Published in final edited form as:
Birth Defects Res C Embryo Today. 2012 June ; 96(2): 132–152. doi:10.1002/bdrc.21013.

Computational Models for Mechanics of Morphogenesis

Matthew A. Wyczalkowski[1,*], **Zi Chen**[1,*], **Benjamen A. Filas**[2], **Victor D. Varner**[3], and **Larry A. Taber**[1]

[1]Department of Biomedical Engineering, Washington University, St. Louis, MO 63130

[2]Department of Ophthalmology and Visual Sciences, Washington University School of Medicine, St. Louis, MO 63130

[3]Department of Chemical and Biological Engineering, Princeton University, Princeton, NJ 08544

Abstract

In the developing embryo, tissues differentiate, deform, and move in an orchestrated manner to generate various biological shapes driven by the complex interplay between genetic, epigenetic, and environmental factors. Mechanics plays a key role in regulating and controlling morphogenesis, and quantitative models help us understand how various mechanical forces combine to shape the embryo. Models allow for the quantitative, unbiased testing of physical mechanisms, and when used appropriately, can motivate new experimental directions. This knowledge benefits biomedical researchers who aim to prevent and treat congenital malformations, as well as engineers working to create replacement tissues in the laboratory. In this review, we first give an overview of fundamental mechanical theories for morphogenesis, and then focus on models for specific processes, including pattern formation, gastrulation, neurulation, organogenesis, and wound healing. The role of mechanical feedback in development is also discussed. Finally, some perspectives are given on the emerging challenges in morphomechanics and mechanobiology.

Keywords

morphogenesis; development; morphomechanics; mechanobiology

1 Introduction

The importance of mechanics in developmental biology is well established. Biomechanical forces are the bridge that connects genetic and molecular-level events to tissue-level deformations that shape the developing embryo (Koehl, 1990; Taber, 1995). In addition, feedback from the cellular mechanical environment affects gene expression (Farge, 2003) and differentiation (Engler et al., 2006). Hence, mechanical forces are not only a proximal cause of morphogenesis, but they also play a regulatory role (Vogel and Sheetz, 2006; Wozniak and Chen, 2009; Mammoto and Ingber, 2010).

During the last two decades, there has been increasing appreciation for the value of computational models in studying the mechanics of morphogenesis (Taber, 1995; Brodland, 2006; Rauzi et al., 2008; Davidson, 2008). This interest is being facilitated by dramatic increases in computing power and the growing availability of quantitative experimental data,

Corresponding Author: Zi Chen, Department of Biomedical Engineering, Washington University, St. Louis, MO 63130, Phone: (314) 935-9563, Fax: (314) 935-7448, chen.z@seas.wustl.edu.
*These authors contributed equally to this work.

which are needed to inform and test models (Grashoff et al., 2010; Martin et al., 2010; Martin, 2010; Sato et al., 2010; Meng and Sachs, 2011; Rauzi and Lenne, 2011). It is important to recognize, however, that mathematical models never can include all of the complexities inherent in biological systems. Rather, they are utilitarian, *ad hoc* constructions designed to help understand some aspect of the system.

This review is intended to give readers an overview of various aspects of modeling efforts in morphogenesis. We first provide a brief summary of some of the mechanical theories for morphogenesis, the principles of constructing computational models, and the challenges encountered therein. We then turn our attention to a selection of morphogenetic phenomena and describe models that have been used to simulate the underlying mechanisms. Finally, we provide our perspective on the challenges and future directions in modeling morphogenesis.

Admittedly, this topic covers a vast literature, so it is impossible to include everything. For example, chemical and molecular biological effects are crucial topics, but they are not considered in detail here. In addition, although all models must be supported by experiments, only a limited set of experimental work that has stimulated or assisted relevant modeling efforts is included in this review. Also, several important topics, such as bone development and cell migration, are not addressed. We do not claim expertise in all areas discussed and apologize for any inappropriate omissions or misplaced emphasis.

2 Background

While the role of mechanics in morphogenesis has been recognized since the late 19th century, in the latter half of the 20th century its impact on the field of organismal development was eclipsed by the prominence and success of genetic and molecular biological approaches (Davidson and Keller, 2007; Hutson and Ma, 2008; Keller et al., 2008). Recent advances in experimental and computational technologies, however, have revived interest in the mechanical aspects of morphogenesis. In this section, we briefly summarize basic mechanisms of embryonic development and present an overview of the main theories and types of models used to simulate them.

2.1 Basic Morphogenetic Mechanisms

There are two principal tissue types in the developing embryo, mesenchyme and epithelia, which are distinguished by the interactions of cells with each other and with the extracellular matrix (ECM) (Trinkaus, 1984; Bard, 1990; Davies, 2005). Mesenchymal tissue contains significant ECM within which cells are embedded. Such cells form adhesive contacts within the ECM via focal adhesions, and exert strong traction forces which pull on and deform the matrix (Harris et al., 1980). Cellular motility typically involves proteolysis and remodeling of the matrix with only transient cell-cell contacts (Friedl, 2004). The dispersion, migration, and condensation of mesenchymal cells is guided by a variety of signals, including chemical gradients (chemotaxis), adhesion gradients (haptotaxis), and stiffness gradients (durotaxis) (Davies, 2005).

Epithelia are polarized sheets of cells with an apical side facing away from the body (or toward an inner lumen) and a basal side contacting the basal lamina and ECM. The cells in an epithelium are attached to one another apically through belt-like adherens junctions, where extracellular E-cadherin modulates cell-cell adhesion and the cytoskeleton facilitates the transmission of forces through and between cells (Lodish et al., 2004). Because of strong cell-cell contacts, epithelia move collectively as sheets during morphogenesis (Bard, 1990).

In some cases, coordinated contraction of the apical actomyosin bands bends an epithelium by constricting the apex and giving the cells a wedge-like profile (Martin, 2010). Besides

passive stretching, planar changes in geometry can be caused by cell intercalation, as cells exchange neighbors in the plane of the epithelium. This process produces convergent extension, with the epithelium shortening along one axis and extending in the other (Keller et al., 2008). In addition, epithelia are said not to tolerate a free edge (Trinkaus, 1984), and, in the case of embryonic wound healing and certain morphogenetic events, develop supracellular actomyosin cables at the leading edge. These cables contract to close the wound with a “purse string” like mechanism, in some cases aided by filopodia. Other epithelial morphogenetic mechanisms include branching of tubes, fusion of sheets, and inflation of sealed vesicles via hydrostatic pressure (Davies, 2005). Both mesenchyme and epithelia can change shape and volume through cellular proliferation, growth, and apoptosis.

2.2 Theories and Models for Morphogenesis

A number of quantitative theories have been proposed to account for morphogenetic phenomena, where mechanical and biochemical processes interact in complex ways to sculpt a developing organism (Urdy, 2012). The role of such theories is to help explain observed behavior based on biologically and physically plausible mechanisms, and to make experimentally testable predictions (Maini, 2004). In this section, we first briefly discuss two early theories, largely based on biochemistry, which have had a major impact on the field. While the remainder of this review focuses primarily on mechanical theories and models, it is important to keep in mind that developmental processes must simultaneously obey the laws of mechanics, thermodynamics, and biochemistry.

An early theory for morphogenesis was proposed by Turing (1952), who suggested that spatial pattern is created by reactions between two morphogens — a slowly diffusing short-range activator and a rapidly diffusing long-range inhibitor (Kondo and Miura, 2010). Since the appearance of that paper, a number of investigators have used variations of this reaction-diffusion model to study pattern formation (Gierer and Meinhardt, 1972; Murray, 2003a; Kondo and Miura, 2010). Although morphogens are generally understood to play a prominent role during development (Wolpert, 1969; Howard et al., 2011), from an experimental perspective the applicability of the Turing mechanism to pattern formation has been limited (Kondo and Miura, 2010).

The second theory involves cell sorting. The Differential Adhesion Hypothesis (DAH), proposed by Steinberg (1963), is based on the observation that embryonic cells, when disaggregated and allowed to recoalesce, behave in ways that strikingly resemble the behavior of immiscible fluids. Cells of one type aggregate so as to minimize their surface area, and different cell types, when mixed together, sort themselves into distinct homogeneous clusters, one of which may engulf the other. Steinberg (1963) postulated that this behavior is governed by differences in cell-cell adhesion, and that cell mixtures undergo a phase separation such that the final configuration corresponds to the minimum interfacial and surface free energies (Forgacs and Newman, 2005; Lecuit and Lenne, 2007). A wide range of predictions made by the DAH have been experimentally validated (Steinberg, 2007), and a number of computer simulations based on this theory or its modifications have supported its conclusions (Brodland, 2004 and references therein).

In both of these theories, tissue deformation is generally regarded as a downstream consequence of chemical patterns generated by other means. In mechanical theories, on the other hand, deformations themselves are the pattern, and the interplay of stress, material properties, and deformation becomes the central consideration (Koehl, 1990). For more than a century, investigators have speculated about and proposed numerous theories regarding physical mechanisms of morphogenesis. Many early researchers used physical simulacra of embryonic tissue to test their ideas by analogy (Weiss, 1939; Thompson, 1942). Lewis (1947), for instance, constructed a physical model of an epithelium using brass bars and

rubber bands to investigate hypotheses about the mechanical forces driving invagination. In the modern era, computational models have largely replaced physical models, but the emphasis on mechanical forces as the proximal cause of morphogenesis remains (Clausi and Brodland, 1993; Taber, 1995; Hutson and Ma, 2008).

Perhaps the most elementary biomechanical approach is to model tissue as a network of 1-dimensional (1D) elastic elements (springs), viscous elements (dashpots), and contractile elements (Koehl, 1990). These models can provide insight into basic mechanical behavior, but generally do not include other potentially important characteristics, such as resistance to shear.

More commonly, mechanical behavior of soft tissue is analyzed using the principles of continuum mechanics, whereby tissue is treated as a continuous material rather than a collection of discrete particles. The concepts of stress (force per unit area) and strain (relative change in length or angle) are central ideas in the theory. These quantities must obey equilibrium, geometric compatibility, mass conservation, and constitutive (stress-strain) equations (Lai et al., 2009).

Oster et al. (1983) and Murray and Oster (1984) presented a continuum mechanics-based theory that can be used to simulate both mesenchymal and epithelial morphogenesis. In this theory, a tissue is treated as a mixture of cells and matrix. The basic continuum equations are modified to allow for cell migration and proliferation, active stresses exerted by cells on the matrix, and passive stresses exerted by the matrix on the cells. Most applications of this theory have assumed that deformations are small, which is not a valid approximation for many morphogenetic processes.

To handle arbitrarily large deformations, the nonlinear growth theory of Rodriguez et al. (1994) has been used to effectively simulate a number of morphogenetic processes (Muñoz et al., 2007; Taber, 2009; Varner et al., 2010; Ambrosi et al., 2011; Filas et al., 2012). For example, active contraction can be simulated by negative growth with a corresponding increase in stiffness. Here, the kinematic (strain) equations of continuum mechanics are modified to include volumetric growth. The basic idea in this theory is that uniform growth of tissue produces a change in volume without generating stress. However, growth restricted by external supports or surrounding tissue, e.g., differential growth, will produce stress. One limitation of this theory is that it does not distinguish between growth caused by cell division and that caused by hypertrophy (Taber, 1995).

Models based on the Murray-Oster and growth theories described above treat embryonic tissue as an elastic or viscoelastic solid. Another class of continuum models assumes that the tissue behaves as a viscous liquid. Indeed, for the relatively long time scales of morphogenesis, embryonic tissue can undergo permanent deformation analogous to viscous flow (Phillips et al., 1977; Forgacs et al., 1998), and cells in some early morphogenetic processes move in eddy-like patterns. Lubkin and Li (2002) treat both epithelia and mesenchyme as fluids of different viscosities, while Pouille and Farge (2008) and Fleury (2011) model morphogenetic movements in terms of classical two-dimensional hydrodynamical flow.

The large and complex changes in geometry that occur during embryogenesis pose a challenge to investigators aiming to solve problems in morphomechanics, and finite-element methods are a key tool for obtaining solutions to continuum mechanical problems. In this method, a complex body is divided into geometrically simple subdomains (finite elements) over which the strain field can be approximated by simple functions. The governing equations are solved for the entire assemblage of elements by enforcing compatibility and

continuity between elements (Reddy, 1993; Bathe, 1996). Such tools and theories have been applied to a wide range of morphogenetic processes, as we now describe.

3 Gastrulation

Gastrulation is a highly orchestrated event featuring dramatic cell deformations and migrations to establish the multi-layered body plan of the embryo. This process establishes the three principal germ layers (endoderm, ectoderm, and mesoderm). The process of gastrulation varies across species, possibly reflecting differences in environment and early embryonic morphology (Bard, 1990; Leptin, 2005).

Although much has been learned about the genetic and molecular aspects of gastrulation (Leptin, 1995; Nikolaidou and Barrett, 2004; Dawes-Hoang et al., 2005), less is known about the mechanics of this process. To help elucidate this important biomechanical event, a number of theoretical models have been proposed for gastrulation in organisms such as sea urchin and *Drosophila*. The initial stages of sea urchin gastrulation involve axisymmetric invagination of a fluid-filled spherical shell (Davidson et al., 1995), while gastrulation (or ventral furrow formation) in *Drosophila* involves the creation of a groove along the relatively flat side of an ellipsoidally shaped embryo (Lye and Sanson, 2011). These differences in geometry influence the mechanics considerably (Conte et al., 2008; Taber, 2008).

In one of the earliest such investigations of morphogenesis, Odell et al. (1981) presented a 2-dimensional (2D) model for an epithelium that treats each cell as a viscoelastic truss-like element with a contractile apex. In a circular ring of cells, a specified contraction in one cell apex (simulated by a shortening of the stress-free length) stretches neighboring cells, which themselves contract if stretched beyond a critical amount. With appropriately chosen parameter values, this response produces a wave of contraction that generates a local invagination, which resembles morphogenetic processes such as ventral furrow formation and neurulation. While the authors suggest that this model can be used to simulate sea urchin gastrulation, a cylindrical model is not appropriate for the spherical sea urchin embryo. Nevertheless, this model was ahead of its time, and the mechanical feedback it incorporates has since gained increasing attention in studies of morphogenesis (Beloussov, 1998, Beloussov, 2008; Pouille et al., 2009; Kornikova et al., 2010).

Regional variations in mechanical properties can strongly affect morphogenetic shape change. For example, an epithelium that spreads uniformly on a relatively soft substrate may buckle if constrained by a stiffer substrate. However, relatively little is known about the mechanical properties in embryos, potentially making it difficult to distinguish between multiple mechanisms that produce similar shapes. To illustrate this point, as well as to provide guidance for future experiments, Davidson et al. (1995) used spherical finite element models to test five possible mechanisms for sea urchin invagination (Fig. 1):

1. apical constriction/basal expansion within a circular region;
2. cell tractoring, as cells in a ring at the outer edge of the invaginating region emit protrusions (rods) that contract and pull the ring radially inward, buckling the cells inside the ring;
3. circumferential contraction of an actomyosin ring surrounding the invaginating region buckles cells inside the ring;
4. apico-basal contraction causing cells in the invaginating region to spread and buckle due to constraints from surrounding tissue; and

5. regional swelling in the apical lamina (located between the cells and an outer hyaline layer), with constrained expansion causing the invaginating region to bend inward.

