

Spatial Organization of Microtubule-Organizing Centers and Microtubules

JOHN B. TUCKER

Department of Zoology, St. Andrews University, St. Andrews, Fife KY16 9TS, United Kingdom

Do microtubule-organizing centers (MTOCs)¹ really organize microtubules? How is MTOC action integrated with other procedures for spatial control of microtubules? These are crucial questions so far as further progress in understanding the organization of the cytoplasmic matrix and the integration of cellular function are concerned. For example, it has been pointed out that microtubules may act as the overall organizers of the cytoskeleton by providing temporary scaffolds to organize the other components of the cytoplasm. It has also been suggested that the pericentriolar MTOC of an animal tissue cell might be considered to be the "command post" of such a cell. Furthermore, cytoskeletal organization can be transmitted across cell-surface membranes between adjacent tissue cells (see reference 1, pp. 600–604). Hence, MTOCs may have some impact on the organization of tissues as well as that of individual cells (54). Current indications of the extent to which the structural organization of MTOCs influences the spatial organization of microtubules are considered below.

What Is a Microtubule-Organizing Center?

Most of a eukaryotic cell's microtubules usually grow out from one or more special sites so that initiation of microtubule assembly is often largely confined to a few discrete intracellular localities (6, 53). These sites have been described as microtubule-initiating sites (32), microtubule-nucleating sites (45), microtubule centers (31), microtubule-nucleating centers (44) and microtubule-organizing centers (30). Microtubule-organizing center has proved irresistible to many investigators and has been widely adopted as a general term for sites where the nucleation of microtubule assembly apparently takes place. However, the extent to which MTOCs include spatial instructions for organizing microtubule arrays is only just beginning to emerge.

MTOCs vary considerably in appearance and location (Figs. 1–4). They seem to exhibit a diversity of overall organization that sets them apart from other "ubiquitous cell organelles." In many cases, MTOCs consist of concentrations of dense

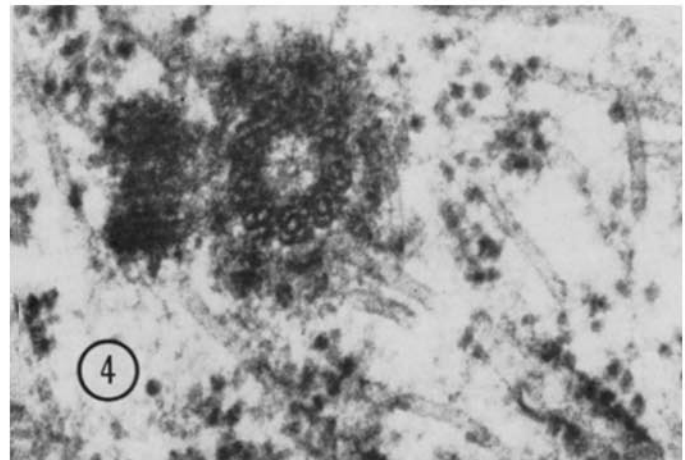
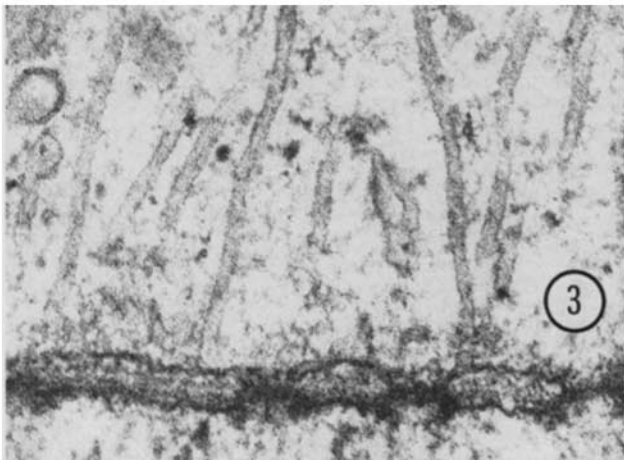
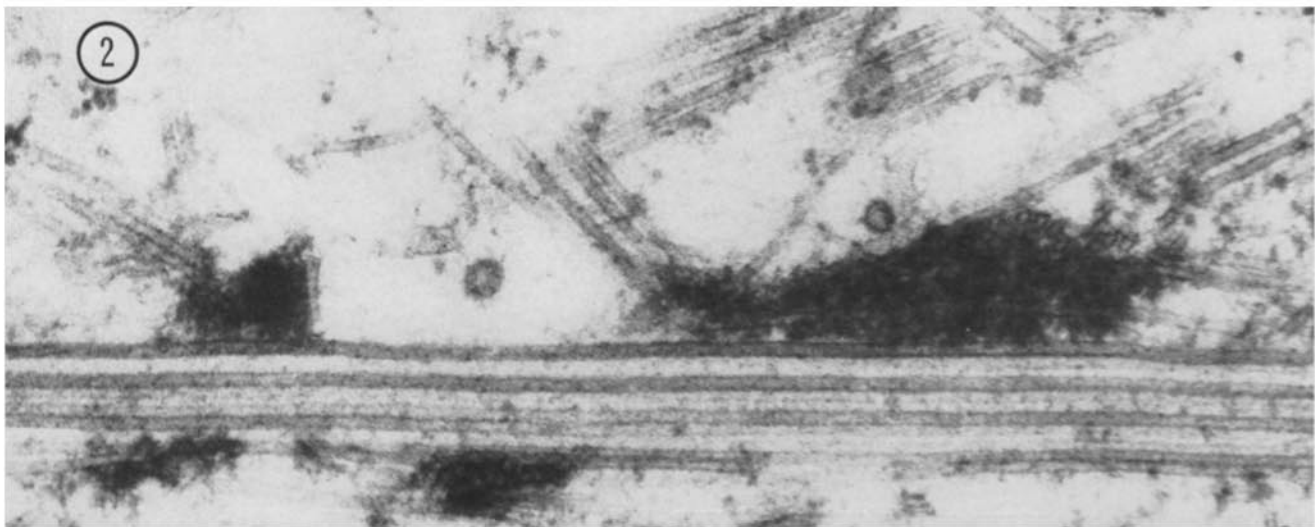
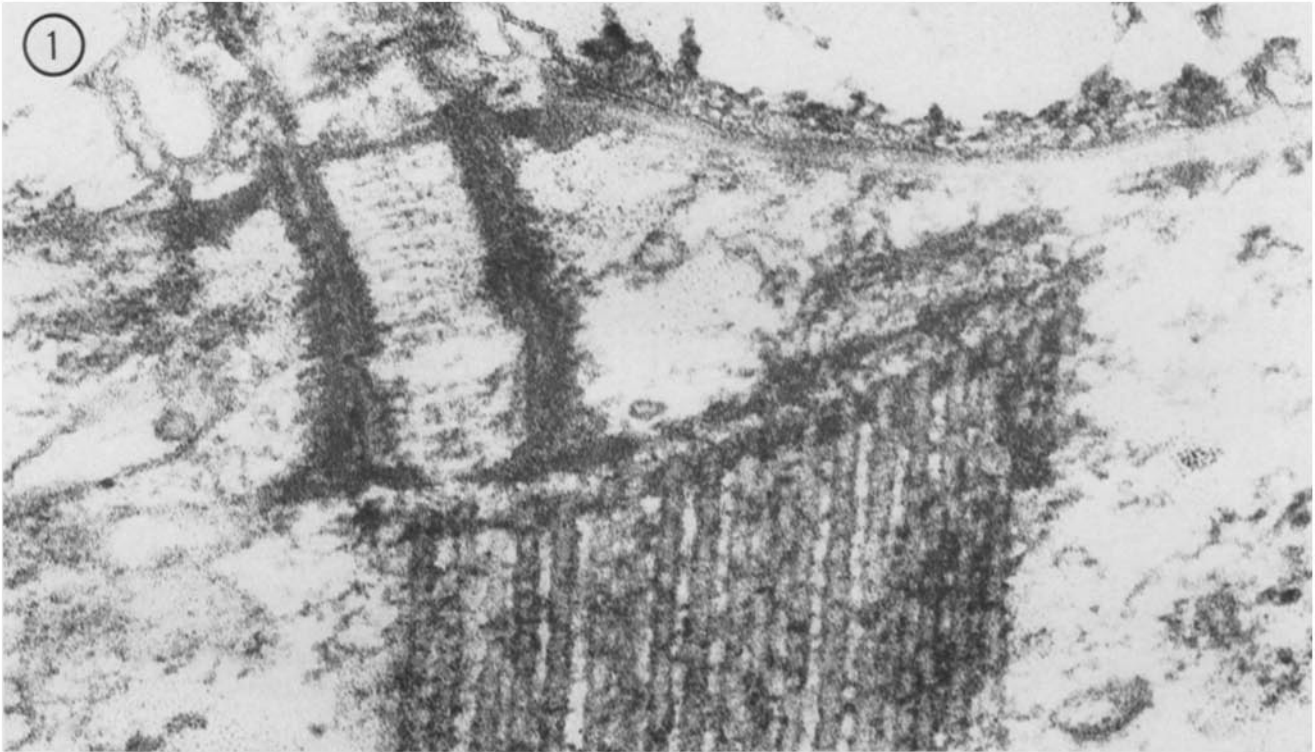
material. This material can occur, for example, as irregularly shaped clumps (Fig. 2), groupings of more or less finely divided particles (29, 40), ring-shaped concentrations of an apparently fibrous nature (37), or compact, flat laminated plaques (Fig. 1), depending on the particular MTOC in question. In certain situations, these materials are attached to, or positioned closely against, other cell components such as centrioles (Fig. 4), basal bodies (Fig. 1), microtrabeculae (33), nuclear envelopes (Fig. 3), and the cell-surface membrane (17).

