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THE CORTICAL TRACTOR MODEL FOR EPITHELIAL FOLDING: APPLICATION TO THE NEURAL PLATE[1]

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ABSTRACT We propose a "cortical tractor model" to account for the shaping and folding of epithelial tissues. We suggest that a particular type of cortical flow common to both mesenchymal and epithelial cells allows epithelial cells to interdigitate with one another while maintaining the integrity of their apical seals. Indeed we propose that the cortical tractoring builds these apical seals by cycling intracellular adhesive junctions. The boundaries between cellular domains with differing adhesive characteristics may organize movements amongst the epithelial cells to produce some of the characteristic changes of shape of the tissue. We apply the model to the phenomenon of neurulation and demonstrate that it can account for neural plate elongation and rolling into a tube.

INTRODUCTION

During early development, embryonic cells appear to fall into two broad morphological classes; epithelial and mesenchymal. Epithelial cells array themselves into sheets and may present a very regular "paving stone" appearance

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when viewed from the apical side. Epithelia form the external and internal boundary layers of embryos. Mesenchymal cells are more amorphous, migratory cells that move about in spaces between and on epithelial layers. Form-shaping movements of the embryo involve the coordinated activities of both cell types. An epithelial layer folds, rolls, invaginates and deforms, while retaining its integrity as a connected sheet. Mesenchymal cells migrate independently and aggregate into patterned collections of cells.

Classification of embryonic cells into these two catagories is based largely on the gross differences in appearance between epithelia and mesenchyme. These distinctions have proven so conceptually appealing that it is easy to downplay the fact that cells frequently interconvert between the two forms. For example, from the boundary of the epithelial epidermis and neural plate, mesenchymal neural crest cells emerge. As another example, epithelial cells of the epiblast of amniote embryos converge to the primitive streak at the midline where many epithelial cells detach during focal contractions (C. Stern, personal communication) and spread out below the epiblast as migrating mesenchymal cells. Some of these condense into somites and become epithelial again, and then later the epithelial somite again breaks up into mesenchyme (1).

In this paper we shall blur the distinction between the two types of cells still further by proposing a model for epithelial morphogenesis that imparts to epithelial cells certain "mesenchymal" behavioral characteristics. In particular, we shall propose a mechanism by which epithelial cells can fold, roll, and change their neighbors. This mechanism not only ensures maintenance of the all-important apical seal that insulates the embryo from the external environment, but actually constructs that seal at the cell apices.

The sorts of phenomena to which this model may apply include epiboly in amphibian embryos, many movements during amphibian gastrulation, convergence and ingression of the primitive streak of birds, neurulation, the formation and invagination or evagination of epidermal placodes, and eye-cup morphogenesis. In this paper we shall use elongation and tube formation in the neural plate of the newt embryo as our example of how active cortical tractoring of cells in an epithelial tissue may be organized to produce large-scale morphogenetic deformations.

THE CORTICAL TRACTOR MODEL

The model that we present here departs from previous views of how epithelial layers are constructed. Rather than treating the cells that constitute the layer as static entities, we propose that they are constantly cycling their surfaces in a certain way. It is only on the short time scale of visual observations - or in the frozen rictus of the scanning electron microscope - that epithelial cells convey the appearance of stationary cobblestones that can at most contract and relax their apical surfaces. We shall assume that epithelial cells are rather more "mesenchymal-like" than heretofore presumed. To make clear exactly what we mean, we briefly discuss mesenchymal cell motions as we present the postulates of our model of epithelial cell behavior.

Motile Cells Cycle Their Cortex

A cell moving across a substratum clearly exhibits several features. First, the cell puts out protuberances (eg. filopodia, lamellipodia) at its leading edge. When one of these appendages attaches to the substratum, contraction of the cortex pulls the cell forward a small increment. This cycle of spreading, or extension, followed by attachment and contraction, is accomplished by internal flows of cytoplasm, generally forward through the center of a lamellipodium and rearward in the cortex. If a time-lapse movie could be taken of the cortical flow, the net average motion would be the double fountain movement of cytoplasm shown schematically in Fig. 1. Oster (2) has previously presented a model for lamellipodial motion based on a cycle of events involving solation of the actomyosin cortical gel followed by osmotic expansion and active contraction of the cortex. However, our discussion here does not depend on the detailed assumptions of that model, but only on the average cortical flow pattern, whatever the driving mechanism. Therefore, the first postulate of our model is:

I. *Cell motion is characterized by a "cortical tractor" that produces a time-averaged cortical flow from the leading to the trailing surface, as shown in Fig. 1.*

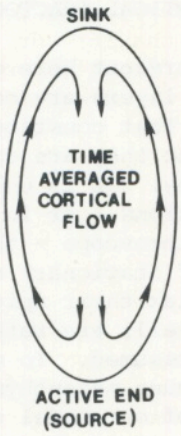

FIGURE 1. Schematic of the cortical tractor mechanism. The actomyosin cortex flows as indicated by the arrows, and cell-to-cell adhesion structures are dragged along by the flow of the cortex.

This sort of fountainoid motion of the cortex has been suggested by a number of workers (eg. 3-7). Marking experiments have shown that the cortical flow is reflected on the surface of the cell, indicating an intimate connection between the flowing cortex and the plasma membrane (8). It is also necessary for us to postulate that the adhesion structures that anchor the cell to the substratum and to its neighboring cells are cycled along with the cortex (9):

II. *Adhesion and junctional molecules are inserted at the site of the cortical "source" and flow with the cortex to be resorbed at the cortical "sink", unless stabilized by bonding with the substratum, or with another cell surface.*

Thus a moving cell attaches to its surroundings via attachment sites that are continuously being inserted at the leading edge, dragged posteriorly, and there resorbed.

