The septin cortex at the yeast mother–bud neck

Amy S Gladfelter*, John R Pringle† and Daniel J Lew‡

A specialized cortical domain is organized by the septins at the necks of budding yeast cells. Recent findings suggest that this domain serves as a diffusion barrier and also as a local cell-shape sensor. We review these findings along with what is known about the organization of the septin cortex and its regulation during the cell cycle.

Addresses
*Department of Molecular Microbiology, Biozentrum, University of Basel, Klingelbergstrasse 70, CH-4056 Basel, Switzerland
†Department of Biology and Program in Molecular Biology and Biotechnology, University of North Carolina, Chapel Hill, North Carolina 27599-3280, USA
‡Department of Pharmacology and Cancer Biology, Duke University Medical Center, Durham, North Carolina 27710, USA;
e-mail: daniel.lew@duke.edu

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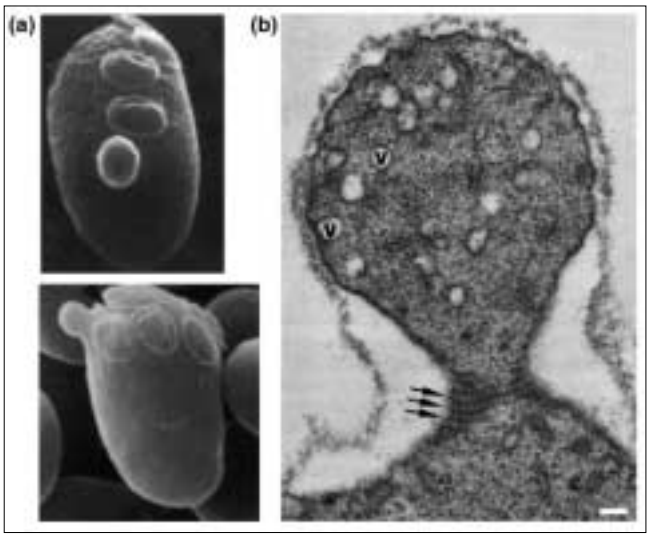
Abbreviation
GFP green fluorescent protein

Figure 1



The yeast mother–bud neck. (a) Scanning electon micrographs of haploid yeast cells. Note the bud scars from previous cell divisions. Adapted, with permission, from Chant and Pringle [59]. (b) Thin-section transmission electron micrograph of a small-budded cell. Note the alternating dark and light striations (arrowed) that underlie the plasma membrane. These striations disappear in septin-mutant strains at restrictive temperature. Adapted, with permission, from Byers and Goetsch [6]. The bar in (b) represents 0.1μm.

Introduction

The ability to specialize specific domains of the cell cortex is crucial to the function of almost all cell types. Notable examples include the distinct apical and basolateral membrane domains of epithelial cells, the pre- and post-synaptic membranes of neurons, the leading edges of migrating cells, and the cleavage furrows of dividing cells. In the budding yeast *Saccharomyces cerevisiae*, a specialized cortical domain is established that separates the bud, which will grow into a new daughter cell, from the pre-existing mother cell. The neck between mother and bud will become the site of cytokinesis and septum formation at the end of the cell cycle.

Throughout the process of bud formation, the cell cortex at the neck is clearly differentiated from the cortex at other parts of the cell, both inside and outside the plasma membrane (Figure 1). Outside, a chitin-rich ring (the eventual 'bud scar') forms in the cell wall on the mother side of the neck [1–4]. Inside, a family of evolutionarily conserved proteins called septins [5] form a highly ordered cortical domain that can be seen as a set of ~20 evenly-spaced striations spanning the neck by electron microscopy [6,7]. Although the neck is unique to budding yeasts, septins are present widely, if not ubiquitously, in animal cells, in which they may also serve to organize cortical domains, including regions concerned with cytokinesis [8–12] or with regulated secretion [13,14].

The septin-containing cortex in yeast is thought to act as a scaffold, recruiting many other proteins to the neck, where they can perform their varied functions (reviewed by Longtine *et al.* [5]). Well-established functions for the septin cortex include synthesis of the chitin ring and septum, bud-site selection and cytokinesis [15–17]. The ability of the septins to recruit chitin synthases, bud-site-selection landmarks, actomyosin ring components and other proteins to the neck is perhaps sufficient to account for their role in these processes. However, recent findings have documented novel roles for the septins that appear to entail more than simply recruiting other proteins to the neck. These studies highlight longstanding questions regarding how the septin cortex is organized and how it is regulated during the cell cycle. In this review, we discuss these recent findings with a focus on the regulation of septin organization and its implications for septin function.