How "Organized" Are Microtubules?

Very exact spatial and numerical precision are achieved during the assembly of certain microtubule arrays. A particularly clear example of this is provided by the microtubule arrays in an organism that is generally regarded as a relatively simple and primitive eukaryote, namely, the phytoflagellate *Chlamydomonas reinhardtii* (Fig. 5). In addition to flagellar axonemes with a 9 + 2 microtubule configuration and basal bodies, which each have nine microtubule triplets, there are four flagellar rootlets. The rootlets emanate from the vicinity of the organism's two flagellated basal bodies, where nucleation of rootlet microtubule assembly probably takes place. Two of the rootlets each include two microtubules. The other two rootlets each possess four microtubules that have a 3 over 1 arrangement near the basal bodies. Furthermore, the rootlets project from the basal body region at well-defined angles with respect to each other (4-50°-2-130°-4-50°-2). Flagellar microtubules and rootlet microtubules are differentiated from each other. The dynein-associated microtubules of the flagellar axonemes are involved in active bending movements, while the rootlet microtubules seem to be concerned with defining the positions of certain surface structures in the cell body (26). In addition, there are substantial indications that most of the α -tubulins in the rootlet microtubules are different from those in the flagellar microtubules (25).

The situation in *Chlamydomonas* highlights two important aspects of the control of microtubule organization. Spatial control (microtubule number, packing, location, orientation, polarity) and control of microtubule differentiation (in terms of molecular composition).

¹ Abbreviations used in this paper: MAP, microtubule-associated protein; MTOC, microtubule-organizing center.



A Multinucleating Element Hypothesis for Control of Microtubule Differentiation

Some microtubule arrays are made up of exactly specified numbers of microtubules. For example, there are 9 outer microtubule doublets and 2 central microtubules in cilia and flagella (Fig. 7), 9 microtubules in the haptonemata of certain phytoflagellates (23), 3, 6, 12, or 27 microtubules in the axopodial axonemes of certain heliozoans, depending on the species in question (4, 8), 12 rootlet microtubules in *Chlamydomonas* (Fig. 5), and 24 cortical microtubules in the sporozoites of *Eimeria* (37). Such specifications are probably achieved because the MTOCs that nucleate assembly of these arrays each apparently possess a precisely specified number of components called microtubule-nucleating elements (52). The suggestion is that each nucleating element nucleates the assembly of one microtubule (Fig. 6). There are indications that certain MTOCs include RNA and/or certain proteins that are involved during the nucleation of microtubule assembly (6). However, the molecular nature of nucleating elements remains to be elucidated.

The multinucleating element hypothesis (55, 56) predicts that there are several different types of nucleating elements. It is proposed that each type nucleates microtubule assembly using a different selection of tubulins and microtubule-associated proteins (MAPs). Thereafter, further microtubule assembly and elongation might proceed only by using the same combination of tubulins and MAPs as those that participated in the initial nucleation step, provided no marked changes in the cytoplasmic microenvironment occur in the immediate vicinity of an elongating microtubule. Thus, a MTOC might determine which type, or types, of microtubule grows out from its surface under a particular set of conditions. In addition, a nucleating element might determine how many protofilaments are included in the wall of the microtubule it nucleates. Most microtubules are composed of 13 protofilaments. Widespread exceptions to this are the "incomplete" B and C tubules of cilia, flagella, basal bodies, and centrioles (12). However, "complete" microtubules with more, or fewer, than 13 protofilaments have been reported for certain microtubule arrays in representatives of four animal phyla, including a mammal (11, 12, 38). There are indications from studies of *in vitro* microtubule assembly that control of protofilament number resides in MTOCs (39).