Cortical Activity Can Be Triggered by Ionic Stimuli

As shown by experiments with ionophores and channel-plugging molecules, cell motility can be stimulated by ionic leaks, especially calcium, and inhibited by blocking

ionic channels (10, 11). The model suggested by Oster (2) provides one possible mechanism for this behavior, however, we need not be more specific here than that motility can be ionically triggered. Thus our third postulate is:

III. *Local ionic conditions can "activate" any face of a cell, which thence becomes the leading edge; that is, the cortical source in Fig. 1.*

This postulate accords with the observation that cells can "contact inhibit" one another. When the leading membrane of a cell becomes closely attached to the membrane of another cell, the ionic leak, which stimulates motility, may become sealed, thus paralysing cell movement. However, we will not be specific about the cellular or molecular mechanisms that regulate the intercellular ionic conditions; that such mechanisms exist is amply evident, but how they are mediated is yet unclear. Furthermore, the proximal signal for activating a cell's surface may be other, nonionic, chemical messengers. For example, chemotactic agents that bind to specific receptors on a cell's surface may initiate the ionic leak that triggers motile activity. For the purposes of our discussion here we can remain vague about the specific extracellular molecular trigger that activates a cell's surface.

Epithelial Layers Are Dynamic Structures

For the final ingredient in our model we reconsider the nature of an epithelial layer. Microscopic examination of the surface of an epithelium reinforces the impression of a static paving of cells. The individual cells of an epithelium are bonded firmly along their apical edges to form a molecular seal that insulates the interior of the embryo from the exterior environment. The epithelial layer as a whole can deform by folding, bending, rolling into tubes, invaginating or evaginating. The occasional evidence of apical bundles of microfilaments has led to the postulate that constriction of these filaments could generate the force that drives these deformations (12-14). Here we augment that model dramatically by postulating the following:

IV. *Epithelial cells continuously cycle their cortical cytogel in a manner analogous to motile mesenchymal cells, with the added constraint that their apical boundaries remain firmly attached.*

The implications of Postulate IV can be appreciated by considering the sequence of events shown schematically in Fig. 2. An epithelial layer consists of an array of cells, all joined at their apical boundaries. The basal surfaces of each cell may be active, so that each cell's cortex undergoes the tractor motion. This means that each cell is, in effect, attempting to crawl downward on its neighbors by adhesion of its surface and contraction of its cortex. However, if each cell undergoes the same motion, then no net traction can be developed between adjacent cells, and thus no net motion transpires. That is, if adjacent cells attempt to crawl on one another with equal intensity in the same direction, then no relative motion can take place. Moreover, since the adhesion structures are being continuously added in the basal region, they will be carried apically by the cortical flow, finally piling up at the apical seal.

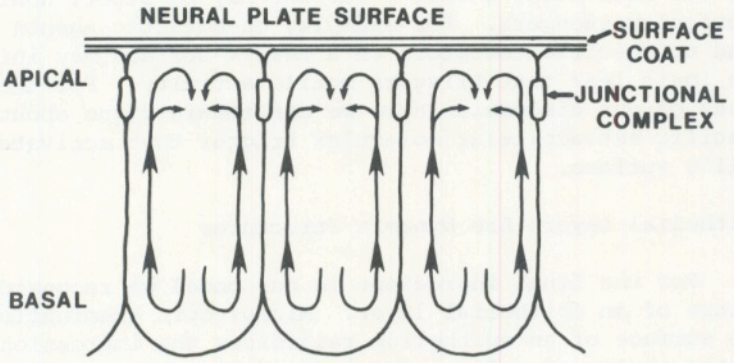

FIGURE 2. Diagram of a section of the neural epithelium. Each cell undergoes the cortical tractor motion (arrows). In this case, the cells are active basally.

Microfilament bundles, usually found attached to surface adhesion structures, linking them together, would be concentrated at the apical end of a cell by cortical flow. Contraction of these filament bundles could then rapidly contract the apex of the cell. In this way, our cortical tractor model subsumes the previous models of Baker and Schroeder (12), Burnside (13), Jacobson and Gordon (19), and Odell, et al. (14).

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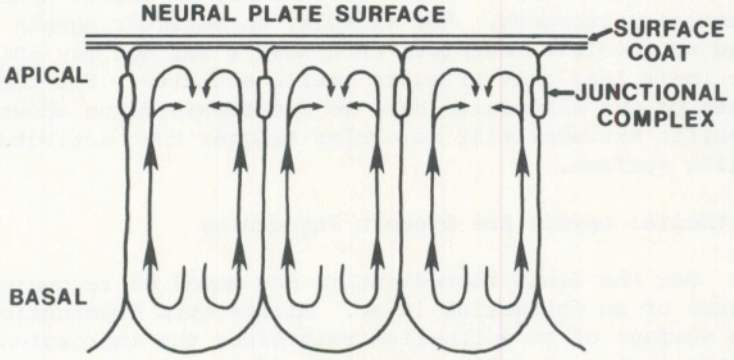

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Thus according to the cortical tractor model, an epithelial layer is a dynamic structure, each cell continually cycling its cortical cytogel in a basal-to-apical fountain flow. When each cell cycles with nearly equal intensity, there is no net relative motion, and the appearance of a static structure is maintained.