The septin cortex as diffusion barrier

In a beautiful set of experiments, Takizawa and colleagues [18••] demonstrated that the septin cortex serves as a diffusion barrier for integral membrane proteins. They first identified an mRNA that was transported to the bud tip by the actin cytoskeleton and then found that the encoded transmembrane protein, Ist2p, was localized exclusively to the bud plasma membrane. They suggested that translation, translocation and transport of Ist2p to the plasma membrane all occurred within the bud and, furthermore, that once at the membrane, Ist2p could not diffuse into the mother

portion of the cell. Using GFP-tagged Ist2p, they monitored fluorescence recovery after photobleaching of a spot on the bud membrane and concluded that Ist2p was freely diffusible within the plane of the membrane, indicating that the restriction to the bud was due to a diffusion barrier at the neck. In septin-mutant cells, this barrier was eliminated and Ist2p spread throughout the mother, as well as the bud membrane. The septin cortex's role as a diffusion barrier may explain why it is so highly ordered — perhaps the striations observed by electron microscopy correspond to the physical barriers to diffusion through the neck.

This barrier role of the septins invites comparison to tight junctions in animal cells, which exhibit clear 'ridges' of transmembrane proteins when examined by freeze-fracture electron microscopy. Thus far, no analogous structures have been described at the yeast neck. Given that the septins themselves seem to be cytoplasmic proteins that are only peripherally associated with the plasma membrane, it seems possible that the barrier in yeast only blocks the migration of membrane-associated molecules with sizable cytoplasmic domains and not that of all membrane components, including lipids. In this regard, it is interesting that another recent study suggested that the neck serves as a diffusion barrier for proteins that are peripherally associated with the plasma membrane [19•]. However, this same study found that cortical actin patches (large mobile disc-shaped structures containing F-actin that are associated with the cytoplasmic face of the plasma membrane [20,21]) were able to migrate from the bud side of the neck to the mother. This astonishing feat would seem to imply that the barrier is highly selective and perhaps able to open to let the patches through. Alternatively, the patches may transiently abandon the plasma membrane to travel through the neck interior. Support for the latter possibility is provided by recent findings on actin patches in fission yeast, which do appear to make excursions away from the membrane into the cell interior [22]. The degree to which the septin cortex forms an effective barrier to the diffusion of various cellular constituents is clearly an issue worthy of further investigation. It seems likely that an understanding of this issue will require elucidation of the higher-order organization of the septins and associated proteins within this domain.

