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Current Opinion in Cell Biology

Pattern formation in centrosome assembly

Robert Mahen and Ashok R Venkitaraman

A striking but poorly explained feature of cell division is the ability to assemble and maintain organelles not bounded by membranes, from freely diffusing components in the cytosol. This process is driven by information transfer across biological scales such that interactions at the molecular scale allow pattern formation at the scale of the organelle. One important example of such an organelle is the centrosome, which is the main microtubule organising centre in the cell. Centrosomes consist of two centrioles surrounded by a cloud of proteins termed the pericentriolar material (PCM). Profound structural and proteomic transitions occur in the centrosome during specific cell cycle stages, underlying events such as centrosome maturation during mitosis, in which the PCM increases in size and microtubule nucleating capacity. Here we use recent insights into the spatio-temporal behaviour of key regulators of centrosomal maturation, including Polo-like kinase 1, CDK5RAP2 and Aurora-A, to propose a model for the assembly and maintenance of the PCM through the mobility and local interactions of its constituent proteins. We argue that PCM structure emerges as a pattern from decentralised self-organisation through a reaction–diffusion mechanism, with or without an underlying template, rather than being assembled from a central structural template alone. Self-organisation of this kind may have broad implications for the maintenance of mitotic structures, which, like the centrosome, exist stably as supramolecular assemblies on the micron scale, based on molecular interactions at the nanometer scale.

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Introduction

Cellular form and function are dependent on massive supramolecular protein assemblies, which form by the aggregation of many nanometre-scale protein components. One such organelle—the centrosome—has important roles in mitotic spindle function, asymmetric cell division and cilia formation [1]. Microtubule-based structures called centrioles form the core of the centrosome and these are surrounded by an area of dense protein matrix termed the pericentriolar material (PCM; Figure 1a). Like other mitotic structures, the centrosome is not membrane delimited, and instead its contents mix freely with the cytoplasm. Centrosomal morphology and constituents change during particular cell cycle phases, and at the onset of mitosis the PCM increases in size and microtubule nucleating capacity, a process termed centrosome maturation [2]. Defects in centrosomal structure contribute to a diverse range of human diseases including microcephaly, cancer and a group of disorders called ciliopathies, which result from defective cilia function [3].

Here we will review recent work into how key regulators of centrosome PCM structure are temporally and spatially regulated. This leads us to suggest a dynamic model for PCM formation, wherein centrosomal components undergoing decentralised diffusion and reaction collectively cause self-organised PCM assembly. This principle of reaction–diffusion may provide a simplifying explanation for the general problem of how biological information contained in proteins on the nanometer scale can initiate and maintain the formation of aggregated structures which are orders of magnitude larger, on the micron scale.

Key molecular machinery of centrosome maturation

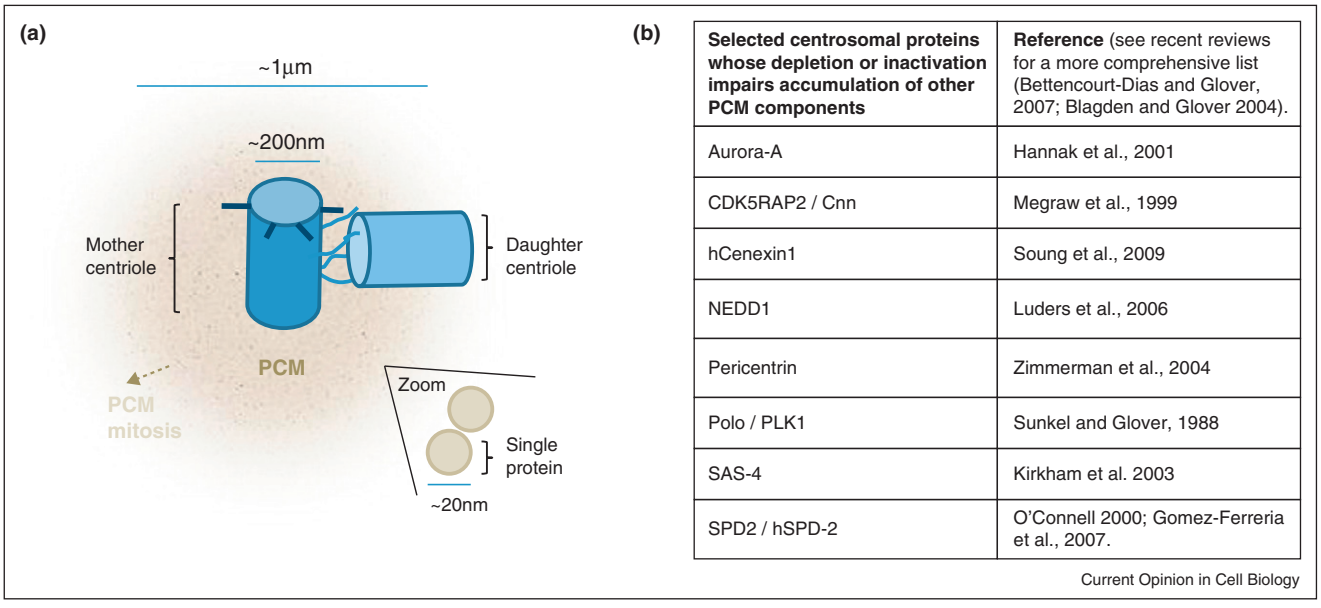
The PCM is thought to be a fibrous matrix, composed of coiled-coil proteins. However, its ultrastructure is far less well defined than that of centrioles, which have clearly visible morphology by electron microscopy [4], and the molecular arrangement of PCM proteins is largely unknown. Functions of the PCM include microtubule nucleation from γ-tubulin ring complexes [5], and influencing the control of centriole number [6]. The expansion in size and microtubule nucleating capacity of the PCM during mitosis is influenced by a growing list of proteins, whose depletion or inactivation has been shown to interfere with centrosome maturation (Figure 1b). Since insights have come from different model organisms, and the proteins involved in general have many conserved functions [1], we will refer interchangeably to protein homologues from different species throughout the review. A common theme amongst papers describing these proteins is that they are hypothesised to form specific interactions within the PCM which anchor other factors. For example NEDD1 is a component of the γ-tubulin ring complex which is thought to interact

Glossary
Centrosome: The major microtubule organising centre in the cell.
Centrosome maturation: The increase and size and microtubule nucleating capacity of the centrosome during mitosis.
Diffusion: The motion of particles produced by the inherent thermal energy in matter, described by a random walk.
Diffusion limited aggregation (DLA): A mechanism of fractal pattern formation through Brownian motion driven particle clustering.
Pericentriolar material (PCM): Protein material of centrosomes surrounding the centrioles of unknown ultrastructure.
Reaction–diffusion: Model of self-organised pattern formation based on the mobility of the particles and their interactions, first described by Turing (1951).
Self-organisation: Model of pattern formation for complex systems out of thermodynamic equilibrium, based on dynamic components.
Stigmergy: Model of self-organised pattern formation stating that the behaviour of the agents in a system is influenced by the structure they build. Formulated by Grasse (1959).

directly with γ-tubulin to allow its anchoring at the centrosome [7–9].