The Cortical Tractor Model Allows Epithelial Cells to Change Neighbors Without Breaking the Apical Seal

The cortical tractor model can explain how cells can interdigitate while still maintaining the integrity of the apical seal. Consider the situation illustrated in apical view in Fig. 3a: cells A and C are in contact with each other, and cells B and D are separated. If at some later time we observe that cells B and D are in contact, then the bonds between A and C must have been severed. How could this neighbor shift take place without breaking the apical seal? According to the cortical tractor model, the answer lies below the surface. Suppose that the cortex source of cell B shifts laterally toward cell D. Thus cell B puts out lamellipodia on its basal and lateral surfaces and the lateral lamellipodia contact the neighboring cells, eventually contacting cell D near its basal end. This lamellipodial surface area, along with its adhesive structures, is swept apically by the cortical tractor. Thus cell B interposes itself between its neighbors from underneath as shown in Fig. 3b, and new attachments between cells B and D are swept

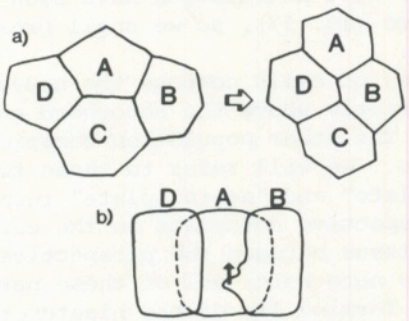


FIGURE 3. (a) Apical view of four epithelial cells as they change neighbors. (b) Lateral view of the cortical tractor mechanism showing cell B interdigitating between cells A and C. Cell B is laterally and basally active.

from their basal to apical ends. Viewed from the top, it appears as if cell B is actively crawling between cells A and C, giving the appearance that A-C bonds are being broken. However, the basal to apical flow of cortical cytogel - which carries the junctional structures - inserts junctions as fast as they are recycled at the apex. The net result is that cell B can appear to interdigitate itself between A and C, and bring about a neighbor exchange without breaking the apical seal.

A cell population, wherein each cell tractors its cortex in a fountainoid such as we have described, can behave quite counterintuitively. Recently, Odell and Bonner (15) have shown that the crawling of the *Dictyostelium discoideum* grex can be explained by the concerted action of a population of amoebae, each of which crawls on its neighbors. Indeed, much of the morphogenesis of *D. discoideum* can be interpreted in terms of the cortical tractor model, including the formation of the grex, its motion, the distribution and orientation of the stalk and spore cells in the grex, and the erection of the fruiting body. Here we apply the same physical analysis to model the shaping and rolling of the neural plate. We show next how the type of cell dynamics described by the cortical tractor model could drive the process of neurulation in newt embryos.

NEURULATION AND THE CORTICAL TRACTOR MODEL

The details of newt neurulation have been reviewed recently by Jacobson (16, 17), so we shall present only a brief summary here.

Two populations of cells compose the neural plate. One population lies directly above the notochord and is called "notoplate" (16). The other population comprises the rest of the neural plate. We will refer to these two populations of cells as "notoplate" and "neural plate" respectively (Fig. 4). The prospective notoplate in the early gastrula is a crescent of tissue between the prospective neural plate and the prospective notochord; all of these parts lying in sequence above the forming lip of the blastopore (Fig. 4). As gastrulation proceeds, the notochord involutes around the blastopore lip, converges toward and elongates along the midline. The notoplate does not involute, but rather converges toward and elongates along the midline in concert with the notochordal tissue below it. As this happens, the

neural plate is shaped around the notoplate, which comes to occupy the midline of the emerging spinal cord and brain up through the mesencephalon (Fig. 4).

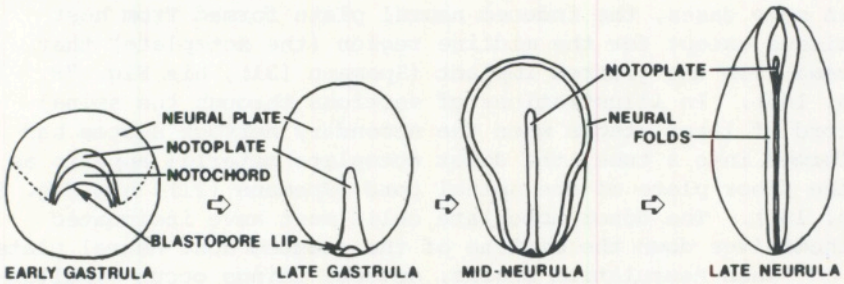


FIGURE 4. The parts referred to in the text are labelled on these drawings of a rear view of an early gastrula, and dorsal views of late gastrula to late neurula of newt embryos. The notochord shown above the dorsal lip of the blastopore of the early gastrula has involuted around the blastopore lip and lies beneath the notoplate in subsequent stages. Figures are approximately to scale. An early gastrula is about 2.5 mm in diameter.

Besides the profound differences in behavior that have been described between notoplate and the rest of the neural plate (19), there are other observations that suggest the notoplate is quite a different population of cells from the rest of the neural plate. For the most part, the notoplate does not differentiate into nervous tissue. It occupies the same position as the floor plate of the spinal cord and brain. The fate of the floor plate is to become a raphe (a seam) between the basal plates of the spinal cord and the brain stem (which is precisely what one would expect to be the ultimate consequence of the cell behavior proposed here for the notoplate).

The notoplate may not be a part of the induced neural plate, but may be more closely related to the chordamesoderm, and be induced along with that tissue. As shown in Fig. 4, the position of the notoplate in the early gastrula is contiguous with the prospective notochord in the dorsal lip of the blastopore. In the famous experiments of Spemann and Mangold (20) in which a piece of dorsal lip of the blastopore of an early gastrula of *Triton cristatus* was transplanted

into the ventral ectoderm of a *Triton taeniatus* host embryo, a second neural plate was induced in the ventral ectoderm by the implanted dorsal lip material. Pigmentation differences between the donor and host species allowed the tissues of the two species to be distinguished from one another. At least in some cases, the induced neural plate formed from host tissue except for the midline region (the notoplate) that came from the grafted implant (Spemann (21), his Fig. 78, p. 144). In illustrations of sections through the spinal cord of later stages when the secondary nervous system had formed into a tube, the donor notoplate material appears as the floor plate of the spinal cord (Spemann (21), his Fig. 80, p. 146). The donor notoplate cells must have insinuated themselves down the midline of the induced host neural plate.

