization, but it is needed for filamentation in matrix-embedded conditions (13). Thus, there may be environment-specific polarity-determining factors in hyphae. Components that directly interact with the actin cytoskeleton also play important roles in the control of *C. albicans* polarity. The type I myosin encoded by *MYO5* is required for hyphal growth but not for budding; surprisingly, the polarized distribution of cortical actin patches can be disrupted in specific *myo5* mutants without blocking hyphal development (100). The type V myosin encoded by the *MYO2* gene is also required for development of true hyphae but not for budding (132), and loss of the Wiskott-Aldrich syndrome protein Wal1 (126) and the Sla2 protein (101) also disrupts hyphal development without these proteins being essential for viability during yeast-form growth.

Spitzenkorper and Hyphal Development

In inherently filamentous fungi such as *Aspergillus* and *Neurospora* there is a distinct structure called a Spitzenkorper (**Figure 1**) near the tip of the growing cell that is implicated in polarized growth (125). This Spitzenkorper is believed to direct secretion to the growing tip and thus has a function similar to that of the polarisome of the budding yeast (51). However, it does not appear that the switch from yeast to hyphal growth in *C. albicans* occurs by the transfer of regulatory control from a polarisome to a Spitzenkorper, as both elements have been identified in hyphal cells (37) and, as noted, hyphal cells lacking polarisome components do not generate normal hyphae. Thus, the Spitzenkorper appears to be one of the unique characteristics of the hyphal growth pattern in *C. albicans*, and it may function in conjunction with, instead of in place of, the polarisome (37). Because the Spitzenkorper is only found in actively growing hyphae of filamentous fungi, care must be taken when using it as a diagnostic feature for the true hyphal growth mode in *C. albicans*; the mere absence of the structure in a polarized cell may reflect a poor growth rate or nongrowing cell. Individual growth rates and localization of Spitzenkorper markers need to be monitored in the same cells.

Defining the molecular characteristics of the Spitzenkorper, and the relationship between this element and the polarisome, will be an important step in both defining polarity control in hyphal cells and specifying the distinction between hyphal and yeast-form growth. Because the Spitzenkorper is identified primarily as a vesicle-rich structure currently visualized in living cells through staining with dyes such as FM4–64, the protein/enzymatic components of the Spitzenkorper are as yet poorly characterized. In *C. albicans* the polarisome proteins Spa2p and Bud6p are generally not associated with the Spitzenkorper, whereas Bni1p and, when overexpressed, a fraction of Cdc42p are apparently associated with this structure (37). In filamentous fungi, components of the polarisome that function at the hyphal tip, but are not involved in the formation or stability of the Spitzenkorper, have been identified (124). The Golgi structure of hyphal *C. albicans* cells is fundamentally relocalized to the growing tip of the cell, and this organellar redistribution is dependent on the formin Bni1p (104). It does not appear that this repositioned Golgi represents the Spitzenkorper, but the ability to identify cellular localization of membranes through staining with lipophilic dyes, and the ability to localize proteins through green fluorescent protein (GFP) tagging, will permit the identification of genes required for Spitzenkorper formation and positioning and the proteins that colocalize with the Spitzenkorper. These approaches applied in both *C. albicans* and in filamentous fungi should provide rapid insights into the organization of the Spitzenkorper and its relationship to other polarity-related functions in the cell. They will also allow a more direct investigation of whether the Spitzenkorper-like structure in *C. albicans* is required for rather than associated with polarized growth of hyphae. To date, treatments that perturb the integrity of the Spitzenkorper, including deletion of polarisome genes or exposure to methyl benzimidazol-2-yl-carbamate (37), also disturb polarized growth, but the growth defect may not be due to a direct effect on the Spitzenkorper-like structure itself.

Spitzenkorper: membraneous structure at the growing tip of hyphal cells

GFP: green fluorescent protein

Nuclear Positioning and Division

In addition to the presence of the Spitzenkorper, other characteristics distinguish a *C. albicans* cell initiating a bud from those initiating a germ tube that elaborate into hyphae. A critical difference is the arrangement of the septum and the division of the chromosomes. In yeast cells, as in cells of *S. cerevisiae*, the dividing nucleus is positioned to the neck between the mother and daughter cells, and the nuclear division partitions one chromosomal complement into the mother cell and the other into

Cytokinesis: process of separating two cells after mitosis has segregated the genetic information

APSES (Asm1p, Phd1p, Sok2p, Efg1p, and StuAp): group of structurally related transcription factors

cAMP: cyclic AMP

the daughter cell (**Figure 1**). This division takes place across the septum structure, which ultimately defines the site of cytokinesis (120). A molecularly similar event occurs in pseudohyphal cells—the nuclear division also occurs between the mother and daughter cell, and although the dividing cells are elongated relative to the yeast cells, the behavior of the dividing nucleus and the organization of the septum are similar.

The cellular behavior of the nucleus is distinct in unbudded yeast cells that are elaborating a germ tube while in the G1 phase of the cell cycle. In these cells the nuclear division and the position of the initial septum take place in the elongating germ tube rather than at the junction between the mother cell and the polarizing structure (46). The nucleus migrates into the extending germ tube, divides across the site of the incipient septum, and then one nucleus migrates back into the mother cell and the other moves out toward the tip of the elongating germ tube (**Figure 1**). This complex movement of the nucleus during the initial steps of hyphal development is orchestrated by microtubules (46). The dividing cell appears to be committed to elaborate a germ tube or a bud prior to any morphological distinction in the emerging polarized structure. However, the cell fate commitment appears to be reversible. Yeast cells in later cell cycle stages can be induced to form hyphae from their established buds (53). In these cells, the first nuclear division occurs across the bud neck, where there is also a constriction due to the initial bud emergence from the yeast cell. Thus, defining the initiation signals and how those signals are regulated by external conditions will be an important goal of the next period of investigation of the yeast-to-hyphal transition.