Drosophila Cnn (or its mammalian orthologue, CDK5RAP2) and the mitotic serine/threonine kinases Polo-like kinase 1 (PLK1) and Aurora-A (reviewed in [2,10]) play a central role in PCM maturation. CDK5RAP2/Cnn is a component of the PCM which is required for the normal recruitment of many other PCM factors, including γ-tubulin, with which it interacts via its amino-terminus [11–13], and pericentrin with which it interacts via its carboxyl-terminus [14,15]. The importance of CDK5RAP2/Cnn to centrosome maturation is underscored by its identification in a whole genome siRNA screen in *Drosophila* as one of only two factors whose depletion is able to completely prevent maturation [16]; the other being Polo (PLK1 in mammalian cells). Indeed, PLK1 inactivation in a range of different genetic and chemical systems has consistently established that PLK1 kinase activity is required for the normal localisation of many other PCM components [2], including hSPD-2 (also known as CEP192), pericentrin, CDK5RAP2/Cnn, and NEDD1 [16,17]. The exact mechanisms allowing PLK1 recruitment to the centrosome in mammalian cells are still unclear, but in *Caenorhabditis elegans* at least, SPD-2 may be a key upstream binding partner [18••]. Two recent papers have shown a link between cell cycle progression and centrosome maturation; CDK1 is able to create binding sites for the phospho-binding domain of PLK1 on both the mother centriole component hCenexin1, and NEDD1 [19,20]. PLK1 activity during mitosis may modify centrosomes significantly before the time of their maturation, by licensing daughter centrioles to form PCM in the following interphase of the cell cycle [21••]. However, the molecular targets of PLK1 which regulate this process remain to be determined. Similarly to PLK1, Aurora-A activity is required for the enrichment or localisation of multiple centrosomal factors which have roles in maturation, including LATS2 [22] and CDK5RAP2/Cnn [23] (see [10] for a review). Aurora-A is thought to be targeted and activated at centrosomes by multiple mechanisms, including binding to hSPD-2 [24], *Drosophila* Ajuba [25] and Arcp1b [26].

Figure 1



Selected centrosomal proteins whose depletion or inactivation impairs accumulation of other PCM components	**Reference** (see recent reviews for a more comprehensive list (Bettencourt-Dias and Glover, 2007; Blagden and Glover 2004).
Aurora-A	Hannak et al., 2001
CDK5RAP2 / Cnn	Megraw et al., 1999
hCenexin1	Soung et al., 2009
NEDD1	Luders et al., 2006
Pericentrin	Zimmerman et al., 2004
Polo / PLK1	Sunkel and Glover, 1988
SAS-4	Kirkham et al. 2003
SPD2 / hSPD-2	O'Connell 2000; Gomez-Ferreria et al., 2007.

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The centrosome is a supramolecular assembly consisting of many nanometre scale protein components. **(a)** Cartoon schematic of a centrosome consisting of two centrioles (blue) surrounded by a region of protein termed the pericentriolar material (PCM; brown). The PCM expands in size and microtubule nucleating capacity during mitosis, a process termed maturation (represented by soft brown edges). **(b)** The table lists a selection of centrosomal proteins implicated in γ-tubulin accumulation during centrosome maturation; see recent reviews [1,2] for a comprehensive list.

Overall, the considerations summarised above suggest firstly that the enrichment of a core module of proteins is necessary for PCM assembly and maturation, and secondly that protein phosphorylation by the mitotic kinases PLK1 and Aurora-A has an important role in promoting this recruitment [2].

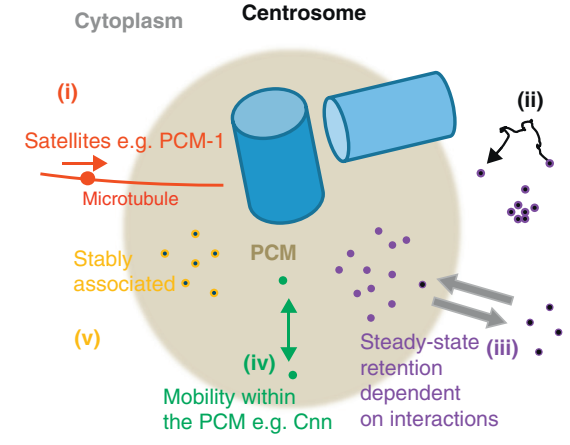
Mechanisms of recruitment and turnover of centrosomal PCM proteins

Given that centrosome maturation is enacted by the enrichment of numerous factors into the organelle, how are these proteins assembled therein? One mechanism of transport to and from the centrosome is along the microtubules which emanate from it (Figure 2ai; [27,28]). Dynein motors transport non-membrane bound aggregations of some centrosomal proteins ('satellites'), including PCM-1, OFD1 and BBS4, and are required for normal centrosomal targeting of pericentrin [29,30], and possibly CDK5RAP2/Cnn [31].

Moreover, because the centrosome is not enclosed by a lipid membrane, cytoplasmic proteins are also able to freely to assemble in it through diffusion (Figure 2aii and aiii). Indeed, a range of centrosomal factors including PLK1 [32,33], Aurora-A [34] and CDK5RAP2/Cnn [35••] have been shown to rapidly exchange between the centrosome and cytoplasm in living cells by fluorescence recovery after photobleaching (FRAP), with half maximal recovery times in the order of seconds (Figure 2b). This suggests that dynamically diffusing pools of protein in the cytoplasm stochastically collide with the centrosome, and then reside there for a period of time whose duration may depend on protein-protein interactions within the organelle (Figure 2aiii–v).

A pulsed-SILAC labelling approach [36•] reveals the turnover of the entire proteomic complement of the centrosome over a 20 h period. Centrosomal proteins exhibit a range of turnover rates, with PLK1, Aurora-A and CDK5RAP2 showing close to 100% exchange in 20 h, in contrast to other components such as γ-TuRC subunits, which had <40% turnover in the same period. The class of centrosomal proteins showing a high exchange rate (>75% in 20 h) was enriched in protein kinases. As

Figure 2



(b)

Steady state behaviour of selected centrosomal factors measured by fluorescence recovery after photobleaching (FRAP)			
Centrosomal protein	**Half time of fluorescence recovery after photobleaching (s)**	**% Recovery of fluorescence recovery after photobleaching**	**Reference**
Aurora-A	2-3	100	Stenoien et al., 2003
CDK5RAP2 / Cnn	25	70	Ge et al., 2010
PLK1	9	85	Mahen et al., 2011; Kishi et al., 2009.
PLK4	11	20	Cizmecioglu et al., 2010
SAS-4/CPAP	-	Stably associated during mitosis, exchanges during interphase	Leidel and Gonczy 2003; Kirkham et al., 2003; Dammermann et al., 2008
hSPD-2	mins	-	Zhu et al., 2008
γ-tubulin	30-60 mins	-	Khodjakov and Rieder 1999; Stenoien et al., 2003.

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Mechanisms of recruitment and turnover of centrosomal proteins. **(ai)** Aggregations of proteins including PCM-1 ('satellites') are transported along microtubules to the centrosome using the motor protein dynein. **(aii)** Cytosolic proteins may exist in a diffusible pool and stochastically encounter the centrosome through Brownian motion. A protein in the cytoplasmic pool may be contained in heterogeneous complexes. **(aiii, aiv)** Some centrosomal factors undergo continuous exchange at the centrosome. The rate of turnover may be dependent on reactivity, as interactions transiently retain them within the centrosome, and diffusivity, both in the cytoplasm and PCM. **(av)** Other PCM proteins may be more stably associated and not exchange over a timescale of seconds or minutes. **(b)** The table shows the steady state behaviour of selected centrosomal factors measured by fluorescence recovery after photobleaching (FRAP).

yet, biochemical analyses and live cell imaging have not been closely correlated in parallel. Despite this caveat, however, available data suggest a model wherein certain PCM factors are steady-state residents based on low affinity interactions, whereas others exist in more stably associated structures, a notion that is supported by the observation that some PCM factors are resistant to harsh salt stripping, whilst others are not [37].