A Template Hypothesis for Spatial Control

The dense material of MTOCs usually appears to have an amorphous composition when examined using electron mi-

croscopy (Figs. 1, 2, and 4). This is most disappointing. It reveals little about the molecular organization of MTOCs. Nevertheless, the dense MTOC material may play a crucial role if it ever represents a compact and highly ordered fabric that binds a particular number of nucleating elements together

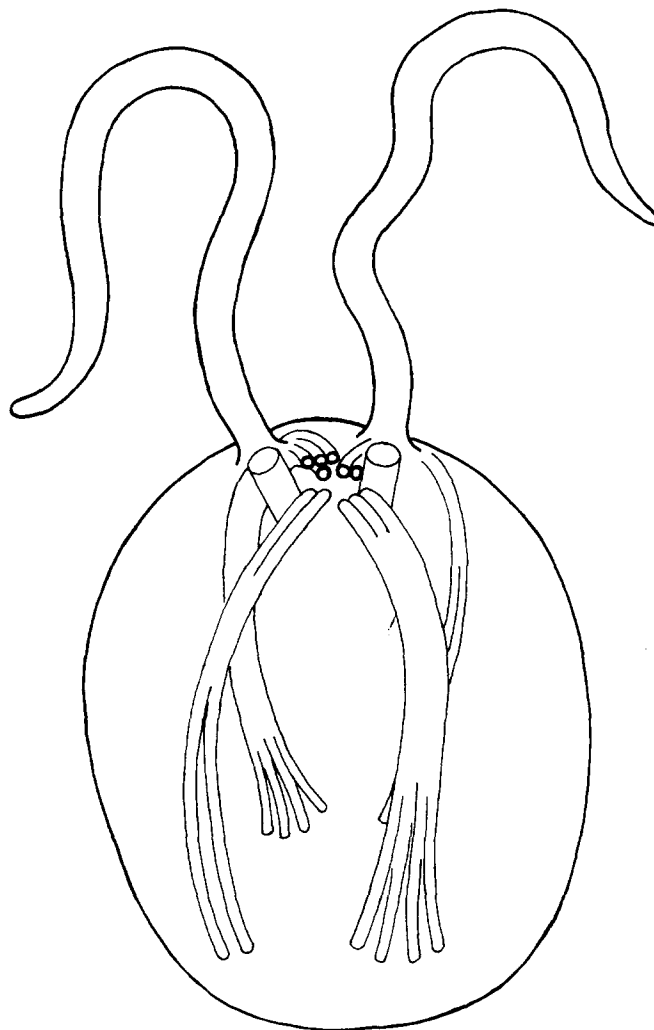


FIGURE 5 Schematic diagram showing the positioning of the 4-2-4-2 microtubular rootlet array with respect to the two flagella and their basal bodies in *Chlamydomonas reinhardtii*. Two immature basal bodies and striated fibers that are included in this structural complex have been omitted for clarity. The exact arrangement of rootlets in the posterior portion of the cell is uncertain. Based on the micrographs of Ringo (35) and Goodenough and Weiss (14).

FIGURES 1-4 Examples of variation in the form and spatial organization of MTOCs. (1) Longitudinal section through the proximal portion of a developing cytopharyngeal microtubule bundle in the ciliate *Nassula*. Well-aligned and evenly spaced microtubules grow out from a compact, flat laminated MTOC attached to the proximal portion of a basal body. Reproduced from reference 48. $\times 130,000$. (2) Longitudinal section through the basal portion of a developing axopodial axoneme in the heliozoan *Echinospaerium nucleofilum*. The MTOC is an irregularly shaped clump of dense material. Initially, microtubules extend out from the MTOC in an apparently random variety of orientations. Subsequently they become aligned and regularly spaced with respect to each other; this has taken place so far as some of the microtubules shown are concerned. Reproduced from reference 18. $\times 85,000$. (3) Section through the nuclear envelope of the heliozoan *Actinophrys sol* during the assembly of new axopodial axonemes. Microtubules extend out in a radial array from the surface of the organism's centrally positioned nucleus. They then become more closely packed and aligned as the morphogenesis of discrete axonemes proceeds. Reproduced from reference 18. $\times 90,000$. (4) Section through the pericentriolar MTOC of an epidermal cell in the developing leg of the dipteran *Calliphora erythrocephala*. Microtubules project out from dense pericentriolar material that is closely applied to the sides of centrioles. $\times 95,000$.

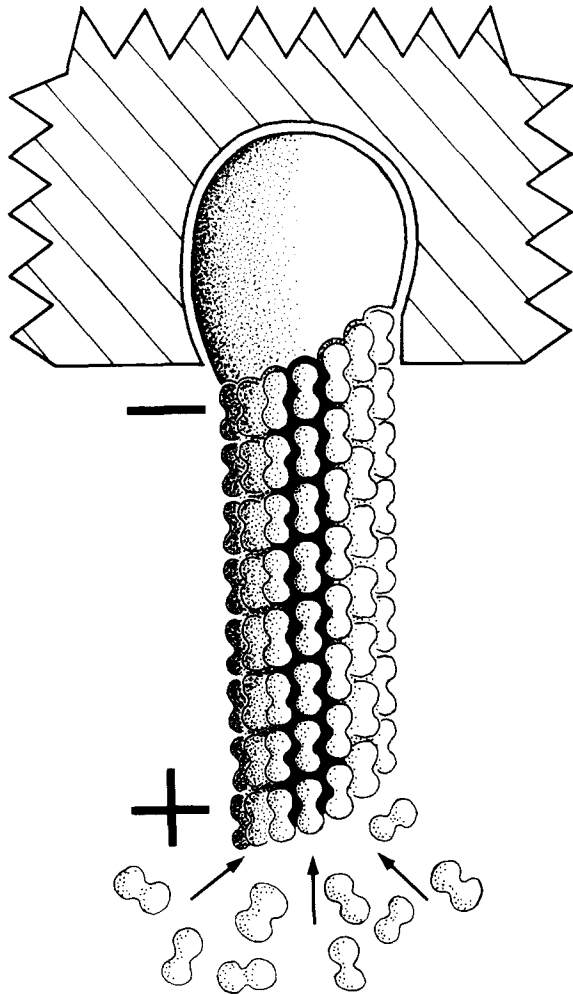


FIGURE 6 Schematic diagram showing the arrangement of tubulins and protofilaments in the first-formed portion of a microtubule shortly after it starts to assemble. Microtubule elongation proceeds as a consequence of the addition of tubulin heterodimers to the tips of protofilaments at the plus end of the microtubule. The nucleating element that initiated this assembly sequence caps the minus end of the microtubule and is embedded in some of the dense material (crosshatched) of the MTOC. The bilobular topography of the outer surfaces of the tubulins probably reveals the positions of the α - and β -tubulins in each heterodimer. Modified from Tucker (56).

in a regularly spaced pattern and anchors them firmly at particular orientations. In such situations, MTOCs would possess a considerable capacity for organizing microtubule arrangement. They could establish microtubule number, spacing, orientation, and the pattern of microtubule packing, as well as ensuring that adjacent microtubules have the same polarity.