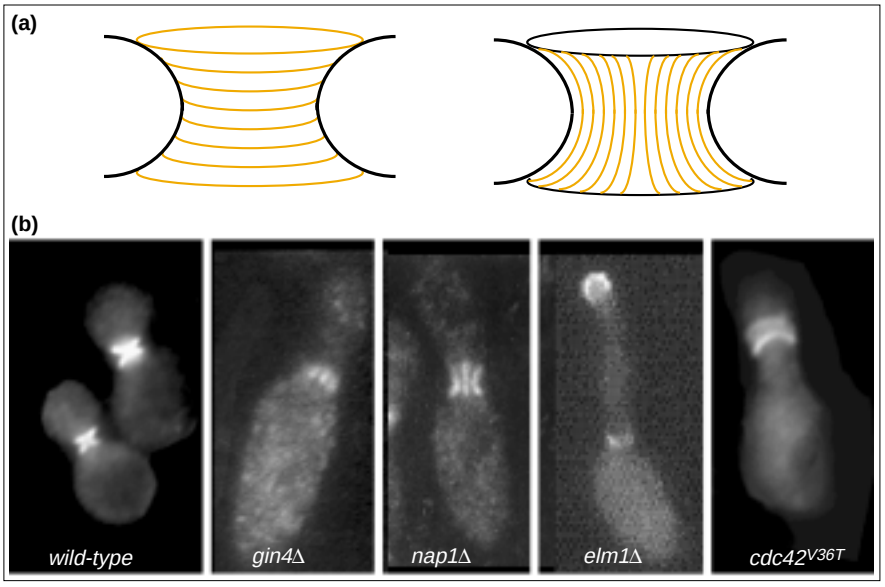
Regulators of septin organization

How are the septins organized within the neck cortex? This fundamental question remains largely unanswered. When Byers and Goetsch first examined the neck cortex by electron microscopy [6,7], they hypothesized that the striations represented helical filaments that wrapped around the neck (Figure 2a). Consistent with this proposal, overproduction of certain proteins can induce cells to elaborate extensive rings and spirals of septins into the bud [23•,24]. Subsequent studies demonstrated that purified septin complexes could polymerize to form filaments *in vitro* [25,26], and this capability seems likely to be important for the higher-order organization of the septin cortex *in vivo* (but see Field and Kellogg [27] for a stimulating discussion on this point). These studies also led Field *et al.* [25] to propose an alternative model for septin organization in which polymerized septin filaments were oriented along the mother–bud axis and associated laterally to form a collar around the neck (Figure 2a). Support for this model has come from observations on mutant strains lacking factors important for septin organization. The first such factor to be identified was the protein kinase Gin4p [28], which is co-localized with septins at the neck (Figure 4). Mutant cells lacking Gin4p frequently display aberrant septin staining, in which several thick septin-containing bars are oriented along the mother–bud axis (Figure 2b). Similar bars have also been observed more recently in cells lacking the Gin4p-binding protein Nap1p or the Cdc42p-activated kinase Cla4p [29••]. An attractive interpretation of the mutant phenotype is that Gin4p, Nap1p and Cla4p are required for regular lateral interactions between septin filaments that bring them into register to form the proposed collar. Without these factors, improper lateral interactions could cause the collapse of the collar into thick bundles of filaments, seen as bars.

Gin4p, Nap1, and Cla4p do not constitute a simple linear pathway for septin organization, because the septin defects in deletion mutants lacking any two of these proteins are significantly more severe than those observed in the single mutants [29••] (Figure 2b). However, even cells lacking all three proteins do generate patches of septin-containing cortex at the neck, indicating that additional pathways exist to recruit and/or assemble septins. Recently, Bouquin *et al.* [30•] showed that the protein kinase Elm1p, like Gin4p, is localized to the septin cortex and plays a role in septin organization. However, rather than displaying septin bars at the neck, cells lacking Elm1p frequently displayed ectopic septin localization near the bud tip, in addition to an irregular septin-containing zone at the neck (Figure 2b). A similar septin localization has been observed in some strains deprived of both Cla4p and the related kinase Ste20p [31,32•]. Yet another pattern of septin mislocalization was seen in cells containing a particular mutant allele of *CDC42*, which frequently displayed ectopic rings of septin staining partway into the bud [33•] (Figure 2b). Together, these studies indicate that many genes contribute to the normal organization of septins within the septin cortex and that a variety of curious aberrations can be produced in mutant strains. However, the basis for these aberrations and the exact role played by each protein in septin organization remain mysterious.

Interestingly, Gin4p kinase activation appears to require the septins, and it may involve recruitment of Gin4p to the neck [34]. In addition, Gin4p kinase activity is cell-cycle-regulated, with a peak during mitosis [35]. Full activation of Gin4p requires Nap1p, Cla4p and Elm1p, and these findings have been interpreted as evidence for the existence of a signaling pathway initiated by the mitotic form of the cyclin-dependent kinase Cdc28p and culminating in Gin4p activation [34–37]. However, an alternative interpretation

Figure 2



Septin organization at the neck. (a) Models of septin filament organization. Filaments have been suggested to wrap around the neck (left, [6]) or to align along the mother–bud axis (right, [25]). In the model on the left, the dark striations in Figure 1b are thought to represent individual 10 nm diameter filaments, whereas, in the model on the right, the light and dark stripes represent inherent repeat units within the filaments that are laterally associated in register with each other. (b) Aberrant septin organization in mutant strains. Note the septin bars in the *gin4Δ* and *nap1Δ* mutants, bud-tip-localized septins in the *elm1*Δ mutants, and misplaced septin rings within the buds of the *cdc42*^{V36T} mutants. Adapted, with permission, from Longtine *et al.* [29••], Bouquin *et al.* [30•] and Gladfelter *et al.* [33•].

of these results could be that the defective septins in *nap1*, *gin4* or *elm1* mutants fail to stimulate full Gin4p activation. As Gin4p kinase activity, in turn, contributes to septin organization [28], these findings suggest a bidirectional communication between septins and Gin4p that could contribute to the higher-order structure of the septin cortex. It will be very interesting to determine how septins affect Gin4p activation and whether septin organization, like Gin4p activity, is regulated during mitosis.