Each hypothesis was carefully evaluated relative to available data, and ranges of material parameters were determined that would be required for each model to function as proposed. In a subsequent study, measurements appeared to rule out the apical constriction and contractile ring mechanisms (Davidson et al., 1999).

Recently, the Miodownik group has used the continuum growth theory of Rodriguez et al. (1994) to simulate active changes in cell shape in a series of models for ventral furrow formation (Muñoz et al., 2007; Conte et al., 2008, 2009). In these models, cell dimensions change by specifying positive or negative growth along particular directions, while cell wedging occurs via an apico-basal growth gradient. Taken together, these models include apical constriction and basal elongation in the invaginating mesodermal region, and apico-basal shortening with transverse extension in the ectoderm outside this region. The models were used to study the effects of various combinations of these cell shape changes, as well as 3-dimensional (3D) ellipsoidal geometry and constraints imposed by the surrounding vitelline membrane and internal fluid.

The 3D ellipsoidal model yielded global shape changes similar to those observed in experiments, and confirmed the important role of the vitelline membrane and yolk in gastrulation (Fig. 2). Importantly, invagination in the 3D model was found to be less sensitive to variations in the ratio between apical constriction and apico-basal elongation than the 2D model of a transverse section, suggesting that the 3D model is more robust to perturbations (Conte et al., 2008). Moreover, this 3D model revealed that yolk flow would be generated from the center of the embryo toward the anterior and posterior ends, leading to global compression and expansion of the embryo, resembling the motion of an accordion.

Using a variation of the previous 2D model, Conte et al. (2009) investigated multiple combinations of invagination mechanisms, including ectodermal spreading, and correlated cell shape changes, with regional data on gene expression. While a redundancy in mechanisms leads to more robust development, this feature also makes it very difficult, if not impossible, to identify the principal driving forces in a morphogenetic event. Additional experiments, perhaps similar to those suggested by Davidson et al. (1995), can be used to rule out some of the possible mechanisms (Conte et al., 2009). Mutations that affect certain mechanisms can also help determine how local cellular or molecular level activities affect global changes in shape (Davidson, 2008; Martin et al., 2010).

Recently, Allena et al. (2010) presented a similar 3D finite-element model for a *Drosophila* embryo, using essentially the same growth theory to simulate morphogenesis. The authors simulated three concurrent morphogenetic movements in early gastrulation: ventral furrow formation, cephalic furrow formation, and germ band extension. Their results suggest that the number of active deformation modes could be less than suggested by experiments, e.g., apical constriction alone, without apico-basal elongation, could drive invagination.

In the models discussed thus far, any molecular underpinnings for the most part are included only in a cursory manner. Two recent models take important steps toward a more explicit accounting of molecular mechanisms. In the first study, Sherrard et al. (2010) used new data to propose a two-step process for invagination during ascidian gastrulation. Their 2D model for the embryo consists of coupled layers of endoderm and ectoderm, and an explicit dependence on Rho, a cellular regulatory signal. First, Rho-dependent apical contraction makes the endodermal cells wedge-shaped and pulls the ectoderm around them (Fig. 3). Then, a Rho-independent basolateral contraction shortens and spreads the endodermal cells,

while their apical sides remain contracted and relatively stiff. This second step drives invagination.

In the second study, Driquez et al. (2011) investigated how recently identified contractile oscillations in an epithelium (Martin et al., 2010) can lead to a ratchet-like but sustained contractile force. These two contractile modes are associated with the molecular signals Snail and Fog, respectively, with the latter mediated by stretch. Accordingly, the model is based on an idea similar to the stretch-activation mechanism proposed by Odell et al. (1981), whereby a cell apex contracts when stretched beyond a specified threshold. These researchers used relatively simple 1D and 2D spring-dashpot models to represent the apical side of an epithelium; invagination was not modeled explicitly. In the simulation, a region containing a specified stochastic distribution of relatively small oscillating contractions eventually coordinates in phase to stretch a single cell beyond its critical length. This triggers a sustained contraction of that cell, and these responses eventually transform into a collective constriction. In the 2D model this mechanism produces a wave of contraction similar to that observed *in vivo*.

4 Cell Rearrangement and Pattern Formation

The last two models of the previous section are examples of multi-scale models for morphogenesis, as they consider tissue-level effects in response to both cellular and molecular-scale events. Multiscale models have also been proposed for studies of cell rearrangement within tissue. Here, we consider specific applications to cell sorting, convergent extension, and branching morphogenesis. All of these processes generate pattern in the embryo.

4.1 Cell Sorting

According to the Differential Adhesion Hypothesis (Steinberg, 1963), differences in adhesion strength between cells drive the sorting and engulfment of one tissue by another. In a finite-element cell-level model, Chen and Brodland (2000) extended this hypothesis by including contractile forces along cell boundaries (see also Brodland, 2002). Here, individual cells are represented as two-dimensional polygons, with the mechanical contributions of apical actomyosin contraction and intercellular adhesion represented collectively by one equivalent interfacial tension acting tangent to the cell edge. The cytoplasm is taken as a viscous fluid. In this model, strengthening adhesion works to lengthen a boundary, while additional contraction shortens it. The computer algorithm keeps track of the changing shapes and positions of the cells. In simulations for various boundary conditions, applied loadings, and mixtures of cell types, the model produces dynamic rearrangements of cells and changes in cell geometry that are in general agreement with experimental observations for cell remodeling and sorting behaviors (Fig. 4) (Chen and Brodland, 2000; Brodland and Chen, 2000).

4.2 Convergent Extension

During convergent extension (CE), epithelia narrow in one direction and extend in another (Keller et al., 2000; Brodland, 2006; Lye and Sanson, 2011). This occurs, for example, during gastrulation, neurulation, and epiboly. In many instances, cell rearrangement in the form of intercalation drives this process, e.g., cells move mediolaterally between neighboring cells, causing the neighbors to separate and extend the tissue in the axial direction.

As a precursor to later models developed by Brodland and colleagues, Weliky and Oster (1990) proposed a 2D network model for cell rearrangement, with cells represented by interconnected polygons. In this model, intracellular pressure drives swelling and

protrusions, and tension is generated at cell boundaries. The motion of each node depends on the forces exerted on it, and cell geometry is updated to ensure geometric compatibility. This model was used to simulate epiboly in fish, with the results showing that stress relaxation could result from cell rearrangements in the epidermal enveloping layer. Soon afterwards, Weliky et al. (1991) modified this model to include other features of cell motility, including directional persistence, tension-induced inhibition of protrusion, and contact inhibition. This improved model was then used to study tissue extension, cell rearrangement, cell-tissue boundary interactions, and parallel cell alignment.

At the cellular level, some experimental data suggest that cells emit lamellipodia which squeeze between two neighbors to contact a more distant cell (Brodland, 2006; Keller et al., 2008). The protrusions then contract, pulling the originating and target cells together to drive intercalation and CE. Using this idea, Brodland and Veldhuis (2006) modified their cell-level finite-element algorithm to simulate this lamellipodial activity in a model for CE. The results illustrate possible “mechanical pathways” of CE across different length scales, i.e., lamellipodia produce directional, contractile forces at a sub-cellular level that drive cell intercalation, which in turn results in global convergence and extension of epithelia.

Using experiments and a similar model, Rauzi et al. (2008) investigated the mechanics of germ band elongation (GBE) in *Drosophila* embryos and showed that cell intercalation could be driven by anisotropic cortical tension. The cell networks in this model are 2D and the cell contact lines form junctions. The total energy of the network is the sum of line energy (due to line tension), area elastic energy (caused by fluctuating cell area), and cortical elastic energy (stretching of cell periphery). During intercalation, two three-way vertices are brought into contact to minimize energy, generating a temporary four-way vertex, followed by a topological change in the junction called a T1 transition, which drives the cell shape changes. In the simulations, T1 transitions occur when the length between adjacent junctions becomes sufficiently small, and the cell network eventually reaches an equilibrium configuration where GBE is produced. Here, anisotropy of cortical tension is shown to control cell shape changes during intercalation. This study suggests that anisotropy of cortical tension at the subcellular level, regulated by myosin II activity, can drive the coordinated cell shape changes and tissue deformations during CE, and that contractile fluctuations observed *in vivo* can play an important role in intercalation.

4.3 Branching

Experiments have suggested that mechanics plays an important role in the initiation and progression of branching, which occurs during the formation of blood vessels, organs such as the lung and kidney, and mammary and salivary glands (Davies, 2005; Affolter et al., 2009). For example, *in vitro* experiments indicate that branching is triggered in regions of high mechanical stress (Nelson et al., 2005; Gjorevski and Nelson, 2010). Moreover, cells cultured on a gelled substrate exert tractions on the matrix and surrounding cells, eventually forming aggregate clusters between which lines of tension appear (Fig. 5A). Cells then migrate along these lines of tension, forming cellular cords and leading to the development of a network structure resembling capillary blood vessels (Vernon et al., 1995; Vailhé et al., 2001; Murray, 2003b). Such patterns are observed for a wide variety of cells and substrates, and since the geometric characteristics of the network are sensitive to substrate stiffness, mechanical effects appear to play a major role in this process (Murray, 2003b).

Manoussaki et al. (1996) presented a mechanical model for vascular network formation based on the Murray-Oster theory for mesenchymal morphogenesis (Oster et al., 1983). In this model, cells exert an isotropic traction whose magnitude rises, peaks, and decays with cell density. The viscoelastic matrix, which is anchored to a stiff substrate (the dish), carries cells along as it deforms, and the cells actively crawl on the matrix in a direction biased by

the orientation of matrix strain. Manoussaki et al. (1996) found that patterns form only if the initial cell density is not too high, and the strength of the traction exerted by each cell exceeds a critical value. The patterns generated in numerical simulations of this system resemble those observed *in vitro*, both in their temporal evolution and general appearance (Fig. 5B).

A number of researchers have extended this model (see Ambrosi et al., 2005, for a review). Namy et al. (2004) included the effects of haptotaxis and the fibrous nature of the substrate. Combining experiments with finite-element simulations, the authors explored the effect of cell traction forces and the mechanical resistance of the matrix on network topology, and were able to reproduce patterns resulting from a thickness gradient in the substrate. Holmes and Sleeman (2000) investigated the role of cellular traction and matrix viscoelasticity on the vascularization of tumors. Together with haptotaxis, their model includes a diffusing tumor-generated chemical signal which induces chemotaxis and mitosis in the endothelial cells. The model provides excellent agreement with experimental results and captures a number of relevant features of angiogenesis.

Tosin et al. (2006) distinguished between mesenchymal motion of cells, where matrix adhesion and traction play a dominant role, and amoeboid-type motion, which is characterized by its speed, directional persistence, and chemotactic nature (Friedl, 2004; Ambrosi et al., 2005; Painter, 2009). In some experimental protocols (Serini et al., 2003), cells undergo amoeboid-type motion for several hours before anchoring to the substrate and proceeding with network formation as discussed above. To account for the two types of motion and the transition between them, Tosin et al. (2006) constructed a model consisting of a cell layer and a substrate layer. The cell layer, modeled as an elastic fluid, is initially independent of the substrate to simulate amoeboid-type chemotactic motion; a coupling term then transfers stress from the elastic substrate to the cell layer, thus capturing the transition to mesenchymal motion. The model yields realistic network topologies and predicts a characteristic length that depends on diffusion and decay of a chemical signal, a feature that purely mechanical models cannot capture (Ambrosi et al., 2005).

Computational models have also been proposed for the branching of epithelial tubes in mesenchymal tissue. While the epithelium is known to play an important role in branching morphogenesis, Wan et al. (2008) presented a model that explored the possible contribution of active contraction of the mesenchyme. Both tissue types were modeled as fluids of different viscosities, with contractile cells embedded in the mesenchyme. Using a 3D model of a spherical lobe of epithelial tissue, the authors found that a narrow band of contractile mesenchyme is capable of generating a cleft, a critical step in initiating a branch point, and that the relative viscosities of the mesenchyme and epithelium have a strong effect on cleft geometry.

5 Head Fold Formation

The early avian embryo is initially organized as a flat disc of cells which sits atop the yolk at the animal pole of the egg (Gilbert, 2000). The first structure to break this planar geometry is the head fold, which arises as a crescent-shaped fold at the anterior end of the elongating neural plate (NP) (Fig. 6A,B). The head fold constitutes the first bounding body fold (Patten, 1971) and initiates both heart and brain development (Stalsberg, 1969; Schoenwolf and Smith, 2000). Although researchers have long speculated about the physical forces involved in head fold morphogenesis (Balfour, 1881; Shore and Pickering, 1889; Stalsberg and DeHaan, 1968), the problem has only recently been quantitatively investigated (Varner et al., 2010).

Varner et al. (2010) employed a continuum mechanical framework in combination with the Rodriguez et al. (1994) theory for volumetric growth to model the mechanics of head fold morphogenesis. Their results suggest that, much like the problem of dorsal closure in *Drosophila* (Kiehart et al., 2000), multiple forces work in tandem to drive the deformations observed during head fold formation. These forces are ectodermal in origin, and include several physical mechanisms typically associated with neurulation: convergent extension in the NP, cell wedging along the NP border, and transverse shortening outside the NP (Colas and Schoenwolf, 2001; Lawson et al., 2001; Moury and Schoenwolf, 1995).

These driving forces were simulated in a 3D model consisting of two parallel layers representing the blastoderm and the vitelline membrane, separated by a narrow space (Fig. 6C). With the material properties estimated by microindentation, the model yields tissue morphology consistent with observations (Fig. 6D). In addition, stress and strain fields given by the model are in reasonable agreement with experimental measurements, and the model is able to predict the abnormal head fold geometries produced by local dissections that relieve stress (Varner et al., 2010). This study shows that all three driving forces, as well as contact with the vitelline membrane, are required for proper head fold formation.

6 Neurulation and Brain Development

Following head fold development, the process of neurulation begins, as a longitudinal invagination in the neural plate creates the neural tube. The anterior end of this tube then expands to create the primitive brain, while the rest becomes the spinal cord. The brain next subdivides into a series of vesicles and begins a period of rapid growth driven by increasing cerebrospinal fluid pressure (Desmond and Jacobson, 1977). Cells lining the ventricles proliferate, differentiate, and migrate from the inner cavity to form the characteristic layers of the mature brain. During the later stages of development in most large mammals, the cerebral cortex folds into a highly convoluted shape to accommodate this rapid expansion.