Certain compact MTOCs do seem to act as templates by providing quite detailed spatial instructions in the form of firmly bound, well-oriented, patterned arrays of nucleating elements. For example, when cytopharyngeal microtubules start to grow out from a laminated MTOC (Fig. 1) in the ciliate *Nassula* they are precisely spaced out and well aligned with each other as they start to assemble. Analyses of perturbations induced in the pattern of microtubule packing by heat shocks (49) and exposure to colchicine (57) support the suggestion that the MTOC acts as a microtubule-nucleating template (28). New microtubules are also positioned in a highly ordered fashion when the assembly of basal bodies and centrioles is initiated (Fig. 7). The microtubules extend out from a ring or circular plate of dense material in some instances (3, 9, 19). This material seems to represent the MTOC and to act as a template—rather than the central “cartwheel,” which is not present during the assembly of certain centrioles (10).

Decentralized Generation of Order

It has been established that some MTOCs do not act as templates; they do not precisely define the spacing and orientation of the microtubules they nucleate. A striking example of this occurs during the assembly of the 12-sectored, double-spiral microtubule array (Fig. 8) found in certain heliozoan axonemes. Axonemal microtubules are not aligned or regularly spaced when they start to assemble (Figs. 2 and 3), in spite of the extremely high degree of order that is finally achieved. Hence, the MTOCs in question do not act as templates. Pattern is apparently generated by a self-linkage procedure when intertubule links connect microtubules together (18, 44).

The microtubules that grow out from some MTOCs (including some of those that act as templates) are linearly differentiated (55, 56). The microtubules bear different types and arrangements of links along particular portions of their

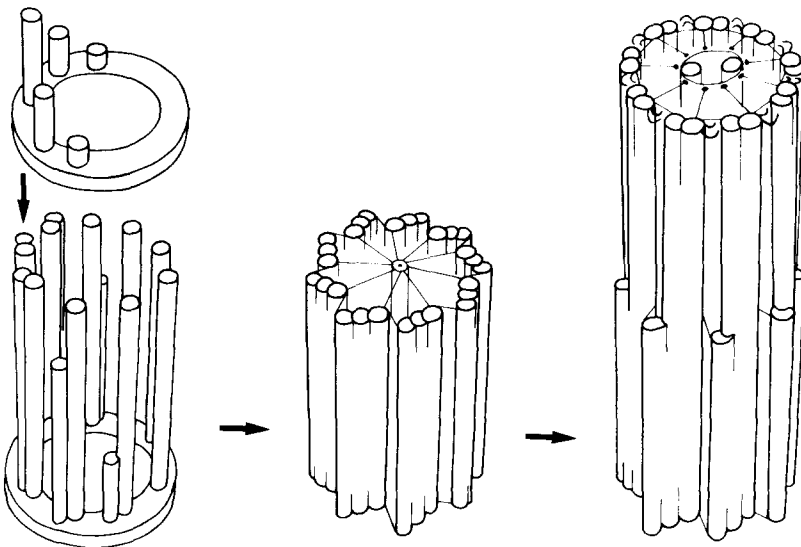


FIGURE 7 Diagrammatic representation of the assembly sequence for centrioles and basal bodies, culminating (to the right of the illustration) in the formation of a 9 + 2 axoneme (only the basal portion of a mature axoneme is included). The central cartwheel is shown extending to the distal end of the centriole to reveal its configuration, but in most cases it is usually confined to the proximal portion of a centriole or basal body.

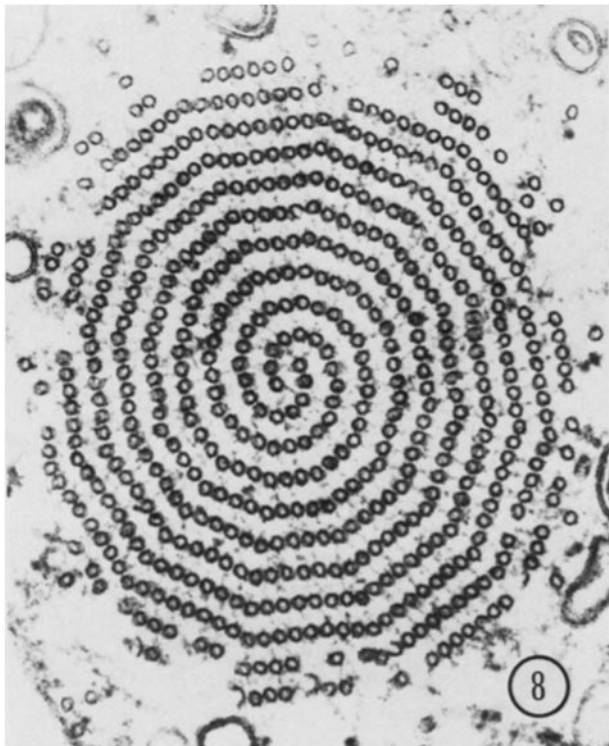


FIGURE 8 Cross section through the axopodial axoneme of a heliozoan that exhibits the complex 12-sectored, double-spiral pattern of microtubule packing. Previously unpublished micrograph courtesy of Dr. J. C. R. Jones. $\times 80,000$.

lengths. This is especially evident, for example, in the tentacular axonemes and cytopharyngeal microtubule bundles of the ciliates *Tokophrya* and *Nassula*, respectively (47, 51, 58). Linear microtubule differentiation is also displayed during ciliogenesis and flagellogenesis (Fig. 7). The nine microtubule doublets of a ciliary or flagellar axoneme result from further assembly and elongation of the A and B microtubules of each microtubule triplet in a basal body. Axonemal doublets are interconnected in a very different way than they are at levels where they form a part of basal body triplets.

