ARCHENTERON ELONGATION IN THE SEA URCHIN EMBRYO IS A MICROTUBULE-INDEPENDENT PROCESS

JEFFREY D. HARDIN

Graduate Group in Biophysics and Medical Physics and Department of Zoology, University of California, Berkeley, California 94720

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Earlier studies using colchicine (L. G. Tilney and J. R. Gibbins, 1969, J. Cell Sci. 5, 195-210) had suggested that intact microtubules (MTs) are necessary for archenteron elongation during the second phase of sea urchin gastrulation (secondary invagination), presumably by allowing secondary mesenchyme cells (SMCs) to extend their long filopodial processes. In light of subsequently discovered effects of colchicine on other cellular processes, the role of MTs in archenteron elongation in the sea urchin, Lytechinus pictus, has been reexamined. Immunofluorescent staining of ectodermal fragments and isolated archenterons reveals a characteristic pattern of MTs in the ectoderm and endoderm during gastrulation. Ectodermal cells exhibit arrays of MTs radiating away from the region of the basal body/ciliary rootlet and extending along the periphery of the cell, whereas endodermal cells exhibit a similar array of peripheral MTs emanating from the region of the apical ciliary rootlet facing the lumen of the archenteron. MTs are found primarily at the bases of the filopodia of normal SMCs. β-Lumicolchicine (0.1 mM), an analog of colchicine which does not bind tubulin, inhibits secondary invagination, indicating that the effects previously ascribed to the disruption of MTs are probably due to the effects of colchicine on other cellular processes. The MT inhibitor nocodazole (5-10 µg/ml) added prior to secondary invagination does not prevent gastrulation or spontaneous exogastrulation, even though indirect immunofluorescence indicates that cytoplasmic MTs are completely disrupted in drug-treated embryos. Transverse tissue sections indicate that a comparable amount of cell rearrangement occurs in nocodazole-treated and control embryos. Significantly, SMCs in nocodazole-treated embryos often detach prematurely from the tip of the gut rudiment and extend abnormally large broad lamellipodial protrusions but are also capable of extending long slender filopodia comparable in length to those of control embryos. These results indicate that cytoplasmic MTs are not essential for either filopodial extension by SMCs or for the active epithelial cell rearrangement which accompanies archenteron elongation during sea urchin gastrulation.


INTRODUCTION

The sea urchin gastrula is a convenient model system for studying the roles of cell motility and changes in cell shape during embryonic development. Sea urchin gastrulation occurs in two stages (Dan and Okazaki, 1956). Primary invagination involves the inward bending of the vegetal plate to form a “table-topped” archenteron (Okazaki, 1975). During secondary invagination this invagination elongates to form a tube that is longer but smaller in diameter. Secondary invagination is accompanied by the rearrangement of epithelial cells in the wall of the archenteron (Ettensohn, 1985); this rearrangement actively lengthens the gut rudiment (Hardin and Cheng, 1986).

Although the basic morphogenetic movements of gastrulation have been described in increasing detail, it has proven more difficult to unravel exactly how the cytoskeleton functions in these movements. In particular, several studies have focused on the role of microtubules (MTs) in later stages of sea urchin development. In a pioneering study, Gibbins et al. (1969) used transmission electron microscopy to follow changes in MT structure during primary mesenchyme cell ingression, migration, and association to form the calcareous spicules of the pluteus larva. Tilney and Goddard (1970) analyzed the polymerization of MTs following their disruption in the ciliated ectoderm of Arbacia punctulata blastulae and described a radiating array of MTs originating near the basal body and extending along the periphery of these cells. Most important in the context of the present study, Tilney and Gibbins (1969) found that colchicine and hydrostatic pressure could at least partially inhibit the second phase of gastrulation, and they therefore concluded that MTs were necessary for secondary invagination to occur. MTs were found at the bases of filopodia extended by secondary mesenchyme cells in untreated embryos, and they were presumed to stabilize these filopodia as they extended across the blastocoel and attached to the blastocoel roof. It was believed that the filopodia would then contract to pull the archenteron across the blastocoel.