Signaling Pathways

Our growing appreciation of the complex machinery involved in directing polarized growth has been paralleled by an improved understanding about the signaling pathways involved in transferring information about the environmental state that ultimately controls the yeast or hyphal morphology. These environmental conditions can vary considerably, but a standard trigger of hyphal development is the combined addition of serum to rich growth medium and the elevation of the growth temperature to 37°C (130). However, nutrient-poor media such as Lee's or more chemically defined inducers such as *N*-acetyl-glucosamine, together with a rise in temperature to 37°C, are also suitable for inducing a high frequency of hyphal growth. An important consideration is that improved characterization of the hyphal, yeast, and pseudohyphal states can permit a more exact definition of the role of specific genes and pathways in cell morphogenesis. Studies in which filamentous growth was described without specification to the hyphal or pseudohyphal form must be re-evaluated to distinguish functions that are common to both states from those common to only one state.

Much of the analysis of the yeast-hyphal transition has been concerned with transcriptional control and the relevant transcription factors (**Table 1**). The initial observation that the combined loss of the Cph1p and Efg1p transcription regulators blocked the hyphal transition in most tested conditions, and led to reduced virulence, focused interest on these two transcription factors and their controlling networks (81). Subsequently, a large number of transcription regulators have been reported to play roles, either positive or negative, in the yeast-to-hyphal transition. An important goal has been to connect these regulators to signaling pathways transferring information from the external environment and to establish the regulatory relationships among the various transcription factors.

The APSES protein Efg1p plays a central role in the general control of morphogenesis (42); in the yeast-hyphal transition the regulatory circuit leading through Efg1p is linked to cAMP metabolism. The role of the cAMP-mediated signaling pathway has been well established in the yeast-to-hyphal

Table 1 Transcriptional regulators of morphogenesis

State	Factor	Loss-of-function phenotype	Reference(s)
Opaque state	Wor1p	Blocks opaque state	136
	Efg1p	Activates subset of functions	115
	Tup1p	Modulates subset of functions	103
Hyphal state	Rim101p	Block in alkali-induced hyphae	40
	Czf1p	Block in matrix-induced hyphae	26
	Flo8p	Block in serum-induced hyphae	29
		Derepresses embedded hyphae	29
	Efg1p	Block in serum-induced hyphae	42
		Derepresses embedded hyphae	49
	Hap5p	Block in hyphae on Spider medium	62
	Efh1p	Synergizes with *efg1*	42
	Ace2	Defect in hypoxia-triggered hyphae	95
	Mcm1p	Activates hyphae	106
	Ash1p	Reduces filaments on Spider medium	57
	Cph2p	Moderate inhibition of hyphae	75
	Tec1p	Suppresses serum-induced hyphae	107
Chlamydospores	Rim101p	Affects proper timing	99
	Nrg1p	Derepresses formation on Staib agar	119
	Efg1p	Blocks chlamydospore formation	114
Pseudohyphal state	Tup1p	Constitutive pseudohyphae	24
	Nrg1p	Constitutive pseudohyphae	25, 96
	Fkh2p	Constitutive pseudohyphae	19
	Tcc1p	Constitutive pseudohyphae	67
	Rfg1p	Constitutive pseudohyphae	69
	Ssn6p	Moderately pseudohyphal	56
	Ace2p	Inappropriate pseudohyphae	68
	Rap1p	Inappropriate pseudohyphae	22
Mating projections	Cph1p	Mating defective	33

switch, as a variety of mutations that affect the pathway influence hyphal development (9, 34, 64, 77, 87). In particular, cells defective in the adenylyl cyclase Cdc35p are competent to grow as yeast cells but are unable to form hyphae under standard inducing conditions (105). Transcription profiling suggests that Efg1p and Cdc35p control a set of genes implicated in the yeast-to-hyphal transition, but both control significant nonoverlapping sets of other genes, consistent with each regulator influencing different functions separately from hyphal development (50). In addition, the Flo8p transcription factor, which is essential for serum-induced hyphal development, controls a subset of Efg1p-regulated genes (29). The regulation of Efg1p function through this pathway is suggested to be through direct phosphorylation by the cAMP-dependent protein kinase (23), and interaction with other regulators such as Flo8p and Czf1p may explain the ability of Efg1p to act both as an activator and as a repressor, depending on the conditions. As yet the direct relationships between the changes in gene expression and the changes in cell shape have not been established, and this will be an important area of future studies.

MAP kinase: mitogen-activated protein kinase

Other pathways that transmit external signals to the transcription machinery, including a MAP kinase signaling pathway, an external pH sensing pathway and a matrix-sensing pathway, have also been identified as playing roles in hyphal development under specific conditions. The MAP kinase pathway signals in opaque-form cells to direct mating, but initial studies established a minor role in yeast-hyphal signaling under specific nutrient conditions (38, 71, 76). The target of the MAP kinase cascade is proposed to be the Cph1p transcription factor on the basis of comparison with *S. cerevisiae*, and Cph1p provided a residual level of virulence in the *efg1* mutant background (81). The phenotypes of the various signaling mutants in limiting hyphal development under specific conditions were similar to those of the transcription factor mutant, consistent with a common molecular role.