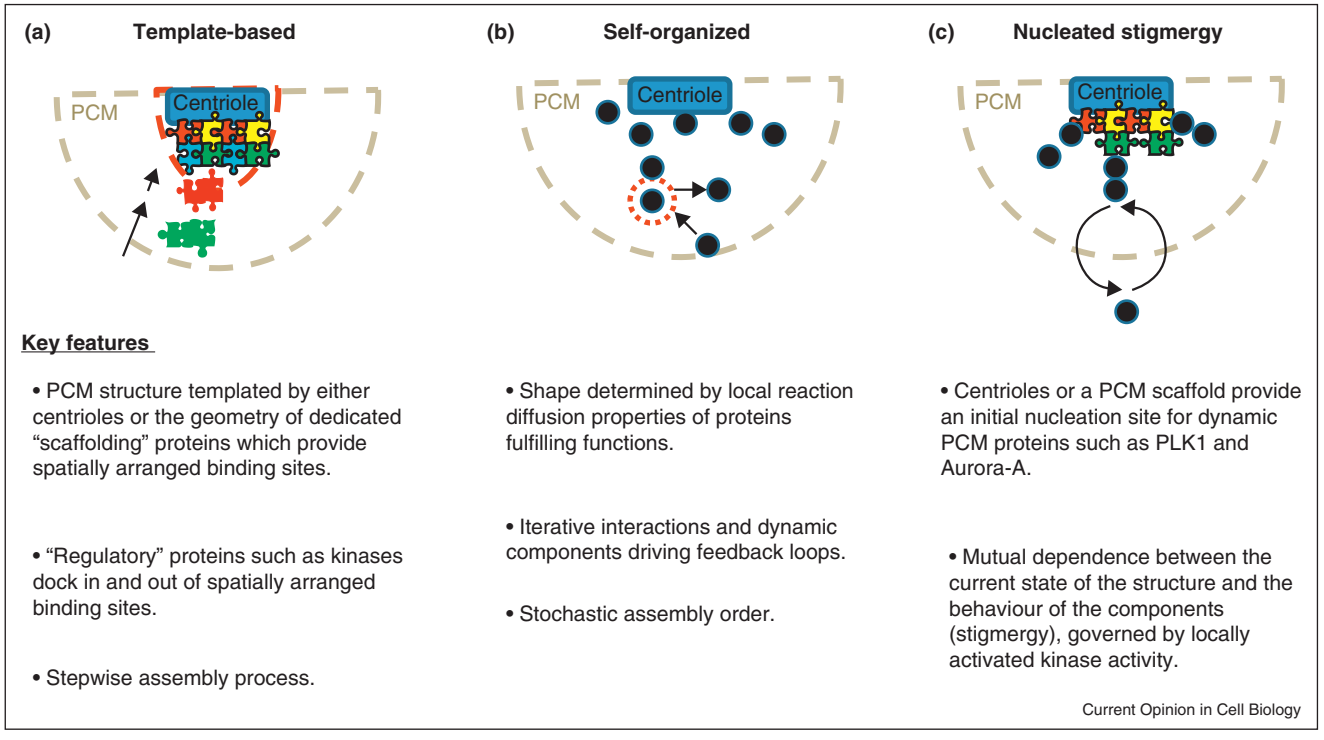
Template-based PCM organisation

How, then, might information transfer across biological scales explain the assembly and maintenance of the supramolecular structure of the PCM from its dynamic protein components? Recent proteomic studies [36•,38] have confirmed earlier work [39], by showing that the centrosome harbours >100 different protein species. The total number of proteins within each centrosome is unknown but is probably in the thousands, and it is a challenge to understand how these molecules can robustly assemble into a single complex structure whose function emerges from the collective properties of the system. For instance, what determines the overall shape and size of the PCM when it is an order of magnitude larger than its constituent parts? Here we consider two different hypothetical models for PCM assembly, which we refer to as 'template-based' or 'self-organised'.

Centrioles are located inside the PCM cloud and their assembly in general precedes that of the PCM, so one possibility (the template-based model) is that the centrioles provide a template in the form of spatially arranged binding sites from which the PCM is built outwards (Figure 3a). Indeed, it is clear that centrioles have a role in organising the PCM: perturbations thought to specifically target centrioles have also been shown to result in PCM disassembly [40,41]. Furthermore, mother and daughter centrioles organise PCM to a different extent [21••,42].

SAS-4 is a core centriolar and PCM protein which becomes stably associated during mitosis and whose levels have been shown to affect both centriole and PCM size [42,43•,44]. Although it is not known exactly how SAS-4 mediates this effect, it has recently been found in conserved complexes also containing CDK5RAP2/Cnn and Asterless (Asl)/CEP-152 [45•,46••,47••]. Whereas recombinant SAS-4 can bind

Figure 3



'Template-based' and 'self-organised' hypothetical models of PCM organisation. **(a)** Template-based: the centrioles may provide binding sites for specific PCM factors such as Asl, Cnn or SAS-4 (represented by jigsaw pieces), which are then able to assemble further PCM components analogous to a jigsaw. PCM shape and size would consequently be determined by the arrangement of binding sites and the geometry of constituent proteins (red dashed line). **(b)** Self-organised: the combined reaction–diffusion behaviour of PCM components acting at the local level (represented by the red dashed line) may result in self-organised structure formation at the level of the whole organelle (brown dashed line). **(c)** Nucleated stigmergy: a central template such as the centrioles or PCM matrix may provide a nucleation site around which dynamic components such as PLK1 and Aurora-A assemble, with mutual dependence between the current state of the structure and the behaviour of the components.

to centrosomes stripped of PCM by high salt treatment, CDK5RAP2/Cnn or Asl cannot, suggesting that SAS-4 might act as a scaffold to assemble other PCM components within the centrosome. Similarly, the centriole component Asl is a good candidate for providing a template for PCM formation, since it is required for the normal maintenance of the PCM [48], and binds PLK4 and SAS-4 at its amino and carboxyl-termini respectively [46••,47••]. Strikingly, expression of a PLK4-binding mutant of Asl leads to MTOCs which are devoid of centrioles at their core [47••], implying that Asl could structurally link centrioles and the PCM. CDK5RAP2/Cnn has also been suggested as a link between centrioles and the PCM; in Cnn or CDK5RAP2 null cells the two structures partially detach from each other, and Cnn is known to attach to many PCM components [13,49,50]. SPD-2/CEP192 has also been implicated in both centriolar duplication and PCM recruitment during maturation [51–53], and elevated levels of SPD-2 in *C. elegans* result in larger centrosomes with increased growth kinetics [18••]. Most of these key regulators of centrosomal structure are involved in both centriole and PCM biogenesis, highlighting the close relationship between the two structures, and providing evidence that the centrioles could serve as a template for PCM assembly.

