system function is striking. Specifically, failures in mirror neuron system development should result in action understanding and imitation deficits which, in turn, would lead to impaired self-other representation, social and communicative deficits, and ultimately empathic, language, and emotional failures.

Despite this promising take on ASD etiology, the abnormal direct-matching mechanism described here is obviously one among many presumably abnormal processes in ASD. Indeed, the mirror neuron system hypothesis of ASD does not exclude the possibility that other cognitive processes also participate in the complex pathophysiology of ASD. Although the integrity of the mirror neuron system seems to be critical for action understanding, mirror neuron system failures probably do not account for all of the reported social impairments of ASD. Moreover, mirror neuron system function and its neural network are not entirely abnormal in ASD. As shown by Dapretto *et al*. [16], children with ASD are able to imitate facial expressions and display patterns of activity in the amygdala similar to those of healthy participants.

It is important to note that despite enormous efforts in the last decade to pinpoint the specific causes of ASD, the gold standard in diagnosing ASD still rests on behavioral observation; no biological or genetic marker exists as of yet. As such, abnormalities in mirror neuron system neural substrates — the inferior frontal and parietal areas — may be important cues in the diagnosis of ASD. From this knowledge, diagnostic markers, and ultimately therapeutic targets for treatment that would allow for early intervention, could be developed. The critical step that needs to follow these exciting results is to establish whether the reported abnormalities in mirror neuron system function have any clinical value.

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Plasmid Segregation: A New Class of Cytoskeletal Proteins Emerges

The discovery that a plasmid-partitioning ATPase forms astral cytoskeletal structures both unveils a new family of cytoskeletal proteins and suggests that cytoskeletal involvement is a universal feature of DNA segregation.

Zemer Gitai

The ability to propagate genetic information faithfully is a prerequisite for evolutionary success. To this end, many bacterial plasmids encode their own machinery to ensure their proper segregation and subcellular positioning. A recent study [1] finds that a member of the most common class of these plasmid-partitioning proteins assembles into cytoskeleton-like filaments, and that these filaments can focus into asters that strikingly resemble those found in eukaryotic mitotic spindles. These insights further our understanding of both the specifics of plasmid partitioning,

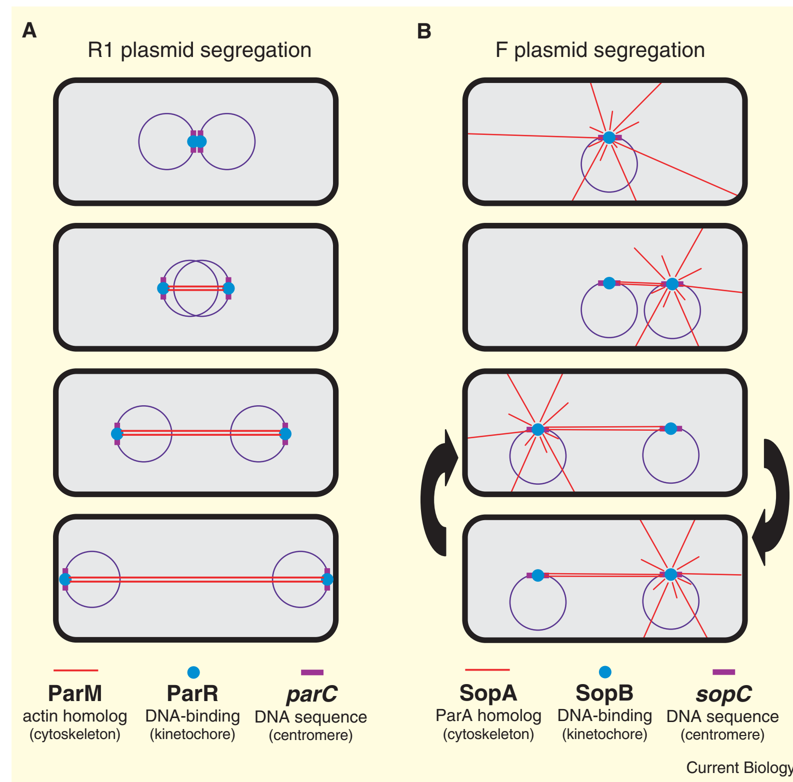

Figure 1. Both classes of plasmid segregation mechanisms involve cytoskeletal proteins.