However, recent evidence suggests that a reexamination of the role of MTs during secondary invagination is in order. First, as Tilney and Gibbins (1969) were quick...
to acknowledge, their results using colchicine were variable, and the use of hydrostatic pressure is problematic, since high pressure also disrupts microfilaments. Second, since the time of this early study, colchicine has been found to exert effects on other cellular processes, including disruption of membrane ion transport (Bos and Emmelot, 1974), osmoregulation (Beebe et al., 1979), and nucleoside uptake (Mizel and Wilson, 1972). The availability of lumicolchicine, a derivative of colchicine that does not bind tubulin but presumably exhibits the MT-independent effects of colchicine (Wilson and Friedkin, 1967), has now made it possible to learn whether the effect of colchicine on gastrulation is due to its action on MTs or some other cellular process. Indeed, several early experiments utilizing colchicine have been reinterpreted as a result, including the effects of the drug on melanophore movements in teleosts (Lambert and Fingerman, 1976) and on the control of the timing of the cell cycle in echinoderm eggs (Sluder et al., 1986). Finally, filopodial contraction during secondary invagination is probably less important in lengthening the gut rudiment than active rearrangement of epithelial cells in the wall of the archenteron (Ettenson, 1985; Hardin and Cheng, 1986).

In light of these findings, the role of MTs during secondary invagination has been reexamined here, using the more specific inhibitor of tubulin polymerization, nocodazole. Although nocodazole has some long-term effects on cellular processes apparently unrelated to MTs (De Brabander et al., 1975), it is not toxic to sea urchin embryos (Karp and Solursh, 1985). Experiments using nocodazole and β-lumicolchicine, combined with indirect immunofluorescent staining of MTs, indicate that intact cytoplasmic MTs are not necessary for the extension of filopodia by secondary mesenchyme cells or for the active epithelial cell rearrangement and cell shape changes that occur during secondary invagination.

MATERIALS AND METHODS

Procurement of embryos. Gametes of Lytechinus pictus (Marinus Biologicales, Winchester, CA) were collected “dry” by intracoelomic injection of isotonic KCl, fertilized, and cultured in Millipore-filtered seawater (MFSW) as described previously (Hardin and Cheng, 1986). Cultures were maintained either in large beakers with stirring paddles at 16°C or in 6- or 20-ml aliquots in tissue culture dishes; embryos were collected for experimental manipulation by gentle hand centrifugation.

Microscopy. Living embryos were attached to coverslips coated with a solution of 1 mg/ml poly-L-lysine HBr (Sigma). The coverslips were then attached to microscope slides at the edges with silicone sealant (Dow Corning) and viewed using an Olympus differential interference contrast (DIC) microscope and either a 20× or 40× Olympus DIC objective.

Tissue sectioning. Embryos were fixed and collected as described previously (Hardin and Cheng, 1986); fixed embryos were dehydrated in increasing concentrations of ethanol, cleared in Histosol (National Diagnostics), and embedded in Paraplast embedding medium (Mo-ject Scientific). Embedded embryos were sectioned at a thickness of 8 µm, stained with hematoxylin and eosin, mounted, and viewed using DIC optics.

Drug treatments. For lumicolchicine experiments, embryos were collected at the early gastrula stage and placed in 0.1 mM β-lumicolchicine (Sigma) dissolved in MFSW. For nocodazole experiments, a nocodazole stock solution was added to early gastrulae cultured in tissue culture dishes. The stock solution consisted of 2 mg/ml nocodazole (Sigma) in pure dimethyl sulfoxide (DMSO); a sufficient amount of the stock solution was added to achieve a final concentration of 5-10 µg/ml nocodazole. Parallel control experiments using an equivalent amount of DMSO without nocodazole were also performed.