The external pH is one of many signals important for regulating the yeast-to-hyphal transition. Many of the players in a pathway transmitting the pH signal to the intracellular machinery are homologs of the components identified in *S. cerevisiae* that also regulate response to pH. The main transcription factor is Rim101p (40), which is proteolytically activated by the Rim13p protease (78); loss of Rim101p function blocks alkaline-induced hyphal development. Rim20p and Rim8p act upstream of the proteolytic step (40), as do several membrane proteins, including Dfg16p, that may act as pH sensors (12). The processing and activation of Rim101p also require members of the endosomal complex required for transport (ESCRT) protein module (35, 133). A large number of genes are under control of the Rim101p factor (20), and connecting the expression of these genes to the morphological regulatory system is necessary to understand the link between pH sensing and morphogenesis.

The regulation of hyphal growth in a matrix appears regulated by a complex interplay of the transcription factors Czf1p, Efg1p, Flo8p, and other signals. Czf1p is encoded by an autoinhibited gene required for hyphal formation when *C. albicans* cells are growing in embedded conditions (26). In contrast, the Efg1p/cAMP pathway that is positively required for serum-induced hyphae must be inhibited to allow hyphal development in these conditions, because *efg1*, *cdc35*, and *flo8* mutants result in derepression of hyphal development during matrix-embedded growth (29). The upstream regulatory region of *CZF1* is large, and it appears that transcriptional control integrates a variety of signals to regulate *CZF1* expression (123), similar to *WOR1* in the regulation of the opaque state (136). Both Czf1p and Flo8p may physically interact with Efg1p (29, 49), and Czf1p serves to permit hyphal growth during embedded growth by relieving the inhibitory action of Efg1p under these conditions (123). Separating the roles of the physical matrix conditions from the changes in aeration associated with embedded hyphal development is important to establish a clear picture of this regulatory circuit.

Although the modulation of the transcription program of the cell is an obvious consequence of the transmission of external signals that direct morphological changes, these are not the only outputs of the signaling pathways. For example, kinases are key components of many upstream signaling modules that regulate transcription, and their phosphorylation targets are not limited to transcription factors. Defining the important substrates of such critical components as the cAMP-dependent protein kinases is essential to fully understanding the regulation of morphological transitions. Overall, it is critical that researchers establish how these signaling pathways control the machinery directing polarized growth. Whether the polarisome is a direct target of signaling pathways, whether Cdc42p activity or localization is regulated through the transmission of external hyphae-inducing conditions, or whether the Spitzenkorper forms in direct response to the environment must be addressed. Ultimately the link between signaling and morphogenesis must be made at the level of the growth pattern itself, not solely at the

spectrum of gene expression correlated with this pattern.

Roles of Cyclins and Cyclin-Dependent Kinases in Morphological Control

The central cell cycle regulatory kinase, as well as a group of regulatory cyclin molecules, has been identified in *C. albicans*, and several of these molecules appear to have specific roles in morphogenetic control. The initial identification of functional cyclins and the cyclin-dependent kinase was through analysis of *C. albicans* gene functions in *S. cerevisiae* (108, 129), but genome-sequencing efforts have now allowed the comprehensive identification of the cyclin/kinase gene families in the fungal pathogen (21). As in the model yeast *S. cerevisiae* there is a single cyclin-dependent kinase that is the key regulator of cell cycle progression, and this kinase interacts with a variety of cyclins to control the G1, S, and mitotic transitions, as well as aspects of morphogenesis (90).

Ccn1p is a member of the G1 class of cyclins on the basis of both structural and expression pattern similarity to G1 cyclins of yeast and is essential for the maintenance, but not initiation, of hyphal growth under specific nutrient conditions (84). The cyclin Hgc1p also shows structural similarity to the G1 cyclin class but was expressed only in hyphal cells. Hgc1p is required for hyphal development under a variety of tested conditions (134), and Hgc1p expression is limited to the apical, growing cell of the developing hyphae (127). Another G1 class cyclin, Cln3p, is required for budding but not initiation of hyphal growth, as regulated shut-off of Cln3p expression leads to cell enlargement followed by initiation of germ tube extension (8, 32). These data suggest an important regulatory connection between the G1 cyclins and morphogenetic control, but this connection seems inconsistent with the observation that initiation of germ tube formation was apparently cell cycle independent (53). Based on expression of Hgc1p, which peaks in the G1 phase (127), perhaps cells are more primed to form hyphae during G1.

B-type cyclins and the cyclin-dependent protein kinase Cdc28p are also implicated in control of morphogenesis, but not directly in control of the yeast-to-hyphal transition. Loss of the B-type cyclin Clb4p caused pseudohyphal growth, whereas shut-off of the essential cyclin Clb2p resulted in more highly polarized filaments called elongated buds (18). Similar filamentation has been seen in mutants that lack function in other essential cell cycle components (7). Although these filaments superficially resemble hyphae, they are discussed separately. Regulated shut-off of *CDC28* expression leads to diverse types of filamentation (121), including pseudohyphae, true hyphae, and elongated buds.

Currently the data linking cyclins and cell morphogenesis have focused primarily on the role of cell cycle regulators in the transition between the yeast and hyphal states. It is important to extend these observations to other aspects of morphogenesis. For example, cell cycle regulation is coupled to the development of the mating projection during mating, and the control of cyclin function is likely important for this process. Similarly, cell cycle control in the development of the chlamydospore is a critical component of the process. Overall the link between the cyclin-regulated function of the cyclin-dependent protein kinase and cellular morphogenesis is expected to be strong, and the challenge is to make this link in each of the unique morphological forms of *C. albicans* and then to establish the details of the regulatory circuit.