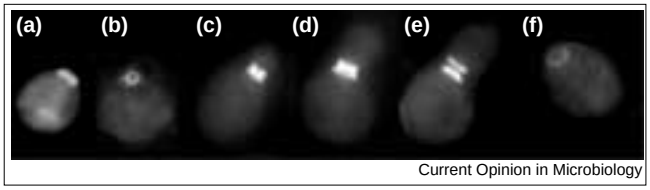
The septin cortex through the cell cycle

As judged either by immunofluorescence or by the use of GFP-tagged septins, a discrete ring containing five septins forms in late G1 phase, 15 minutes or so before the emergence of a visible bud (Figure 3) [34,38–42]. (Two other *S. cerevisiae* septins are expressed only during sporulation [43,44].) Following bud emergence, the ring broadens into an hourglass-shaped cortical zone at the neck, and the septins remain at the neck throughout the cell cycle. During cytokinesis, the hourglass splits into two rings that remain on both mother and daughter cells and can persist until a new ring forms in the next cell cycle. These observations raise a host of interesting questions. What causes the septin ring to form? How does the ring change into an hourglass during bud emergence? How does the septin cortex remain at the neck while growth is directed into the bud? How does the hourglass split into two rings during cytokinesis? What causes the rings that remain after cytokinesis to eventually disassemble?

Assembly of the septin ring requires the GTPase Cdc42p [42,45], which concentrates in a cortical patch at the presumptive bud site in response to cell cycle cues during late G1 [45–47]. Cdc42p is required for polarization of the actin cytoskeleton as well as formation of the septin ring, but actin polarization and septin assembly are independent events, in that each can occur in the absence of the other [45,48]. It is not clear whether Cdc42p employs separate effector pathways to trigger these events, and the effectors linking Cdc42p to septin-ring assembly remain mysterious (although Cla4p is likely to be one of these, see above). The recent identification of *cdc42* alleles with septin-specific defects promises to accelerate progress in this area [33•].

The spreading of the septins from the ring in unbudded cells to the hourglass in budded cells may occur in response to the local cell-shape change that accompanies bud formation. Consistent with this view, cells that are prevented from forming buds by disruption of the actin cytoskeleton appear to retain septin rings for prolonged periods without spreading or other obvious changes [48]. Interestingly, the striations observed at the neck by electron microscopy were not detected before bud emergence, and did not appear fully developed until significantly after bud emergence [6,7]. Although the difficulty of visualizing these structures makes it dangerous to draw firm conclusions,

Figure 3



Septin localization through the cell cycle. (a,b) Septins initially assemble into a planar ring at the incipient bud site. (c,d) Upon bud emergence, septin staining extends into an hourglass shape at the neck. (e) At about the time of cytokinesis, the septin hourglass splits into two rings, (f) which remain, following cell separation, as an 'old' ring that is fainter and broader than the initial ring.