Indirect immunofluorescence. Two protocols were used. For whole-mount preparations, a procedure modified from Harris et al. (1980) was used. Embryos were attached to coverslips coated with poly L-lysine; embryos were then quickly transferred to 100% ice-cold methanol for 6-10 sec and immediately placed in phosphate-buffered saline (PBS) at 22°C. Alternatively, for viewing isolated archenterons, embryos were placed in 0.5-1 ml ice-cold methanol for 6-10 sec in a 15-ml conical centrifuge tube. PBS (8-10 ml) at 22°C was then quickly added, and the embryos were fragmented by 2-3 passes through a loose Dounce homogenizer (15 ml, A pestle; Kontes Glass Co., Vineland, NJ). The fragments were then attached to poly-L-lysine-coated coverslips. In both cases, specimens on coverslips were rinsed three times in PBS, and primary antibody (a monoclonal antibody against sea urchin flagellar tubulin; Asai et al., 1982) was applied to the coverslips for 1 hr at 37°C in a humid chamber. The preparation and characterization of this antibody have been described previously (Asai et al., 1982) Specimens were rinsed three times in PBS, and secondary antibody (FITC-conjugated goat anti-mouse, Cappel Laboratories, Cochranville, PA) was applied for 1 hr at 37°C. The specimens were then rinsed three times in PBS and mounted under coverslips in 90% glycerol, 10% PBS, 100 mg/ml 1,4-diazobicyclo-(2,2,2)octane (Aldrich), an antibleaching agent (Wordeman et al., 1986). Mounted specimens were viewed using a Nikon Diaphot fluorescence microscope equipped with either a Nikon 40× fluorescence or a Zeiss 63× Neofluar water-immersion objective and a Nikon 2.5× or 5× camera ocular. Specimens were photographed with a Nikon FX-35A camera using Technical Pan 2415 35-mm film (Kodak).
RESULTS

Methanol Fixation

The procedures presented here show that methanol is an acceptable and convenient fixative for use in indirect immunofluorescence studies of sea urchin embryos. Glutaraldehyde, the fixative of choice for preserving MT structure, results in unacceptable autofluorescence even following reduction with sodium borohydride (Hollenbeck and Cande, 1985), and as a result it was not used in this study. Methanol has been used extensively to study the cleavage divisions of the sea urchin embryo (Harris et al., 1980; Balczon and Schatten, 1983), and in the one study that compared MT fixation in methanol- and glutaraldehyde-fixed preparations, no appreciable difference was found in MT morphology (Balczon and Schatten, 1983). In addition, cellular morphology of methanol-fixed embryos compares favorably with glutaraldehyde-fixed embryos at the gastrula stage (J. Hardin, unpublished observations). In at least one case, in the ectoderm, a direct comparison can be made between the two fixation procedures (cf. Tilney and Goddard, 1970, and Results), and in this case both procedures yield identical results. Methanol thus seems to be a reliable fixative for use in studying the distribution of MTs during sea urchin gastrulation.

Microtubule Morphology in the Ectoderm

When indirect immunofluorescent staining is performed on whole-mount preparations or ectoderm fragments at the gastrula stage, a pattern very similar to that found by Tilney and Goddard (1970) for the blastula stage is observed. MTs can be seen radiating away from the region of the ciliary rootlet (i.e., the point of insertion of the cilium into the cell body) on the apical surfaces of ectoderm cells in a characteristic pattern (Fig. 1A). In profile, MTs can be seen extending along the periphery of each cell toward the inner basal surface (Fig. 1B). The distribution of MTs in the ectoderm does not appear to change during gastrulation.