Opaque/White: two morphologically distinct cell/colony forms of *C. albicans*

THE WHITE-OPAQUE SWITCH

Although the dramatic change between the yeast and hyphal state is a well-studied hallmark of *C. albicans*, phenotypic switching, in particular the white-opaque transition, has also been an area of extensive investigation. The white-opaque switch was initially identified as a cellular and colony morphology

transition that was limited to specific strains. The switch was epigenetic—each state was stable but capable of transitioning to the other state at a frequency that was higher than the standard mutation rate, and the frequency could be modified by external conditions such as temperature (110). White-form cells are of the classic ovoid shape characteristic of budding yeasts such as *S. cerevisiae*, and they form domed colonies that are a creamy color. Opaque-form cells are elongated and have a cell wall that has frequent pits (3), unlike the relatively smooth cell wall surface of white-form cells. Opaque-form cells generate colonies that are flattened and more gray than the colonies generated by white-form cells (110). The viability of opaque-form cells is reduced relative to white-form cells under many growth conditions, and this allows the opaque-form cells to be distinguished as forming colonies that can be more readily stained by vital dyes such as phloxine B.

Recent evidence has established that this white-opaque transition is intimately coupled to the sexual mating process in *C. albicans*. The ability to switch to the opaque state depends on whether the cells are homozygous for the *MTL* locus that controls cell type; most *C. albicans* cells are heterozygous for the *MTL* locus (*MTL***a**/*MTL*α) and thus are unable to switch (92). The inability of *MTL* homozygous strains to switch is regulated by the **a**1/α2 repressor. This heterodimeric repressor is derived from the **a**1 protein encoded by the *MTL***a** locus, and the α2 protein encoded by the *MTL*α locus, and thus can only be generated in heterozygous strains. A key regulatory role of this repressor is to control the expression of another transcription factor, the product of the *WOR1* gene (55, 136). The Wor1p transcription factor (**Table 1**) appears to be a primary controller of the opaque state; ectopic expression of Wor1p efficiently induces the opaque state in *MTL* homozygotes, and a pseudo-opaque state can be triggered by activating Wor1p expression even in *MTL* heterozygotes (136).

The regulation of Wor1p explains the epigenetic characteristics of the opaque state. Wor1p is autoregulated; chromatin immunoprecipitation experiments suggest as many as five binding sites for Wor1p in its own promoter region (136). The current data suggest that once Wor1p expression is established, it tends to remain on, with a positive-feedback loop keeping the cell in the opaque state. However, once Wor1p levels drop below a critical threshold, this positive-feedback loop is disrupted and the cell establishes a stable white state. It could be expected that the circuit itself is temperature sensitive because high temperatures lead to the switching of the opaque form to the white form. Whether this is due to direct temperature sensitivity of the Wor1 protein itself, or to some other aspect of the regulatory circuit, remains to be established.

There are many genes whose expression switches between cells in the white form and cells in the opaque form (74). Many of these genes are likely to control the distinct physiologies of the two different cell types: White-form cells express genes characteristic of a fermentative life style, whereas opaque-form cells show characteristics of an oxidative metabolism (74). However, many genes regulated differently between the two cell types presumably define the different growth patterns and cell surface structures exhibited by the white- and opaque-form cells. Analysis of opaque-specific genes for the controllers of morphological patterning is an important step in the analysis of the opaque-phase cells.

The Efg1p transcription regulator that is a critical positive controller of the yeast-to-hyphal transition and the Tup1p negative regulator of pseudohyphal development also play roles in the white-opaque switch (**Table 1**). Efg1p is highly expressed in white-form cells but not in opaque-form cells (114); this expression is driven from a strong, white-phase-specific promoter (73). Loss of *EFG1* expression causes otherwise white-form cells to take on some of the morphologies of opaque-form cells, in particular the elongated cell

shape, and to express some opaque-specific genes (114, 117). Ectopic overproduction of Efg1p shifts opaque-form cells to the white morphology (115), and thus repression of *EFG1* expression appears necessary to establish the opaque state. However, certain structural characteristics of the opaque state, in particular the surface pimples, are not found in the *efg1*-null strains. Therefore it appears that Efg1p may regulate a subset of the genes involved in establishing the opaque state.

The Tup1p transcriptional regulator also influences the opaque state. Loss of Tup1p, which creates a constitutive pseudohyphae-like state in white-form cells (24), also dramatically affects the morphology of opaque-form cells (103). Loss of *tup1* deregulates expression of some phase-specific genes but still permits the establishment of a mating-competent cell type. Thus, like loss of Efg1p, loss of Tup1p appears to modify some, but not all, characteristics of opaque-form cells. *MTL* homozygous cells deleted for *TUP1* have the ability to switch among a variety of cell and colony morphologies; whether this pattern of switching is influenced by Wor1p remains to be established.

These experiments have identified a number of transcriptional regulators that play critical roles in the establishment of the opaque state; however, we do not know the details of how these regulators control morphology. Some early experiments characterized the actin cytoskeleton in opaque-phase cells (3), but the spatial and temporal regulation of the polarity machinery has not been extensively examined. The ability to form the surface pimples characteristic of the opaque state must be under regulatory control, but this process is currently enigmatic. The ability to manipulate the opaque state through modulation of the Wor1p transcription regulator should make the state more amenable to investigation, and so future studies directed at identifying the control elements that establish the unique morphological characteristics of opaque-form cells should provide rapid progress in defining the functional bases for this distinctive morphological state.