When neurulation begins, several things occur simultaneously. The surface of the neural plate reduces as the apical surfaces of the cells that compose the plate contract, the cells of the neural plate get taller (i.e. more columnar), and the cells of the notoplate rearrange to elongate the midline of the neural plate. These activities reshape the disc-shaped neural plate into a keyhole shape (19). At the keyhole stage, neural folds appear at the boundary between the neural plate and the epidermis. The length of this boundary remains constant up to the keyhole stage, but the boundary elongates extensively as the neural plate subsequently folds into a tube. During the period of neural tube formation, which commences at the keyhole stage, the neural plate elongates very rapidly, and the cells of the plate continue to reduce their apical surfaces and to get taller (the plate gets thicker) (22).

Cell behavior, as described by the cortical tractor model, could be responsible for most of the activities that are observed during the shaping of the neural plate and the rolling of the plate into a tube. If the cells that compose the single-layered neural plate in urodele and amniote embryos are active at their basal ends, then the cortical tractors of the cells would function as shown in Fig. 2. Since the apical surfaces of the cells are bound together by junctional complexes, and some embryos, such as the newt, have a surface coat, the cells are not free to move, but are held in place by their apical surfaces. If the rate of tractoring in the apical junctions is the same as in the basal ends of the cells, then no net motion will occur. If the rate of tractoring in the apical junctions is slightly lower than the rate of tractoring toward the basal end of the cells, then the end result of cortical tractoring could be

to elongate the cells, and this could account for the observation in newt embryos that neural plate cells get taller during neurulation. If the rate of tractoring of the apical junctions is much slower than the rate of tractoring toward the basal ends of the cells, or if there is no recycling of the junctional structures, then the end result could be drawing out of the apical end, bottle-cell formation, or the traction of the basal end could be enough to break the cells free of the epithelium. This scenario does not eliminate specific contraction of microfilament bundles arranged as pursestrings around the apical surfaces of the cells, as suggested by others (12, 13), but it better accounts for the observation that cell elongation and apical constriction keep pace with each other during much of neurulation (19).

Elongation of the Notoplate

In contrast to the rest of the neural plate, the cells of the notoplate do not get taller during neurulation through to the midneurula keyhole stage (19), but they elongate thereafter. There is intimate contact between the basal surfaces of the notoplate cells and the cells of the notochord (Fig. 5), while more lateral neural plate cells hang free in the extracellular space, which is probably filled with extracellular matrix.

We propose that the notoplate cells would have their basal faces inactivated by contact with the notochord cells, and that their apical surfaces, which are covered by the surface coat, are normally inactive. Only the lateral faces of the notoplate cells could have active cortical tractors.

In accordance with this assumption, tangential sections through the midline of the neural plate (through the notoplate) of a newt neurula reveal numerous long lamellipodia extending up to three or four cell ranks from the lateral faces of notoplate cells (Fig. 6). Thus we suggest that notoplate cells can interdigitate between one another according to the cortical tractor mechanism described above. (It may be relevant to note that Cooper and Keller (23) have shown in tissue culture that it is not unusual for cells to move perpendicular to their long axes.)