Changes in brain size and shape are driven by the dynamic interplay between mechanical forces and cellular processes. For example, during early brain development, rising cerebrospinal fluid pressure increases cell proliferation rates (Desmond et al., 2005), while perturbations in wall stress induce abnormal cytoskeletal activity (Filas et al., 2011). Proper brain development requires the careful regulation of mechanical forces, and computational models have emerged as useful tools to quantitatively test whether hypothesized morphogenetic mechanisms are consistent with physical law.

6.1 Neurulation

Neurulation has been one of the most intensively studied problems in the developing embryo. Due to the prevalence of congenital defects resulting from impaired neural tube closure (e.g., spina-bifida and anencephaly), as well as the complex physical processes involved in shaping the tube, this problem has received considerable attention from the modeling community.

As discussed earlier, Odell et al. (1981) used a 2D model for epithelial invagination to simulate neurulation, showing that certain parameter values produce a closed tube. Later, Brodland and colleagues (Clausi and Brodland, 1993; Brodland and Clausi, 1994) developed 2D cell-level models for neurulation using apical constriction as the primary driving force. These models were used to test the effects of varying epithelial thickness, apical force, and axial elongation, as well as the effects of external forces on neural tube morphology (Brodland and Clausi, 1995). While not directly verified with experiments, some elements of neural tube morphogenesis, including the formation of hinge points, thickening of the neural plate, and neural tube closure were captured in these simulations (Fig. 7).

Although neurulation involves the coordinated bending of a cellular sheet, Jacobson and Gordon (1976) showed that cells exchange neighbors within the epithelium as it deforms. To account for this phenomenon, Jacobson et al. (1986) proposed a cortical tractor model to explain the motile behavior of epithelial cells. Expanding on earlier ideas for cell crawling (Oster, 1984), the authors argued that intracellular fluid flow generates shear stresses that affect neighboring cell movements. Since the cells remain attached at the apical surface of the epithelium, dissimilar flow rates can cause cell columnarization and epithelial rolling movements, similar to those observed during neurulation. However, such a mechanism has not yet been verified experimentally.

More recent efforts have focused on adapting neurulation models to include multi-scale effects and more realistic 3D geometries (Chen and Brodland, 2008; Brodland et al., 2010). In these studies, triangular patches of epithelium are subdivided into active and passive regions and mapped to 3D embryo reconstructions. Morphogenesis is driven primarily by the interfacial tension between cells. With realistic mechanical properties and experimental geometries, these models reproduce normal phenotypes for amphibian neurulation quite well. If incorrect parameters for interfacial tension (generated by contractile actomyosin bundles) and mediolateral tension (generated by oriented lamellipodia) are used in the simulation, abnormal morphologies result.

6.2 Primary Brain Vesicle Morphogenesis

Until recently, no models for early brain morphogenesis had been proposed. Here, we discuss a model that was used to study differences in the shape of the early brain tube between species (Filas et al., 2012), differences which may arise from variations in neurulation mechanisms (Lowery and Sive, 2004; Harrington et al., 2009). For example, brain tubes of amphibians and fish are initially cylindrical in shape with a relatively closed, slit-like lumen, while those of chicken, mouse, and humans are rounded and comparatively open. Despite these differences, the primary vesicles (forebrain, midbrain, and hindbrain) subsequently form across vertebrates to subdivide the brain tube.

For amphibians and fish, the lumen must differentially open to form the brain vesicles, and Filas et al. (2012) simulated this process using a tubular model with an initially slit-like cross section. The theory of Rodriguez et al. (1994) was used to simulate local circumferential contraction at two mediolateral hinge points, as suggested by contractile protein expression patterns. The tube opens into a diamond shape, consistent with normal midbrain development in these species. Interestingly, Filas et al. (2012) also found that enhancing contractile activity through drug treatment transforms the relatively round brain cross sections of chick embryos into morphologies reminiscent of frog and fish, which are regionally characterized by cross-sectional shapes of diamonds, triangles, and slits. The model showed that local contraction in regions of highest actin density can transform a circular tube into these various shapes (Fig. 8). Much like the neurulation models (Odell et al., 1981; Clausi and Brodland, 1993; Chen and Brodland, 2008), these simulations suggest a significant role for actomyosin-based contraction in regulating early brain morphogenesis.

6.3 Cortical Folding

Relatively little is known about the forces that drive brain folding. Cortical folding is an extremely important problem, as abnormal folding patterns are associated with a number of neurological disorders, including autism, schizophrenia, and mental retardation. During the last few decades, several mathematical models have been proposed for the folding process.

Richman et al. (1975) modeled the cerebral cortex as a growing bi-layered shell. They assumed that the outer layer grows faster than the inner layer, creating compression in the

outer layer that causes buckling and the appearance of folds (Fig. 9). This hypothesis is consistent with experiments showing that isolated brains fold without the constraint provided by the skull (Barron, 1950). With parameters estimated from available data, their analysis showed that the predicted wavelength of the buckling pattern is consistent with that of normal brains. In addition, when geometry and growth rates were adjusted according to the data, the wavelengths matched reasonably well those found in brains with abnormally small folds (microgyric) or fewer folds (lissencephalic).

More recently, Toro and Burnod (2005) developed a 2D model involving truss-like elements forming an elastoplastic cortical layer, with circumferential growth stipulated and constrained by radial fibers. This growth works in combination with regionally prescribed variations in mechanical properties to produce folds, and the authors investigated the effects of various asymmetries. Nie et al. (2010) proposed a similar model, extended to three dimensions, in which folds are driven by both circumferential cortical growth and radial growth of underlying tissue, consistent with recent evidence suggesting that cortical folding involves growth in multiple directions (Reillo et al., 2011). However, while these models produce realistic folding patterns, to our knowledge there has been no attempt to test these mechanisms using other data, e.g., stresses and strains. Such tests are important to distinguish between multiple mechanisms.

Recent imaging studies have shown that folding occurs in a spatially and temporally phased manner (Neal et al., 2007; Kroenke et al., 2009). To capture these phenomena, Xu et al. (2010) developed a modified version of the differential growth model proposed by Richman et al. (1975), with growth included via the theory of Rodriguez et al. (1994). In a layered circular disk representing a brain slice, circumferential growth is first prescribed in a local region of the outer cortical layer, causing the formation of a single bulge. Repeating this sequence in neighboring regions generates a series of folds, similar to normal development, with stress distributions being in agreement with experimental results (Xu et al., 2010).

7 Heart Development

Cardiac development has been one of the most intensively studied problems in the developing embryo (Taber, 2006). Nevertheless, the physical mechanisms that create the heart remain poorly understood. Most work in this area has focused on cardiac looping, which is the first major morphological sign of left-right asymmetry in the embryo.

The heart begins as a relatively straight tubular structure consisting of a thick layer of extracellular matrix (cardiac jelly) sandwiched between an inner endocardial layer and an outer myocardial layer, which is a two-cell-thick epithelium of developing cardiomyocytes. During the first phase of looping, called c-looping, the heart tube (HT) acquires the shape of a c-shaped tube with convex curvature normally oriented toward the right side of the embryo (Männer, 2000). The dorsal mesocardium (DM) is a longitudinal structure that initially connects the dorsal side of the heart to the foregut before rupturing during c-looping. Following c-looping, further deformation (s-looping) then creates the basic pattern of the future heart, with the process of septation eventually dividing this curved tube into two ventricles and two atria (Romanoff, 1960). Here, we focus primarily on c-looping, which historically has received the most attention from researchers in cardiac morphogenesis.

It is important to realize that c-looping involves a combination of ventral bending and rightward torsion (or rotation). Although this has been known for decades (Butler, 1952), a failure to appreciate these two deformation components has led to considerable confusion in interpreting experimental results (Männer, 2000; Taber, 2006). Recent work has suggested that the bending component of c-looping is intrinsic to the HT, while torsion is driven mainly by extrinsic forces (Taber, 2006).

Since the early 1900s, researchers have proposed numerous hypotheses for the mechanisms of cardiac bending. The first mechanistic hypothesis for looping apparently was proposed by Patten (1922), who suggested that the HT is forced to bend simply because it outgrows the space that it occupies. This hypothesis, however, was later contradicted by the finding that the HT bends when isolated in culture (Butler, 1952; Manning and McLachlan, 1990), even when drugs are used to inhibit any contractile wounding response induced by dissection (Rémond et al., 2006). Other hypotheses that have been explored include bending driven by differential growth (Sissman, 1966; Stalsberg and DeHaan, 1969), remodeling in response to hemodynamic forces (Hove et al., 2003; Spitzer and Lev, 1951), active changes in myocardial cell shape (Latacha et al., 2005; Manasek et al., 1972), and cardiac jelly swelling (Manasek et al., 1984b). However, none of these mechanisms are consistent with all available data (Taber, 2006), and this problem is still being debated in the literature.

Theoretical models have been used to test some of these and other ideas. Taber et al. (1995) represented the combined HT and DM as a bilayered beam, with growth simulated using the theory of Rodriguez et al. (1994). Two bending mechanisms were studied. In the first, the DM is assumed to be initially under longitudinal tension. Then, as it ruptures it shortens like a rubber band and pulls the HT into a bent configuration with the DM located along the inner curvature (as observed experimentally). To simulate this mechanism, tension was generated in the DM by contraction, and the HT was assumed to grow in response to stresses induced by the bending.

The second mechanism involves active cell shape change. Here, contraction in the HT was assumed to occur in the circumferential direction, consistent with observed orientations of actin filaments in the myocardium (Itasaki et al., 1991), and incompressibility causes these cells to extend axially. In this model, the DM was taken as a passive structure, which limits deformation of adjacent cells and causes the HT to bend with the DM again lying along the inner curvature.

Both models showed that the proposed hypotheses are physically plausible and consistent with most data available at the time. However, later experiments with myosin inhibitors apparently have ruled out a contractile mechanism for bending (Rémond et al., 2006), although it still remains possible that a mechanism other than contraction drives changes in cell shape.

In this regard, Latacha et al. (2005) speculated that actin polymerization supplies forces that cause regional changes in myocardial cell shape. This idea stemmed from experiments in which actin polymerization was disrupted both globally and locally by various drugs. Based on these experiments, as well as known changes in cell shape during looping (Itasaki et al., 1989; Manasek et al., 1984a; Shiraishi et al., 1992), tubular finite-element models were developed for the isolated HT. In these models, cells were assumed to grow longer circumferentially and shorter axially near the inner curvature, while growing in both directions near the outer curvature. Predicted bending patterns were consistent with data for both normal and chemically perturbed looping of isolated hearts (Latacha et al., 2005; Ramasubramanian et al., 2006).

Recent experiments have suggested that torsional component of c-looping is driven largely by (1) forces applied by the omphalomesenteric veins (OVs) at the caudal end of the HT, with the left OV pushing with more force than the right OV; and (2) compressive forces applied to the ventral surface of the HT by a membrane called the splanchnopleure (SPL). The OV forces rotate the HT slightly toward the right, and the SPL enhances this rotation (Voronov et al., 2004). Both sets of forces are required for normal looping.

To date, models for the torsional aspect of looping have included only the effects of the OV forces, which experiments suggest are generated by a combination of growth on the cranial side of the OVs and contraction on the caudal side (Ramasubramanian et al., 2006, 2008). In these models, the HT and OVs are treated as tubes consisting of an inner layer of cardiac jelly and an outer layer of myocardium. Results were compared to measured strains and stresses that develop during normal looping, as well as the effects of various perturbations in embryos in which the SPL was dissected away before looping. In one notable experiment, the left OV, right OV, or both OVs were removed before looping began. On subsequent culture, the heart rotated toward the right, left, and right, respectively. A model used to simulate these experiments (without modifying parameters) predicts these results and the observed abnormal morphology relatively accurately (Fig. 10) (Ramasubramanian et al., 2008).

Although some questions have been answered for the looping problem, many remain open. For example, few researchers have considered how these forces are regulated in time and space, and s-looping has received relatively little attention.

8 Gut Looping

In amniotes, the gut tube includes three segments: the foregut, midgut, and hindgut. A primary loop forms in the midgut on the ventral side driven by elongation of the gut tube, followed by a sequence of rotation, bending, and twisting to properly position and shape the future intestine (Davis et al., 2008). Data have indicated that differential growth drives aspects of this process. Although there are many studies on the genetic aspects of gut looping and the associated left-right asymmetry (Horne-Badovinac et al., 2003; Davis et al., 2008; Taniguchi et al., 2011), the mechanics of looping morphogenesis remain incompletely understood (Savin et al., 2011).

Kurpios et al. (2008) used a 2D spring-dashpot-like model to investigate the asymmetric tilting of the gut. They simulated the dynamics of the dorsal mesentery and gut using a model of highly damped lattice cells driven by mechanical forces. Each cell is represented by seven interconnected nodes in a hexagonal lattice, and the ECM is taken as an elastic medium with a given local volume. In this model, cell-cell adhesion forces, elasticity of the ECM, and intracellular forces are included to account for the damped dynamics of the cell network. The model yields asymmetric changes in cell shape that tilt the gut tube in the appropriate direction. More significant tilting occurs when both the epithelial cells change shape and the mesenchymal ECM increases volume on the right side (e.g., through ECM swelling).

Although a number of studies have been devoted to interpret the initial looping (Horne-Badovinac et al., 2003; Davis et al., 2008; Taniguchi et al., 2011), the mechanical origin of subsequent looping has been described only recently (Savin et al., 2011). The embryonic gut in vertebrates has a highly conserved number and size of loops in a given species, and similar looping patterns occur across species from avians to mammals. Through a series of experiments, Savin et al. (2011) first ruled out some possible mechanisms that could drive gut looping, such as constraints due to the body cavity and differential growth at certain locations of the gut tube. Further dissection experiments suggested that differential growth between the gut and the mesentery is indispensable for looping, as the gut straightens into a long, uncoiled tube when dissected away, while the freed mesentery contracts, suggesting it is under tension. Motivated by these observations, the authors proposed a theory for gut looping which minimizes the stretching energy in the mesentery, as well as the bending energy in the gut tube; this theory then yields a scaling law for the wavelength of the gut loop.

Predictions from this theory were tested through experiments on chick embryos, physical models, and *in silico* simulations (Fig. 11). In the computer simulation, the mesentery is modeled as a discretized isotropic membrane composed of a hexagonal lattice of linear elastic springs, and the gut tube is modeled as a membrane strip. The physical parameters (thickness, stiffness, strain, etc.) were obtained through experimental measurements. Results from both the computational and physical models match very well with experimental observations of looped gut tubes (Savin et al., 2011).

9 Dorsal Closure and Wound Healing

Actomyosin cables are found in a variety of contexts, including embryonic wound healing and cytokinesis (Kiehart, 1999; Sonnemann and Bement, 2011). Here, we discuss two problems that involve contractile rings: dorsal closure in *Drosophila* embryos and wound healing in embryonic epithelia.