Linear microtubule differentiation and self-linkage are two aspects of microtubule organization that presumably are not under *direct* MTOC jurisdiction. MTOCs might, however, at least in theory, define which particular modes of self-linkage and linear differentiation microtubules are potentially capable of adopting in response to interactions that impinge upon them as they elongate. This could be because of control over microtubule composition imparted *via* the particular nucleating elements that MTOCs provide.

Structural Organization of MTOCs

Virtually identical double-spiral patterns of axonemal microtubule packing (Fig. 8) are generated by the heliozoans *Echinospaerium nucleofilum* and *Actinophrys sol*. However, the axonemal MTOCs of these two organisms are very different in structure and appearance: dense clumps (Fig. 2) of juxtannuclear material in multinucleate *E. nucleofilum* and the outer surface of the nuclear envelope (Fig. 3) in mononucleate *A. sol* (18). Hence, the overall form of an MTOC is not always directly related to the spatial organization of the microtubule array it initiates. This conclusion is pertinent to a point raised

earlier, namely, the extensive range of variation in the form of MTOCs in cells generally (Figs. 1–4).

The form of MTOCs may vary because most, perhaps all, of the dense MTOC material detected by electron microscopy represents material that is involved in anchoring nucleating elements rather than the nucleating elements themselves. In cases in which MTOCs do not act as templates, the shape and disposition of the dense anchoring material may be largely defined by whether there is a requirement for the MTOC to be situated in a particular cell locality and by how the MTOC is attached to other cell components (plasmalemma, microtrabeculae, nucleus, centrioles, etc.) in order to achieve this. For example, there is no readily detectable dense MTOC material associated with the nuclear envelope of *A. sol* (Fig. 3). The nuclear envelope is structurally more substantial than nuclear envelopes in general (27). It apparently suffices as a sturdy substrate for the attachment of nucleating elements, and this may account for the lack of much additional dense MTOC anchoring material.

Pericentriolar MTOCs of Metazoan Tissue Cells: Organizers or Initiators?

Most types of metazoan tissue cells possess one main MTOC, a juxtannuclear pericentriolar MTOC. As a consequence, this MTOC is of special interest. Nucleating elements are apparently included in dense material that usually surrounds a pair of centrioles situated at right angles to each other (Fig. 4) (16, 29). The “pericentriolar cloud” often appears to consist of rather diffuse and irregular groupings of granules and/or clumps of dense material (24). In some cases, reasonably discrete dense bodies called satellite bodies are present, and densely staining basal feet are attached in a fairly

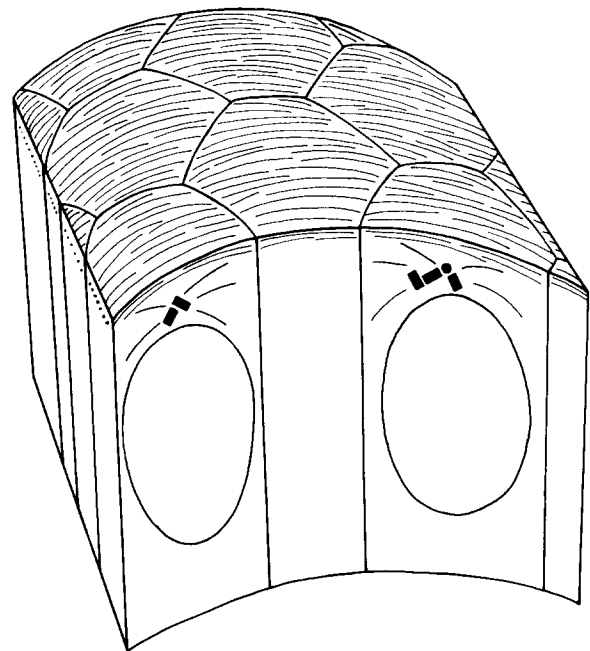


FIGURE 9 Schematic diagram showing a portion of the tubeshaped epidermis of a developing blowfly leg tarsomere. In at least some of the cells the centrioles (blocked in black) have replicated so that the pericentriolar MTOC surrounds four centrioles. Centrioles are situated near the cell apices, above nuclei, and close to the level of a subsurface layer of aligned microtubules (thin lines). These microtubules are oriented at right angles to the longitudinal axis of the developing leg.

regular fashion around certain centrioles (2). Pericentriolar MTOCs seem to play an important part in the temporal control of microtubule organization by initiating the assembly of fresh sets of microtubules at different points in the cell cycle and in response to changes in certain physiological conditions (21, 41–43). Current failure to detect and/or preserve order in the arrangement of the pericentriolar materials in most cell types does not encourage confidence in the additional possibility of detailed control of microtubule distribution by pericentriolar MTOCs (5), but it is too early to dismiss it entirely. For example, there have been demonstrations that the locations of pericentriolar MTOCs are correlated in some instances with cell polarity so far as cell migration and certain other polarized and cytoskeletally mediated functions are concerned (13, 15, 20, 22, 36).

Recent examinations of supracellular microtubule alignment in certain tissues indicate that the alignments in question are mainly controlled by interactions between microtubules and other components of the cytoplasmic matrix that take place outside of the immediate surroundings of the pericentriolar MTOCs and that perhaps include cell-surface interac-

tions between cell neighbors (54). For example, much of the developing leg of the blowfly *Calliphora* is basically a tube-shaped epidermal sheet. At one stage in leg morphogenesis large numbers of aligned microtubules are situated just beneath the outermost surface of the sheet. They are nearly all oriented so that they run at right angles to the longitudinal axis of the elongating leg. In each cell the pericentriolar MTOC is situated slightly below the aligned microtubule array (Fig. 9). The ends of microtubules are not concentrated around the pericentriolar MTOC to the extent that would be anticipated if most of the aligned microtubules were connected to the MTOC (cf. Figs. 10 and 11). This is also mainly the case during earlier stages of leg morphogenesis when subsurface microtubules are not well aligned, but in a few instances some microtubules project from the pericentriolar MTOCs (Fig. 4). It seems that the microtubules in question either move away from pericentriolar MTOCs after their assembly has been nucleated or that the assembly of most of these microtubules is nucleated elsewhere. Whichever situation obtains, control of microtubule alignment is apparently effected by activities that are not directly associated with the