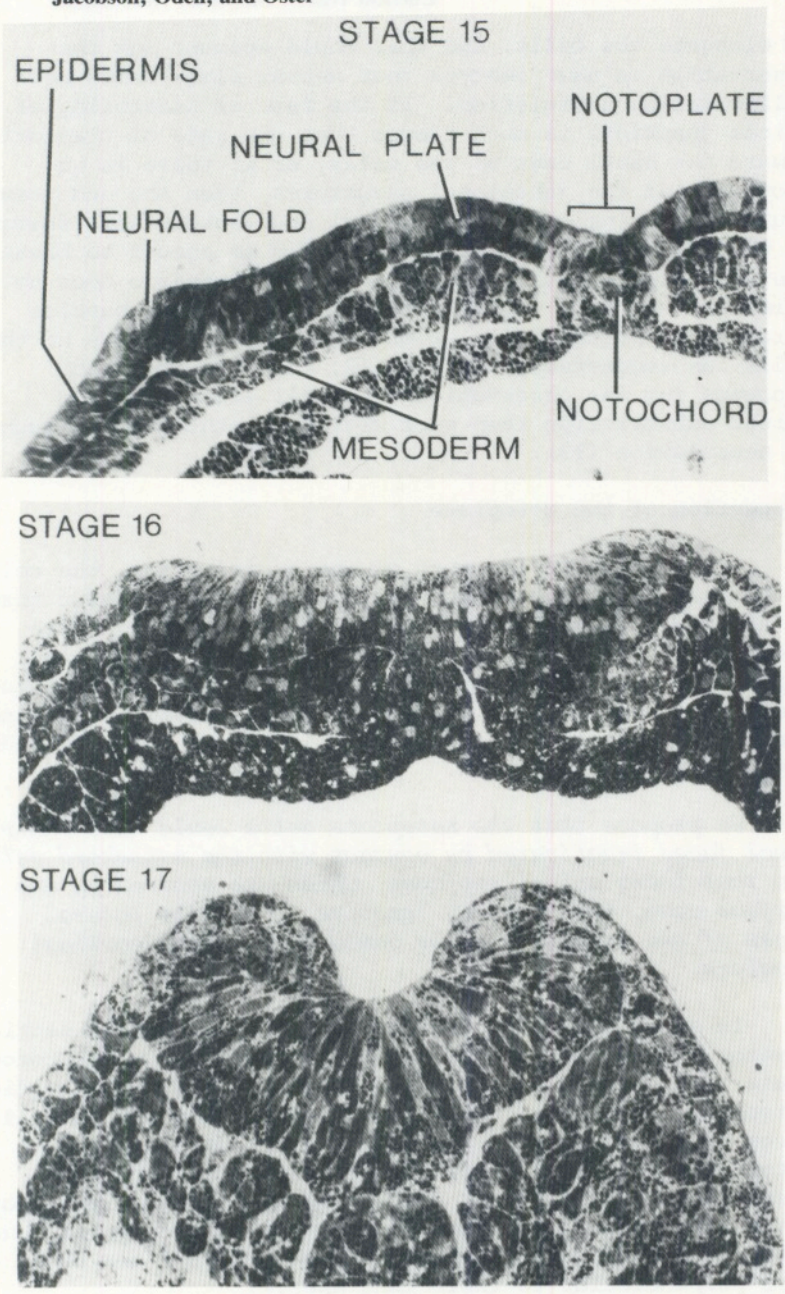


FIGURE 5. (Continued on the next page.)

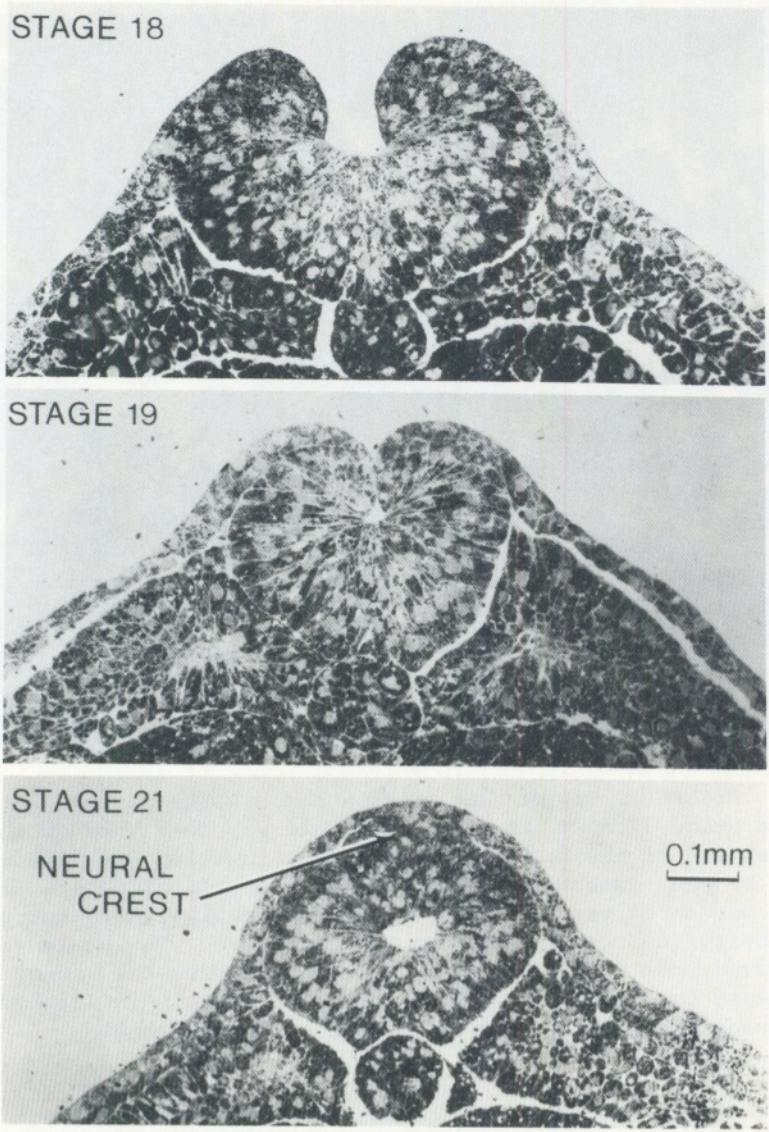


FIGURE 5. The neural plate, and the tissues that underlie it, are shown in cross section at stages through the period of neural fold formation and rolling of the plate into a tube, in newt embryos. These plastic sections (2μm thick) are from the region where spinal cord joins the brain. All are at the same magnification.