9.1 Dorsal Closure

During dorsal closure, the amnioserosa, an eye-shaped area of epithelial cells, with a canthus at each corner, is enveloped by a sheet of lateral epidermis which spreads from each side of the embryo to meet at the dorsal midline (Fig. 12A). The two leading edges (LE) of the epidermis then fuse where they meet (Kiehart et al., 2000; Gorfinkiel et al., 2011). Forces that drive this process include contraction of the amnioserosa, tension in an actomyosin cable at the LE, and filopodial tractions that pull apposing edges of the LE together in a zipper-like mechanism (Fig. 12). Laser wounding experiments, which ablate the tissue with subcellular resolution, have shown that these mechanisms work redundantly in dorsal closure, with no single mechanism absolutely required (Kiehart et al., 2000; Hutson et al., 2003).

Hutson et al. (2003) used laser dissection and modeling to study the forces involved in dorsal closure. The model considers the force balance at the midpoint of the LE and includes stresses in the amnioserosa and lateral epidermis, tension in the curved actin cable, and a viscous drag force dependent on the rate of closure (Fig. 12B). The zipping of the canthi was modeled kinematically. By rapidly ablating the amnioserosa (thereby excluding its contribution from the force balance) and observing the recoil of the leading edge and its subsequent equilibrium, the authors estimated the relative contributions of the various forces. With appropriate parameter values obtained through parametric fitting, the resulting model reproduced the closure of both native embryos and those with the amnioserosa removed, as well as for a genetic mutant (*myospheroid*). This model was later refined to consider anterioposterior and mediolateral asymmetry (Peralta et al., 2007, Peralta et al., 2008).

A second generation model for dorsal closure by Layton et al. (2009) considers the balance of forces along the entire LE. Both the amnioserosa and the actin cable generate force by active contraction, and the lateral epidermis is assumed to exert a constant and uniform tension. Zipping is specified kinematically as in Hutson et al. (2003). Control, amnioserosa removal, and mutant experiments were used to determine parameter values, and the model then predicted relatively well the results from additional laser ablation experiments, provided that amnioserosa contraction was upregulated.

Solon et al. (2009) observed that amnioserosa cells undergo pulsatile apical constrictions which displace the LE. To investigate the possible role of this activity in dorsal closure, the authors constructed a 2D network model of the amnioserosa and leading edge, with the cells represented as viscoelastic polygons. The active force generated at cell boundaries is assumed to be triggered when tension reaches a critical value, similar to the stretch-

activation mechanism of Odell et al. (1981). This model reproduces correlations in pulsing activity between neighboring cells, as well as pulsatile arrest following the loss of tissue tension from laser ablation.

9.2 Embryonic Wound Healing

The cytoskeletal machinery that operates during dorsal closure, i.e., the contractile ring and filopodia, is also employed in the healing of embryonic epithelial wounds (Wood et al., 2002; Garcia-Fernandez et al., 2009). Embryonic wounds differ from adult wounds in significant ways. For example, fewer cell types are involved, minimal inflammation occurs, and they heal quickly and without scarring (Jacinto et al., 2001; Martin and Parkhurst, 2004; Sonnemann and Bement, 2011).

Martin and Lewis (1992) studied wound healing in the skin of chick embryos. After dissecting away a patch of epidermis and a thin layer of underlying mesenchyme, they observed that the epidermis moved actively over the mesenchyme until the lesion closed, leaving no scar. No filopodia were seen, and fluorescent staining revealed a thick cable of actin at the leading edge of the epithelium, supporting the hypothesis that contraction of an actomyosin “purse string” in part drives the closing of the wound. While these observations form the basis for some models, it should be noted that not all embryonic wounds share these features. Although an actomyosin ring is observed in *Xenopus* (frog) embryos, for example, it appears that contraction of the underlying cells and protrusive activity of the remaining epithelial cells are primarily responsible for wound healing (Davidson et al., 2002). Significantly, the wound does not become more round as it heals, in contrast to observations in the chick (Martin and Lewis, 1992) and mouse (McCluskey and Martin, 1995).

Murray and Sherratt (Sherratt et al., 1992; Murray, 2003a) used the Murray-Oster theory to model embryonic wound healing. Here, the embryonic skin consists of a cell layer attached to a basal lamina, which is represented by an elastic foundation (springs). This study focuses on the aggregation and alignment of actin into an actomyosin cable in the cell layer, and considers the quasi-equilibrium state when the actin cable halts epithelial expansion following wounding. At that point, epithelial tension is balanced by elastic restoring forces originating in the basal lamina. With this model, Sherratt and et al. (1992) were able to reproduce the experimentally observed retraction of the epithelium, and the actin concentration at the wound edge bore a close resemblance to the actin cable observed experimentally.

Sadovsky and Wan (2007) extended the Murray-Sherratt model to investigate the dynamics of wound closure following the quasi-equilibrium state described above. In this model, the dynamics of tissue deformation is given by the balance of forces originating in both the epithelium and the mesenchyme. Contraction in the epithelium is a function of asymmetry in a wounding signal, which the authors argue is most plausibly mechanical stress, and the mesenchyme contracts at a rate which depends on the velocity of the epithelium. Utilizing a specialized version of the Murray-Sherratt model and a radially symmetric wound, the authors reproduced the initial and intermediate stages of wound closure.

10 Mechanical Feedback in Morphogenesis

It has become increasingly clear that mechanical stress, in addition to driving the tissue deformations responsible for morphogenesis, also plays a role in the control of cellular behavior in the embryo, including differentiation, proliferation, and contractility (Vogel and Sheetz, 2006; Hutson and Ma, 2008; Wozniak and Chen, 2009; Levayer and Lecuit, 2012). In the *Drosophila* embryo, for instance, mechanical stress has been shown to affect both

gene expression (Farge, 2003; Desprat et al., 2008) and downstream signaling pathways (Pouille et al., 2009) which govern the production of forces driving gastrulation. Interestingly, Pouille et al. (2009) showed that gastrulation in mutant *Drosophila* embryos can be rescued by local mechanical indentation. Results such as these suggest that mechanical feedback is involved in epigenetic control of morphogenesis (Brouzés et al., 2004).

As discussed earlier, perhaps the first model for morphogenesis to include mechanical feedback is the invagination model of Odell et al. (1981), in which cells contract in response to stretch. More recently, Ramasubramanian and Taber (2008) proposed a different mechanism, where contraction and growth are controlled by changes in the value of a "target stress" as specified by genetic activity. In their finite-element models, an increase or decrease in target stress triggers contraction or growth, respectively. Based on this idea, models were used to simulate bending of epithelia, including invagination in cylindrical and spherical shells.

For several decades, the developmental biologist Lev Beloussov has studied the role of mechanical feedback in morphogenesis. His experimental observations suggest that morphogenesis is regulated in part by mechanical stress (Beloussov, 1998), leading to his Hyper-Restoration Hypothesis (HRH). According to HRH, embryonic tissue reacts to mechanical perturbations in a way that tends to restore, but overshoot, the original stress (Beloussov, 1998; Beloussov and Grabovsky, 2006). This implies that embryos have the capability to self-assemble to some extent, presumably governed by morphogenetic laws and controlled by mechanical feedback loops, assuring a degree of robustness against perturbations (Taber, 2008). Here, it is worth noting that HRH is based on the view that tissue shortening elicits a contractile response, while stretch triggers relaxation and active lengthening. This behavior is opposite to the stretch activation response postulated by Odell et al. (1981). Realizing this discrepancy, Beloussov (1998) suggested that HRH applies to large deformations, while stretch-activated contraction occurs when stretch perturbations are relatively small.

Although some efforts have been geared toward obtaining quantitative support for HRH (Beloussov and Grabovsky, 2006, Beloussov and Grabovsky, 2007), most available experimental data are qualitative. For instance, Beloussov et al. (1975) performed dissection experiments on amphibian embryos to map changes in stress distributions during development. Both rapid, passive deformations and slower, active deformations were observed. From the results of these experiments, Belintsev et al. (1987) proposed a phenomenological model that accounts for active and passive deformations of cells and mechanical feedback to address the collective polarization and pattern formation in epithelia. From these studies, Beloussov and colleagues (Beloussov et al., 1994; Beloussov, 1998; Beloussov and Grabovsky, 2006; Beloussov, 2008) suggested conceptual models based on HRH to explain a variety of morphogenetic events such as gastrulation, neurulation, and convergent extension. Experiments by Kornikova et al. (2010), where artificially bent explants (suprablastoporal regions of *Xenopus laevis*) continued to bend after release of loads also seem to support HRH.

Recently, Taber (2008) used finite-element models to explore the theoretical limitations of HRH, as embodied in a feedback law which includes an overshoot. The problems investigated include stretching, bending, and invagination of epithelial sheets, as well as neurulation and sea urchin gastrulation. In each model an initial perturbation, e.g., a small contraction (presumably dictated by genes), was used to initiate the HRH response. The results were mixed, with HRH producing realistic morphogenesis in some cases but not in others. Hence, while mechanical feedback likely is involved in these processes, genes also

may participate by making “mid-course corrections” (Taber, 2008) or by producing new perturbations at appropriate times to guide the creation of proper morphology.

Biological systems clearly must obey the laws of physics as expressed mathematically. However, it is not at all clear that mathematical laws for biology exist. Recently, Taber (2009) examined this issue using a generalization of HRH. The key idea was to assume that the feedback law depends on the rate of change in stress. For example, a slow stretch induces growth, whereas a rapid stretch elicits a contractile response. Finite-element models for growth of a pressurized artery, sea urchin gastrulation, wound healing, axon stretching, and other examples improved on the predictions of HRH, but still yielded somewhat inconsistent results when compared to available experimental data. Therefore, the existence of mathematical laws in biology remains an open question (Taber, 2009; Ambrosi et al., 2011).

11 Concluding Remarks

Despite numerous computational and experimental studies, it remains incompletely understood how the genetic information encoded in DNA is translated, through mechanics, into three-dimensional functional shapes. Moreover, relatively little is known about how epigenetic factors, in turn, interact with genetics through mechanical feedback to regulate morphogenesis. Clearly, molecular biology has triggered a revolution in studies of developmental processes. Many developmental biologists, however, now realize that a complete understanding of how the embryo is constructed also requires studies of the nuts and bolts of morphogenesis (i.e., mechanics), which has been largely neglected during the last few decades. Because development is a complicated process which can defy physical intuition, computational models for morphogenesis are receiving increasing attention.

While the importance of mechanics in morphogenesis is well established, a detailed accounting of the stresses and strains that drive the associated tissue deformations is still far from complete (Davidson et al., 2009). Sophisticated mathematical models notwithstanding, advances in mechanobiology have been closely connected to technological advances (Eyckmans et al., 2011), and this will likely hold true for the foreseeable future. Such advances have been rapid and impressive, however. The revolution in *in vivo* fluorescence microscopy is revealing the localization and dynamics of cellular components with unprecedented spatial and temporal resolution (Martin, 2010; Sato et al., 2010). Laser ablation techniques (Rauzi and Lenne, 2011, and references therein) allow for precise mechanical perturbations of cells and subcellular structures, and are a tool of outstanding utility in probing mechanical forces at those scales. Finally, a new generation of FRET-based single molecule stress sensors (Grashoff et al., 2010; Meng and Sachs, 2011) open the possibility of measuring stress with piconewton sensitivity at the subcellular level. Taken together, our understanding of the mechanics of certain cellular and morphogenetic systems is progressing at an accelerating pace, and results from such model systems will continue to serve as a guide to the general principles underlying morphogenesis.

For a number of reasons, creating and testing computational models for morphomechanical processes is a challenging endeavor. The mechanical properties of embryos can be surprisingly variable, complicating the comparison between experimental and numerical results. Critical morphogenetic events often use multiple redundant mechanisms which must first be identified, and not all such mechanisms need or can be incorporated into a model. Problems in development, as in much of biology, frequently involve large numbers of unknown parameters, and the response of the system can be highly nonlinear. In constructing such models, a delicate balance must be struck between realism with excessive complexity on the one hand, and transparency but over-simplification on the other.

We perceive two major themes in the modeling of morphogenesis which we feel hold particular promise. One is the explicit incorporation of mechanical and chemical feedback into models of morphogenesis. Such feedback, which has been demonstrated in a number of experimental systems (Fernandez-Gonzalez et al., 2009; Ren et al., 2009) has long been recognized as a rich source of morphogenetic pattern (Murray and Oster, 1984). Intuition and reductionist methods typically fail for such systems, and new experimental approaches, computer modeling, and mathematical analysis will all play a role in the description and understanding of the behavior of such systems.

The development of multi-scale models of morphomechanics (Blanchard and Adams, 2011) is perhaps even more significant. Quantitative data at multiple spatial scales are becoming increasingly available (Blankenship et al., 2006; Martin et al., 2010), and recent models provide an excellent beginning to capturing multi-scale effects (e.g., Stylianopoulos and Barocas, 2007; Chen and Brodland, 2008; Sherrard et al., 2010; Driquez et al., 2011). Still, much remains to be done. Biological phenomena include the interaction between biochemistry, genes, and mechanics, and it appears likely that tomorrow's models will need to incorporate all of these aspects to gain a comprehensive understanding of the physical mechanisms of morphogenesis.

Acknowledgments

This work was supported by grants R01 GM075200 and R01 NS070918 (LAT), F32 GM093396 (MAW), and T90 DA022871 (BAF) from the National Institutes of Health, the American Academy of Mechanics Founder's Award from the Robert M. and Mary Haythornthwaite Foundation (ZC), as well as grant 09PRE2060795 from the American Heart Association (VDV).

References

Affolter M, Zeller R, Caussinus E. Tissue remodelling through branching morphogenesis. Nat Rev Mol Cell Bio. 2009; 10:831–842. [PubMed: 19888266]

Allena R, Mouronval AS, Aubry D. Simulation of multiple morphogenetic movements in the *Drosophila* embryo by a single 3d finite element model. J Mech Behav Biomed Mater. 2010; 3:313–323. [PubMed: 20346899]

Ambrosi D, Ateshian GA, Arruda EM, Cowin SC, Dumais J, Goriely A, Holzapfel GA, Humphrey JD, Kemkemer R, Kuhl E, Olberding JE, Taber LA, Garikipati K. Perspectives on biological growth and remodeling. J Mech Phys Solids. 2011; 59:863–883. [PubMed: 21532929]

Ambrosi D, Bussolino F, Preziosi L. A review of vasculogenesis models. Comput Math Method M. 2005; 6:1–19.

Balfour, FM. A treatise on comparative embryology. London: MacMillan and Co; 1881.

Bard, J. Morphogenesis: the cellular and molecular processes of developmental anatomy. New York: Cambridge University Press; 1990.

Barron DH. An experimental analysis of some factors involved in the development of the fissure pattern of the cerebral cortex. J Exp Zool. 1950; 113:553–573.

Bathe, KJ. Finite Element Procedures. Englewood Cliffs, NJ: Cambridge University Press; 1996.

Belintsev BN, Beloussov LV, Zaraisky AG. Model of pattern formation in epithelial morphogenesis. J Theor Biol. 1987; 129:369–394. [PubMed: 3455468]

Beloussov, LV. The Dynamic Architecture of a Developing Organism: An Interdisciplinary Approach to the Development of Organisms. Dordrecht, the Netherlands: Kluwer; 1998.