Figure 4

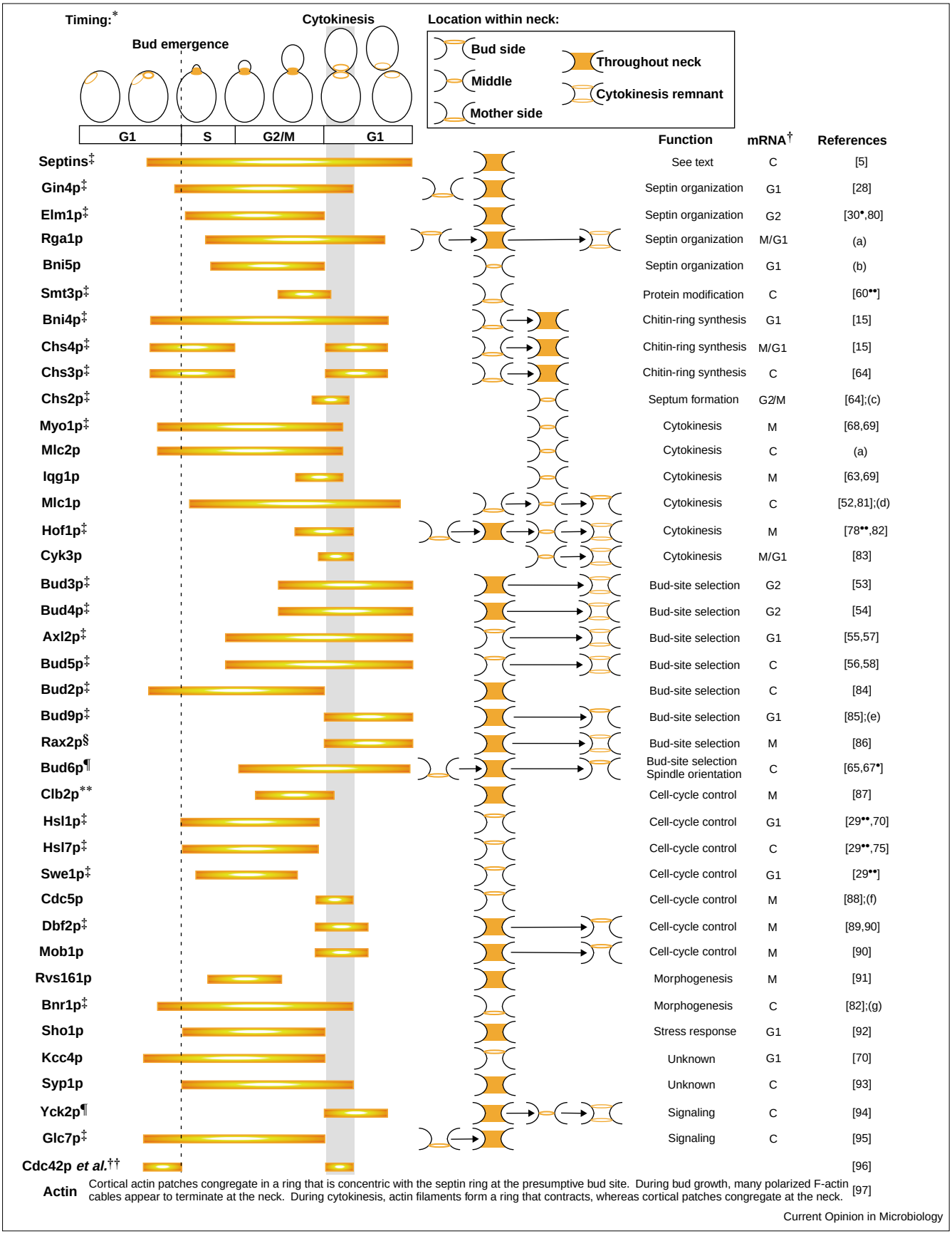

Timing:*
Bud emergence
Cytokinesis
G1
S
G2/M
G1
Location within neck:
Bud side
Middle
Mother side
Throughout neck
Cytokinesis remnant
Function
mRNA†
References
Septins‡ | See text | C | [5]
Gin4p‡ | Septin organization | G1 | [28]
Elm1p‡ | Septin organization | G2 | [30•,80]
Rga1p | Septin organization | M/G1 | (a)
Bni5p | Septin organization | G1 | (b)
Smt3p‡ | Protein modification | C | [60••]
Bni4p‡ | Chitin-ring synthesis | G1 | [15]
Chs4p‡ | Chitin-ring synthesis | M/G1 | [15]
Chs3p‡ | Chitin-ring synthesis | C | [64]
Chs2p‡ | Septum formation | G2/M | [64];(c)
Myo1p‡ | Cytokinesis | M | [68,69]
Mlc2p | Cytokinesis | C | (a)
Iqg1p | Cytokinesis | M | [63,69]
Mlc1p | Cytokinesis | C | [52,81];(d)
Hof1p‡ | Cytokinesis | M | [78••,82]
Cyk3p | Cytokinesis | M/G1 | [83]
Bud3p‡ | Bud-site selection | G2 | [53]
Bud4p‡ | Bud-site selection | G2 | [54]
Axl2p‡ | Bud-site selection | G1 | [55,57]
Bud5p‡ | Bud-site selection | C | [56,58]
Bud2p‡ | Bud-site selection | C | [84]
Bud9p‡ | Bud-site selection | G1 | [85];(e)
Rax2p§ | Bud-site selection | M | [86]
Bud6p¶ | Bud-site selection
Spindle orientation | C | [65,67•]
Clb2p** | Cell-cycle control | M | [87]
Hsl1p‡ | Cell-cycle control | G1 | [29••,70]
Hsl7p‡ | Cell-cycle control | C | [29••,75]
Swe1p‡ | Cell-cycle control | G1 | [29••]
Cdc5p | Cell-cycle control | M | [88];(f)
Dbf2p‡ | Cell-cycle control | M | [89,90]
Mob1p | Cell-cycle control | M | [90]
Rvs161p | Morphogenesis | M | [91]
Bnr1p‡ | Morphogenesis | C | [82];(g)
Sho1p | Stress response | G1 | [92]
Kcc4p | Unknown | G1 | [70]
Syp1p | Unknown | C | [93]
Yck2p¶ | Signaling | C | [94]
Glc7p‡ | Signaling | C | [95]
Cdc42p *et al.*†† | | | [96]
Actin | | | [97]
Cortical actin patches congregate in a ring that is concentric with the septin ring at the presumptive bud site. During bud growth, many polarized F-actin cables appear to terminate at the neck. During cytokinesis, actin filaments form a ring that contracts, whereas cortical patches congregate at the neck.
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Figure 4 legend