Microtubule Morphology in Isolated Archenterons

Although it is possible to obtain good images of MTs in whole mounts of gastrulae, fluorescence of ectodermal MTs obscures the MT morphology of endoderm cells in the archenteron. To circumvent this problem, fixed and extracted embryos were homogenized in a loose Dounce homogenizer and the resulting tissue fragments were attached to poly-L-lysine-coated coverslips (see Materials and Methods). The embryos often shear apart preferentially, yielding archenteron fragments, whole archenterons, or vegetal plates with attached archenterons (Fig. 2). Such fragments allow improved antibody labeling and resolution of the MT morphology of individual cells in the archenteron. The overall morphology of such fragments is indistinguishable from that of their counterparts in intact embryos, and the results obtained from these fragments have always been checked, as far as possible, with corresponding whole-mount preparations.

The distribution of MTs in the cells of the archenteron

Fig. 1. Microtubule morphology in the ectoderm. (A) Apical staining pattern. Note the array of microtubules radiating away from the region of the basal body/ciliary rootlet. ci, cilium. (B) Profile view of ectodermal fragments. Note the microtubules extending away from the apical region along the periphery of each cell (arrow). Scale bar = 5 μm.
wall has some similarities to that in ectoderm cells (Fig. 2). MTs appear to originate from one or perhaps several MT organizing centers located at the apical (luminal) surfaces of endoderm cells, a location which corresponds to the sites of the rootlets of the short stubby cilia which characterize the archenteron at this time (Morrill and Santos, 1985). The MTs form a basketlike network extending along the periphery of each cell and thus conveniently outline cell boundaries in the archenteron (Figs. 2B and 2C). This basic pattern does not change appreciably during secondary invagination, although the "baskets" of MTs do change their shape somewhat to correspond to the flattened shape that the cells in the archenteron adopt by the end of gastrulation (Hardin and Cheng, 1986). The endoderm cells of the early gastrula (Fig. 2A) gradually lose their columnar organization as gastrulation proceeds, adopting first a slanted stacked appearance (Fig. 2B) and finally exhibiting the almost squamous morphology of the late gastrula (Figs. 2D and 2E). In addition, mitotic figures can occasionally
be seen in the wall of the gut rudiment (Fig. 2D), but no more than one or two mitotic figures per specimen have ever been observed.

It is also possible to visualize individual secondary mesenchyme cells at the tip of the archenteron in isolated fragments. On the basis of transmission electron micrographs, Tilney and Gibbins (1969) reported that in some secondary mesenchyme cells MTs are found at the bases of the filopodia, but that MTs are rarely present at the tips of these extensions. Indirect immunofluorescence confirms this result; MTs can be seen along the periphery of SMCs and extending into the broad bases of filopodial processes, but they are usually not observed in the long slender portions of the filopodia which comprise most of their length (data not shown).

Lumicolchicine Inhibits Archenteron Elongation

Because an earlier study used colchicine to depolymerize MTs (Tilney and Gibbins, 1969), control experiments using the colchicine analog β-lumicolchicine were first performed to determine whether the results of this earlier study were due to the specific effects of colchicine on tubulin or to other effects of the drug on the embryo. When 0.1 mM β-lumicolchicine is applied to embryos 2 hr prior to the onset of secondary invagination, dramatic effects are seen on the further progress of invagination. In over 90% of the embryos examined, archenteron elongation is markedly inhibited (Fig. 3). Many embryos exhibit a bent shortened gut rudiment (Fig. 3A) or an outward buckling of the vegetal plate with a short archen-

![Figure 3](image-url)