Beloussov LV. Mechanically based generative laws of morphogenesis. Phys Biol. 2008; 5:15009.

Beloussov LV, Dorfman JG, Cherdantzev VG. Mechanical stresses and morphological patterns in amphibian embryos. J Embryol Exp Morphol. 1975; 34:559–574. [PubMed: 1082486]

Beloussov LV, Grabovsky VI. Morphomechanics: goals, basic experiments and models. Int J Dev Biol. 2006; 50:81–92. [PubMed: 16479477]

Beloussov LV, Grabovsky VI. Information about a form (on the dynamic laws of morphogenesis). BioSystems. 2007; 87:204–214. [PubMed: 17030084]

Beloussov LV, Saveliev SV, Naumidi II, Novoselov VV. Mechanical stresses in embryonic tissues: patterns, morphogenetic role, and involvement in regulatory feedback. Int Rev Cytol. 1994; 150:1–34. [PubMed: 8169076]

Blanchard GB, Adams RJ. Measuring the multi-scale integration of mechanical forces during morphogenesis. Curr Opin Genet Dev. 2011; 21:653–663. [PubMed: 21930371]

Blankenship JT, Backovic ST, Sanny JSP, Weitz O, Zallen Ja. Multicellular rosette formation links planar cell polarity to tissue morphogenesis. Dev Cell. 2006; 11:459–70. [PubMed: 17011486]

Brodland GW. The differential interfacial tension hypothesis (DITH): a comprehensive theory for the self-rearrangement of embryonic cells and tissues. J Biomech Eng. 2002; 124:188–197. [PubMed: 12002128]

Brodland GW. Computational modeling of cell sorting, tissue engulfment, and related phenomena: A review. Appl Mech Rev. 2004; 57:47–76.

Brodland GW. Do lamellipodia have the mechanical capacity to drive convergent extension? Int J Dev Biol. 2006; 50:151–155. [PubMed: 16479484]

Brodland GW, Chen HH. The mechanics of cell sorting and envelopment. J Biomech. 2000; 33:845–851. [PubMed: 10831759]

Brodland GW, Chen X, Lee P, Marsden M. From genes to neural tube defects (NTDs): insights from multiscale computational modeling. HFSP J. 2010; 4:142–152. [PubMed: 21119766]

Brodland GW, Clausi DA. Embryonic tissue morphogenesis modeled by FEM. J Biomech Eng. 1994; 116:146–155. [PubMed: 8078321]

Brodland GW, Clausi DA. Cytoskeletal mechanics of neurulation: insights obtained from computer simulations. Biochem Cell Biol. 1995; 73:545–553. [PubMed: 8703425]

Brodland GW, Veldhuis JH. Lamellipodium-driven tissue reshaping: a parametric study. Comput Methods Biomech Biomed Engin. 2006; 9:17–23. [PubMed: 16880153]

Brouzés E, Supatto W, Farge E. Is mechano-sensitive expression of twist involved in mesoderm formation? Biol Cell. 2004; 96:471–477. [PubMed: 15380614]

Butler, JK. PhD thesis. University of Texas; 1952. An experimental analysis of cardiac loop formation in the chick.

Chen HH, Brodland GW. Cell-level finite element studies of viscous cells in planar aggregates. J Biomech Eng. 2000; 122:394–401. [PubMed: 11036563]

Chen X, Brodland GW. Multi-scale finite element modeling allows the mechanics of amphibian neurulation to be elucidated. Phys Biol. 2008; 5:15003.

Clausi DA, Brodland GW. Mechanical evaluation of theories of neurulation using computer simulations. Development. 1993; 118:1013–1023.

Colas JF, Schoenwolf GC. Towards a cellular and molecular understanding of neurulation. Dev Dyn. 2001; 221:117–145. [PubMed: 11376482]

Conte V, Muñoz JJ, Miodownik M. A 3d finite element model of ventral furrow invagination in the *Drosophila* melanogaster embryo. J Mech Behav Biomed Mater. 2008; 1:188–198. [PubMed: 19627783]

Conte V, Munoz JJ, Baum B, Miodownik M. Robust mechanisms of ventral furrow invagination require the combination of cellular shape changes. Phys Biol. 2009; 6:016010. [PubMed: 19342769]

Davidson LA. Integrating morphogenesis with underlying mechanics and cell biology. Curr Top Dev Biol. 2008; 81:113–133. [PubMed: 18023725]

Davidson LA, Ezin AM, Keller R. Embryonic wound healing by apical contraction and ingression in *Xenopus laevis*. Cell Motil Cytoskel. 2002; 53:163–176.

Davidson LA, Keller R. Measuring mechanical properties of embryos and embryonic tissues. Method Cell Biol. 2007; 83:425–439.

Davidson LA, Koehl MA, Keller R, Oster GF. How do sea urchins invaginate? using biomechanics to distinguish between mechanisms of primary invagination. Development. 1995; 121:2005–2018. [PubMed: 7635048]

Davidson LA, Oster GF, Keller RE, Koehl MA. Measurements of mechanical properties of the blastula wall reveal which hypothesized mechanisms of primary invagination are physically plausible in the sea urchin *Strongylocentrotus purpuratus*. Dev Biol. 1999; 209:221–238. [PubMed: 10328917]

Davidson LA, von Dassow M, Zhou J. Multi-scale mechanics from molecules to morphogenesis. Int J Biochem Cell Biol. 2009; 41:2147–2162. [PubMed: 19394436]

Davies, JA. Mechanisms of Morphogenesis: The creation of biological form. Elsevier; 2005.

Davis NM, Kurpios NA, Sun X, Gros J, Martin JF, Tabin CJ. The chirality of gut rotation derives from left-right asymmetric changes in the architecture of the dorsal mesentery. Dev Cell. 2008; 15:134–145. [PubMed: 18606147]

Dawes-Hoang RE, Parmar KM, Christiansen AE, Phelps CB, Brand AH, Wieschaus EF. Folded gastrulation, cell shape change and the control of myosin localization. Development. 2005; 132:4165–4178. [PubMed: 16123312]

Desmond ME, Jacobson AG. Embryonic brain enlargement requires cerebrospinal fluid pressure. Dev Biol. 1977; 57:188–198. [PubMed: 863106]

Desmond ME, Levitan ML, Haas AR. Internal luminal pressure during early chick embryonic brain growth: descriptive and empirical observations. Anat Rec A Discov Mol Cell Evol Biol. 2005; 285:737–747. [PubMed: 15977221]

Desprat N, Supatto W, Pouille PA, Beaurepaire E, Farge E. Tissue deformation modulates twist expression to determine anterior midgut differentiation in *Drosophila* embryos. Dev Cell. 2008; 15:470–477. [PubMed: 18804441]

Driquez B, Bouclet A, Farge E. Mechanotransduction in mechanically coupled pulsating cells: transition to collective constriction and mesoderm invagination simulation. Phys Biol. 2011; 8:066007. [PubMed: 22120059]

Engler AJ, Sen S, Sweeney HL, Discher DE. Matrix elasticity directs stem cell lineage specification. Cell. 2006; 126:677–689. [PubMed: 16923388]

Eyckmans J, Boudou T, Yu X, Chen CS. A hitchhiker's guide to mechanobiology. Dev Cell. 2011; 21:35–47. [PubMed: 21763607]

Farge E. Mechanical induction of twist in the *Drosophila* foregut/stomodeal primordium. Curr Biol. 2003; 13:1365–1377. [PubMed: 12932320]

Fernandez-Gonzalez R, Simoes SdM, Röper JC, Eaton S, Zallen JA. Myosin II dynamics are regulated by tension in intercalating cells. Dev Cell. 2009; 17:736–743. [PubMed: 19879198]

Filas B, Oltean A, Beebe D, Okamoto R, Bayly P, Taber L. A potential role for differential contractility in early brain development and evolution. Biomech Model Mechanobiol. 2012 In Press.

Filas BA, Bayly PV, Taber LA. Mechanical stress as a regulator of cytoskeletal contractility and nuclear shape in embryonic epithelia. Ann Biomed Eng. 2011; 39:443–454. [PubMed: 20878237]

Fleury V. A change in boundary conditions induces a discontinuity of tissue flow in chicken embryos and the formation of the cephalic fold. Eur Phys J E. 2011:34.

Forgacs G, Foty RA, Shafrir Y, Steinberg MS. Viscoelastic properties of living embryonic tissues: a quantitative study. Biophys J. 1998; 74:2227–2234. [PubMed: 9591650]

Forgacs, G.; Newman, SA. Biological physics of the developing embryo. New York: Cambridge University Press; 2005.

Friedl P. Prespecification and plasticity: shifting mechanisms of cell migration. Curr Opin Cell Biol. 2004; 16:14–23. [PubMed: 15037300]

Garcia-Fernandez B, Campos I, Geiger J, Santos AC, Jacinto A. Epithelial resealing. Int J Dev Biol. 2009; 53:1549–1556. [PubMed: 19247953]

Gierer A, Meinhardt H. A theory of biological pattern formation. Kybernetik. 1972; 12:30–39. [PubMed: 4663624]

Gilbert, SF. Developmental biology. 6. Sunderland, Mass: Sinauer Associates; 2000.

Gjorevski N, Nelson CM. Endogenous patterns of mechanical stress are required for branching morphogenesis. Integr Biol. 2010; 2:424–434.

Gorfinkiel N, Schamberg S, Blanchard GB. Integrative approaches to morphogenesis: lessons from dorsal closure. Genesis. 2011; 49:522–533. [PubMed: 21162075]

Grashoff C, Hoffman BD, Brenner MD, Zhou R, Parsons M, Yang MT, McLean MA, Sligar SG, Chen CS, Ha T, Schwartz MA. Measuring mechanical tension across vinculin reveals regulation of focal adhesion dynamics. Nature. 2010; 466:263–266. [PubMed: 20613844]

Grumbling G, Strelets V. FlyBase: anatomical data, images and queries. Nucleic Acids Res. 2006; 34:D484–8. [PubMed: 16381917]

Harrington MJ, Hong E, Brewster R. Comparative analysis of neurulation: first impressions do not count. Mol Reprod Dev. 2009; 76:954–65. [PubMed: 19653285]

Harris AK, Wild P, Stopak D. Silicone rubber substrata: a new wrinkle in the study of cell locomotion. Science. 1980; 208:177–179. [PubMed: 6987736]

Holmes MJ, Sleeman BD. A mathematical model of tumour angiogenesis incorporating cellular traction and viscoelastic effects. J Theor Biol. 2000; 202:95–112. [PubMed: 10640431]

Horne-Badovinac S, Rebagliati M, Stainier DYR. A cellular framework for gut-looping morphogenesis in zebrafish. Science. 2003; 302:662–665. [PubMed: 14576439]

Hove JR, Köster RW, Forouhar AS, Acevedo-Bolton G, Fraser SE, Gharib M. Intracardiac fluid forces are an essential epigenetic factor for embryonic cardiogenesis. Nature. 2003; 421:172–177. [PubMed: 12520305]

Howard J, Grill SW, Bois JS. Turing's next steps: the mechanochemical basis of morphogenesis. Nat Rev Mol Cell Bio. 2011; 12:392–398. [PubMed: 21602907]

Hutson MS, Ma X. Mechanical aspects of developmental biology: perspectives on growth and form in the (post)-genomic age. Phys Biol. 2008; 5:015001. [PubMed: 18403828]

Hutson MS, Tokutake Y, Chang MS, Bloor JW, Venakides S, Kiehart DP, Edwards GS. Forces for morphogenesis investigated with laser microsurgery and quantitative modeling. Science. 2003; 300:145–149. [PubMed: 12574496]

Itasaki N, Nakamura H, Sumida H, Yasuda M. Actin bundles on the right side in the caudal part of the heart tube play a role in dextro-looping in the embryonic chick heart. Anat Embryol. 1991; 183:29–39. [PubMed: 2053708]

Itasaki N, Nakamura H, Yasuda M. Changes in the arrangement of actin bundles during heart looping in the chick embryo. Anat Embryol. 1989; 180:413–420. [PubMed: 2619084]

Jacinto A, Martinez-Arias A, Martin P. Mechanisms of epithelial fusion and repair. Nat Cell Biol. 2001; 3:E117–23. [PubMed: 11331897]

Jacobson AG, Gordon R. Changes in the shape of the developing vertebrate nervous system analyzed experimentally, mathematically, and by computer simulation. J Exp Zool. 1976; 197:191–246. [PubMed: 965908]

Jacobson AG, Oster GF, Odell GM, Cheng LY. Neurulation and the cortical tractor model for epithelial folding. J Embryol Exp Morphol. 1986; 96:19–49. [PubMed: 3805983]

Keller R, Davidson L, Edlund A, Elul T, Ezin M, Shook D, Skoglund P. Mechanisms of convergence and extension by cell intercalation. Philos T R Soc B. 2000; 355:897–922.

Keller R, Shook D, Skoglund P. The forces that shape embryos: physical aspects of convergent extension by cell intercalation. Phys Biol. 2008; 5:015007. [PubMed: 18403829]

Kiehart DP. Wound healing: The power of the purse string. Curr Biol. 1999; 9:R602–5. [PubMed: 10469588]

Kiehart DP, Galbraith CG, Edwards KA, Rickoll WL, Montague RA. Multiple forces contribute to cell sheet morphogenesis for dorsal closure in *Drosophila*. J Cell Biol. 2000; 149:471–490. [PubMed: 10769037]

Koehl MAR. Biomechanical approaches to morphogenesis. Sem Dev Biol. 1990; 1:367–378.

Kondo S, Miura T. Reaction-diffusion model as a framework for understanding biological pattern formation. Science. 2010; 329:1616–1620. [PubMed: 20929839]

Kornikova ES, Troshina TG, Kremnyov SV, Beloussov LV. Neuro-mesodermal patterns in artificially deformed embryonic explants: a role for mechano-geometry in tissue differentiation. Dev Dyn. 2010; 239:885–896. [PubMed: 20140909]

Kroenke CD, Taber EN, Leigland LA, Knutsen AK, Bayly PV. Regional patterns of cerebral cortical differentiation determined by diffusion tensor MRI. Cereb Cortex. 2009; 19:2916–2929. [PubMed: 19363145]

Kurpios NA, Ibañes M, Davis NM, Lui W, Katz T, Martin JF, Izpisúa Belmonte JC, Tabin CJ. The direction of gut looping is established by changes in the extracellular matrix and in cell:cell adhesion. P Natl Acad Sci Usa. 2008; 105:8499–8506.

Lai, WM.; Rubin, D.; Krempl, E. Introduction to Continuum Mechanics. 4. Butterworth-Heinemann; 2009.