However, one difficulty of a template-based model (Figure 3a), is that some positional information must be encoded within PCM proteins relative to the whole structure. This is analogous to the problem of how a single cell can know its position within a tissue or organism: unique pairwise interactions between cells (or in this case, between PCM proteins) are unlikely. In whole organisms, diffusible factors called morphogens, emitted from a source, give positional information based on concentration [54]. Gradients of protein activity have been observed around mitotic structures such as the spindle midzone [55], and also within the spindle emanating from the centrosomes (discussed further below in the section entitled *Self-organised and stigmergic PCM assembly*). However, no gradient of activity of centrosomal factors has thus far been reported to set the size of the PCM itself. Elegant recent work in *C. elegans* embryos has suggested that a far simpler mechanism may in fact limit centrosome size and may also provide a solution to the positional information problem [18••]. Centrosome size was observed to scale with cell size, and could also be altered by changing the levels of SPD-2. Therefore, an alternative and simpler mechanism underlying the potential template function of centrioles is that PCM components accumulate around them until rate-limiting components such as SPD-2 run out, at which point no more PCM expansion can occur. It has recently become clear that regulating the levels of centrosomal factors through degradation is crucial for control of centriole biogenesis; PP2A counteracts PLK4 autophosphorylation to prevent its proteasome-mediated degradation during mitosis [56,57]. Whether similar degradational mechanisms also control PCM structure is less clear at this time, but remains possible, considering that protein phosphatase PP2A appears to be required for maturation in flies [16].

Thus, the centrioles might exert a dominant guiding influence on PCM structure in a template-based model, with the nature of the interactions between PCM proteins determining their position. A conceptually similar principle may be applicable to centriole biogenesis, rather than the PCM. SAS-6, a component of the core centriole biogenesis machinery, can oligomerise *in vitro* into a nine-fold symmetrical cartwheel structure similar to that seen in the centriole *in vivo* [58••,59••], and co-overexpression of SAS6 and Ana2 causes tubules resembling the cartwheel in *Drosophila* spermatocytes [60]. This is compelling evidence that at least the structure of this part of the centriole results primarily from the geometry of a single protein. Similarly, elegant cryo-electron microscopy structural studies have shown that γ-tubulin containing complexes are able to promote microtubule nucleation by forming a platform of the correct shape, onto which microtubule subunits can assemble [61]. Centriole assembly is mediated by a conserved pathway of essential factors governing a stepwise assembly process, coordinated in part by post-translational modifications (reviewed in [62]). Exciting recent work has demonstrated how PLK4 is upstream of SAS-5 and SAS-6, and SAS-5 assembly at the centrosome is promoted by dephosphorylation through PP2A [47,63,64]. It is not clear at present whether such a sequential enrichment and hierarchy of core factors also governs PCM assembly or maturation however, and it is still possible that it is controlled by a distributed network (see section entitled *Key molecular machinery of centrosome maturation*).

Although the considerations in this section suggest that the centrioles exert an influence over PCM structure and morphology, consistent with a template-based model (Figure 3a), there are serious gaps in our understanding of how exactly how such a model might work. Recent super-resolution imaging of the PCM proteins SAS-4, Asl and CEP-63 shows that they form a toroid shape around centrioles [45•,65], hinting at a regular ultrastructure connecting the PCM-centriole interface. However, the existence and exact identity of a direct physical linkage between the centrioles and the PCM remains unclear. Pairwise interactions between important PCM proteins have been partially elucidated, but whether PCM assembly as a whole constitutes a stepwise assembly process (Figure 3) similar to centriole biogenesis [62] is unclear. Given these points, the notion that the overall shape and size of the PCM is constructed from spatially arranged binding sites, or from the geometry of its protein constituents warrants further investigation.

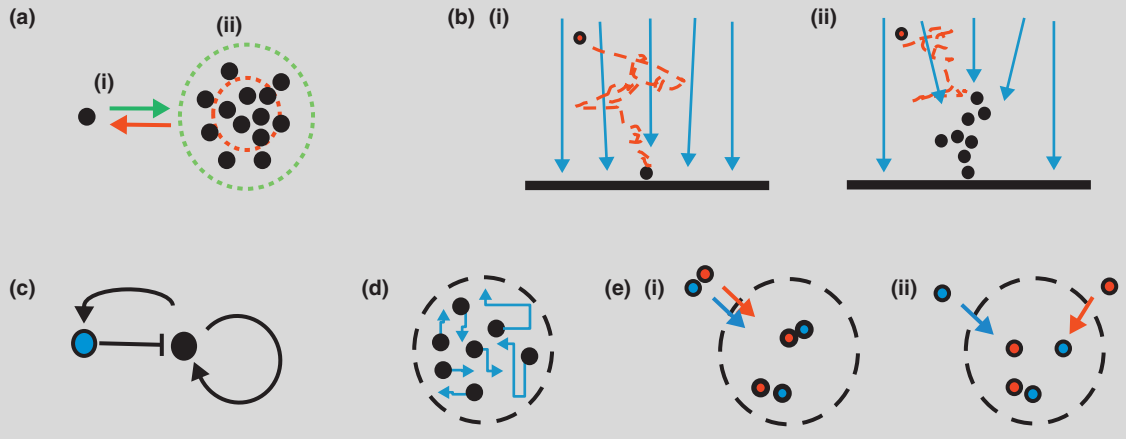
Self-organised and stigmergic PCM assembly

There is, however, another major gap in current evidence supporting a template-based model for PCM assembly. If centrioles do create a template for PCM formation then one central prediction is that the PCM should not form in the absence of the template. However, the overexpression of various PCM components, such as hSPD-2/CEP192 [66], hCenexin1 [20], CDK5RAP2 [13] and Cnn [23] causes the formation of multiple extracentriolar foci which are able to nucleate microtubules. Similarly, expression of a PLK4 binding mutant of Asl or SAS-4, causes the formation of acentriolar structures reminiscent of PCM by both electron and fluorescence microscopy [47••,67]. Although there are tight controls which restrict centrosome biogenesis in normal cell cycles [1], whole centrosomes can form *de novo* in certain circumstances [68], suggesting that the presence of another centrosome is not strictly necessary for building the entire organelle. Furthermore, after laser ablation of centrosomes in S phase arrested cells, *de novo* centrosome assembly precedes by the formation of a cloud of PCM before centrioles appear [69]. These considerations argue that PCM assembly is not templated by centrioles.

An alternative but less well explored model of PCM formation posits that it self-organises from the combined reaction–diffusion properties of its constituents (Figure 3b). In contrast to a template-based model of pattern formation, in which the pattern is determined based on the template or scaffold, in a self-organisation model the global pattern might arise internally in the system from the dynamic decentralised interactions of the agents. The dynamic steady-state nature of many PCM proteins (see Figure 2 and the section entitled *Mechanisms of recruitment and turnover of centrosomal PCM proteins*) is consistent with self-organisation. For example, Cnn shows rapid exchange dynamics in living *Drosophila* cells and its rate of exchange is highest in the PCM immediately adjacent to the centrioles rather than at the exterior of the PCM [35••]. Dynamic behaviour of this nature is difficult to reconcile with a template-based model wherein the position of spatially arranged binding sites promotes PCM assembly.