**Fig. 3.** The effects of 0.1 mM β-lumicolchicine on archenteron elongation. The drug was added 2 hr prior to the onset of secondary invagination.
(A) Many embryos exhibit a flaccid short archenteron. (B) Archenteron elongation ceases when the gut rudiment is one half to two-thirds of the way across the blastocoel, even though differentiation and spicule formation are unaffected. sp, spicule rudiment. (C) Control embryo at same time as (A) and (B). Scale bar = 25 μm. (D) Microtubules in the ectoderm of drug-treated embryos (compare with Fig. 1A). (E) Microtubules in the archenteron of a drug-treated embryo viewed from the animal pole (compare with Fig. 2A). Note the normal appearance of the MTs, which can be seen along the periphery of each cell (arrows). Scale bar = 5 μm.
teron, resembling so-called “enteroexogastrulae” (Dan and Okazaki, 1956; data not shown). Others have relatively normal morphology, except that the tip of the archenteron never reaches the animal pole. Instead, the tip of the gut rudiment stops one-half to two-thirds of the way across the blastocoel, remaining there through subsequent development (Fig. 3B). Moreover, embryos treated with β-lumicolchicine show normal MT morphology (Figs. 3D and 3E), so the effects of the drug on invagination are not due to any effect on MTs but instead represent an effect on some other cellular process. Since Tilney and Gibbins used concentrations of colchicine up to 50 times greater than the concentration of lumicolchicine used in this study, it is likely that many of the effects of colchicine on invagination that they observed were not due to the specific effects of the alkaloid on MTs.

**Nocodazole Depolymerizes Microtubules but Does Not Inhibit Archenteron Elongation**

In order to determine unambiguously whether or not MTs are necessary during the second phase of gastrulation, the synthetic MT inhibitor nocodazole (De Brabander et al., 1975) was used to study the effects of depolymerization of cytoplasmic MTs on gastrulation. To ensure penetration of the drug throughout the embryo, a rather high dose of the drug (5-10 µg/ml) was applied

**Fig. 4.** The effects of 10 µg/ml nocodazole on archenteron elongation. The drug was added 2 hr prior to the onset of secondary invagination. (A) Midgastrula. Note the prominent filopodia (fp) as well as the broad lamellipodial protrusions (lp). (B) Late gastrula. (C) Spontaneous exogastrula. Scale bar = 25 µm. (D) Indirect immunofluorescent staining of ectoderm cells. There is no evidence of intact cytoplasmic microtubules; nuclei (nu) appear dark, and the surrounding cytoplasm (cyt) exhibits punctate staining. (E) Staining pattern in the archenteron prior to the onset of secondary invagination, viewed from the animal pole (compare with Fig. 2A). MTs again appear to be disrupted. Scale bar = 5 µm.
to the embryos 2 hr prior to the onset of secondary invagination. This dose is 5- to 10-fold higher than that reported to inhibit mitosis in sea urchin embryos (Karp and Solursh, 1985).

When nocodazole is applied in the manner just described, the effects of the drug are rapid. Embryos begin showing abnormal swimming behavior within 5 min, losing the ability to swim directionally, and instead spin rapidly in tight circles. Although the drug is apparently affecting the MT-based ciliary machinery of the ectoderm cells, and presumably their cytoplasmic MTs as well, these embryos gastrulate essentially normally (Figs. 4A and 4B). In particular, nocodazole-treated embryos undergo epithelial cell rearrangement to an extent comparable to DMSO controls as judged by transverse tissue sections (Fig. 5). The flattening of cells in the wall of the archenteron during secondary invagination also occurs (Fig. 6). DMSO-treated embryos and untreated controls behave identically (data not shown). Occasionally, untreated embryos will spontaneously exogastrulate, and embryos treated with nocodazole are also capable of spontaneous exogastrulation (Fig. 4C). Since exogastrulation is accompanied by active cell rearrangement (Hardin and Chen, 1986), such active rearrangement is apparently unaffected by the drug.

Since archenteron elongation occurs normally in the presence of nocodazole, it is of interest to show that the drug is actually depolymerizing cytoplasmic MTs before secondary invagination begins. When drug-treated embryos are examined by indirect immunofluorescence prior to the onset of secondary invagination, all of their cells display diffuse cytoplasmic staining surrounding a dark nucleus, with no evidence of intact cytoplasmic MTs (Figs. 4D and 4E).