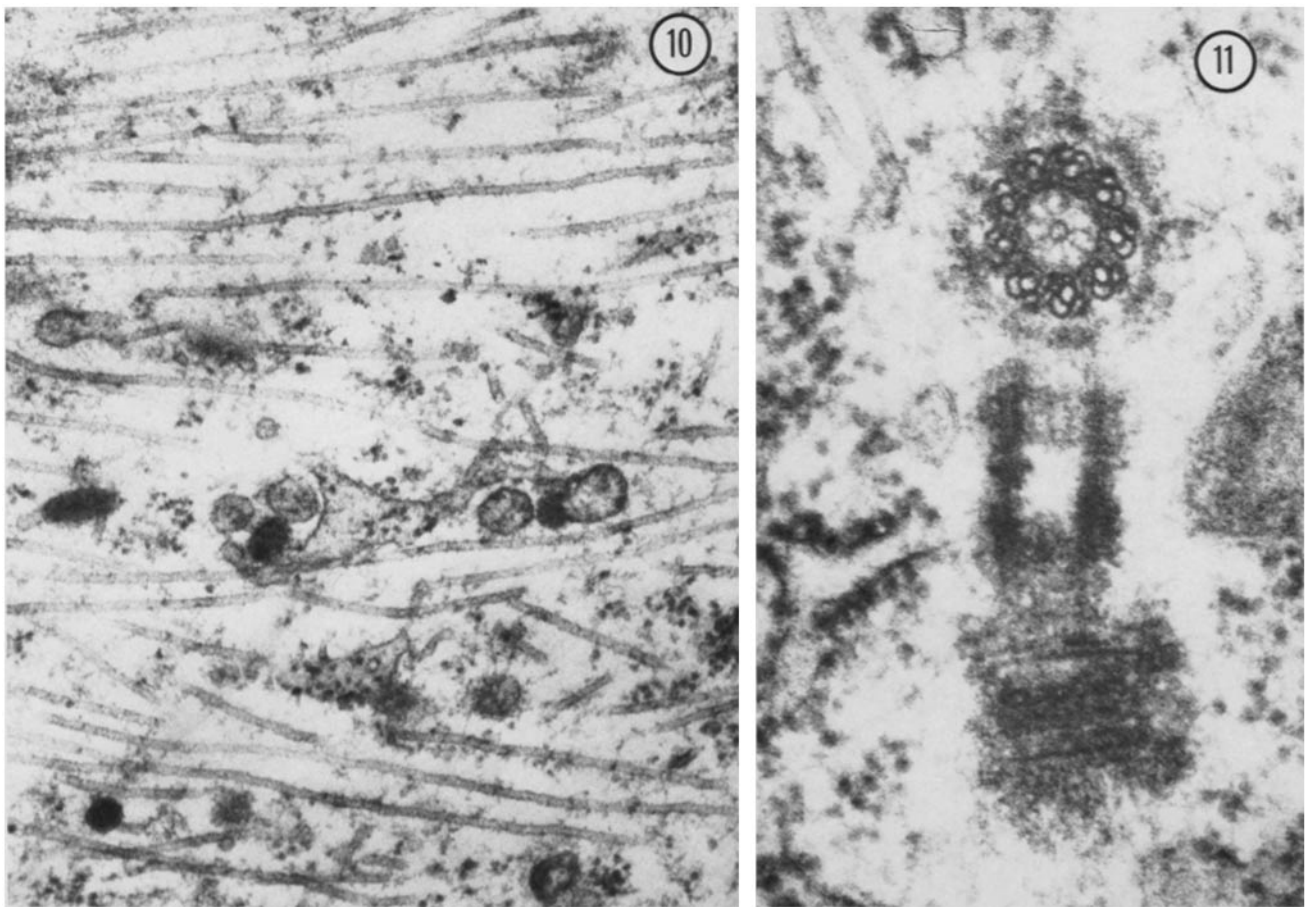


FIGURE 10 Section grazing the apical surface of part of a tarsomeric epidermal cell in a developing leg of the blowfly *Calliphora erythrocephala* at the level of the aligned layer of microtubules (cf. Fig. 9). $\times 54,000$.

FIGURE 11 Section through three of a tarsomeric epidermal cell's four centrioles at the stage in leg development at which the layer of aligned microtubules is present (Fig. 9). Centrioles are positioned in the classical orthogonal arrangement for mother-daughter centriole pairs often found in metazoan tissue cells. Few microtubules are located in the vicinity of the centrioles, especially when comparison is made with the spatial concentration of microtubules in the subsurface layer (Fig. 10). Clumps of densely staining material grouped in a fairly regular fashion around the centrioles are particularly evident for the transversely sectioned centriole. The centrioles are somewhat atypical insofar as the microtubules of each of their nine triplets are grouped in a cloverleaf arrangement rather than forming a straight row (cf. Fig. 7). Previously unpublished micrograph courtesy of Sandra Anderson. $\times 103,000$.

pericentriolar MTOCs. This is also probably the case during microtubule alignment in elongating myoblasts (59) and goldfish scale scleroblasts (7). In the scleroblasts, microtubules radiate out from a region near the cell center, where the pericentriolar MTOC is presumably located. Certain microtubules turn abruptly into alignment with each other (and those in neighboring cells) at points 5–20 μm from the cell center.

It is important to realize that studies of highly ordered microtubule arrays have undeniably revealed a striking combination of versatility and spatial sophistication for effecting microtubule-connection to adjacent structures (Fig. 7) (50, 52, 53). It is reasonable to suppose that microtubules in general may have a potential that is equivalent to that of highly ordered arrays for effecting spatially detailed structural interactions. Such interactions may be essential during the “decentralized organization” of microtubules that have been initiated by pericentriolar MTOCs.

MTOCs—Current Status

Microtubules interact with a wide range of cytoplasmic components (1, 53), including microfilaments, intermediate filaments, and microtrabeculae. However, the extent to which microtubules organize other cytoskeletal components, compared with the degree to which microtubules are themselves organized by interactions with such components, remains to be resolved (54). Hence, the possibility that MTOCs represent command posts for controlling the organization of the cytoplasmic matrix and the integration of cellular function is still uncertain. MTOCs may only be valuable subordinates, albeit essential, in the cytoplasmic command hierarchy. Their importance definitely depends on how extensively they organize microtubules.

Some MTOCs apparently make a direct structural contribution to the spatial organization of the microtubules they nucleate, but others do not seem to do so. Furthermore, interactions between microtubules and other components of the cytoplasmic matrix at points distant from MTOCs are essential during the organization of many microtubule arrays. Is it sensible to retain the term microtubule-organizing center for all sites at which microtubule assembly is initiated? There is a case for continuing to do so, at least until more is known about MTOC function and composition. For example, even where MTOCs seem to be “initiators” rather than “spatial organizers,” they could nevertheless exert considerable control over a cell’s microtubule layout. They may still be “organizing centers” in the sense that they specify when, how many, and what type during the initiation of microtubule assembly. Such specifications could have considerable impact on the subsequent spatial organization of microtubules (induced in response to situations and conditions encountered as microtubules grow, or migrate, out from MTOCs).

