Implications of domain growth in morphogenesis

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Abstract. Domain growth can play an important role in pattern formation during early embryonic development. By considering the Turing reaction-diffusion model for pattern formation, we show how growth can influence patterning. We consider how growth affects mode selection and robustness of patterns. Specifically, we investigate ligament patterns in arcoid bivalves.

1 Introduction

Uncovering the mechanisms underlying the formation of spatiotemporal pattern in biological systems has been the goal of experimentalists and theoreticians alike for a number of decades. One approach assumes that pattern formation is a self-organising or emergent property of the system being investigated. Several theoretical models for spatial pattern formation have been proposed (see [1] for review). Perhaps the most well-studied of these is the reaction-diffusion theory first proposed by Turing in 1952 [2] and its later extensions [3] in which it was shown that a system of reacting chemicals, stable in the absence of diffusion, could be driven unstable by diffusion. Turing proposed that cells would respond to specific concentrations of chemicals (he coined the term *morphogen* for these chemicals) and differentiate accordingly. Since the appearance of Turing's seminal paper, a number of reaction-diffusion models have been proposed for pattern formation. These all rely on the diffusion-driven instability mechanism proposed by Turing, differing only in the terms that govern reaction kinetics [1].

Turing structures have been found in chemistry but there is still no conclusive demonstration that they occur in living organisms (see [4] for review) although morphogens are now known to exist ([5]). Recently, Kondo and Asai [6] observed that pigmentation patterns in fish changed qualitatively due to growth of the individual. They showed that the resulting changes in patterns were consistent with those exhibited by a Turing reaction-diffusion model simulated on a growing domain. This finding renewed interest in this subject area, which was first prompted in 1979 when Newman and Frisch [7] proposed

that the sequential formation of skeletal elements in the chick limb could be accounted for by considering the effects on a Turing model of the changes in size and geometry of the progress zone, a specialised zone of undifferentiated cells situated at the growing tip of the limb bud.

Kulesa *et al.*, 1996 [8] investigated the spatiotemporal sequence of tooth primordia initiation in the growing alligator jaw via a Turing mechanism augmented with a further equation for the diffusion and degradation of a control chemical and showed that this modified Turing model exhibited pattern formation similar to that observed experimentally. More recently, Chaplain *et al.*, 2001 [9] computed the solution to the Turing model on a growing spherical surface, with application to cancerous tumour growth.

Several such studies simply examine the steady states of a particular Turing reaction-diffusion model for different values of the scaling parameter, or they incorporate growth in an *ad hoc* manner. Recently, we have reformulated the model on a growing domain from first principles and we have started to study the properties of the patterns exhibited by the model. In this paper, we briefly review some of this work. In Section 2, we present the model equations and describe some of the different types of behaviour observed on a spatially one-dimensional domain due to growth. In Section 3 we illustrate the potential uses of the model with an application to ligament patterns in arcoid bivalves, and we discuss future research directions in Section 4.

2 The effects of growth on a one-dimensional domain

The two species Turing reaction-diffusion model takes the general form:

$$\frac{\partial u}{\partial t} = D_u \nabla^2 u + f(u,v), \tag{1}$$

$$\frac{\partial v}{\partial t} = D_v \nabla^2 v + g(u,v), \tag{2}$$

where $u(\underline{x},t)$ and $v(\underline{x},t)$ are the concentrations of chemicals u and v at spatial position $\underline{x}$ and time t, and the kinetic functions f and g describe the nonlinear reaction between the chemicals. Typical boundary conditions on the spatial domain are either zero flux (Neuman) or fixed (Dirichlet). It can be shown [10], using Reynold's Transport Theorem, that on a growing domain, the model becomes

$$\frac{\partial u}{\partial t} + \nabla \cdot (\underline{a}u) = D_u \nabla^2 u + f(u,v), \tag{3}$$

$$\frac{\partial v}{\partial t} + \nabla \cdot (\underline{a}v) = D_v \nabla^2 v + g(u,v), \tag{4}$$

where $\underline{a}(\underline{x},t)$ is the velocity field generated by the distributed source term $S(\underline{x},t)$ such that $\nabla \cdot \underline{a} = S(\underline{x},t)$. If all cells have the same rate of proliferation throughout the domain then $S(\underline{x},t) = r$ is constant over the domain, and in

one spatial dimension the domain length is given by $L(t) = L_0 e^{rt}$, where L_0 is the initial length of the interval.