Latacha KS, Rémond MC, Ramasubramanian A, Chen AY, Elson EL, Taber LA. Role of actin polymerization in bending of the early heart tube. Dev Dyn. 2005; 233:1272–1286. [PubMed: 15986456]

Lawson A, Anderson H, Schoenwolf GC. Cellular mechanisms of neural fold formation and morphogenesis in the chick embryo. Anat Rec. 2001; 262:153–168. [PubMed: 11169910]

Layton AT, Toyama Y, Yang GQ, Edwards GS, Kiehart DP, Venakides S. *Drosophila* morphogenesis: tissue force laws and the modeling of dorsal closure. HFSP J. 2009; 3:441–460. [PubMed: 20514134]

Lecuit T, Lenne PF. Cell surface mechanics and the control of cell shape, tissue patterns and morphogenesis. Nat Rev Mol Cell Bio. 2007; 8:633–644. [PubMed: 17643125]

Leptin M. *Drosophila* gastrulation: from pattern formation to morphogenesis. Annu Rev Cell Dev Biol. 1995; 11:189–212. [PubMed: 8689556]

Leptin M. Gastrulation movements: the logic and the nuts and bolts. Dev Cell. 2005; 8:305–320. [PubMed: 15737927]

Levayer R, Lecuit T. Biomechanical regulation of contractility: spatial control and dynamics. Trends Cell Biol. 2012; 22:61–81. [PubMed: 22119497]

Lewis WH. Mechanics of invagination. Anat Rec. 1947; 97:139–156. [PubMed: 20284907]

Lodish, H.; Berk, A.; Matsudaira, P.; Kaiser, CA.; Krieger, M.; Scott, MP.; Zipursky, SL.; Darnell, J. Molecular Cell Biology. 5. New York: W.H. Freeman and Company; 2004.

Lowery LA, Sive H. Strategies of vertebrate neurulation and a re-evaluation of teleost neural tube formation. Mech Dev. 2004; 121:1189–1197. [PubMed: 15327780]

Lubkin SR, Li Z. Force and deformation on branching rudiments: cleaving between hypotheses. Biomech Model Mechanobiol. 2002; 1:5–16. [PubMed: 14586703]

Lye CM, Sanson B. Tension and epithelial morphogenesis in *Drosophila* early embryos. Curr Top Dev Biol. 2011; 95:145–187. [PubMed: 21501751]

Maini PK. Using mathematical models to help understand biological pattern formation. C R Biol. 2004; 327:225–234. [PubMed: 15127893]

Mammoto T, Ingber DE. Mechanical control of tissue and organ development. Development. 2010; 137:1407–1420. [PubMed: 20388652]

Manasek FJ, Burnside MB, Waterman RE. Myocardial cell shape change as a mechanism of embryonic heart looping. Dev Biol. 1972; 29:349–371. [PubMed: 4120601]

Manasek, FJ.; Isobe, Y.; Shimada, Y. Congenital Heart Disease: Causes and Processes. Mount Kisco, NY: Futura Publishing; 1984a. The embryonic myocardial cytoskeleton, interstitial pressure, and the control of morphogenesis; p. 359-376.

Manasek, FJ.; Kulikowski, RR.; Nakamura, A.; Nguyenphuc, Q.; Lacktis, JW.; Zak, R. Growth of the heart in Health and Disease. New York: Raven Press; 1984b. Early heart development: a new model of cardiac morphogenesis.

Männer J. Cardiac looping in the chick embryo: a morphological review with special reference to terminological and biomechanical aspects of the looping process. Anat Rec. 2000; 259:248–262. [PubMed: 10861359]

Manning A, McLachlan JC. Looping of chick embryo hearts in vitro. J Anat. 1990; 168:257–263. [PubMed: 2323996]

Manoussaki D, Lubkin SR, Vernon RB, Murray JD. A mechanical model for the formation of vascular networks in vitro. Acta Biotheor. 1996; 44:271–282. [PubMed: 8953213]

Martin AC. Pulsation and stabilization: contractile forces that underlie morphogenesis. Dev Biol. 2010; 341:114–125. [PubMed: 19874815]

Martin AC, Gelbart M, Fernandez-Gonzalez R, Kaschube M, Wieschaus EF. Integration of contractile forces during tissue invagination. J Cell Biol. 2010; 188:735–49. [PubMed: 20194639]

Martin P, Lewis J. Actin cables and epidermal movement in embryonic wound healing. Nature. 1992; 360:179–183. [PubMed: 1436096]

Martin P, Parkhurst SM. Parallels between tissue repair and embryo morphogenesis. Development. 2004; 131:3021–3034. [PubMed: 15197160]

McCluskey J, Martin P. Analysis of the tissue movements of embryonic wound healing DiI studies in the limb bud stage mouse embryo. Dev Biol. 1995; 170:102–114. [PubMed: 7601301]

Meng F, Sachs F. Visualizing dynamic cytoplasmic forces with a compliance-matched FRET sensor. J Cell Sci. 2011; 124:261–269. [PubMed: 21172803]

Moury JD, Schoenwolf GC. Cooperative model of epithelial shaping and bending during avian neurulation: autonomous movements of the neural plate, autonomous movements of the epidermis, and interactions in the neural plate/epidermis transition zone. Dev Dyn. 1995; 204:323–337. [PubMed: 8573723]

Muñoz JJ, Barrett K, Miodownik M. A deformation gradient decomposition method for the analysis of the mechanics of morphogenesis. J Biomech. 2007; 40:1372–1380. [PubMed: 16814298]

Murray, J. Mathematical Biology II: Spatial Models and Biomedical Applications. Springer; 2003a. Epidermal wound healing; p. 441-490.

Murray JD. On the mechanochemical theory of biological pattern formation with application to vasculogenesis. C R Biol. 2003b; 326:239–252. [PubMed: 12754942]

Murray JD, Oster GF. Generation of biological pattern and form. Math Med Biol. 1984; 1:51–75.

Namy P, Ohayon J, Tracqui P. Critical conditions for pattern formation and in vitro tubulogenesis driven by cellular traction fields. J Theor Biol. 2004; 227:103–120. [PubMed: 14969709]

Neal J, Takahashi M, Silva M, Tiao G, Walsh CA, Sheen VL. Insights into the gyrification of developing ferret brain by magnetic resonance imaging. J Anat. 2007; 210:66–77. [PubMed: 17229284]

Nelson C, Jean R, Tan J, Liu W, Sniadecki N, Spector A, Chen C. Emergent patterns of growth controlled by multicellular form and mechanics. P Natl Acad Sci Usa. 2005; 102:11594–11599.

Nie J, Guo L, Li G, Faraco C, Stephen Miller L, Liu T. A computational model of cerebral cortex folding. J Theor Biol. 2010; 264:467–78. [PubMed: 20167224]

Nikolaidou KK, Barrett K. A rho gtpase signaling pathway is used reiteratively in epithelial folding and potentially selects the outcome of Rho activation. Curr Biol. 2004; 14:1822–1826. [PubMed: 15498489]

Odell GM, Oster G, Alberch P, Burnside B. The mechanical basis of morphogenesis. I.~epithelial folding and invagination. Dev Biol. 1981; 85:446–462. [PubMed: 7196351]

Oster GF. On the crawling of cells. J Embryol Exp Morph. 1984; 83 (Suppl):329–364. [PubMed: 6533244]

Oster GF, Murray JD, Harris AK. Mechanical aspects of mesenchymal morphogenesis. J Embryol Exp Morph. 1983; 78:83–125. [PubMed: 6663234]

Painter KJ. Modelling cell migration strategies in the extracellular matrix. J Math Biol. 2009; 58:511–543. [PubMed: 18787826]

Patten BM. The formation of the cardiac loop in the chick. Am J Anat. 1922; 30:373–397.

Patten, BM. Early embryology of the chick. New York: McGraw-Hill; 1971.

Peralta XG, Toyama Y, Hutson MS, Montague R, Venakides S, Kiehart DP, Edwards GS. Upregulation of forces and morphogenic asymmetries in dorsal closure during *Drosophila* development. Biophys J. 2007; 92:2583–2596. [PubMed: 17218455]

Peralta XG, Toyama Y, Kiehart DP, Edwards GS. Emergent properties during dorsal closure in *Drosophila* morphogenesis. Phys Biol. 2008; 5:015004. [PubMed: 18403825]

Phillips HM, Steinberg MS, Lipton BH. Embryonic tissues as elasticoviscous liquids. II. Direct evidence for cell slippage in centrifuged aggregates. Dev Biol. 1977; 59:124–134. [PubMed: 561007]

Pouille PA, Ahmadi P, Brunet AC, Farge E. Mechanical signals trigger myosin II redistribution and mesoderm invagination in *Drosophila* embryos. Sci Signal. 2009; 2:ra16. [PubMed: 19366994]

Pouille PA, Farge E. Hydrodynamic simulation of multicellular embryo invagination. Phys Biol. 2008; 5:015005. [PubMed: 18403824]

Ramasubramanian A, Latacha KS, Benjamin JA, Voronov DA, Ravi A, Taber LA. Computational model for early cardiac looping. Ann Biomed Eng. 2006; 34:1355–1369.

Ramasubramanian A, Nerurkar NL, Achtien KH, Filas BA, Voronov DA, Taber LA. On modeling morphogenesis of the looping heart following mechanical perturbations. J Biomech Eng. 2008; 130:061018. [PubMed: 19045547]

Ramasubramanian A, Taber LA. Computational modeling of morphogenesis regulated by mechanical feedback. Biomech Model Mechanobiol. 2008; 7:77–91. [PubMed: 17318485]

Rauzi M, Lenne PF. Cortical forces in cell shape changes and tissue morphogenesis. Curr Top Dev Biol. 2011; 95:93–144. [PubMed: 21501750]

Rauzi M, Verant P, Lecuit T, Lenne PF. Nature and anisotropy of cortical forces orienting *Drosophila* tissue morphogenesis. Nat Cell Biol. 2008; 10:1401–1410. [PubMed: 18978783]

Reddy, JN. An introduction to the finite element method. New York: McGraw-Hill, Inc; 1993.

Reillo I, de Juan Romero C, García-Cabezas MÁ, Borrell V. A role for intermediate radial glia in the tangential expansion of the mammalian cerebral cortex. Cereb Cortex. 2011; 21:1674–1694. [PubMed: 21127018]

Rémond MC, Fee JA, Elson EL, Taber LA. Myosin-based contraction is not necessary for cardiac c-looping in the chick embryo. Anat Embryol. 2006; 211:443–454. [PubMed: 16636777]

Ren Y, Effler JC, Norstrom M, Luo T, Firtel RA, Iglesias PA, Rock RS, Robinson DN. Mechanosensing through cooperative interactions between myosin II and the actin crosslinker cortexillin I. Curr Biol. 2009; 19:1421–1428. [PubMed: 19646871]

Richman DP, Stewart RM, Hutchinson JW, Caviness VS Jr. Mechanical model of brain convolutional development. Science. 1975; 189:18–21. [PubMed: 1135626]

Rodriguez EK, Hoger A, McCulloch AD. Stress-dependent finite growth in soft elastic tissues. J Biomech. 1994; 27:455–467. [PubMed: 8188726]

Romanoff, AL. The avian embryo: structural and functional development. New York: Macmillan; 1960.

Sadovsky A, Wan FYM. The elastodynamics of embryonic epidermal wound closure. Stud Appl Math. 2007; 118:365–395.

Sato Y, Poynter G, Huss D, Filla MB, Czirok A, Rongish BJ, Little CD, Fraser SE, Lansford R. Dynamic analysis of vascular morphogenesis using transgenic quail embryos. PLoS ONE. 2010; 5:e12674. [PubMed: 20856866]

Savin T, Kurpios NA, Shyer AE, Florescu P, Liang H, Mahadevan L, Tabin CJ. On the growth and form of the gut. Nature. 2011; 476:57–62. [PubMed: 21814276]

Schoenwolf GC, Smith JL. Gastrulation and early mesodermal patterning in vertebrates. Methods Mol Biol. 2000; 135:113–25. [PubMed: 10791309]

Serini G, Ambrosi D, Giraudo E, Gamba A, Preziosi L, Bussolino F. Modeling the early stages of vascular network assembly. EMBO J. 2003; 22:1771–1779. [PubMed: 12682010]

Sherrard K, Robin F, Lemaire P, Munro E. Sequential activation of apical and basolateral contractility drives ascidian endoderm invagination. Curr Biol. 2010; 20:1499–1510. [PubMed: 20691592]

Sherratt JA, Martin P, Murray JD, Lewis J. Mathematical models of wound healing in embryonic and adult epidermis. IMA J Math Appl Med Biol. 1992; 9:177–196. [PubMed: 1295928]

Shiraishi I, Takamatsu T, Minamikawa T, Fujita S. 3-D observation of actin filaments during cardiac myofibrinogenesis in chick embryo using a confocal laser scanning microscope. Anat Embryol. 1992; 185:401–408. [PubMed: 1609966]

Shore TW, Pickering JW. The proamnion and amnion in the chick. J Anat Physiol. 1889; 24:1–21.

Sissman NJ. Cell multiplication rates during development of the primitive cardiac tube in the chick embryo. Nature. 1966; 210:504–507. [PubMed: 5960515]

Solon J, Kaya-Copur A, Colombelli J, Brunner D. Pulsed forces timed by a ratchet-like mechanism drive directed tissue movement during dorsal closure. Cell. 2009; 137:1331–1342. [PubMed: 19563762]

Sonnemann KJ, Bement WM. Wound repair: toward understanding and integration of single-cell and multicellular wound responses. Annu Rev Cell Dev Biol. 2011; 27:237–263. [PubMed: 21721944]

Spitzer, A.; Lev, M. The Architecture of Normal and Malformed Hearts. Vol. 45. Springfield, IL: Thomas; 1951.

Stalsberg H. Regional mitotic activity in the precardiac mesoderm and differentiating heart tube in the chick embryo. Dev Biol. 1969; 20:18–45. [PubMed: 5795847]

Stalsberg H, DeHaan RL. Endodermal movements during foregut formation in the chick embryo. Dev Biol. 1968; 18:198–215. [PubMed: 5672880]

Stalsberg H, DeHaan RL. The precardiac areas and formation of the tubular heart in the chick embryo. Dev Biol. 1969; 19:128–59. [PubMed: 4180350]

Steinberg MS. Reconstruction of tissues by dissociated cells. some morphogenetic tissue movements and the sorting out of embryonic cells may have a common explanation. Science. 1963; 141:401–408. [PubMed: 13983728]

Steinberg MS. Differential adhesion in morphogenesis: a modern view. Curr Opin Genet Dev. 2007; 17:281–286. [PubMed: 17624758]

Stylianopoulos T, Barocas VH. Multiscale, structure-based modeling for the elastic mechanical behavior of arterial walls. J Biomech Eng. 2007; 129:611–618. [PubMed: 17655483]

Taber LA. Biomechanics of growth, remodeling, and morphogenesis. Appl Mech Rev. 1995; 48:487.