Pheromone-Mediated Morphological Changes

In addition to directing the morphology of the opaque-form cell itself, the establishment of the opaque state is a prerequisite for the induction of morphological modifications due to the action of mating pheromones. These pheromones are diffusible peptide and lipopeptide molecules that are produced by cells of one mating type and act on cells of the other mating type. The action of the pheromones initiates a spectrum of physiological, morphological, and cell cycle changes that prepare the potential mating partners for cell and nuclear fusion (11). The components controlling the signaling process, including elements regulating both morphological and transcriptional programs as well as the cell cycle, have been extensively documented in *S. cerevisiae*, and the processes in *C. albicans* appear inherently similar (61).

Both *C. albicans* and *S. cerevisiae* produce structurally distinct pheromones from each of the two mating types. *MAT*α cells of the budding yeast and *MTL*α cells from *C. albicans* produce simple peptide pheromones (17, 83, 97, 102), and the *MAT***a** and *MTL***a** cells from the two species produce pheromones that are either proven or predicted to be lipid-modified peptides (28, 41). The identification of the single gene encoding the *C. albicans* α-factor peptide has allowed for the chemical synthesis of the pheromone and the analysis of the behavior of cells in the presence of this factor (17, 83, 102). *C. albicans* cells treated with pheromone modify their gene expression profiles (17, 83), their cell cycle progression (102), and their cellular morphology (83). In keeping with the terminology established for the modified morphology developed by pheromone-treated cells in *S. cerevisiae*, *C. albicans MTL***a** cells treated with α-factor have been termed shmoos (83), even though their actual shapes are unlike either the Al

Biofilm: complex assembly of various morphological forms of *C. albicans* cells embedded in a cell-derived matrix

Capp creation or the characteristic forms of the budding yeast. Mating projections may be a more accurate terminology for the structures formed in *C. albicans* in response to mating pheromone, but the term shmoo provides a convenient if physically inaccurate summary of a complex morphological process (**Figure1**).

The developing shmoo of a pheromone-treated *S. cerevisiae* cell uses the machinery of polarized growth characteristic of budding cells, but it modifies, in response to the pheromone gradient, the direction of polarized growth by overriding the intrinsic rules for initiating a bud (27). Modifications also occur in the arrangement of the septins at the site of the shmoo evagination relative to septin arrangement in a budding cell (43). Overall, the establishment of the polarized growth of the shmoo without the constriction inherent in the dividing bud of a *S. cerevisiae* cell has led some researchers to equate aspects of shmoo formation in budding yeast to the development of a germ tube in *C. albicans* (37). This analogy cannot be taken too far, as proteins such as Far1p that are critical for shmoo formation in the budding yeast are not required for germ tube development in *C. albicans* (5). However, the ability to initiate polarized growth from a nonconstricted point of emergence, the ability to migrate the nucleus into this polarized extension, and the requirement for the actin cytoskeleton to direct the localization of the Cdc42p GTPase (52) are common characteristics of shmooing yeast cells and *C. albicans* cells initiating a germ tube.

A detailed analysis has been undertaken to establish the cell biology of the developing mating projections in cells in a mating mixture consisting of both mating types (82). Only mixtures of cells of opposite mating types were observed to trigger the formation of mating projections, and these projections could be extensive. It has been proposed that a complex interaction between white- and opaque-form cells can lead to biofilm formation that stabilizes pheromone gradients and permits *MTL***a** and *MTL*α cells to signal over long distances (39). This model implies that rare opaque-form cells can find each other and elaborate mating projections that are many cell diameters long. The observation that a subset of the pheromone-induced genes overlap with the hyphae-induced genes (17, 98) is consistent with the possible involvement of extensively elongated structures in the mating process.

Initial studies on the cell biology of the mating process suggested that nuclear fusion was slow or nonfunctional, as the parental genomes could be recovered from the mated cells after karyogamy (82). However, more recent analysis of the process provides strong evidence for efficient nuclear fusion after zygote formation (16). This process occurs with different frequencies in different strains and is highly dependent on the Kar3 motor protein. Thus the typical outcome of the mating process in *C. albicans* is the formation of a stable tetraploid strain, which, because it has re-established the *MTL* heterozygous state, switches to the white form for subsequent proliferation.

The ability to form projections in response to mating pheromones is under genetic control. A variety of mutations influence this process; disruption of the heterotrimeric G-protein and MAP kinase-mediated signaling pathways (33, 86) prevents morphological changes by blocking the transmission of the signal (D. Dignard & M. Whiteway, unpublished data). Further, deletion of a *FAR1* homolog blocks morphological responses (P. Cote, T. Sulea & M. Whiteway, unpublished data); Far1p is not needed for hyphal growth in the white-form cell phase and thus is not required in general for directing polarized growth in response to external signals, but it is necessary for pheromone-mediated mating projections. Overproduction of Far1p also hypersensitizes cells to pheromone-mediated cell cycle arrest and to projection formation in the presence of pheromones. Pheromones also appear to coordinate with nutrient signals in the arrest of opaque-form cells during mating

(15); therefore developing the link between mating-pheromone-induced signaling, nutritional status, and cell cycle regulation is necessary to fully understand pheromone-mediated morphological response.