Recruitment from a diffusible pool is a common feature of many organelles in the nucleus, which like the centrosome are not membrane delimited, and intriguingly, is hypothesised to be a significant determinant of function and structure [70] (see Box 1a). Indeed, the steady state turnover of both Cnn and PLK1 at the centrosome has been shown to correlate with PCM size [33,35••,71••]. This suggests that by PCM structure can be dynamically maintained by modulating the rate of exchange of key proteins [35••]. Although Brownian diffusion can itself be a key pattern forming parameter in both biological and physical systems (see Box1b), the motion of centrosomal proteins in the cytoplasm is almost entirely unstudied. There is evidence that cytoplasmic proteins may be able

Box 1 Hypothetical models of reaction and diffusion driven pattern formation in the centrosome PCM. **(a) and (b)** The size of non-membrane bound steady-state structures inside the cell (shown by the dashed lines) has been hypothesised to be related to the rate of exchange of constituent proteins [70,75]. Increased flux into an organelle might increase its size (green lines), and decreased flux into an organelle might decrease size. Two factors controlling the rate of exchange may be the rate of association with the organelle from a cytoplasmic pool **(i)** and the duration of the interactions within the structure **(ii)**. **(b)** Brownian motion has been shown to be a key pattern forming parameter in both physical and biological systems undergoing a process called diffusion limited aggregation (DLA) [82]. **(i)** Particles (black dots) adhering to a uniform surface initially do so randomly. **(ii)** As each particle takes a circuitous route by diffusion (red dashed line), it is more likely to bind to the accumulating structure, before reaching the original surface. Blue arrows show the average flow of particles. **(c)** In a long-range inhibition—short-range activation mechanism of self-organisation, an activator (black dot) stimulates production of both itself and its own inhibitor (blue dot) [77,80]. Such a scheme creates spatial patterns from initially random heterogeneities. **(d)** Rather than occupying distinct positions, PCM proteins may continually move (denoted by blue arrows), similar to the molecules in a liquid. **(e)** Centrosomal complexes may preassemble in the cytoplasm **(i)** or assembly stochastically on site **(ii)**.



to pass through the PCM relatively unhindered in certain cases (Figure 2d and Box 1d), as an inert tracer, mCherry, is freely mobile within the PCM [33]. Similarly in living *Drosophila* cells cytoplasmic Cnn diffuses through the PCM to its core, binding there before the exterior [35••]. Therefore, molecular crowding *in vivo* does not appear to impede the access of cytoplasmic proteins to the centrosome, and this is consistent with data suggesting that nuclear organelles may be relatively porous in this regard [72].

Self-organising models of spindle formation have been proposed [73], and a gradient of TPXL-1, the protein responsible for targeting and activating Aurora-A, has been shown to emanate from centrosomes to regulate spindle length [43•]. In this case, the gradient of TPXL-1 may be related to both its diffusion coefficient and detachment rate from the centrosome, illustrating how a reaction–diffusion mechanism involving centrosomes may play a role in spindle length control.

In a self-organised model of PCM assembly, organelle morphology would emerge as a collective property of the system, and not from the individual shape or function of a single protein. Consistent with this, although there are >100 different proteins within the centrosome [39], very few are actually essential for centrosome maturation and centriole biogenesis (reviewed in [2,16,62]). This suggests that all centrosomal proteins are 'structural' constituents that may contribute to the overall morphology, no matter how transiently they reside within it. Even Aurora-A, which might be expected to fulfil a regulatory role through very transient phospho-transfer enzymatic reactions, has recently been found to have kinase independent functions [74].

Self-organisation models of cellular organisation are difficult to test experimentally if they state that structure and function cannot be separated [75]. Positive and negative feedback loops, such as local amplification—long-range inhibition (Box 1c) [76–77] are the basis of many self-organising systems. Is there any evidence to suggest such properties within the PCM? A positive feedback loop may exist between Aurora-A activity and its centrosomal recruitment, because binding to hSPD-2 at the centrosome may facilitate further recruitment [24]. Similarly, at kinetochores it is known that PLK1 kinase activity creates binding sites for further PLK1 recruitment, in a cooperative feed forward mechanism between kinase activity and binding [78]. Such a mechanism is also feasible at the centrosome, particularly since PLK1 and Aurora-A are cooperative [79].

Moreover, we recently showed that PLK1 mobility is markedly slowed at the centrosome in comparison to the cytoplasm, apparently due to repetitive transient interactions in a structure which is much larger than the individual protein components [33]. Such a disparity in the diffusional mobilities of system components has been shown to provide the basis for self-organisation [80], and may provide a feedback loop between reactivity and localisation. Indeed, the existence of putative feedback loops, both positive and negative, is not only consistent with self-organisation, but may in the future suggest avenues to stringently test the model. In particular, the self-organisation model predicts that the removal or rewiring of dynamic feedback loops will impinge upon PCM structure. For example, it might prove interesting in future work to interfere with PLK1 'self-priming' of its own binding sites, thereby perturbing a feedback loop without removing key molecular interactions at the centrosome [78], although the inherent technical challenges should not be overlooked.

There may be elements of truth in both the template-based and self-organisation models, since parts of the PCM seem to form a stable core matrix, whilst others associate in a steady-state fashion. We therefore suggest a combined model that might reconcile these observations (Figure 3c), wherein the more dynamic elements of the PCM are nucleated from an initial template. A crucial aspect of this model is the principle of stigmergy [81], which postulates that feedback between the existing structure and the dynamic parts triggers new behaviours in both (Figure 3c). The PCM could then assemble dynamically, as discussed above, wherein the function and form of the organelle are interdependent. One direction for future work based on this model might be to test whether perturbations to the current state of the structure also affect the behaviour of system components, in contrast to a templated model in which the information flow is largely unidirectional, from the template to the parts. Thus, overall we believe that such a combined model will better explain the available data than less integrative views of centrosome PCM assembly, and point to interesting new avenues for future work.

Conclusion

How is the emergent form and function of centrosomal PCM robustly created in time and space? A common theme amongst recent work delineating both centriole and PCM assembly, is that key constituents are carefully regulated both temporally and spatially by recruitment to the centrosome from a diffusible pool. Future work will surely uncover further multitudinous interactions between PCM components, regulated by post-translational modifications governed by enzymatic activity. We suggest that exciting insights may also be possible by determining how these processes interplay with molecular mobility. This principle of reaction–diffusion may provide a simplifying model explaining how many centrosomal proteins combine to create the complex supramolecular structure of centrosomal PCM, in such a way that its functions emerge from the properties and inter-

actions of its components as a whole, rather than from the discrete roles of specific proteins alone.

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References and recommended reading

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

- • of special interest
- •• of outstanding interest

1. Bettencourt-Dias M, Glover DM: **Centrosome biogenesis and function: centrosomics brings new understanding**. *Nat Rev Mol Cell Biol* 2007, **8**:451-463.

2. Blagden SP, Glover DM: **Polar expeditions—provisioning the centrosome for mitosis**. *Nat Cell Biol* 2003, **5**:505-511.

3. Nigg EA, Raff JW: **Centrioles, centrosomes, and cilia in health and disease**. *Cell* 2009, **139**:663-678.

4. Pelletier L, O'Toole E, Schwager A, Hyman AA, Muller-Reichert T: **Centriole assembly in *Caenorhabditis elegans***. *Nature* 2006, **444**:619-623.

5. Raynaud-Messina B, Merdes A: **Gamma-tubulin complexes and microtubule organization**. *Curr Opin Cell Biol* 2007, **19**:24-30.