Taber LA. Biophysical mechanisms of cardiac looping. Int J Dev Biol. 2006; 50:323–332. [PubMed: 16479500]

Taber LA. Theoretical study of Beloussov's hyper-restoration hypothesis for mechanical regulation of morphogenesis. Biomech Model Mechanobiol. 2008; 7:427–441. [PubMed: 17909868]

Taber LA. Towards a unified theory for morphomechanics. Philos T R Soc A. 2009; 367:3555–3583.

Taber LA, Lin IE, Clark EB. Mechanics of cardiac looping. Dev Dyn. 1995; 203:42–50. [PubMed: 7647373]

Taniguchi K, Maeda R, Ando T, Okumura T, Nakazawa N, Hatori R, Nakamura M, Hozumi S, Fujiwara H, Matsuno K. Chirality in planar cell shape contributes to left-right asymmetric epithelial morphogenesis. Science. 2011; 333:339–341. [PubMed: 21764746]

Thompson, DW. On growth and form. Cambridge University Press; Cambridge: 1942.

Toro R, Burnod Y. A morphogenetic model for the development of cortical convolutions. Cereb Cortex. 2005; 15:1900–13. [PubMed: 15758198]

Tosin A, Ambrosi D, Preziosi L. Mechanics and chemotaxis in the morphogenesis of vascular networks. Bull Math Biol. 2006; 68:1819–1836. [PubMed: 16817028]

Trinkaus, JP. Cells into Organs: The Forces That Shape the Embryo. Prentice Hall; 1984.

Turing AM. The chemical basis of morphogenesis. Philos T R Soc B. 1952; 237:37–72.

Urdy S. On the evolution of morphogenetic models: mechano-chemical interactions and an integrated view of cell differentiation, growth, pattern formation and morphogenesis. Biological Reviews. 2012 no–no.

Vailhé B, Vittet D, Feige JJ. In vitro models of vasculogenesis and angiogenesis. Lab Invest. 2001; 81:439–452. [PubMed: 11304563]

Varner, VD.; Taber, LA. On Measuring Stress Distributions in Epithelia. In: Garikipati, K.; Arruda, EM., editors. IUTAM Symposium on Cellular, Molecular and Tissue Mechanics. Springer; Netherlands: 2010. p. 45-54.

Varner VD, Voronov DA, Taber LA. Mechanics of head fold formation: investigating tissue-level forces during early development. Development. 2010; 137:3801–11. [PubMed: 20929950]

Vernon RB, Lara SL, Drake CJ, Iruela-Arispe ML, Angello JC, Little CD, Wight TN, Sage EH. Organized type I collagen influences endothelial patterns during "spontaneous angiogenesis in vitro": planar cultures as models of vascular development. In Vitro Cell Dev Biol Anim. 1995; 31:120–131. [PubMed: 7537585]

Vogel V, Sheetz M. Local force and geometry sensing regulate cell functions. Nat Rev Mol Cell Bio. 2006; 7:265–275. [PubMed: 16607289]

Voronov DA, Alford PW, Xu G, Taber LA. The role of mechanical forces in dextral rotation during cardiac looping in the chick embryo. Dev Biol. 2004; 272:339–350. [PubMed: 15282152]

Wan X, Li Z, Lubkin SR. Mechanics of mesenchymal contribution to clefting force in branching morphogenesis. Biomech Model Mechanobiol. 2008; 7:417–426. [PubMed: 17901991]

Weiss, PA. Principles of development: A text in experimental embryology. Henry Holt and Company, Inc; New York: 1939.

Weliky M, Minsuk S, Keller R, Oster G. Notochord morphogenesis in *Xenopus laevis*: simulation of cell behavior underlying tissue convergence and extension. Development. 1991; 113:1231–1244. [PubMed: 1811939]

Weliky M, Oster G. The mechanical basis of cell rearrangement. I. epithelial morphogenesis during fundulus epiboly. Development. 1990; 109:373–386. [PubMed: 2401201]

Wolpert L. Positional information and the spatial pattern of cellular differentiation. J Theor Biol. 1969; 25:1–47. [PubMed: 4390734]

Wood W, Jacinto A, Grose R, Woolner S, Gale J, Wilson C, Martin P. Wound healing recapitulates morphogenesis in *Drosophila* embryos. Nat Cell Biol. 2002; 4:907–912. [PubMed: 12402048]

Wozniak MA, Chen CS. Mechanotransduction in development: a growing role for contractility. Nat Rev Mol Cell Bio. 2009; 10:34–43. [PubMed: 19197330]

Xu G, Kemp PS, Hwu JA, Beagley AM, Bayly PV, Taber LA. Opening angles and material properties of the early embryonic chick brain. J Biomech Eng. 2010; 132:011005. [PubMed: 20524743]

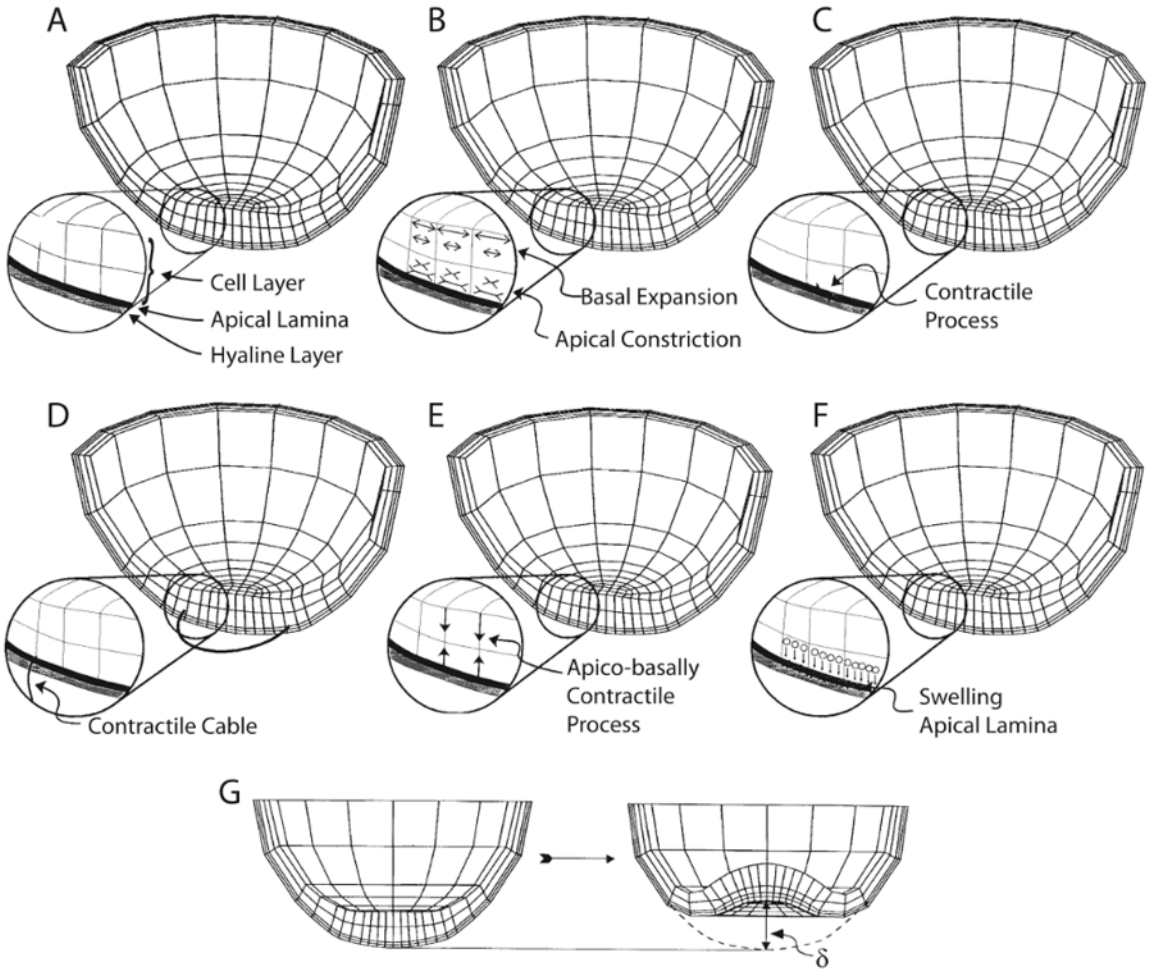

Figure 1.

Representations of various proposed mechanisms in finite element models for sea urchin invagination (Davidson et al., 1995). (A) General features of the models. (B) Apical constriction. (C) Cell tractor. (D) Multicellular contractile ring surrounding invaginating region. (E) Apico-basal contraction. (F) Bending caused by gel swelling. (G) Representative deformation of embryo. From Davidson et al. (1995).

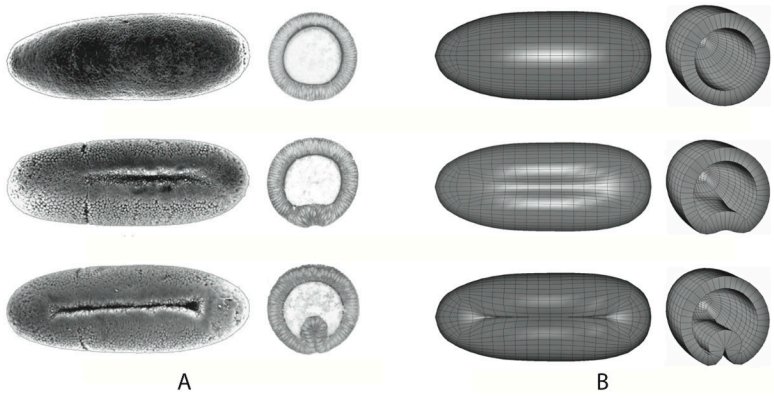

Figure 2.
Experiments and 3D model for *Drosophila* gastrulation. (A) Ventral (from Grumbling and Strelets, 2006) and cross-sectional (from Muñoz et al., 2007) views of ventral furrow formation in experiments. (B) Same views from finite element model. From Conte et al. (2008).

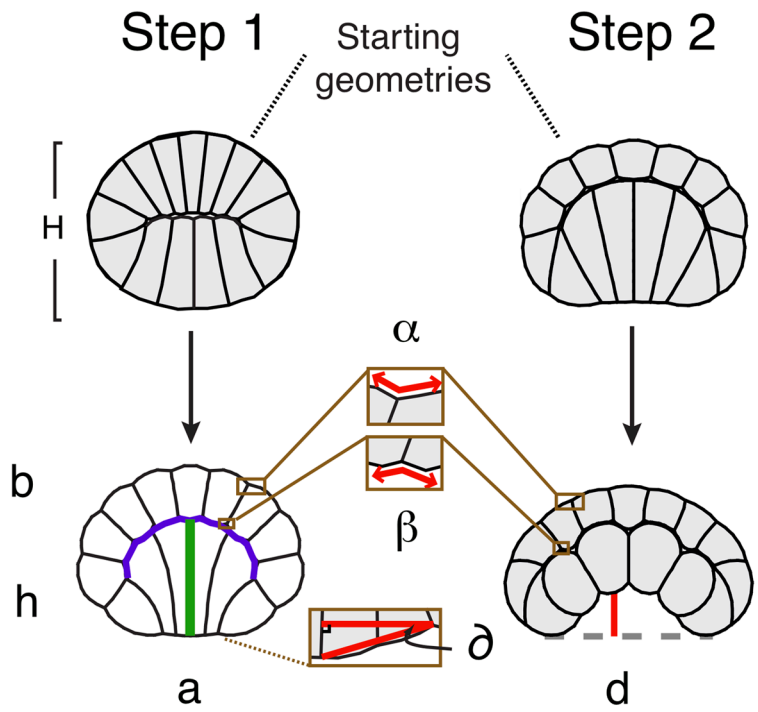

Figure 3.
Two-step process for invagination in ascidian gastrulation. Step 1: Apical constriction results in wedge-shaped cells in endoderm surrounded by ectodermal cells. Step 2: Subsequent apico-basal contraction of endodermal cells results in invagination. From Sherrard et al. (2010).

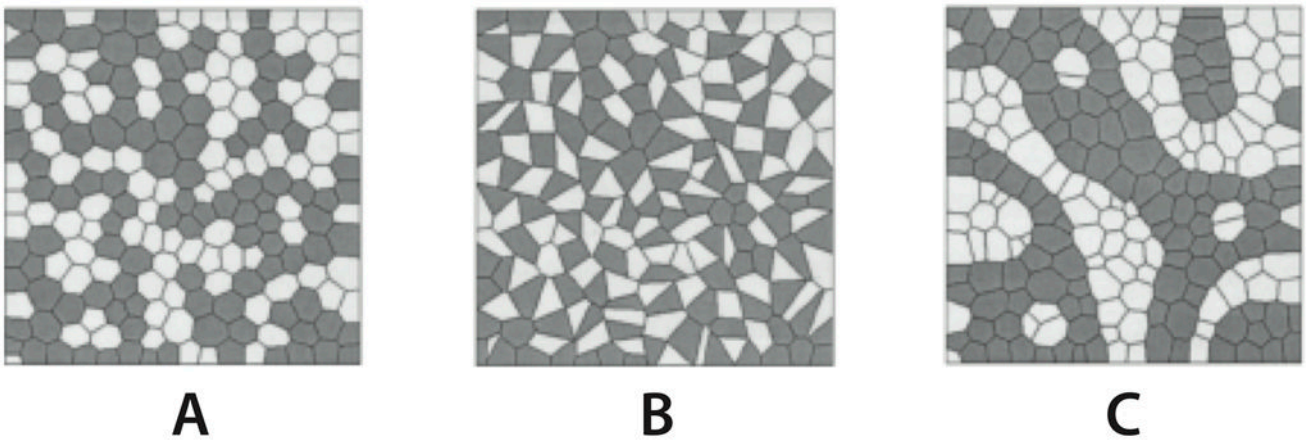

Figure 4.

Cell mixing and sorting in simulations of heterotypic cellular aggregates. (A) Initial configuration for both simulations. A low interfacial tension between light and dark cell types leads to total mixing (B) while sorting occurs with high interfacial tension (C). From Brodland (2002).

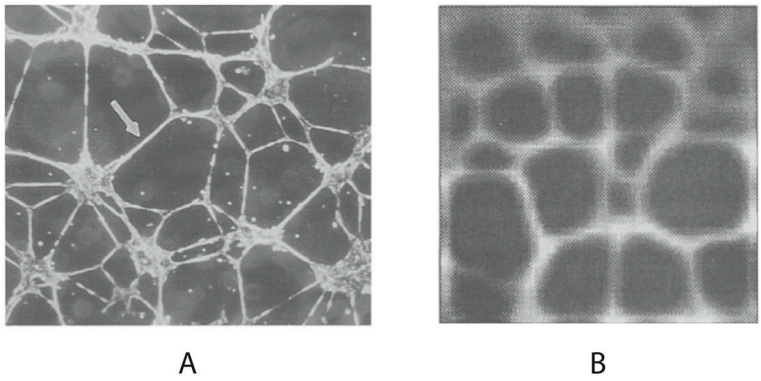

Figure 5.
Pattern formation in vasculogenesis. (A) Endothelial cells cultured on a matrix gel aggregate and form cord-like structures. (B) Computer simulation of this process recapitulates the observed pattern. From Manoussaki et al. (1996).