CHLAMYDOSPORE FORMATION

Chlamydospores of *C. albicans* superficially resemble a stable, resting cell and are distinct from the proliferative forms characterized by the yeast or hyphal states, but there is currently no strong evidence that long-term viability is exhibited by these cells. Initial studies on chlamydospores focused primarily on their structure and the conditions that generated their formation. These investigations established that environments low in oxygen, light, temperature, and nutrients were most conducive to the development of chlamydospores, and that the cells themselves were large, with thick walls (60) and a high lipid (93) and carbohydrate (44) content. Chlamydospores have been observed to germinate under certain conditions (60), and protocols have been developed to purify significant numbers of these spores (44). Media consisting of rice or cornmeal agar supplemented with the detergent Tween 80 are standard conditions for the induction of chlamydospores. These chlamydospores themselves form at the ends of branched filaments or suspensor cells that form under the inducing conditions (**Figure 1**). The formation of chlamydospores under specific conditions is one of the most effective ways to distinguish *C. albicans* from its close relative, *C. dubliniensis* (118).

Current studies are providing a more detailed picture of the regulatory circuits controlling the production of chlamydospores in *C. albicans*. The transcriptional regulator Efg1p (114) and the MAP kinase Hog1p (1) are required for chlamydospore formation. In both cases, filamentation occurred during the microaerophilic conditions, but production of the chlamydospores themselves was blocked. A more general search for functions required for chlamydospore formation was undertaken with a collection of insertion mutants. This approach did not demand the preselection of candidate genes but rather directly screened for defects in chlamydospore formation with a set of 217 genes inactivated through a transposon mutagenesis and mitotic recombination strategy (99). This study identified the *SUV3*, *SCH9*, and *ISW2* genes as essential contributors to the formation of chlamydospores, and *RIM11*, *RIM101*, and *MSD3* were needed for proper timing of their production. The *suv3* and *sch9* mutants were defective in the formation of hyphae-like filaments that serve as the source of the suspensor cells, and the *isw2* mutants formed the suspensor filaments but failed to elaborate the chlamydospores, similar to the situation observed for the *efg1* and *hog1* mutants. However, simple genetic investigations of possible common pathways involving Egf1p, Hog1p, and Isw2p did not provide evidence for coordinate functions, other than induction of chlamydospores, in other pathways (99).

The ability of *C. albicans* to form chlamydospores has been used as a clinical diagnostic tool for this fungal pathogen. The recent identification of the closely related species *C. dubliniensis* established that *C. albicans* and *C. dubliniensis* could be distinguished on the basis of chlamydospore formation in a medium, *Guizotia abyssinica* creatine agar or Staib agar, initially developed for the identification of *Cryptococcus neoformans*. This medium induces chlamydospores in *C. dubliniensis* but not in *C. albicans* (118). Because *C. albicans* and *C. dubliniensis* have similar genomes, efforts were made to identify the elements that generated this difference in chlamydospore generation (119). A library of *C. albicans* sequences was introduced into *C. dubliniensis* and the resulting transformants screened for recombinants that were blocked in spore formation when cultured on Staib agar. This approach identified the transcriptional regulator Nrg1p (**Table 1**) as the key element that differentiated the behavior of *C. albicans*

and *C. dubliniensis*. Increased expression of *NRG1* in *C. albicans* relative to expression in *C. dubliniensis* blocked the ability of Staib agar to induce chlamydospore formation (119), and when an *nrg1* mutant strain of *C. albicans* was tested for chlamydospore formation on Staib agar, the mutant strain was induced as efficiently as *C. dubliniensis*. This result establishes the level of expression of *NRG1* as a major determinant for the production of chlamydospores under the inducing conditions established by Staib agar. Defects in downregulation of *NRG1* expression are predicted to interfere with chlamydospore formation, and Rim101p, which is necessary for the proper timing of chlamydospore formation (99), is a repressor of *NRG1* expression (85). However, the *NRG1* expression level is not the only determinative factor, because the null mutant is not constitutive for chlamydospore formation, just permissive under the signaling regime of Staib agar.

A careful analysis of the cell cycle dynamics of chlamydospore formation has provided strong evidence for the uniqueness of the chlamydospore relative to yeast-form cells, pseudohyphal cells, and hyphal cells. The positioning of the dividing nucleus has been an important marker for the characterization of the cellular morphology pattern, with the nucleus migrating into the hyphal neck and then re-entering the mother cell during hyphal division, and dividing across the mother-bud junction in yeast and pseudohyphal cells (120). The chlamydospore shows yet another pattern of nuclear division. During the formation of the chlamydospore, the nucleus divides within the suspensor cell and then one daughter nucleus migrates into the chlamydospore (89).

This unique patterning of nuclear division is paralleled by the arrangement of septins during the formation of the chlamydospore. Initially the septins were found at the chlamydospore necks, similar to the situation for budding cells, but later in the development of the spores, the septins were localized throughout the plasma membrane. Extensive filamentous structures were formed by the later-stage septins; deletions of the *CDC10*- or *CDC11*-encoded septins perturbed the proper development of the chlamydospore and the suspensor cells (89). These distinctive behaviors of the chlamydospore confirm that they are a unique cellular form; however, their physiological role is still somewhat obscure. There is no evidence for meiosis occurring within the spore (89), although the septin filaments are somewhat suggestive of the modified septin structures observed in the developing spores in *S. cerevisiae* (45). Furthermore, the chlamydospores have not been shown to represent a stable resting state. The formation of these structures is characteristic of the pathogenic *C. albicans* and *C. dubliniensis*; therefore their function may be somehow related to the commensal/opportunistic pathogen lifestyle of these organisms.