6. Loncarek J, Hergert P, Magidson V, Khodjakov A: **Control of daughter centriole formation by the pericentriolar material**. *Nat Cell Biol* 2008, **10**:322-328.

7. Luders J, Patel UK, Stearns T: **GCP-WD is a gamma-tubulin targeting factor required for centrosomal and chromatin-mediated microtubule nucleation**. *Nat Cell Biol* 2006, **8**:137-147.

8. Manning JA, Shalini S, Risk JM, Day CL, Kumar S: **A direct interaction with NEDD1 regulates gamma-tubulin recruitment to the centrosome**. *PLoS ONE* 2010, **5**:e9618.

9. Haren L, Remy MH, Bazin I, Callebaut I, Wright M, Merdes A: **NEDD1-dependent recruitment of the gamma-tubulin ring complex to the centrosome is necessary for centriole duplication and spindle assembly**. *J Cell Biol* 2006, **172**:505-515.

10. Barr AR, Gergely F: **Aurora-A: the maker and breaker of spindle poles**. *J Cell Sci* 2007, **120**:2987-2996.

11. Megraw TL, Li K, Kao LR, Kaufman TC: **The centrosomin protein is required for centrosome assembly and function during cleavage in Drosophila**. *Development* 1999, **126**:2829-2839.

12. Choi YK, Liu P, Sze SK, Dai C, Qi RZ: **CDK5RAP2 stimulates microtubule nucleation by the gamma-tubulin ring complex**. *J Cell Biol* 2010, **191**:1089-1095.

13. Fong KW, Choi YK, Rattner JB, Qi RZ: **CDK5RAP2 is a pericentriolar protein that functions in centrosomal attachment of the gamma-tubulin ring complex**. *Mol Biol Cell* 2008, **19**:115-125.

14. Buchman JJ, Tseng HC, Zhou Y, Frank CL, Xie Z, Tsai LH: **Cdk5rap2 interacts with pericentrin to maintain the neural progenitor pool in the developing neocortex**. *Neuron* 2010, **66**:386-402.

15. Wang Z, Wu T, Shi L, Zhang L, Zheng W, Qu JY, Niu R, Qi RZ: **Conserved motif of CDK5RAP2 mediates its localization to centrosomes and the Golgi complex**. *J Biol Chem* 2010, **285**:22658-22665.

16. Dobbelaere J, Josue F, Suijkerbuijk S, Baum B, Tapon N, Raff J: **A genome-wide RNAi screen to dissect centriole duplication and centrosome maturation in Drosophila**. *PLoS Biol* 2008, **6**:e224.

17. Haren L, Stearns T, Luders J: **Plk1-dependent recruitment of gamma-tubulin complexes to mitotic centrosomes involves multiple PCM components**. *PLoS ONE* 2009, **4**:e5976.

18. •• Decker M, Jaensch S, Pozniakovsky A, Zinke A, O'Connell KF, Zachariae W, Myers E, Hyman AA: **Limiting amounts of centrosome material set centrosome size in *C. elegans* embryos**. *Curr Biol* 2011, **21**:1259-1267.
The authors use automated tracking and analysis of centrosomes in *C. elegans* to show that centrosomal size scales with cell size and propose a limiting component model, hypothesising that total amounts of SPD-2 might limit centrosomal growth.

19. Zhang X, Chen Q, Feng J, Hou J, Yang F, Liu J, Jiang Q, Zhang C: **Sequential phosphorylation of Nedd1 by Cdk1 and Plk1 is required for targeting of the gammaTuRC to the centrosome**. *J Cell Sci* 2009, **122**:2240-2251.

20. Soung NK, Park JE, Yu LR, Lee KH, Lee JM, Bang JK, Veenstra TD, Rhee K, Lee KS: **Plk1-dependent and -independent roles of an ODF2 splice variant, hCenexin1, at the centrosome of somatic cells**. *Dev Cell* 2009, **16**:539-550.

21. •• Wang WJ, Soni RK, Uryu K, Tsou MF: **The conversion of centrioles to centrosomes: essential coupling of duplication with segregation**. *J Cell Biol* 2011, **193**:727-739.
This paper shows that PLK1 dependent modification of centrioles during mitosis licenses them to recruit PCM in the next cell cycle, suggesting a mechanism via which centriole duplication and segregation are coupled.

22. Toji S, Yabuta N, Hosomi T, Nishihara S, Kobayashi T, Suzuki S, Tamai K, Nojima H: **The centrosomal protein Lats2 is a phosphorylation target of Aurora-A kinase**. *Genes Cells* 2004, **9**:383-397.

23. Terada Y, Uetake Y, Kuriyama R: **Interaction of Aurora-A and centrosomin at the microtubule-nucleating site in Drosophila and mammalian cells**. *J Cell Biol* 2003, **162**:757-763.

24. Joukov V, De Nicolo A, Rodriguez A, Walter JC, Livingston DM: **Centrosomal protein of 192 kDa (Cep192) promotes centrosome-driven spindle assembly by engaging in organelle-specific Aurora A activation**. *Proc Natl Acad Sci USA* 2010, **107**:21022-21027.

25. Sabino D, Brown NH, Basto R: **Drosophila Ajuba is not an Aurora-A activator but is required to maintain Aurora-A at the centrosome**. *J Cell Sci* 2011, **124**:1156-1166.

26. Molli PR, Li DQ, Bagheri-Yarmand R, Pakala SB, Katayama H, Sen S, Iyer J, Chernoff J, Tsai MY, Nair SS *et al.*: **Arpc1b, a centrosomal protein, is both an activator and substrate of Aurora A**. *J Cell Biol* 2010, **190**:101-114.

27. Zimmerman W, Doxsey SJ: **Construction of centrosomes and spindle poles by molecular motor-driven assembly of protein particles**. *Traffic* 2000, **1**:927-934.

28. Young A, Dictenberg JB, Purohit A, Tuft R, Doxsey SJ: **Cytoplasmic dynein-mediated assembly of pericentrin and gamma tubulin onto centrosomes**. *Mol Biol Cell* 2000, **11**:2047-2056.

29. Dammermann A, Merdes A: **Assembly of centrosomal proteins and microtubule organization depends on PCM-1**. *J Cell Biol* 2002, **159**:255-266.

30. Lopes CA, Prosser SL, Romio L, Hirst RA, O'Callaghan C, Woolf AS, Fry AM: **Centriolar satellites are assembly points for proteins implicated in human ciliopathies, including oral-facial-digital syndrome 1**. *J Cell Sci* 2011, **124**:600-612.

31. Lee S, Rhee K: **CEP215 is involved in the dynein-dependent accumulation of pericentriolar matrix proteins for spindle pole formation**. *Cell Cycle* 2010, **9**:774-783.

32. Kishi K, van Vugt MA, Okamoto K, Hayashi Y, Yaffe MB: **Functional dynamics of Polo-like kinase 1 at the centrosome**. *Mol Cell Biol* 2009, **29**:3134-3150.

33. Mahen R, Jeyasekharan AD, Barry NP, Venkitaraman AR: **Continuous polo-like kinase 1 activity regulates diffusion to maintain centrosome self-organization during mitosis**. *Proc Natl Acad Sci USA* 2011, **108**:9310-9315.