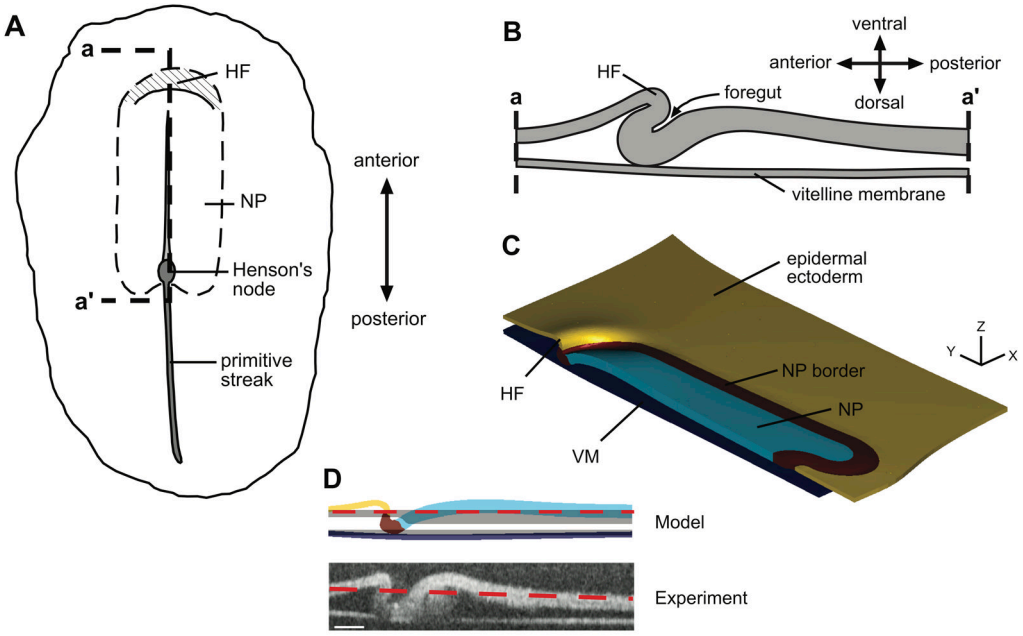

Figure 6.

Head fold formation in chick embryo. (A) Ventral aspect of the embryo illustrating the head fold (HF) and neural plate (NP) regions. (B) Out-of-plane structure of the head fold shown in sagittal section. (C) 3D finite element model for HF formation (VM = vitelline membrane). (D) Correspondence between experiment and model in sagittal section. From Varner and Taber (2010).

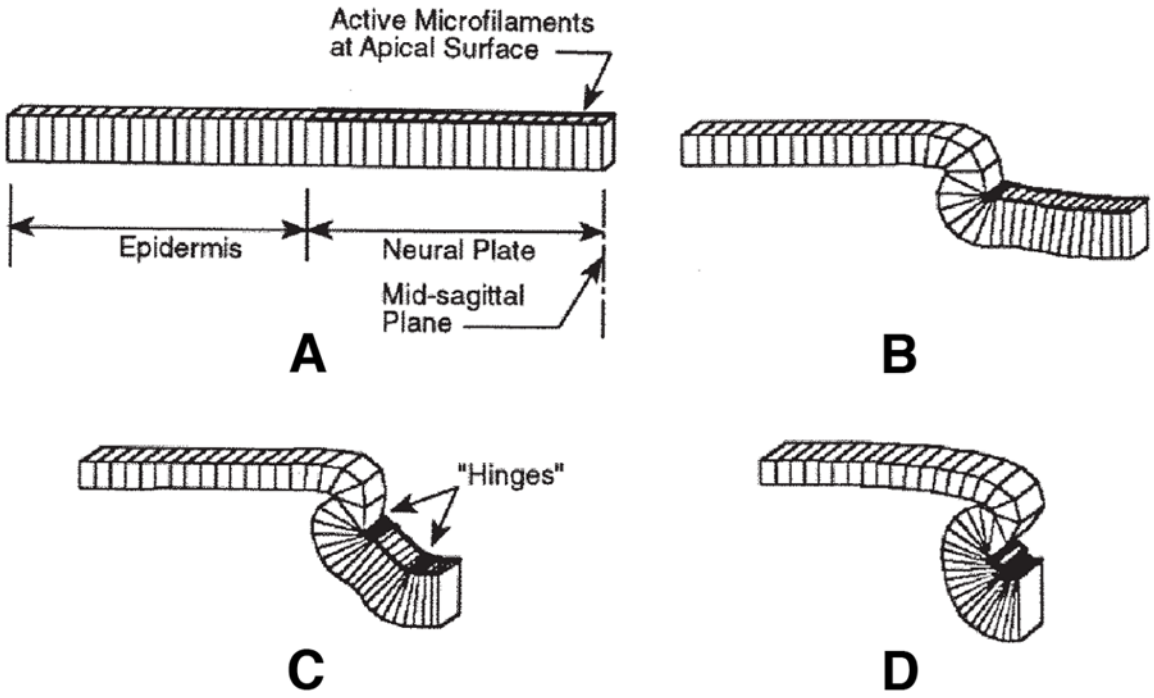

Figure 7.
Simulation of neurulation via apical contraction. (A) The model considers one half of a symmetric transverse strip of ectoderm. As contraction occurs in a length-dependent fashion, a neural ridge-like structure develops, followed by narrowing and thickening of the neural plate as hinge points develop (B–C), and closure of the neural tube (D). From Clausi and Brodland (1993).

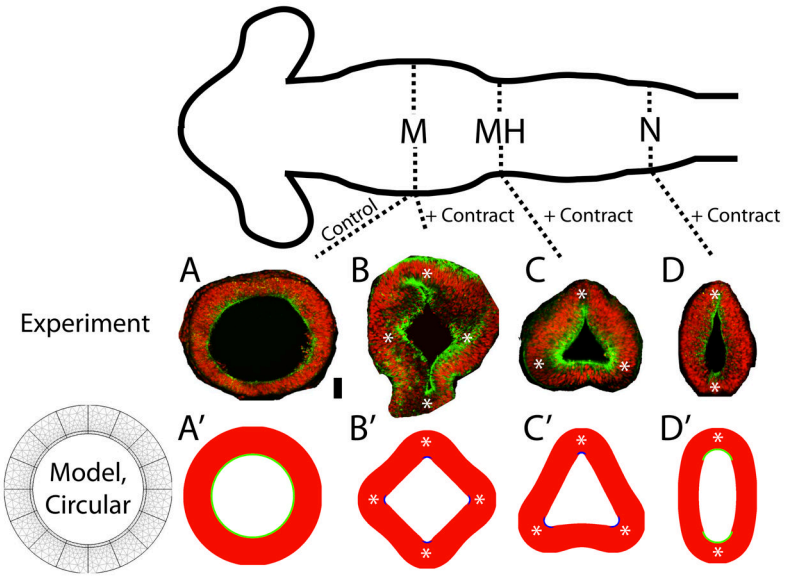

Figure 8.

Brain morphogenesis driven by local apical contraction. (A–D) Cross-sectional views of the chick brain and F-actin distribution (green) at the midbrain (M), mid-hindbrain boundary (MH) and posterior neuroepithelium (N). Normal midbrains (A) have a relatively round shape and uniform F-actin distribution, while enhancing contraction results in cross-sections with sharp corners and high F-actin density (asterisk in B–D). (A′–D′) Computational models of corresponding cross-sections. From Filas et al. (2012).

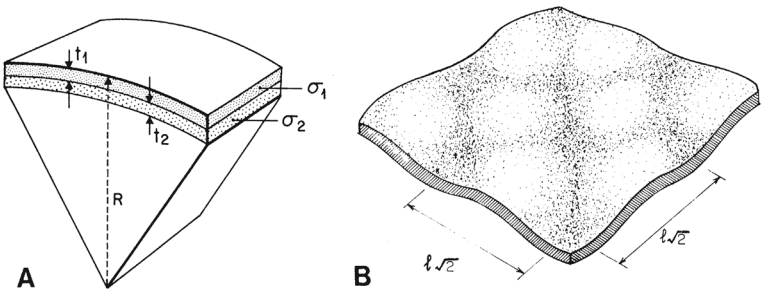

Figure 9.
Schematic of model for brain folding (Richman et al., 1975). (A) Bilayer model for the cerebral cortex. (B) Relatively rapid growth of cortex causes buckling. From Richman et al. (1975).

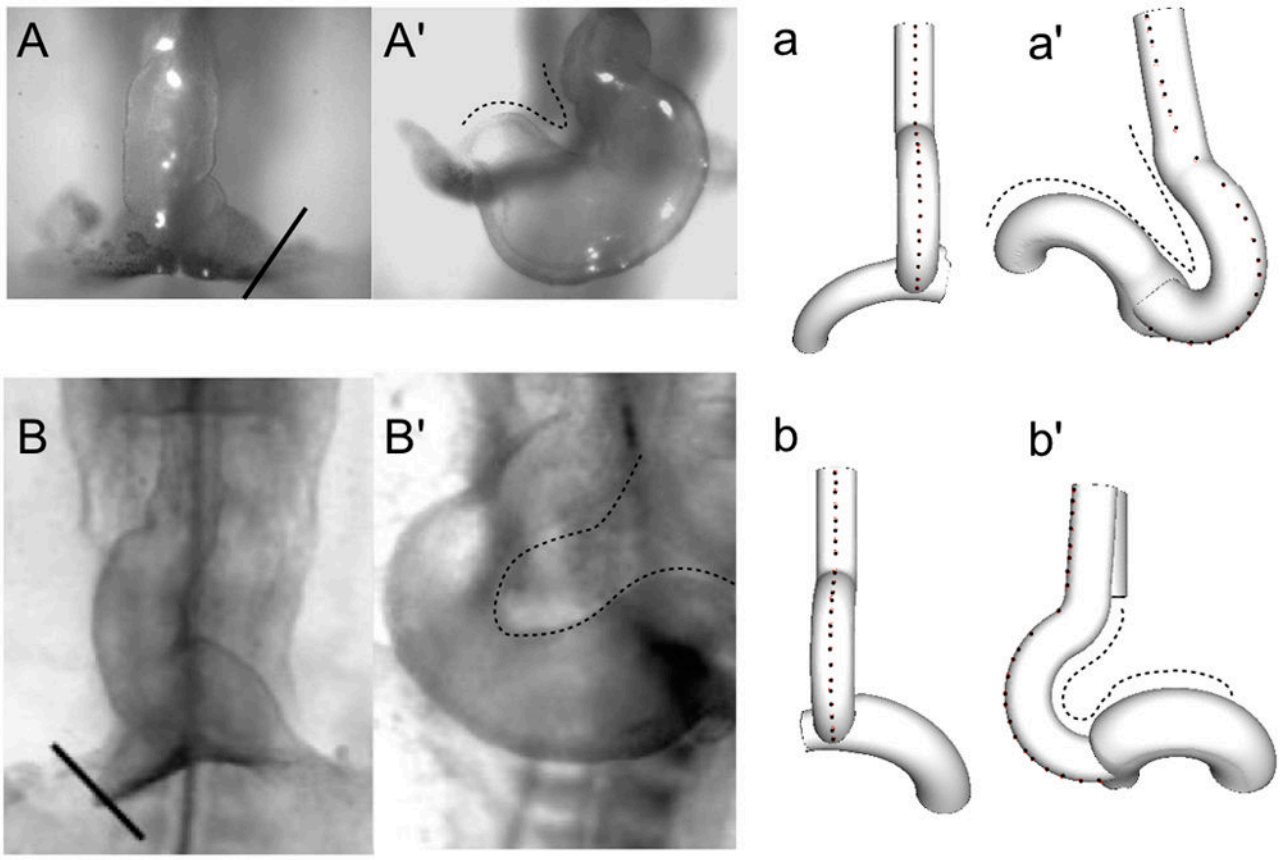

Figure 10.
Effects of atria removal on cardiac looping in chick embryo. Left and right atria removed in top and bottom panels, respectively. Initial (first column) and final (after 12 hours, second column) heart configurations in experiment, with corresponding simulation results (third and fourth columns). Removal of the left and right atria results in leftward and rightward rotation, respectively, in both experiment and model. From Ramasubramanian et al. (2008).

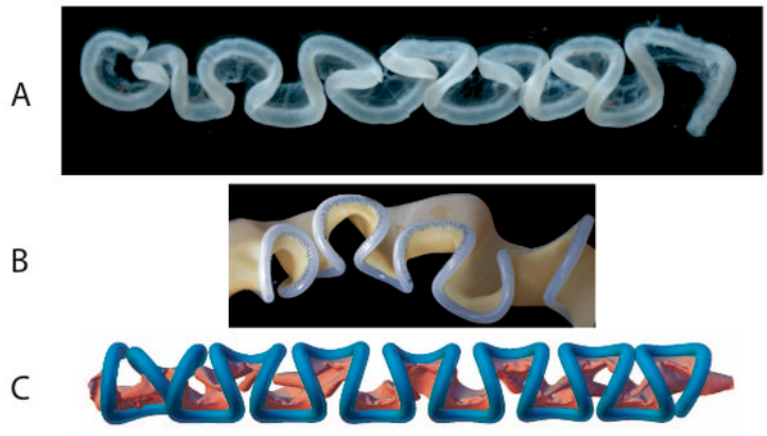

Figure 11.

Gut looping patterns. (A) Experimental chick gut morphology after 16 days incubation. (B) A pre-stretched rubber sheet is stitched to the side of a straight rubber tube which, upon release, deforms into a shape mimicking the looped gut. (C) A computer simulation reproduces the gut looping morphology. From Savin et al. (2011).

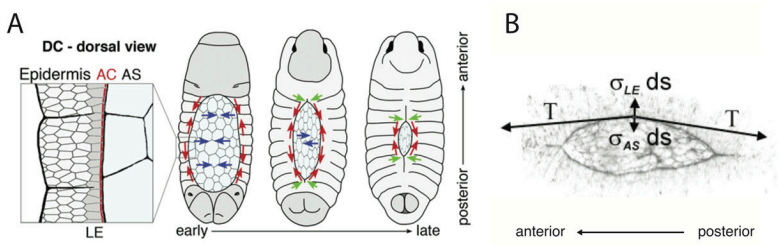

Figure 12.

(A) Schematic of dorsal closure (DC) in *Drosophila* embryo. Actin cable (AC), leading edge (LE) of the lateral epidermis, and amnioserosa (AS) are illustrated (left inset), as are the forces generated by AC contraction (red), AS contraction (blue), and zipping (green) over the course of dorsal closure. From Solon et al. (2009). (B) Force balance diagram at one point of the LE, showing the contributions of the amnioserosa ($\sigma_{AS}ds$), the lateral epidermis ($\sigma_{LE}ds$) and the tension of the actin cable (T). From Hutson et al. (2003).