Proteins that localize to the neck cortex. Synthesizing the available data on protein localization to the neck is complicated by the facts that the reagents and protocols employed to visualize the proteins have not been uniform and that detailed characterizations of the timing or asymmetry of localization were frequently not a primary goal of the investigators. (This figure will therefore undoubtedly contain errors, and the authors strongly encourage investigators to contact them with corrections or new information.) *To enhance the clarity of the figure, the cytokinesis remnant is indicated only in the second G1 interval, following cytokinesis. The grey bar indicates the time between disassembly of the mitotic spindle and completion of the septum. †Timing of mRNA accumulation is drawn from the genomic study of Spellman *et al.* [62]. C, constitutively expressed. ‡Localization of these proteins to the neck has been shown to be eliminated in septin-mutant cells shifted to restrictive temperature. §Rax2p persists at old division sites even after septins and other proteins have disassembled. ¶Localization of these proteins to the neck is not eliminated in septin-mutant cells shifted to restrictive temperature. **Localization of Clb2p to the neck is only detected when its ability to concentrate in the nucleus is impaired. ††A large number of proteins localize to sites of cell growth together with Cdc42p. These include polarity establishment proteins, exocyst components and cell-wall biosynthetic enzymes. These proteins are found in a patch at the presumptive bud site in late G1, around which the septins form a ring. They then part company temporarily with the septins, localizing to the bud tip in small buds and more diffusely within larger buds, before returning to the neck at the time of cytokinesis. Letters in parentheses in the References column of the figure correspond to the following: (a) E Bi, personal communication; (b) M Longtine, K Lee, personal communication; (c) I-C Yu, JR Pringle, unpublished data; (d) A Ragnini-Wilson, personal communication; (e) L Schenkman, JR Pringle, unpublished data; (f) C Hardy, personal communication; (g) M Longtine, personal communication.

it seems likely that the transition of septins from a ring to an hourglass entails a major change in the organization of this cortical domain.

The splitting of the hourglass into two rings at the end of the cell cycle also coincides with a dramatic change in local cell shape as cells undergo cytokinesis. Interestingly, the neck striations seem to disappear before cytokinesis [6,7], and a recent study [49••] provided compelling evidence that the splitting of the septin hourglass is not simply a consequence of cell division. Mutants lacking the acto-myosin ring component Iqg1p/Cyk1p are unable to undergo cytokinesis, but septin splitting occurs nevertheless. The controlling trigger for mitotic exit and cytokinesis is thought to be the GTPase Tem1p, which stimulates a signaling pathway called the 'mitotic exit network' to inactivate the mitotic form of Cdc28p [50,51]. Tem1p also appears to play a more direct role in cytokinesis by promoting actomyosin-ring contraction, probably through binding to Iqg1p/Cyk1p [52]. Intriguingly, it appears that Tem1p is also required for splitting of the septin hourglass, even when its role in triggering Cdc28p inactivation is bypassed [49••]. This suggests that Tem1p promotes septin splitting through a novel pathway, and it will be fascinating to learn how this works.