Although intact cytoplasmic MTs do not seem to be crucial for the rearrangement and flattening of the cells in the archenteron during secondary invagination, there are some alterations in cellular morphology induced by nocodazole. Most notably, secondary mesenchyme cells (SMCs) often exhibit abnormally broad flat lamellipodial protrusions (Fig. 4A). In addition, some SMCs leave the tip of the archenteron prematurely, adopting this broad lamellar shape as they migrate through the blastocoel. However, many SMCs are still capable of extended long narrow protrusions comparable in length to the filopodia extended by SMCs in control embryos. In fact, many of the broad protrusions taper, ending as long slender processes (Fig. 4A). Thus, although intact cytoplasmic MTs seem to aid in the formation of filopodial protrusions by SMCs, they do not seem to be essential for this process, despite the fact that they are usually found at the bases of filopodia in normal embryos (see above).

**DISCUSSION**

The powerful techniques of immunocytochemistry have been used for years to study the role of the cytoskeleton in motility and cell shape changes in tissue culture cells, but it has proven more difficult to apply these same techniques to postcleavage stage embryos. Recent studies have included localization of contractile proteins during mouse and chick neurulation (Sadler et al., 1982; Lee and Nagle, 1985), localization of actin and tubulin during early development in *Drosophila* (Karr and Alberts, 1986), and an investigation of the role of MTs in the spreading of the epiblast during chick gastrulation (Mareel et al., 1984). These techniques have also been used to study the role of MTs during plant morphogenesis (Lloyd, 1986). However, no immunofluorescence studies have been made of the changes in the cytoskeleton during sea urchin gastrulation. Furthermore, the role of the cytoskeleton in epithelial cell rearrangement...
has been unexplored. This study investigates some of these questions.

**Drug Studies of the Role of Microtubules in Gastrulation**

This study shows that MTs are not crucial for secondary invagination and suggests that caution should be exercised when interpreting the results of experiments employing MT inhibitors, particularly colchicine. Many of the early studies of the role of MTs in morphogenesis used colchicine, which has since been shown to have side effects, particularly at the cell membrane (see Introduction). The inhibition of secondary invagination by lumicolchicine argues that the effects of both lumicolchicine and colchicine on gastrulation are due to some shared property or properties other than the ability to bind tubulin. Therefore one cannot conclude from earlier colchicine studies (Tilney and Gibbins, 1969) that secondary invagination depends on intact cytoplasmic MTs. Furthermore, the occurrence of secondary invagination despite the demonstrated disruption of MTs with nocodazole, a MT inhibitor with a greater specificity and fewer side effects than colchicine (De Brabander et al., 1975), suggests that in *Lytechinus pictus*, secondary invagination, and the cell rearrangements that accompany it, are largely MT-independent.

Studies using nocodazole also require cautious interpretation, especially when cells are exposed to the drug for an extended period of time (De Brabander et al., 1975). This is particularly important when using nocodazole to study a developmental process such as sea urchin gastrulation, which occurs over several hours, as opposed to the short-term exposures commonly used to study tissue culture cells. Because nocodazole did not prevent secondary invagination and the efficacy of the drug could be confirmed by examining the integrity of cytoplasmic MTs, the unambiguous conclusion can be drawn that intact MTs are not crucial for archenteron elongation.