Finally, there is the question of whether MTOCs can be usefully defined as sites at which microtubule assembly is nucleated *in vivo*? This question is complicated by indications that there are certain components of “organellar size,” such as some kinetochores, which apparently help to organize microtubules but which may not nucleate their assembly *in vivo* (34, 46). Such components are not included in the definition of an MTOC given above. If one takes the view that the term MTOC should be employed for all components that bind to microtubules and contribute to the spatial orga-

nization of microtubules, then the term becomes unhelpfully indiscriminate. These are points for investigators to bear in mind when discussing their various centers and sites of microtubule organization and nucleation.

REFERENCES

1. Alberts, B., D. Bray, J. Lewis, M. Raff, K. Roberts, and J. D. Watson. 1983. *Molecular Biology of the Cell*. Garland Publishing, New York.
2. Albrecht-Buehler, G., and A. Bushnell. 1980. The ultrastructure of primary cilia in quiescent 3T3 cells. *Exp. Cell Res.* 126:427–437.
3. Anderson, R. G. W., and R. M. Brenner. 1971. The formation of basal bodies (centrioles) in the rhesus monkey oviduct. *J. Cell Biol.* 50:10–34.
4. Bardele, C. F. 1977. Organization and control of microtubule pattern in centrohelid heliozoa. *J. Protozool.* 24:9–14.
5. Brinkley, B. R., S. M. Cox, D. A. Pepper, L. Wible, S. L. Brenner, and R. L. Pardue. 1981. Tubulin assembly sites and the organization of cytoplasmic microtubules in cultured mammalian cells. *J. Cell Biol.* 90:554–562.
6. Brown, D. L., M. E. Stearns, and T. H. Macrae. 1982. Microtubule organizing centres. In *The Cytoskeleton in Plant Growth and Development*. C. W. Lloyd, editor. Academic Press, London. 55–83.
7. Byers, H. R., K. Fujiwara, and K. R. Porter. 1980. Visualization of microtubules in cells *in situ* by indirect immunofluorescence. *Proc. Natl. Acad. Sci. USA.* 77:6657–6661.
8. Davidson, L. A. 1982. Ultrastructure, behavior, and algal flagellate affinities of the helioflagellate *Ciliophrys marina*, and the classification of the helioflagellates (Protista, Actinopoda, Heliozoa). *J. Protozool.* 29:19–29.
9. Dippell, R. V. 1968. The development of basal bodies in *Paramecium*. *Proc. Natl. Acad. Sci. USA.* 61:461–468.
10. Dirksen, E. R. 1971. Centriole morphogenesis in developing ciliated epithelium of the mouse oviduct. *J. Cell Biol.* 51:286–302.
11. Eichenlaub-Ritter, U., and J. B. Tucker. 1984. Microtubules with more than 13 protofilaments in the dividing nuclei of ciliates. *Nature (Lond.)*. 307:60–62.
12. Fujiwara, K., and R. W. Linck. 1982. The use of tannic acid in microtubule research. *Methods Cell Biol.* 25A:217–233.
13. Geiger, B., D. Rosen, and G. Berke. 1982. Spatial relationships of microtubule-organizing centers and the contact area of cytotoxic T lymphocytes and target cells. *J. Cell Biol.* 95:137–143.
14. Goodenough, U. W., and R. L. Weiss. 1978. Interrelationships between microtubules, a striated fiber, and the gametic mating structure of *Chlamydomonas reinhardtii*. *J. Cell Biol.* 76:430–438.
15. Gotlieb, A., L. M. May, L. Subrahmanyam, and V. I. Kalnins. 1981. Distribution of microtubule-organizing centers in migrating sheets of endothelial cells. *J. Cell Biol.* 91:589–594.
16. Gould, R. R., and G. G. Borisy. 1977. The pericentriolar material in Chinese hamster cells nucleates microtubule formation. *J. Cell Biol.* 73:601–615.
17. Gunning, B. E. S., A. R. Hardham, and J. E. Hughes. 1978. Evidence for initiation of microtubules in discrete regions of the cell cortex in *Azolla* root-tip cells, and an hypothesis on the development of cortical arrays of microtubules. *Planta (Berl.)*. 143:161–179.
18. Jones, J. C. R., and J. B. Tucker. 1981. Microtubule-organizing centres and assembly of the double-spiral microtubule pattern in certain heliozoan axonemes. *J. Cell Sci.* 50:259–280.
19. Kalnins, V. I., and K. R. Porter. 1969. Centriole replication during ciliogenesis in the chick tracheal epithelium. *Z. Zellforsch. Mikrosk. Anat.* 100:1–30.
20. Kupfer, A., D. Louvard, and S. J. Singer. 1982. Polarization of the Golgi apparatus and the microtubule-organizing center in cultured fibroblasts at the edge of an experimental wound. *Proc. Natl. Acad. Sci. USA.* 79:2579–2583.
21. Kuriyama, R., and G. G. Borisy. 1981. Microtubule-nucleating activity of centrosomes in Chinese hamster ovary cells is independent of the centriole cycle but coupled to the mitotic cycle. *J. Cell Biol.* 91:822–826.
22. Malech, H. L., R. K. Root, and J. I. Gallin. 1977. Structural analysis of human neutrophil migration. Centriole, microtubule, and microfilament orientation and function during chemotaxis. *J. Cell Biol.* 75:666–693.
23. Manton, I. 1968. Further observations on the microanatomy of the haptonema in *Chrysochromulina chiton* and *Prymnesium parvum*. *Protoplasma*. 66:35–53.
24. McIntosh, J. R. 1983. The centrosome as an organizer of the cytoskeleton. In *Spatial Organization of Eukaryotic Cells*. Modern Cell Biology, Vol. 2. J. R. McIntosh, editor. A. R. Liss Inc., New York. 115–142.
25. McKeithan, T. W., P. A. Lefebvre, C. D. Silflow, and J. L. Rosenbaum. 1983. Multiple forms of tubulin in *Polytomella* and *Chlamydomonas*: evidence for a precursor of flagellar α -tubulin. *J. Cell Biol.* 96:1056–1063.
26. Melkonian, M. 1982. The functional analysis of the flagellar apparatus in green algae. In *Prokaryotic and Eukaryotic Flagella*. W. B. Amos and J. G. Duckett, editors. Cambridge University Press, Cambridge. 589–606.
27. Ockelford, C. D., and J. B. Tucker. 1973. Growth, breakdown, repair and rapid contraction of microtubular axopodia in the heliozoan *Actinophrys sol*. *J. Ultrastruct. Res.* 44:369–387.
28. Pearson, P. J., and J. B. Tucker. 1977. Control of shape and pattern during the assembly of a large microtubule bundle. Evidence for a microtubule-nucleating-template. *Cell Tissue Res.* 180:241–252.
29. Peterson, S. P., and M. W. Berns. The centriolar complex. *Int. Rev. Cytol.* 64:81–106.
30. Pickett-Heaps, J. D. 1969. The evolution of the mitotic apparatus: an attempt at comparative ultrastructural cytology in dividing plant cells. *Cytobios.* 3:257–280.
31. Pickett-Heaps, J. D., and S. E. Kowalski. 1981. Valve morphogenesis and the microtubule center of the diatom *Hantzschia amphioxys*. *Eur. J. Cell Biol.* 25:150–170.
32. Porter, K. R. 1966. Cytoplasmic microtubules and their functions. In *Principles of Biomolecular Organization*. G. E. W. Wolstenholme and M. O'Connor, editors. J. and A. Churchill Ltd., London. 308–345.
33. Porter, K. R., and J. B. Tucker. 1981. The ground substance of the living cell. *Sci. Am.* 244:40–51.
34. Rieder, C. L. 1982. The formation, structure, and composition of the mammalian kinetochore and kinetochore fiber. *Int. Rev. Cytol.* 79:1–58.
35. Ringo, D. L. 1967. Flagellar motion and fine structures of the flagellar apparatus in *Chlamydomonas*. *J. Cell Biol.* 33:543–571.