Numerical solution of the above model in one dimension shows that for different reaction kinetics it is possible to obtain, as the domain grows exponentially, either an invariant pattern, mode doubling via insertion or splitting, or mode tripling (see Fig. 1). We have shown numerically that this type of

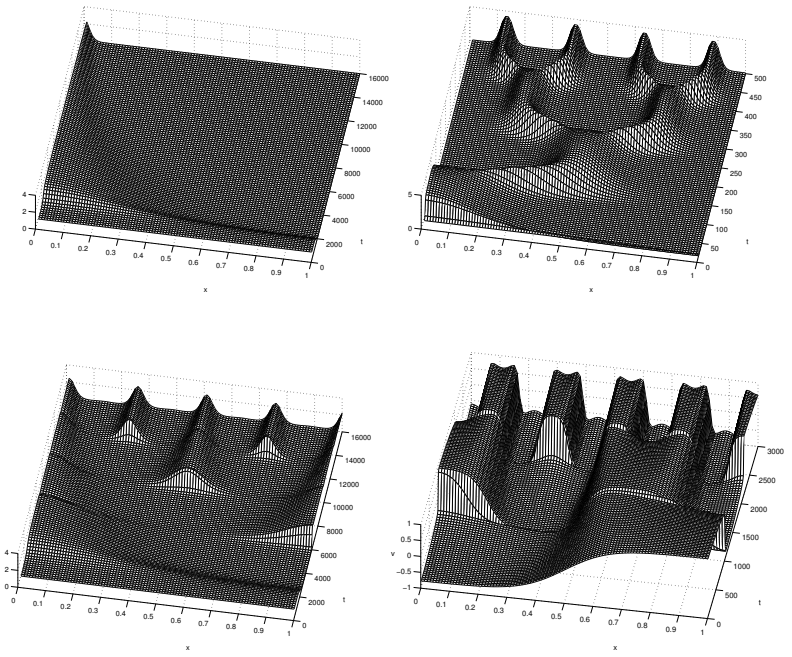

Fig. 1. Invariance, mode doubling (by splitting and insertion), and mode tripling. See text for details. Note that the domain has been rescaled to $[0, 1]$.

behaviour can occur over several orders of magnitude in the growth rate. By introducing a piecewise linear approximation, closed form solutions can be found for steady state patterns for the case where the ratio of diffusion coefficients is small. By considering the existence of these steady-state solutions as the domain length is varied we can predict the point at which the solution ceases to exist. We identify this point with the onset of transition between patterns for the sequence generated on the growing domain. We are thus able to determine the mechanism and timing of transitions between quasi-steady patterns in the sequence and predict the behaviour in terms of the nonlinear kinetics of a particular model [11]. Moreover, we have considered different types of growth, for example, logistic, and linear, and determined if,

and when, mode doubling breaks down [10]. In particular, it can be shown that domain growth greatly enhances the robustness of spatial patterns with a mode which is a multiple of 2, suggesting that domain growth may be a powerful pattern selection mechanism.

3 Application to bivalve ligament morphogenesis

We now consider the application of the Turing model to ligament patterning in bivalve shells (we refer the reader to [12] for full details). The dorsal ligaments of arcoid bivalves typically consist of oblique, lamellar and fibrous sheets, alternating along the hinge so their attachments on the two valves form characteristic chevron patterns (see Fig. 2 for schematic). New elements are added at or near the middle of the growth zone as the ligament expands ventrally. Most Palaeozoic arcoids exhibit this growth pattern. In the Early Cretaceous period, a novel pattern emerged, with vertical strips of lamellar ligament embedded in grooves within the sheet of fibrous ligament that is attached to each valve. New elements are added at each end of the ligament, anteriorly and posteriorly. This distinctive growth pattern is the defining character of the family Noetiidae.