34. Stenoien DL, Sen S, Mancini MA, Brinkley BR: **Dynamic association of a tumor amplified kinase, Aurora-A, with the centrosome and mitotic spindle**. *Cell Motil Cytoskeleton* 2003, **55**:134-146.

35. Conduit PT, Brunk K, Dobbelaere J, Dix CI, Lucas EP, Raff JW:
•• **Centrioles regulate centrosome size by controlling the rate of Cnn incorporation into the PCM**. *Curr Biol* 2010, **20**:2178-2186.
Combined with Ref. [71••], the authors use fluorescence recovery after photobleaching (FRAP) to show that Cnn incorporates into the PCM closest to the centrioles before spreading outwards, and propose that the rate of Cnn incorporation controls centrosome size.

36. Jakobsen L, Vanselow K, Skogs M, Toyoda Y, Lundberg E, Poser I,
• Falkenby LG, Bennetzen M, Westendorf J, Nigg EA *et al.*: **Novel asymmetrically localizing components of human centrosomes identified by complementary proteomics methods**. *EMBO J* 2011, **30**:1520-1535.
This report examines the proteomic composition of the centrosome using mass spectrometry, including a profile of the turnover of centrosomal proteins over a 20 h period.

37. Palazzo RE, Vogel JM, Schnackenberg BJ, Hull DR, Wu X: **Centrosome maturation**. *Curr Top Dev Biol* 2000, **49**:449-470.

38. Muller H, Schmidt D, Steinbrink S, Mirgorodskaya E, Lehmann V, Habermann K, Dreher F, Gustavsson N, Kessler T, Lehrach H *et al.*: **Proteomic and functional analysis of the mitotic Drosophila centrosome**. *EMBO J* 2010, **29**:3344-3357.

39. Andersen JS, Wilkinson CJ, Mayor T, Mortensen P, Nigg EA, Mann M: **Proteomic characterization of the human centrosome by protein correlation profiling**. *Nature* 2003, **426**:570-574.

40. Basto R, Lau J, Vinogradova T, Gardiol A, Woods CG, Khodjakov A, Raff JW: **Flies without centrioles**. *Cell* 2006, **125**:1375-1386.

41. Bobinnec Y, Khodjakov A, Mir LM, Rieder CL, Edde B, Bornens M: **Centriole disassembly in vivo and its effect on centrosome structure and function in vertebrate cells**. *J Cell Biol* 1998, **143**:1575-1589.

42. Kirkham M, Muller-Reichert T, Oegema K, Grill S, Hyman AA: **SAS-4 is a *C. elegans* centriolar protein that controls centrosome size**. *Cell* 2003, **112**:575-587.

43. Greenan G, Brangwynne CP, Jaensch S, Gharakhani J,
• Julicher F, Hyman AA: **Centrosome size sets mitotic spindle length in *Caenorhabditis elegans* embryos**. *Curr Biol* 2010, **20**:353-358.
The authors show that spindle length correlates with centrosome size, possibly owing to control of a TPXL-1 gradient.

44. Dammermann A, Maddox PS, Desai A, Oegema K: **SAS-4 is recruited to a dynamic structure in newly forming centrioles that is stabilised by the gamma-tubulin-mediated addition of centriolar microtubules**. *J Cell Biol* 2008, **180**:771-785.

45. Gopalakrishnan J, Mennella V, Blachon S, Zhai B, Smith AH,
• Megraw TL, Nicastro D, Gygi SP, Agard DA, Avidor-Reiss T: **Sas-4 provides a scaffold for cytoplasmic complexes and tethers them in a centrosome**. *Nat Commun* 2011, **2**:359.
SAS-4 binds Asl, Cnn, D-PLP and CP-190 in the cytoplasm and allows these factors to bind salt stripped centrosomes.

46. Cizmecioglu O, Arnold M, Bahtz R, Settele F, Ehret L, Haselmann-
•• Weiss U, Antony C, Hoffmann I: **Cep152 acts as a scaffold for recruitment of Plk4 and CPAP to the centrosome**. *J Cell Biol* 2010, **191**:731-739.
See Ref. [47••].

47. Dzhindzhev NS, Yu QD, Weiskopf K, Tzolovsky G, Cunha-
•• Ferreira I, Riparbelli M, Rodrigues-Martins A, Bettencourt-Dias M, Callaini G, Glover DM: **Asterless is a scaffold for the onset of centriole assembly**. *Nature* 2010, **467**:714-718.
Papers [46••] and [47••] identify Asterless/CEP-152 as a component of centriole biogenesis which is upstream of PLK4, providing a platform onto which both PLK4 and SAS4 can bind.

48. Varmark H, Llamazares S, Rebollo E, Lange B, Reina J, Schwarz H, Gonzalez C: **Asterless is a centriolar protein required for centrosome function and embryo development in Drosophila**. *Curr Biol* 2007, **17**:1735-1745.

49. Barr AR, Kilmartin JV, Gergely F: **CDK5RAP2 functions in centrosome to spindle pole attachment and DNA damage response**. *J Cell Biol* 2010, **189**:23-39.

50. Lucas EP, Raff JW: **Maintaining the proper connection between the centrioles and the pericentriolar matrix requires Drosophila centrosomin**. *J Cell Biol* 2007, **178**:725-732.

51. Pelletier L, Ozlu N, Hannak E, Cowan C, Habermann B, Ruer M, Muller-Reichert T, Hyman AA: **The *Caenorhabditis elegans* centrosomal protein SPD-2 is required for both pericentriolar material recruitment and centriole duplication**. *Curr Biol* 2004, **14**:863-873.

52. Zhu F, Lawo S, Bird A, Pinchev D, Ralph A, Richter C, Muller-Reichert T, Kittler R, Hyman AA, Pelletier L: **The mammalian SPD-2 ortholog Cep192 regulates centrosome biogenesis**. *Curr Biol* 2008, **18**:136-141.

53. Dix CI, Raff JW: **Drosophila Spd-2 recruits PCM to the sperm centriole, but is dispensable for centriole duplication**. *Curr Biol* 2007, **17**:1759-1764.

54. Wolpert L: **Positional information and the spatial pattern of cellular differentiation**. *J Theor Biol* 1969, **25**:1-47.

55. Fuller BG, Lampson MA, Foley EA, Rosasco-Nitcher S, Le KV, Tobelmann P, Brautigan DL, Stukenberg PT, Kapoor TM: **Midzone activation of aurora B in anaphase produces an intracellular phosphorylation gradient**. *Nature* 2008, **453**:1132-1136.

56. Puklowski A, Homsi Y, Keller D, May M, Chauhan S, Kossatz U, Grunwald V, Kubicka S, Pich A, Manns MP *et al.*: **The SCF-FBXW5 E3-ubiquitin ligase is regulated by PLK4 and targets HsSAS-6 to control centrosome duplication**. *Nat Cell Biol* 2011, **13**:1004-1009.

57. Brownlee CW, Klebba JE, Buster DW, Rogers GC: **The protein phosphatase 2A regulatory subunit twins stabilizes Plk4 to induce centriole amplification**. *J Cell Biol* 2011, **195**:231-243.

58. Kitagawa D, Vakonakis I, Olieric N, Hilbert M, Keller D, Olieric V,
•• Bortfeld M, Erat MC, Fluckiger I, Gonczy P *et al.*: **Structural basis of the 9-fold symmetry of centrioles**. *Cell* 2011, **144**:364-375.
See Ref. [59••].