**The Role of Microtubules in Morphogenetic Shape Changes**

Because nocodazole does not appear to affect the changes in shape that cells in the archenteron undergo during secondary invagination, the general question arises as to what role MTs play in motility and changes in cell shape. The answer to this question is unclear; certain motile events in some cell types seem to depend on MTs, whereas similar processes in other cell types do not. The polarity of motile fibroblasts, but not their protrusive activity, can be disrupted with MT poisons (Vasiliev, 1982); on the other hand, movement of fish keratocytes, including directional movement, does not depend on MTs (Euteneuer and Schliwa, 1984). Similarly, columnarization of neural plate cells during amphibian and chick neurulation may depend on MTs (Burnside, 1973; Karfunkel, 1974), but MTs are not necessary for the elongation of cells in the lens primordium (Beebe et al., 1979). In at least one cultured cell line, MT morphology does not seem to have any direct link to cell shape (De Brabander et al., 1986). Although MT morphology was not examined, one study using colcemid suggests that the shape changes that occur during amphibian gastrulation are largely MT-independent (Cooke, 1973). It is possible that MTs are not directly responsible for determining cell shape in many systems but rather stabilize shape changes which are produced by other intracellular machinery (Hilfer and Searls, 1986). Such a role for MTs would be consistent with the observations presented here for sea urchin gastrulation.

**The Role of Microtubules in Filopodium Formation**

Based on the effects of colchicine it appeared that MTs were necessary for the initiation and stabilization of filopodia extended by secondary mesenchyme cells (Tilney and Gibbins, 1969). In contrast, this study shows that despite disruption of cytoplasmic MTs with nocodazole, filopodia in drug-treated embryos are often as long and as slender as those found in controls. This finding is consistent with what is known about the structure of long slender protrusions in other systems. Microspikes extended by growth cones in neurons contain bundled actin but MTs only insert at their bases (Letourneau, 1983). Similarly, the filopodia of transformed sea urchin coelomocytes are filled with paracrystalline arrays of actin, but no MTs (Edds, 1984). The acrosomal reaction in *Thyone* sperm involves the rapid extension of a long acrosomal process, accompanied by the explosive polymerization of actin (Tilney et al., 1973). and a similar mechanism seems to be operating during the initial stages of microvillus elongation in fertilized sea urchin eggs (Begg et al., 1982; Carron and Longo, 1982). In the sea urchin embryo, filopodial protrusions extended by primary mesenchyme cells contain an actin meshwork but are devoid of MTs except at their bases (Katow and Solursh, 1981). Since actin bundles are found along the length of secondary mesenchyme filopodia while MTs are found primarily at their bases (Tilney and Gibbins, 1969), it is likely that restructuring of actin within the filopodia is the most important process involved in their extension.

However, intact MTs may increase the likelihood that such slender protrusions will form, perhaps by allowing a greater degree of localization of protrusive activity. This may be the case with fibroblasts; colcemid treat-
ment causes fibroblasts to form a symmetrical lamellipodium around the entire cell periphery, instead of in a single location (Vasiliev, 1982). Since the secondary mesenchyme cells of drug-treated sea urchin embryos possess many broad lamellar protrusions in contrast to the slender localized filopodia of the controls, a mechanism similar to that operating in fibroblasts may be operating in secondary mesenchyme cells.

The Role of Microtubules in Cell Rearrangement

Epithelial cell rearrangement is an important phenomenon in many morphogenetic processes (Fristrom, 1976; Keller, 1978; Bode and Bode, 1984; Ettensohn, 1985; Locke, 1985; Keller and Trinkaus, 1987). However, little or no information is available on the role of MTs in these systems. Although MTs are consistently found in cells of the archenteron throughout sea urchin gastrulation, they do not appear to be necessary for the active epithelial cell rearrangement that occurs during secondary invagination (Hardin and Cheng, 1986). Colchicine treatment of Drosophila imaginal discs suggests that MTs are not necessary for epithelial cell rearrangement in this system as well (Fristrom and Fristrom, 1975). MTs may help to stabilize the shapes of cells in the invaginating gut rudiment and the surrounding ectoderm, but they do not appear to be indispensable for the cell rearrangements that occur during secondary invagination. It is still not known what intracellular mechanisms are responsible for epithelial cell rearrangement during sea urchin gastrulation, but the search for such mechanisms must continue at the cytoskeletal level. As more attention is given to the intracellular basis of cellular behavior during sea urchin gastrulation, our understanding of secondary invagination, and epithelial cell rearrangement in general, should be improved.

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