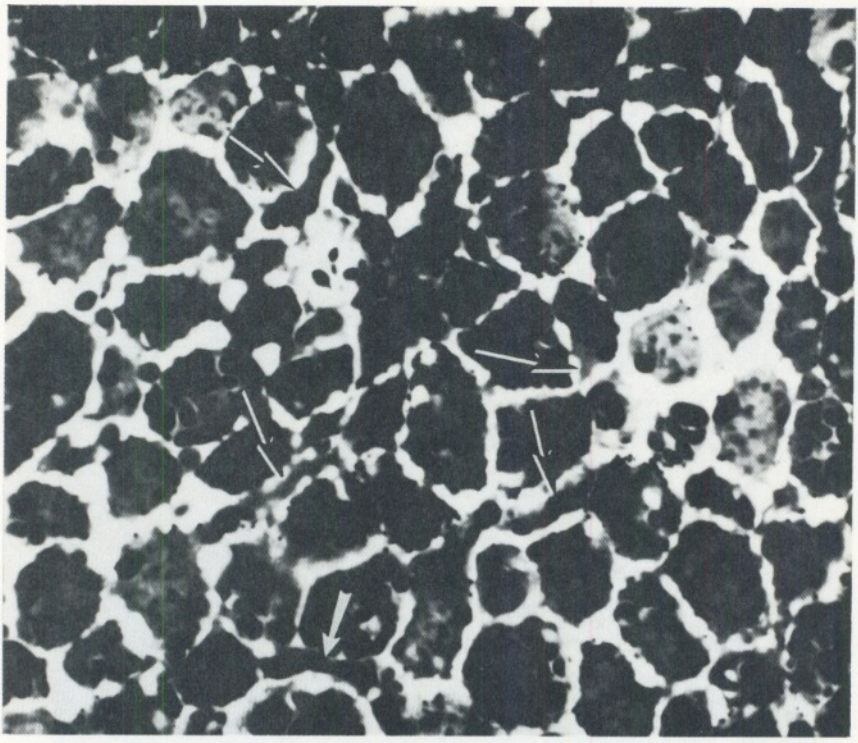


FIGURE 6. Tangential section through the neural plate of a stage 17 newt embryo, about half way down through the thickness of the notoplate. Numerous lamellipodia (arrows) can be seen extending laterally several cell diameters from the bodies of the notoplate cells. The neural plate-notoplate boundary runs across the top of the figure. The plate was removed from underlying tissues (which opened the spaces between the cells some) then fixed immediately in aldehydes, embedded in plastic, and sectioned at 2μm thickness.

If the direction of tractoring were random on the lateral faces of the notoplate cells, the net effect should be a random walk, with cells changing neighbors, but with no net distortion of the notoplate in any direction. However, there are at least two mechanisms by which the boundary between the notoplate and the rest of the neural plate could play a role in organizing these lateral interdigitations amongst the notoplate cells: (a) notoplate cells that contact

neural plate cells stick to them more than they stick to each other (differential adhesion), or (b) contact with a neural plate cell inhibits the activity of that face of the notoplate cell (contact inhibition). Either mechanism would restrict the tractoring to faces not abutting the neural plate. Thus directed tractoring by notoplate cells stuck up against the neural plate boundary can elongate the boundary by cell interdigitation, as shown in Fig. 7.

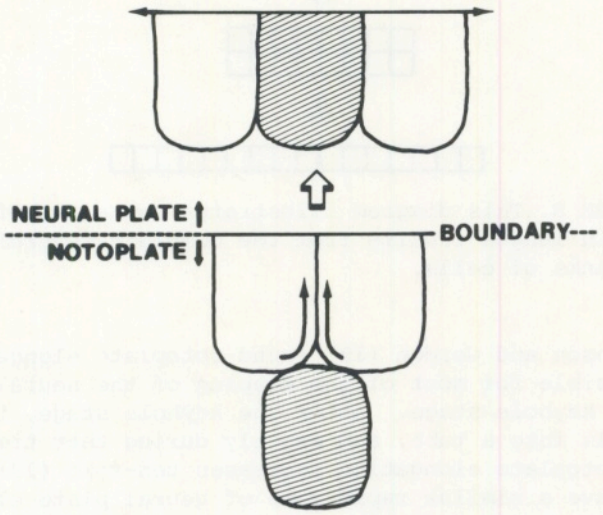


FIGURE 7. This diagram (top view, looking down on the cell apices) illustrates how interdigitation of cells can elongate the boundary at the neural plate/notoplate interface.

Elongation of the notoplate boundaries would stress the adjacent notoplate and neural plate tissue, and would facilitate interdigitation of these adjacent ranks of cells along the long axis of the embryo. The spreading of this effect would elongate the notoplate and neural plate to conform to the elongating notoplate boundaries. Meanwhile, the boundary cells would continue to tractor more cells to the boundary. If no countermanding forces intervene, this process could continue until the notoplate has been reduced to a single rank of cells (i.e. becomes a raphe).

We have measured about a four-fold increase in length

of the notoplate region during neurulation. The minimum number of ranks of notoplate cells required to accomplish this by interdigitation is four (Fig. 8).

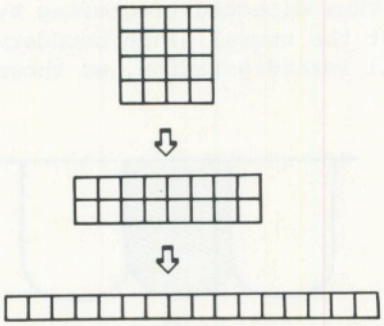


FIGURE 8. This diagram illustrates how a four-fold increase in length results from the complete interdigitation of four ranks of cells.

Jacobson and Gordon (19) found notoplate elongation to be responsible for most of the shaping of the neural plate up to the keyhole stage. After the keyhole stage, the plate rolls into a tube, and exactly during that time the rate of notoplate elongation increases ten-fold (19). Chick embryos have a similar rapid rate of neural plate elongation during tube closure (16-18). As suggested by Jacobson (16, 17, 22), notoplate elongation can help to roll the neural plate into a tube, much as a rubber sheet folds into a tube when stretched along a line (16, 17 22). However, analyses discussed below suggest that this is not the only mechanism that drives the folding of the plate into a tube.

Jacobson and Tam (24) found that while the brain plate of the mouse embryo rolls into a tube, the neural folds elongate considerably, but the midline does not. This led us to look more closely at the neural folds of the newt embryo during neural tube formation. We find that the neural folds begin to elongate considerably just at the stage when the plate begins to fold, and increase in length by 30% by the time the neural tube is completely formed. During this same period, the midline increases by 35%.