(A) The R1 plasmid partitioning (*par*) locus encodes the ParM actin-like homolog (red), the ParR DNA-binding protein (blue), and the *parC* DNA sequence (purple). The ParR–*parC* complex promotes ParM polymerization in between the two plasmids, generating a tension rod that both separates the plasmids and localizes them to the cell poles [6]. (B) The F plasmid *par* locus encodes the SopA deviant Walker-type ATPase (red), the SopB DNA-binding protein (blue), and the *sopC* DNA sequence (purple). SopA forms cytoskeletal filaments that focus into asters in the presence of SopB and *sopC*, and SopA oscillates from plasmid to plasmid [1]. Though the precise mechanism of F plasmid segregation remains unclear, it is possible that each aster pushes off the other plasmid as well as the cell membrane, generating forces that both separate the plasmids and localize them to the quarter-cell positions.

and the general properties of DNA segregation machineries.

Bacterial plasmids are non-essential molecular symbionts. These extrachromosomal circular DNA molecules hijack their bacterial hosts' machineries for functions such as replication, and in return often provide bacteria with benefits such as sexual conjugation, as in the case of F plasmids, or antibiotic resistance, as in the case of R plasmids. Some plasmids, like the high-copy vectors commonly used in molecular cloning, are randomly distributed to the two daughter cells upon cell division. Many naturally occurring low-copy plasmids, however, have active mechanisms that ensure their precise segregation such that each daughter cell inherits an equal number of plasmids [2]. Recent cell biological studies have demonstrated that, despite their small size and lack of intracellular membrane compartments, the insides of bacterial cells show exquisite spatial organization [3]. Plasmids are no exception to this organization, as their segregation machineries also ensure their reproducible spatial and temporal localizations [4,5].

All known low-copy plasmids encode their own segregation and positioning machinery in their *par* cassette, which invariably consists of two protein-coding genes and a centromere-like *cis*-acting DNA sequence. The two *par*-encoded proteins are an ATPase of one of two different ATPase families, and a DNA-binding protein that interacts with the *par* cassette's *cis*-acting DNA sequence [2]. Thus, *par* cassettes can be subdivided into two classes on the basis of their encoded ATPase. One class of *par* ATPase, typified by the F plasmid's SopA, contains a ParA-like deviant Walker-type ATPase motif, while the other, typified by the R1 plasmid's ParM, exhibits sequence similarity to the actin superfamily. The ParM ATPase structurally resembles actin and polymerizes into actin-like filaments that separate R1 plasmids by pushing them apart [6].

The new study by Lim *et al*. [1] suggests that the ParA-like ATPases are also cytoskeletal proteins [1]. They found that purified SopA protein forms ATP-dependent cytoskeletal filaments similar those of actin or the ParM-like class of plasmid partitioning ATPases. SopA filaments could be visualized by the non-specific stain, Nile red, enabling their direct visualization. By performing time-lapse fluorescence microscopy on the assembly of Nile red-stained SopA filaments in the presence of ATP, the authors determined the average rate of filament elongation. This rate of ~0.18 ± 0.05 µm per minute is comparable to that of F plasmid separation, 0.43 µm per minute [7], which is at least consistent with a direct role for SopA elongation in segregating F plasmids.

Armed with the knowledge that SopA forms filaments *in vitro*, Lim *et al*. [1] next sought to determine the subcellular distribution of SopA protein. SopA had previously been shown, by antibody staining of fixed cells, to localize to discrete foci whose formation required the presence of both the F plasmid *par* cassette's DNA-binding protein SopB, and the *cis*-acting DNA sequence *sopC* [8]. Live-cell imaging of SopA-GFP in the presence of SopB and *sopC* confirmed the formation of foci, but revealed a far more complex pattern of SopA dynamics. In addition to cells with tight SopA foci, some cells exhibited other distributions, including a diffuse haze, a directional comet, or a thin tether connecting two foci.