After cytokinesis, the septin ring remains for a time at the site of cell division. A haploid yeast cell normally forms its new bud adjacent to the site of the preceding cell division, and there is ample evidence that septins play a key role in marking the division site by recruiting several bud-site-selection proteins, such as Bud3p, Bud4p, Axl2p/Bud10p and Bud5p [53–58]. However, the old septin ring (and associated proteins) eventually disassembles, and if the cells experience a delay in entering the next cell cycle, they 'forget' this positional landmark [59]. It is unclear whether some specific cell-cycle signal triggers disassembly of the septin structure or if it undergoes a spontaneous decay. Some very interesting observations in this regard were made by Johnson and Blobel [60••], who demonstrated that several of the septins are covalently modified by attachment of the ubiquitin-related protein Smt3p (or small ubiquitin-related modifier [SUMO]) (see also [61]). Smt3p was conjugated to septins during mitosis and was removed at around the time of cytokinesis (Figure 4). Remarkably, cells that were rendered largely unable to conjugate Smt3p to septins (by the heroic expedient of mapping and mutating the conjugation-site lysines in three septin genes) retained old septin rings for prolonged periods, often accumulating multiple such structures over time. This finding suggested that Smt3p influenced the eventual disassembly of the old septin structure (but see Update).

Dynamic and asymmetric character of the septin cortex

The septin cortex plays host to a multitude of arriving and departing proteins during the cell cycle (Figure 4). How is the timing of localization of individual proteins to the neck regulated? Based on genomic analyses of gene expression, 25 of the 37 non-septin proteins known to associate with the septin cortex are the products of genes that are transcribed periodically during the cell cycle [62], so periodic protein accumulation may underlie the timing of neck association. This appears to be true at least for Bud3p [23•] and Iqg1p [63], for which it has been shown that earlier synthesis of the protein during the cell cycle causes premature association with the septin cortex. However, many proteins that are present throughout the cell cycle show restricted and, in some cases, complex, timing of localization to the neck. For instance, the chitin synthase III complex (Chs3p/Chs4p), which is responsible for making the chitin ring, localizes to the septin cortex before bud emergence but departs shortly thereafter and resides in an endosomal compartment until the time of cytokinesis, when it returns to the septin cortex [15,64]. Another example is provided by Bud6p, which functions in both bud-site selection and spindle orientation [65,66]. Bud6p is initially concentrated at the presumptive bud site and the bud tip, and it subsequently begins to accumulate at the neck [65,67•]. The timing of Bud6p neck localization appears to depend on a competition between factors (including Bni1p) that attract Bud6p to the bud tip and other factors (including Bud3p) that attract

Bud6p to the neck [67•]. Surprisingly, the outcome of this competition is different in haploid and diploid cells, with Bud6p accumulating at the neck much earlier in haploids. These examples illustrate the potential for cell-cycle and mating-type cues that affect membrane trafficking, polarized growth, or other processes to determine when particular proteins visit the neck.

In addition to the temporal differences in the composition of the septin cortex through the cell cycle, this membrane domain is spatially differentiated along the mother-bud axis. Although many proteins are present throughout the septin cortex, several are concentrated primarily on the mother side of the neck, some localize to a ring in the center of the neck, and others are located predominantly on the bud side of the neck (see Figure 4). Thus, the composition of the septin cortex varies along the mother–bud axis. How might this asymmetry be achieved? One appealing hypothesis was suggested on the basis of the behavior of Bni4p, which links the chitin synthase III complex to the septins [15]. Bni4p is located on the mother side of the neck during early stages of bud formation, but gradually becomes more symmetrically distributed across the neck during subsequent bud growth (Figure 4). Because of cell-cycle-regulated mRNA accumulation, the bulk of Bni4p appears to be synthesized before bud emergence, and it associates with the septin ring in unbudded cells. DeMarini and colleagues speculated that the septin cortex forming the ring before bud emergence may remain intact at the base (mother side) of the expanded hourglass cortex seen in budded cells. Thus, the pool of Bni4p made during G1 and associated with the ring at that time would remain concentrated on the mother side of the neck, and the lower levels of Bni4p synthesized later in the cell cycle may gradually populate the bud side of the septin cortex. However, more recent studies revealed that other proteins that are initially associated with the septin ring in unbudded cells do not remain on the mother side of the neck as the bud emerges. These proteins include Gin4p, which distributes throughout the neck [28], Myo1p, which localizes to the central ring [68,69], and Kcc4p, which localizes to the bud side of the neck [70]. Thus, there is no systematic correlation between the timing of recruitment to the septin cortex and localization along the mother–bud axis, suggesting that time of assembly cannot, in general, account for the asymmetry of the septin cortex.