59. van Breugel M, Hirono M, Andreeva A, Yanagisawa HA,
•• Yamaguchi S, Nakazawa Y, Morgner N, Petrovich M, Ebong IO, Robinson CV *et al.*: **Structures of SAS-6 suggest its organization in centrioles**. *Science* 2011, **331**:1196-1199.
Ref. [58••] and [59••] show that SAS-6 is able to oligomerise in order to form a nine-fold symmetrical cartwheel structure, revealing that this likely forms the structural basis for the morphology of this part of the centriole *in vivo*.

60. Stevens NR, Roque H, Raff JW: **DSas-6 and Ana2 coassemble into tubules to promote centriole duplication and engagement**. *Dev Cell* 2010, **19**:913-919.

61. Kollman JM, Polka JK, Zelter A, Davis TN, Agard DA: **Microtubule nucleating gamma-TuSC assembles structures with 13-fold microtubule-like symmetry**. *Nature* 2010, **466**:879-882.

62. Azimzadeh J, Marshall WF: **Building the centriole**. *Curr Biol* 2010, **20**:R816-R825.

63. Kitagawa D, Fluckiger I, Polanowska J, Keller D, Reboul J, Gonczy P: **PP2A phosphatase acts upon SAS-5 to ensure centriole formation in *C. elegans* embryos**. *Dev Cell* 2011, **20**:550-562.

64. Song MH, Liu Y, Anderson DE, Jahng WJ, O'Connell KF: **Protein phosphatase 2A-SUR-6/B55 regulates centriole duplication in *C. elegans* by controlling the levels of centriole assembly factors**. *Dev Cell* 2011, **20**:563-571.

65. Sir JH, Barr AR, Nicholas AK, Carvalho OP, Khurshid M, Sossick A, Reichelt S, D'Santos C, Woods CG, Gergely F: **A primary microcephaly protein complex forms a ring around parental centrioles**. *Nat Genet* 2011, **43**:1147-1153.

66. Gomez-Ferreria MA, Rath U, Buster DW, Chanda SK, Caldwell JS, Rines DR, Sharp DJ: **Human Cep192 is required for mitotic centrosome and spindle assembly**. *Curr Biol* 2007, **17**:1960-1966.

67. Peel N, Stevens NR, Basto R, Raff JW: **Overexpressing centriole-replication proteins in vivo induces centriole overduplication and de novo formation**. *Curr Biol* 2007, **17**:834-843.

68. Rodrigues-Martins A, Riparbelli M, Callaini G, Glover DM, Bettencourt-Dias M: **Revisiting the role of the mother centriole in centriole biogenesis**. *Science* 2007, **316**:1046-1050.

69. Khodjakov A, Rieder CL, Sluder G, Cassels G, Sibon O, Wang CL: **De novo formation of centrosomes in vertebrate cells arrested during S phase**. *J Cell Biol* 2002, **158**:1171-1181.

70. Dundr M, Misteli T: **Biogenesis of nuclear bodies**. *Cold Spring Harb Perspect Biol* 2010, **2**:a000711.

71. •• Conduit PT, Raff JW: **Cnn dynamics drive centrosome size asymmetry to ensure daughter centriole retention in Drosophila neuroblasts**. *Curr Biol* 2010, **20**:2187-2192.
See Ref. [35••].

72. Handwerger KE, Cordero JA, Gall JG: **Cajal bodies, nucleoli, and speckles in the Xenopus oocyte nucleus have a low-density, sponge-like structure**. *Mol Biol Cell* 2005, **16**:202-211.

73. Karsenti E, Vernos I: **The mitotic spindle: a self-made machine**. *Science* 2001, **294**:543-547.

74. Toya M, Terasawa M, Nagata K, Iida Y, Sugimoto A: **A kinase-independent role for Aurora A in the assembly of mitotic spindle microtubules in *Caenorhabditis elegans* embryos**. *Nat Cell Biol* 2011, **13**:708-714.

75. Misteli T: **The concept of self-organization in cellular architecture**. *J Cell Biol* 2001, **155**:181-185.

76. Camazine S, Deneubourg J, Franks N, Sneyd J, Theraluz G, Bonabeau E: *Self Organization in Biological Systems*. Princeton University Press; 2001.

77. Gierer A, Meinhardt H: **A theory of biological pattern formation**. *Kybernetik* 1972, **12**:30-39.

78. Park JE, Erikson RL, Lee KS: **Feed-forward mechanism of converting biochemical cooperativity to mitotic processes at the kinetochore plate**. *Proc Natl Acad Sci USA* 2011, **108**:8200-8205.

79. Macurek L, Lindqvist A, Lim D, Lampson MA, Klompmaker R, Freire R, Clouin C, Taylor SS, Yaffe MB, Medema RH: **Polo-like kinase-1 is activated by aurora A to promote checkpoint recovery**. *Nature* 2008, **455**:119-123.

80. Turing AM: **The chemical basis of morphogenesis. 1953**. *Bull Math Biol* 1990, **52**:153-197 [discussion p. 119–152].

81. Grasse PP: **La reconstruction du nid et les coordinations inter-individuelles chez Bellicositermes natalensis et Cubitermes sp. La theorie de la stigmergie: Essai d'interpretation du comportement des termites constructeurs**. *Insectes Sociaux* 1959, **6**:41-81.

82. Witten TA, Sander LM: **Diffusion-limited aggregation, a kinetic critical phenomenon**. *Phys Rev Lett* 1981, **47**:1400-1403.

References only contained in figures

Megraw TL, Li K, Kao LR, Kaufman TC: The centrosomin protein is required for centrosome assembly and function during cleavage in Drosophila. Development 1999, 126:2829-2839.

O'Connell KF, Maxwell KN, White JG: The spd-2 gene is required for polarization of the anteroposterior axis and formation of the sperm asters in the Caenorhabditis elegans zygote. Dev Biol 2000, 222:55-70.

Hannak E, Kirkham M, Hyman AA, Oegema K: Aurora-A kinase is required for centrosome maturation in Caenorhabditis elegans. J Cell Biol 2001, 155:1109-1116.

Zimmerman WC, Sillibourne J, Rosa J, Doxsey SJ: Mitosis-specific anchoring of gamma tubulin complexes by pericentrin controls spindle organization and mitotic entry. Mol Biol Cell 2004, 15:3642-3657.

Sunkel CE, Glover DM: polo, a mitotic mutant of Drosophila displaying abnormal spindle poles. J Cell Sci 1988, 89 (Pt 1):25-38.

Ge X, Frank CL, Calderon de Anda F, Tsai LH: Hook3 interacts with PCM1 to regulate pericentriolar material assembly and the timing of neurogenesis. Neuron 2010, 65:191-203.

Khodjakov A, Rieder CL: The sudden recruitment of gamma-tubulin to the centrosome at the onset of mitosis and its dynamic exchange throughout the cell cycle, do not require microtubules. J Cell Biol 1999, 146:585-596.

Leidel S, Gonczy P: SAS-4 is essential for centrosome duplication in C elegans and is recruited to daughter centrioles once per cell cycle. Dev Cell 2003, 4:431-439.

Witten TA, Sander LM: Diffusion-Limited Aggregation, a Kinetic Critical Phenomenon. Physical Review Letters 1981, 47:1400-1403.