The boundary of the neural plate and epidermis, which becomes the neural fold and crest, could be another organizing boundary for elongation of the neural plate. We assume

that the epidermal cells have inactive basal and apical surfaces, and so can move only tangentially amongst one another. The adjacent neural plate cells are basally (and perhaps laterally) active, and as they tractor, their faces that contact the epidermal cells adhere to them. This would cause the neural plate cells to crawl beneath the epidermal cells, and as their adhering faces become inactived, to preferentially tractor more neural plate cells after them, elongating the plate edge, and drawing epidermal cells along the edge. This would crowd even more adhering plate cells into the forming neural folds. Since the plate cells are tethered apically, their tractoring would tend to pull them under the epidermis, while buckling that epidermis up into an arch, and stretching the plate cells doing the crawling. As the plate cells stretch beneath the epidermis, their apical surfaces will constrict much as the trailing tail of a migrating fibroblast is constricted. Some of the plate cells at the edge of the plate may have their apical surfaces reduced to a point, then pull free of the epithelium and become neural crest cells. Compare Fig. 9, which illustrates these proposed events schematically, to the sections in Fig. 5.

Preliminary Simulations of the Cortical Tractor Model

The assumptions of the cortical tractor model seem to lead quite naturally to the above scenario for neurulation. However, it is important to verify the verbal descriptions we have given by mathematical analyses and/or numerical simulation. Otherwise, we have no way to prove that our verbal scenarios will actually evolve from the mechanical assumptions of the model. These simulations are exceedingly complex, and we have approached them piece-meal, simulating only restricted aspects of the model. A complete simulation of the cortical tractor model is in progress, however, we report here a number of related simulations that give us confidance that the verbal descriptions we have given are indeed valid.

The Mechanical Properties of Cytogel

The basis for the cortical tractor model lies in the physicochemical properties of the cortical cytogel in each epithelial cell. We have constructed a series of detailed models of actomyosin gels, and have applied them to a number of cell motility phenomena (25-31). We feel confident that

our models of cytogel behavior, while certainly not yet a complete description, nevertheless capture enough of the relevant physics to warrent the assumptions of the cortical tractor model. In particular, the basic cycle of ionically triggered solation, osmotic expansion and active contraction appears well verified in a number of experimental systems.

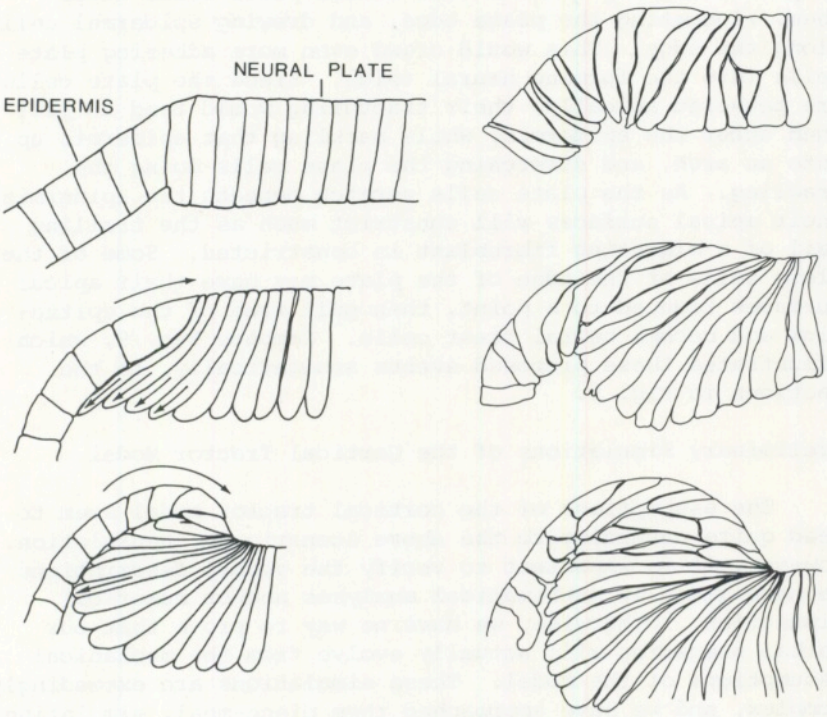


FIGURE 9. The diagrams at left illustrate how we interpret events at the epidermis/neural plate boundary. Neural plate cells tractor on the bottoms of the epidermal cells, pulling them into a fold, and at the same time stretching the neural plate cells until their apical surfaces are points, or even become released. Neural plate cells interdigitate along the boundary (not shown) to elongate the neural folds, and their tractoring produces a rolling moment toward the midline and lifts the folds up out of the plane. The drawings at right are tracings of cells from cross sections of newt neurulae at stages 15, 16, and 17. Actual cell shapes can be compared to the diagrams.

Motile Cells Can Exert Considerable Forces on Their Surroundings

A basic assumption of the cortical tractor model is that the motile behavior of epithelial cells is identical to that of mesenchymal cells, except for the constraint on their motion by their apical junctional structures. The cortical tractor we have imputed to epithelial cells is identical to that observed in motile mesenchymal cells *in vitro*. That motile cells can exert considerable forces on their own is well established (32, 33), and we have assumed that this property is retained by motile cells that are tethered into an epithelial configuration. Thus the neural plate cells should be amply strong to roll the tube, as shown in Fig. 9.

Simulations of Epithelial Sheet Folding

Odell, et al. (14) simulated the rolling of the neural tube employing a model for epithelia wherein the only force generated by the cells was developed by apical microfilament bundles. These simulations demonstrated that apical contraction was sufficient to mimic many aspects of epithelial folding phenomena. In this paper, we have relegated apical contraction to a secondary role in contrast to active crawling; however, it is likely that both mechanisms operate during neurulation. Indeed, our cortical tractor model provides a mechanism for assembling and concentrating microfilament contractile machinery at cell apices, and thus subsumes and enhances our previous work.