Another hypothesis to explain the asymmetry is based on the model that septin filaments are oriented along the mother–bud axis (Figure 2a). If the filaments are intrinsically polar (like actin or tubulin polymers), and if they are all aligned in the same direction, then the septins themselves may provide an intrinsic asymmetry that could be exploited by septin-binding proteins. Thus, asymmetric localization of a septin-associated protein would simply reflect molecular recognition of the underlying septin polarity. However, this hypothesis does not easily account for proteins that localize to a ring in the middle of the neck, and it requires additional assumptions to explain the behavior of proteins, such as Bni4p, whose distribution within the neck changes during the cell cycle.

The septin cortex as cell shape sensor

In the past few years, it has become clear that cell-cycle-checkpoint controls monitor cytoplasmic as well as nuclear events [71]. In *S. cerevisiae*, a morphogenesis checkpoint delays mitosis in response to insults that perturb bud formation [72]. This mitotic delay involves stabilization of the cell-cycle-inhibitory kinase Swe1p [73]. Targeting of Swe1p for degradation involves its interaction with two regulators, Hsl1p and Hsl7p [74]. Intriguingly, these proteins are recruited to the bud side of the septin cortex at the time of bud emergence, and their function in downregulating Swe1p appears to require this localization [29••,70,75]. As discussed above, the septin cortex expands from a ring to an hourglass during bud emergence, and it has been argued that Hsl1p and Hsl7p may monitor this septin reorganization and trigger Swe1p degradation only after a bud has formed [29••]. In this way, the reorganization of the septin cortex in response to bud formation may allow the septins to act as indicators of local cell shape.

In fission yeast, a cytokinesis checkpoint arrests the cell cycle if cytokinesis was not completed successfully in the preceding cell cycle [76,77]. There are hints that cytokinesis may also be monitored in budding yeast. In particular, examination of mutants in which cytokinesis is slowed or prevented indicates that both the septins and cortical actin patches can persist at the neck for longer than usual under these circumstances [68,69,78••,79]. Does this mean that cells 'know' if cytokinesis has been completed, and delay the release of cytoskeletal elements from the neck when more time is required for cell division? In contrast to the mutants referred to above, septin-mutant cells proceed rapidly with the subsequent round of bud formation despite a complete failure of cytokinesis, suggesting that septins must be assembled for any 'cytokinesis sensor' to work. The change in local cell shape that accompanies cytokinesis may well cause alterations of septin organization analogous to those thought to accompany bud emergence. Conceivably, therefore, the septin cortex may allow cells to monitor local shape changes at both of these stages.

Conclusions

The mother–bud neck is a specialized subdomain of the cell cortex organized by the septins. Recent studies indicate that the septin cortex can function as a diffusion barrier and perhaps also as a local cell-shape sensor. To understand these functions, we must learn much more about the detailed architecture of the septin cortex and about its regulation by the cell cycle and by local geometry. The number of proteins known to visit the neck in a septin-dependent manner is large and growing, but our models of septin organization at the neck are still purely speculative. The general problem of how a cortical domain is specified and organized is of great biological importance, and we believe that investigation of septin-dependent

cortical organization will yield many insights in the coming years. Such advances will require integration of the information from molecular genetic approaches with detailed structural information. We hope that the fascinating inroads being made into septin regulation and function will attract more investigators with structural expertise into this area. The rewards should be magnificent.

Update

Recently, Johnson and Gupta [98] identified Siz1p as an E3-type enzyme responsible for conjugation of Smt3p to the septins. Like Smt3p, Siz1p was concentrated on the mother side of the neck during mitosis. A previous study [60••] found that cells in which septins had been mutated at the Smt3p-conjugation-site lysines had a defect in disassembling the old septin rings. However, cells lacking Siz1p showed no discernible defect in septin localization, suggesting that the previous result was due to an alteration of septin structure rather than to the lack of Smt3p conjugation, and the role of Smt3p at the septin cortex remains mysterious.

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Papers of particular interest, published within the annual period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

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