Variation among individuals within populations of a living arcoid, *Limopsis grandis*, includes large, adult shells with vertical strips of lamellar ligament. The variants show how the noetiid growth pattern could have been derived from the chevron pattern, as the domain of ligament growth expanded. The latter can be generated by solving a typical Turing reaction-diffusion model numerically on a growing domain with Dirichlet boundary conditions (see Fig. 2). Moreover, the noetiid growth pattern can simply be derived from the chevron pattern by fixing the value of the morphogen at the centre of the domain. These results indicate that striking differences in form may arise from modest changes in developmental process and they suggest that the evolution of the Noetiidae should now be reassessed.

4 Discussion

There are still many unanswered mathematical questions concerning the patterning properties exhibited by Turing reaction-diffusion models on one spatial dimension. For example, why the mode doubling behaviour breaks down in the exponentially growing case for growth rates that are extremely small or extremely large is still an open question. Our recent analysis shows that this problem may share some similarities with self-replicating dynamics on a large fixed domain [13]. Spike solutions can also undergo the sorts of transitions we have reviewed here and this has been investigated in a number of papers (see [11] for references). We have carried out numerical simulations for the case of a non-uniformly growing domain in order to explore, amongst other things, the possibility of robustly selecting modes which are not a multiple of

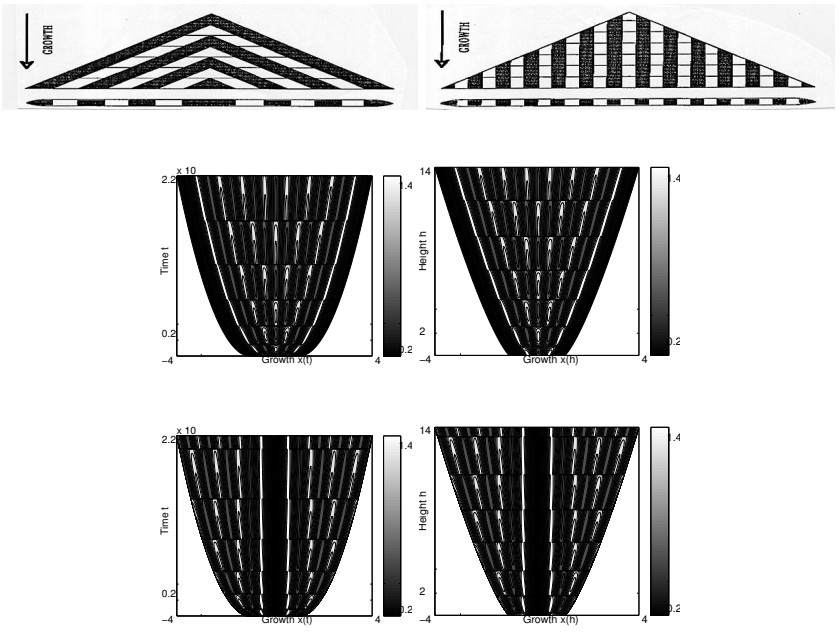

Fig. 2. Top panel: Growth patterns of the chevron ligament typical of *Glycymeris* and the noetiid ligament in vertical, saggital cross-section. These patterns are also observed on the surfaces where the ligament is attached to each valve. Middle panel: Numerical results are consistent with the growth patterns of *Glycymeris* (compare with Top left panel). Bottom panel: Results are consistent with the patterns observed in noetiid ligament growth (compare with Top right panel).

2 [14]. We are unaware of any mathematical analysis for such cases. The problem is further complicated in higher spatial dimensions due to degeneracy of solutions. This is presently under investigation.

We have documented one specific biological application of a Turing model in this paper. There are potentially many others. For example, one of the main criticisms of the Turing model has been that it cannot produce robust patterns. However, our analyses show that domain growth can select certain types of pattern in a very robust manner, at least for one-dimensional domains. In two dimensions, we have recently shown that a Turing model can account for pigmentation patterning on the adult wing of the butterfly *Papilo dardanus* [15]. We carried out these simulations on a geometrically accurate adult wing shape, but it is known that the pigmentation process occurs as the butterfly is growing. We are presently solving the model computationally on a growing domain to see if this has any major implications for the patterns observed on the adult wing.

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