Time-lapse microscopy clarified this confusing picture: nearly all cells were seen to exhibit a similar pattern of SopA oscillation. With a period of roughly 20 min, SopA–GFP appeared as a tight focus at the quarter-cell position, redistributed as a diffuse haze around the three-quarter-cell position, condensed into a tight focus at the three-quarter-cell-position, redistributed as a haze around the quarter-cell position, and then completed the cycle by condensing into a focus at the quarter-cell position. These SopA dynamics were shown to depend on both SopB and *sopC*. The SopA–GFP foci also colocalized with SopB–CFP, suggesting that the SopB–*sopC* complex is involved in forming the SopA structures. These SopA dynamics may be essential for F plasmid segregation, because a mutant that hyperstabilized SopA filaments — as measured by fluorescence recovery after photobleaching (FRAP) — dominantly perturbed plasmid maintenance.

SopA structures form around the SopB–*sopC* complex *in vivo*. Lim *et al*. [1] investigated the effect of SopB protein and *sopC* DNA on SopA filaments *in vitro*. Mixed together, SopA, SopB, *sopC* and ATP produced the first observed example of bacterial radial asters. Rather than forming individual SopA filaments, this complex generated multiple long, thin filaments, the ends of which originated from a single point. Asters failed to form in the absence of any one of SopB, *sopC* or ATP, and a fluorescent labeling experiment verified that the foci of these asters contained SopB and DNA, demonstrating that the SopB–*sopC* complex functions to nucleate or focus the ends of SopA filaments.

The presence of SopA asters *in vivo* still needs to be verified, as the diffuse SopA haze is consistent with, but does not prove, the presence of asters. The main question that now emerges is how does SopA physically move plasmids apart? One could imagine that, during segregation, an aster forms around each of the two plasmids' *sopC* sites and that the two asters push off one another to separate the plasmids. R1 plasmid segregation follows this model, as a ParM filament is nucleated in between the two plasmids, generating a tension rod that pushes the plasmids apart [6] (Figure 1A). This straightforward model does not, however, quite fit the SopA data; as most cells are observed to have only one SopA structure at a time, not two opposing asters. It is possible that SopA does not oscillate during the brief period of actual plasmid separation, accounting for the minority of cells with two SopA structures.

Alternatively, each plasmid might push off the other with unidirectional tension rods that are too small or transient to be detected (Figure 1B). Yet another possibility is that SopA moves F plasmids by an entirely different mechanism, such as by polarized, propulsive polymerization. Regardless of how SopA separates plasmids, it must also localize them, and a radial aster of filaments would be the perfect structure to push against the membrane in equal proportions, driving itself to the quarter cell position (like a focus of an ellipse). That would explain why an aster-like SopA spindle drives F plasmids to the quarter cell, while a rod-like ParM spindle drives R plasmids all the way to the pole (Figure 1).

The nature of the SopA plasmid-to-plasmid localization oscillations must be further explored as well. It is curious that other SopA homologs, such as ParA of plasmid pB171, Soj of *Bacillus subtilis* and MinD of *Escherichia coli*, also have oscillating localizations [4,9–11]. It has been proposed that such oscillations represent a self-organizing mechanism for finding the long axis of the cell [12]. A similar axis-finding function for SopA oscillations might properly align the axis of plasmid segregation with the axis of cell division, thereby ensuring that each daughter cell inherits one plasmid. Alternatively, oscillations could just be a by-product of the assembly kinetics of SopA homologs.