36. Rogers, K. A., M. A. Khoshbaf, and D. L. Brown. 1981. Relationships of microtubule organization in lymphocytes to the capping of immunoglobulin. *Eur. J. Cell Biol.* 24:1-8.
37. Russell, D. G., and R. G. Burns. 1984. The polar ring of coccidian sporozoites: a unique microtubule-organising centre. *J. Cell Sci.* 65:193-207.
38. Saito, K., and K. Hama. 1982. Structural diversity of microtubules in the supporting cells of the sensory epithelium of the guinea pig organ of Corti. *J. Electron Microsc.* 31:278-281.
39. Scheele, R. B., L. G. Bergen, and G. G. Borisy. 1982. Control of the structural fidelity of microtubules by initiation sites. *J. Mol. Biol.* 154:485-500.
40. Schliwa, M. 1978. Microtubular apparatus of melanophores. Three-dimensional organization. *J. Cell Biol.* 76:605-614.
41. Schliwa, M., U. Euteneuer, W. Herzog, and K. Weber. 1979. Evidence for rapid structural and functional changes of the melanophore microtubule-organizing center upon pigment movements. *J. Cell Biol.* 83:623-632.
42. Snyder, J. A., and J. R. McIntosh. 1975. Initiation and growth of microtubules from mitotic centers in lysed mammalian cells. *J. Cell Biol.* 67:744-760.
43. Telzer, B. R., and J. L. Rosenbaum. 1979. Cell cycle-dependent, *in vitro* assembly of microtubules on the pericentriolar material of HeLa cells. *J. Cell Biol.* 81:484-497.
44. Tilney, L. G. 1971. Origin and continuity of microtubules. In *Origin and Continuity of Cell Organelles*. J. Reinert and H. Ursprung, editors. Springer-Verlag, Berlin, 222-260.
45. Tilney, L. G., and J. Goddard. 1970. Nucleating sites for the assembly of microtubules in the ectodermal cells of blastulae of *Arbacia punctulata*. *J. Cell Biol.* 46:564-575.
46. Tippit, D. H., J. D. Pickett-Heaps, and R. Leslie. 1980. Cell division in two large pennate diatoms *Hantzschia* and *Nitzschia*. III. A new proposal for kinetochore function during prometaphase. *J. Cell Biol.* 86:402-416.
47. Tucker, J. B. 1968. Fine structure and function of the cytopharyngeal basket in the ciliate *Nassula*. *J. Cell Sci.* 3:493-514.
48. Tucker, J. B. 1970. Morphogenesis of a large microtubular organelle and its association with basal bodies in the ciliate *Nassula*. *J. Cell Sci.* 6:385-429.
49. Tucker, J. B. 1970. Initiation and differentiation of microtubule patterns in the ciliate *Nassula*. *J. Cell Sci.* 7:793-821.
50. Tucker, J. B. 1971. Spatial discrimination in the cytoplasm during microtubule morphogenesis. *Nature (Lond.)* 232:387-389.
51. Tucker, J. B. 1974. Microtubule arms and cytoplasmic streaming and microtubule bending and stretching of intertubule links in the feeding tentacle of the suctorian ciliate *Tokophrya*. *J. Cell Biol.* 62:424-437.
52. Tucker, J. B. 1977. Shape and pattern specification during microtubule bundle assembly. *Nature (Lond.)* 266:22-26.
53. Tucker, J. B. 1979. Spatial organization of microtubules. In *Microtubules*. K. Roberts and J. S. Hyams, editors. Academic Press, London, 315-357.
54. Tucker, J. B. 1981. Cytoskeletal coordination and intercellular signalling during metazoan embryogenesis. *J. Embryol. Exp. Morphol.* 65:1-25.
55. Tucker, J. B. 1982. Microtubule-organizing centres and assembly of intricate microtubule arrays in protozoans. In *Microtubules in Micro-Organisms*. P. Cappuccinelli and N. R. Morris, editors. Marcel Dekker, Inc., New York, 351-375.
56. Tucker, J. B. 1982. Microtubule-organizing centres and microtubule deployment in Protozoa. *Acta Protozoologica Congress Volume I, Progress in Protozoology*, 45-57.
57. Tucker, J. B., M. Dunn, and J. B. Patisson. 1975. Control of microtubule pattern during the development of a large organelle in the ciliate *Nassula*. *Dev. Biol.* 47:439-453.
58. Tucker, J. B., and J. B. Mackie. 1975. Configurational changes in helical microtubule frameworks in feeding tentacles of the suctorian ciliate *Tokophrya*. *Tissue Cell* 7:601-612.
59. Warren, R. H. 1974. Microtubular organization in elongating myogenic cells. *J. Cell Biol.* 63:550-566.