OTHER GROWTH MODES

Pseudohyphal Growth

C. albicans cells exhibit several patterns of growth that involve extensive cell elongation but do not involve the formation of true hyphae. The pseudohyphal pattern is the best studied of these filamentous growth modes. Pseudohyphal growth involves elongated cells that remain in chains but are separated by true constrictions, and not by septa. It has been controversial whether pseudohyphal growth is an intermediate stage in the yeast-to-hypha transition, but current work suggests that pseudohyphal cells represent a discrete cell growth pattern (120). The absence of a Spitzenkorper, and the positioning of nuclear division across the mother-daughter junction, suggest that pseudohyphal growth is closer to yeast growth than to hyphal growth. Major distinctions from the yeast form are the extended period of polarized growth exhibited by the pseudohyphal cells and the tendency for the cells to remain attached even though proper septa have formed between them.

Some environmental conditions, such as medium rich in phosphate (54) or in alkanes (116), have been reported to stimulate the pseudohyphal pattern of division. In addition, a number of mutants lead to constitutive pseudohyphal growth. Several of these mutations are in genes involved in cell cycle regulation, implicating this process in control of this morphogenetic state. This involvement is logical because the switch from yeast growth to pseudohyphal growth arises from a subtle modulation in the length of time the cell spends in the polarized growth mode relative to the isotropic growth mode. Inactivation of the Fhk2p transcription factor (**Table1**) implicated in the regulation of mitotic cyclins, or of the Fkh2p target cyclin Clb4p, generates cells that remain pseudohyphal under both hyphae-inducing and yeast-growth conditions (18). Loss of Grr1p, an F box protein involved in the degradation of the G1 cyclins Ccn1p and Cln3p, leads to a pseudohyphal growth state (79). Perhaps extension of the G1 phase by stabilizing the G1 cyclins or reducing G2 cyclins can trigger the pseudohyphal state. Another F box protein, Cdc4p, is implicated in morphogenetic control. Loss of Cdc4p generates cells that are even more hyphal than those created by loss of Grr1p, suggesting that a critical target of the Cdc4p version of the SCF (Skp1-Cullin-1/Cdc53-F-box protein) complex is necessary to block hyphal growth (5). In *C. albicans* the Nim1p-related kinases Gin4p and Hsl1p function in the regulation of the pseudohyphal state; mutation of either kinase leads to constitutive pseudohyphae (131).

In addition to cell cycle perturbations that lead to constitutive pseudohyphae, defects in the Tup1p regulatory circuit lead to this phenotype (**Table 1**). Tup1p and the associated protein Ssn6p play important regulatory roles in *S. cerevisiae*; they interact with different DNA binding partners to provide a generic gene repression function (111). For specific transcription modules the functional consequences of loss of Tup1p or Ssn6p can be different; therefore Tup1p and Ssn6p do not provide completely overlapping functions. In *C. albicans* Tup1p inactivation causes cells to proliferate in a pseudohyphal growth mode (24). However, the *ssn6* deletion mutant does not trigger equivalent pseudohyphal development in *C. albicans* (48, 56), although normal morphology is perturbed. The global transcriptional consequences of the *tup1* and *ssn6* mutations are different (48), suggesting that the Ssn6p corepressor is not a critical component of the Tup1p regulatory circuit that represses the pseudohyphal growth pattern. An alternative tetratricopeptide repeat protein, Tccp1p, may play the corepressor role with Tup1p in the repression of pseudohyphal growth, as loss of Tccp1p creates similar phenotypes and affects patterns of gene expression similar to loss of Tup1p (67). It appears the Nrg1p DNA binding protein targets the Tup1p(Tcc1p) corepressor to a variety of promoters involved in regulation of morphogenesis, as loss of Nrg1p function leads to constitutive pseudohyphal growth (48), and microarray analysis suggests that Tup1p and Nrg1p influence distinct but overlapping gene sets. A second DNA binding protein, Rfg1p (66), appears to function as a targeting element for Tup1p(Tcc1p); loss of Rfg1p leads to a similar constitutive pseudohyphal phenotype, but the networks of gene expression modulated by Nrg1p and Rfg1p are different. *NRG1* is repressed by hyphal inducing conditions, and Rfg1p may be shut off post-translationally (65, 96), and thus inactivation of this negative regulatory circuit is a critical component of morphogenetic control in *C. albicans*.

Hyperpolarized Buds

A second occasion in which *C. albicans* cells exhibit a filamentous growth pattern distinct from hyphal growth occurs under certain cellular stresses or in the absence of certain gene products that influence the cell cycle. Treatment of yeast-form cells with the DNA synthesis inhibitor hydroxyurea (HU) resulted in a growth mode in which the bud continued

F box protein: component of the ubiquitination machinery that serves as part of the substrate recognition system

Microarray: collection of nucleotide probes attached to a solid support that allows simultaneous assessment of all the transcripts in the cell

to elongate in the absence of further DNA replication (7). This is different from the behavior of *S. cerevisiae* cells treated with synthesis inhibitors, in which the cells arrest with no further growth of the bud. Treatment of *C. albicans* cells with nocodazole, another chemical that blocks cell cycle progression, also causes elongation of the bud in the absence of further DNA replication (10). Several mutants generate similar bud elongation; inactivation of the *CDC5* gene encoding the *C. albicans* polo kinase (7) and shutoff of the repair gene *RAD52* (2) cause extensive bud growth. Blocking mitotic cyclin degradation by eliminating the destruction boxes of Clb2p or Clb4p traps cells in mitosis and also triggers elongated bud growth (18), as does deletion or overproduction of a stabilized form of Sol1p (5). Regulated repression of the cyclin-dependent kinase Cdc28p (121) and the essential B-type cyclin Clb2p (18) also generates extended bud elongation. In contrast to deletion of other cell cycle genes that cause typical pseudohyphal growth, the elongated bud phenotype seems to be specific for depletion of essential cell cycle genes.