SopA is not the only deviant Walker-type ATPase that can assemble into a cytoskeletal structure *in vitro*: ParF, a partitioning ATPase encoded by the TP228 plasmid, and MinD, a chromosomally encoded regulator of bacterial cytokinesis, both form ATP-dependent filamentous polymers [13,14]. Together, these findings suggest that SopA homologs represent a new family of cytoskeletal elements. If so, it would appear that every plasmid segregation mechanism has a cytoskeletal basis. Use of a cytoskeleton may in fact prove to be a universal feature of DNA segregation. Eukaryotic chromosome segregation is microtubule-based [15]. And recently, bacterial chromosome segregation has been definitively linked to the actin homolog, MreB, in two unrelated bacterial species [16,17]. Many bacterial chromosomes actually encode an actin homolog (MreB) in addition to several SopA homologs, including ParA (Soj in *B. subtilis*), and MinD. MinD and MreB have been shown to represent independent filament systems in *E. coli* [18], and chromosomal *parA* mutants have subtle chromosome segregation and cell cycle progression defects [6]. If the chromosomal ParA proteins also prove to be cytoskeletal, it will be interesting to see if they collaborate with the MreB actin-like cytoskeleton to facilitate chromosome segregation.

If all DNA segregation machineries involve a cytoskeletal structure, what else might they have in common? Both the microtubules of the eukaryotic spindle apparatus and the ParM filaments of the R1 plasmid partitioning complex exhibit dynamic instability, a kinetic tendency to cycle through phases of assembly and disassembly that facilitates their rapid dynamics [15,19]. Such dynamic instability may also lie at the root of SopA's oscillatory localization cycle. It should prove interesting to see if there are conditions in which SopA and MreB also exhibit dynamic instability.

Perhaps the most surprising aspect of the discovery that SopA forms filaments is that we are still uncovering new cytoskeletal elements. What else is out there in bacteria, or for that matter in other

kingdoms? Provocatively, several prion-like proteins have recently been shown to naturally form fibrils that could be beneficial, rather than aberrant, protein isoforms [20]. A broader perspective should promote a more complete understanding of the identities, functions, and relationships of the cytoskeletal superfamily.

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Working Memory: Linking Capacity with Selectivity

Working memory is one of the most intensively studied psychological processes, but little is known about what distinguishes individuals in their working memory capacity. Recent evidence from electroencephalogram recordings suggests that one crucial component of this variation is our ability to exclude irrelevant information.

Daniel Bor and Adrian M. Owen

While establishing links between mental processes and brain regions is undoubtedly important, these results alone tell us little about the psychological mechanisms under study. Neuroimaging, however, has the potential to inform us about psychology as well as neurophysiology. Indeed, it is possible under certain circumstances for neuroimaging to provide a more sensitive measure of psychological mechanisms than cruder behavioural scores can supply. Vogel *et al*. [1] have recently provided an elegant and striking example of this, by using electroencephalogram (EEG) data to link poor working memory performance with the unnecesary retention of irrelevant items.

Despite a plethora of neuroimaging and behavioural studies, comparatively little is known about how we retain information in working memory [2,3]. In the last year, however, three neuroimaging papers [1,4,5] have reported studies which shed light on how the brain supports this process by using novel approaches to index working memory storage capacity. Using functional magnetic resonance imaging (fMRI), Todd and Marois [4] found that activity in the posterior parietal cortex reflected the number of visual items a volunteer was able to retain in working memory. In a related study, Vogel and Machizawa [5] presented subjects with varying numbers of visual stimuli to one half of their visual field. EEG recordings in parietal and occipital cortices showed greater activity in the hemisphere opposite to the attended stimuli, compared to the hemisphere on the same side. Moreover, this difference varied with the number of items that were successfully encoded. They labelled this novel index of working memory storage ‘contralateral delay activity’. Further analyses demonstrated that the contralateral delay activity associated with the increase from two to four items was highly correlated with working memory capacity between subjects. In other words, two items consumed a larger proportion of working memory storage capacity for subjects with poorer working memory.

Most recently, Vogel *et al*. [1] extended this approach by