Does Elongation of the Neural Folds and/or Notoplate Roll the Neural Plate into a Tube?

During neurulation, not only do the neural folds close into a tube, but they rise out of the plane of the neural plate a considerable distance. Previously, Jacobson and Tam (24) and Jacobson (16, 22) have suggested that both rising and rolling can result from the elongation of the neural folds and/or the notoplate. In order to investigate the relative importance of elongation of the neural folds in comparison to the rolling action of the crawling plate cells, we employed a finite element program developed by L. Chang (to appear). Fig. 10 shows a simulation wherein the folds elongated, but no other forces acted. The elongation develops transverse compressive stresses (Poisson buckling)

sufficient to raise the folds upward. However, we were unable to cause the folds to close into a tube. We concluded that an active bending moment must be developed by the plate in order to roll completely into a tube. These bending moments can be generated by crawling, according to the cortical tractor model, or by apical constraction, as previously assumed by Odell, et al. (14) in their simulations, or by both mechanisms acting in concert.

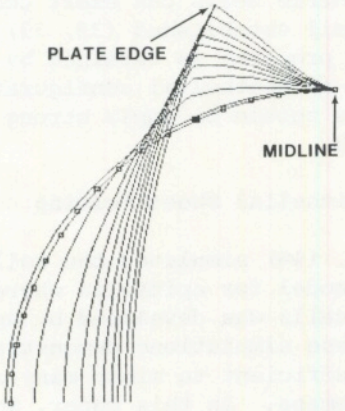


FIGURE 10. Results of simulations in which the only force applied is the elongation of the plate edge (neural fold). The simulations were done using only one quadrant, as shown, since the other quadrants may be reconstructed by symmetry.

Does the Neural Tube Roll From the Centerline Outward, Or From the Edge of the Plate Inwards?

There is also the question of whether the neural tube rolls from the centerline outwards, or from the neural folds inward, or both. Fig. 11 shows a comparison between the two possibilities. The simulations wherein rolling commences at the folds and proceeds inward look more realistic, reinforcing our suspicion that the bending moments that roll the neural plate mostly commence at the plate edge and propagate toward the centerline. However these simulations do not address the issue of whether the bending moments are generated by activation of the basal and lateral faces of the plate cells, or by apical contraction, or both.

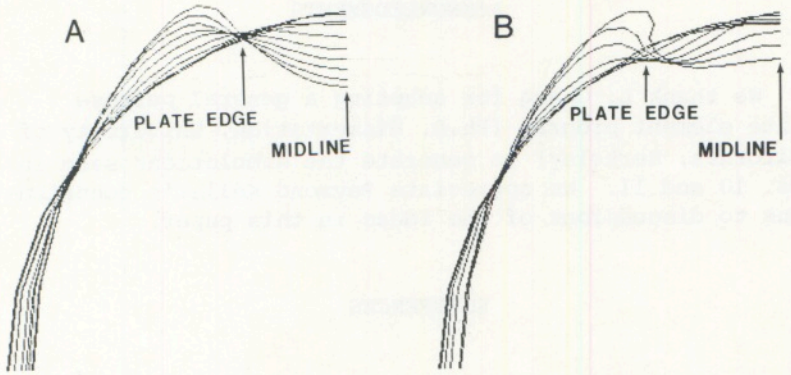
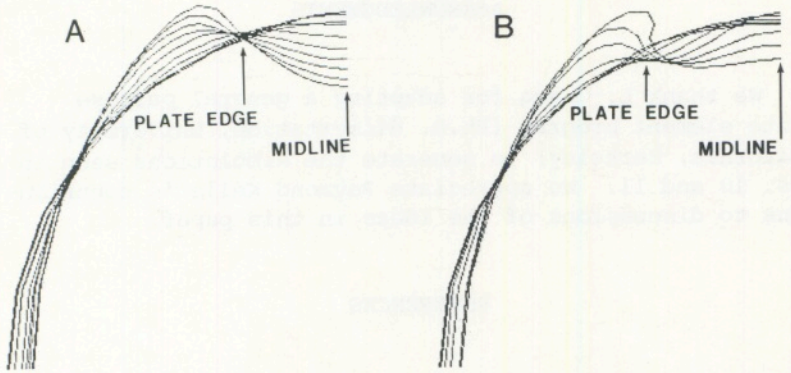


FIGURE 11. A) Results of simulations in which the imposed rolling moment is from the midline to the plate edge. B) Results of simulations in which the imposed rolling moment is from the plate edge to the midline. In neither case is any elongation imposed. These simulations were done using only one quadrant, as shown, since the other quadrants may be reconstructed by symmetry.

It may be difficult to distinguish whether the contraction proceeds from the centerline to the edge, or from the margins inward, because in the previous simulation of Odell et al. (14), the cells were triggered at the centerline, yet rolling commenced at the margins. This was due to purely geometrical effects.

The tractor motion suggested in the cortical tractor model for epithelial morphogenesis can create quite complex epithelial foldings, and many other epithelial movements may be generated by this mechanism that both epithelial cells and mesenchymal cells hold in common. Key features of the cortical tractor model, as applied to epithelia, are that cell movements and neighbor changes can occur amongst the cells of an epithelium without breaking the apical seals; when different populations of cells exist within the tissue layer, each with distinctive adhesive characteristics (or with the ability to contact inhibit one another), then the tractor movements of the cells may be organized to produce orderly epithelial morphogenesis.



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