The elongated buds resemble true hyphae in that they maintain polarized growth at the tip and do not show periodic constricted growth as do pseudohyphae. They also demonstrate nuclear movement out of the mother yeast cell and into the filament. Similar to pseudohyphae, a constriction is present at the junction between the yeast cell and elongated bud, which is consistent with the bud forming prior to the cell cycle arrest and the elongated growth mode. The elongated buds eventually die, which is not unexpected given that they cannot continue the cell cycle. However, the cells alter their transcription program during elongation. For example, time course microarray analysis of HU-treated and *CDC5*-depleted cells demonstrated that the elongated buds express many hyphae-specific genes, but at later stages of elongation. More hyphae-specific genes were turned on in the Cdc5p-depleted cells than in HU-treated cells. The elongated buds may therefore be capable of changing fate and becoming more like true hyphae with continued polarized growth, suggesting that a feedback mechanism may exist between the extent of polarized growth and activation of the hyphal signaling pathways (7).

Although phenotypically similar, the molecular mechanisms underlying elongated bud formation at different cell cycle stages appear to be different. Transcriptional profiling of HU-treated and *cdc5* mutant strains showed different expression patterns. The HU-arrested cells require Ras1p for the extended bud growth, and the *cdc5*-arrested cells require Bub2p function for the formation of the extended buds (6). The different triggers stop the cells at different points in the cell cycle, and the hyperpolarized bud does not arise because the cells are trapped at a point corresponding to normal polarized growth. Expression of the *C. albicans BUD4* (*INT1*) in *S. cerevisiae* triggers hyperpolarized bud growth (4); it will be interesting to establish if the process in *C. albicans* is Bud4p dependent.

The physiological relevance of the elongated bud morphology remains unclear because it has been reported only in response to creating cell cycle mutations or upon exposure to cell cycle-blocking drugs. However, the full suite of cell morphologies that *C. albicans* is capable of achieving in vivo is far from understood. Because the organism can grow in such a morphology that eventually expresses hyphal and virulence-specific factors, an act that requires a large amount of resources, this growth mode may be important for pathogenicity and/or survival in the host. Certain stressful environments in the host could feed into the cell cycle and interfere with DNA replication and/or chromosome segregation. If such interference were linked with triggering a highly polarized growth mode, the organism might escape the immediate stressful environment (7).

CONCLUSIONS

Morphological plasticity is a defining characteristic of the fungal pathogen *C. albicans*. A number of naturally occurring morphological states are adapted to specific physiological functions of this organism. The molecular circuits underlying some of these morphological states have been investigated extensively, whereas others have been studied at the molecular level only recently. Even the regulatory circuits controlling the yeast-to-hyphal transition can be reconsidered in respect to the more precise definitions of hyphal and pseudohyphal cells—-accurate interpretation of the networks will require a accurate determination of the cellular process being regulated. In addition, the roles of the morphological regulators in defining cell forms such as the opaque-form cell and the chlamydospore require more investigation. However, the rapidly developing tools for molecular and genomic studies in *C. albicans* are providing important insights into the specific controls underlying both the unique morphological states and the interrelationships between these states. Because the ability to regulate morphology is a critical component of the virulence of *C. albicans*, a detailed understanding of the roles of the morphological forms and how they are regulated will provide important insights into strategies for controlling this pathogen.

SUMMARY POINTS

1. *C. albicans* has several distinct morphological forms. Improving rules defining the different forms and improving analytical tools are permitting a more sophisticated analysis of the molecular bases for this morphological diversity.
2. When growing in the yeast form, *C. albicans* follows the pattern of growth of obligate yeasts such as *S. cerevisiae*. When growing in the hyphal form, it follows the pattern of obligate hyphal organisms such as *N. crassa*. Understanding the mechanisms through which environmental signals direct the choice of morphology is a central goal of studies on *C. albicans* morphogenesis.
3. The opaque state is controlled by the Wor1p transcription factor and is required for pheromone responsiveness and mating. Morphological changes directed by mating pheromones are a key aspect of the mating process.
4. The chlamydospore forms under adverse conditions through a process of cell division distinct from that of yeast, hyphal, or pseudohyphal growth. The function of the chlamydospore is currently enigmatic; it may represent a resting state but evidence for this is weak.
5. Pseudohyphae have a morphological appearance that falls between that of yeast-form and hyphae-form cells, but their cell division process resembles that of yeast, as the nucleus divides across the mother-daughter junction, and the cells lack a Spitzenkorper. Thus, it does not appear that pseudohyphae are a transitional state, but rather represent a modification of the yeast form of cell division.

DISCLOSURE STATEMENT

The authors are not aware of any biases that might be perceived as affecting the objectivity of this review.

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136. This paper and Reference 55 identify and characterize the Wor1p transcription factor that regulates the opaque state.

RELATED RESOURCES

Li L, Wright SJ, Krystofova S, Park G, Borkovich KA. 2007. Heterotrimeric G protein signaling in filamentous fungi. *Annu. Rev. Microbiol.* 61:423–52