Journal of Theoretical Biology 363 (2014) 344–356

Contents lists available at ScienceDirect

Journal of Theoretical Biology

journal homepage: www.elsevier.com/locate/yjtbi

Spatial and temporal dynamics of cell generations within an invasion wave: A link to cell lineage tracing

Bevan L. Cheeseman [a], Donald F. Newgreen [b], Kerry A. Landman [a,*]

[a] Department of Mathematics and Statistics, University of Melbourne, Victoria 3010, Australia
[b] Murdoch Childrens Research Institute, Royal Children's Hospital, Parkville, Victoria 3052 Australia

HIGHLIGHTS

- Determine spatial-temporal dynamics of cell generations in an invasion wave.
- Develop new PDE model and compare with CA model.
- Organized spatial distribution of cell generations obtained.
- Develop new technique to obtain individual lineage tracings from generation data.
- Provides a potential technique for deducing cell lineage data.

ARTICLE INFO

Article history:
Received 8 April 2014
Received in revised form
6 August 2014
Accepted 7 August 2014
Available online 19 August 2014

Keywords:
Cell invasion
Galton–Watson process
Fisher wave
Agent-based model
Continuum multispecies model

ABSTRACT

Mathematical models of a cell invasion wave have included both continuum partial differential equation (PDE) approaches and discrete agent-based cellular automata (CA) approaches. Here we are interested in modelling the spatial and temporal dynamics of the number of divisions (generation number) that cells have undergone by any time point within an invasion wave. In the CA framework this is performed from agent lineage tracings, while in the PDE approach a multi-species generalized Fisher equation is derived for the cell density within each generation. Both paradigms exhibit qualitatively similar cell generation densities that are spatially organized, with agents of low generation number rapidly attaining a steady state (with average generation number increasing linearly with distance) behind the moving wave and with evolving high generation number at the wavefront. This regularity in the generation spatial distributions is in contrast to the highly stochastic nature of the underlying lineage dynamics of the population. In addition, we construct a method for determining the lineage tracings of all agents without labelling and tracking the agents, but through either a knowledge of the spatial distribution of the generations or the number of agents in each generation. This involves determining generation-dependent proliferation probabilities and using these to define a generation-dependent Galton–Watson (GDGW) process. Monte-Carlo simulations of the GDGW process are used to determine the individual lineage tracings. The lineages of the GDGW process are analyzed using Lorenz curves and found to be similar to outcomes generated by direct lineage tracing in CA realizations. This analysis provides the basis for a potentially useful technique for deducing cell lineage data when imaging every cell is not feasible.

1. Introduction

Cell invasion is common to many areas of cell biology and has been the subject of much mathematical modelling. Examples include epidermal wound healing (Maini et al., 2004; Cai et al., 2007), tumor invasion (Sherratt and Chaplain, 2001) and developmental migratory morphogenesis (Simpson et al., 2007a). Cell invasion usually involves a population of cells which moves and undergoes cell division (cell proliferation) into previously unoccupied substrates or tissues. The population density attains a preferred maximum, called the carrying capacity, behind the wavefront. Continuum models of cell invasion, using partial differential equations (PDEs), typically use the well-known Fisher equation (Fisher, 1937; Murray, 2002). The equation supports

* Corresponding author. Tel.: +61 3 8344 6762; fax: +61 3 8344 4599.
E-mail address: kerryl@unimelb.edu.au (K.A. Landman).

http://dx.doi.org/10.1016/j.jtbi.2014.08.016
0022-5193/

travelling wave solutions characterized by constant shape and constant speed invasion waves (Murray, 2002). Therefore the population-level behavior is highly predictable and stereotyped.

In recent years, many discrete agent-based models, or cellular automata (CA) models, have been used to model collective cellular behavior such as cell invasion processes (Binder et al., 2008; Binder and Landman, 2009; Deutsch and Dormann, 2005; Hatzikirou et al., 2012; Sander and Deisboeck, 2002). Each agent represents a cell and agent movement and proliferation are determined by probabilities on a regular lattice. From such local rules, simulations of the total population are highly predictable and advance like the Fisher wave (Hatzikirou et al., 2012; Landman et al., 2012; Simpson et al., 2010). An informative feature that a CA model allows over a PDE model is that individual agents can be labelled and tracked, as implemented in current imaging techniques (Boulina et al., 2013; Egawa et al., 2013; Ridenour et al., 2012; Steen et al., 2010; Nishiyama et al., 2012). The CA model results demonstrate that individual agent contributions are highly variable, within this predictable advancement of the total population wave (Landman et al., 2012).

A powerful technique for understanding how cells behave within an animal model is cell lineage tracing. A single cell is marked (labelled) within complex tissues and this mark is inherited by all progeny. Therefore the contributions of an individual cell lineage to a biological process can be traced within a population of cells. Such work has already had a significant impact on stem and progenitor cell research and on developmental biology (Amat and Keller, 2013; Kretzschmar and Watt, 2012; Krzic et al., 2012; Livet et al., 2007; Lichtman et al., 2008; McMahon et al., 2008; Naik et al., 2013; Pan et al., 2013; Olivier et al., 2010; Tomer et al., 2012).

In developmental processes, cell movement and division can be affected by spatially distributed microenvironmental requirements, such as growth factors or nutrients, and the availability and organization of embryonic tissue space. An archetypical process of cell colonization by an invasion wave is the development of the enteric nervous system (ENS). Enteric neural crest (ENC) cells migrate from the foregut as a strictly timetabled wave to colonize the whole of the gastrointestinal tract, forming the ENS (Druckenbrod and Epstein, 2005, 2007; Young et al., 2004). ENC cells are motile and proliferate to a carrying capacity density (Newgreen et al., 1980), governed by competition for resources (e.g. space, growth factor, nutrient, Uesaka et al., 2013). Failure of the invasion process results in Hirschsprung Disease, a relatively common and potentially fatal human birth defect, where the terminal intestine fails to perform peristalsis due to the absence of the ENS.

Recent experimental work on the development of the ENS has uncovered enormous spatial and stochastic variability in the contributions of individual cells within the total ENS cell population (Cheeseman et al., 2014). In the biological experiments, we found a single ENC cell that accounted for approximately one-third of all ENS descendants, but most cells contributed very few descendants to the total ENS population. These experimental results can be understood using agent-based models and econometric data analysis tools (Cheeseman et al., 2014). A consistent and persistent dynamic of lineage tracings emerges. A small proportion of otherwise identical initial cells (agents) accounts for a substantial proportion of the final population of cells (agents) in the colonization and invasion process. We named these individuals as *superstars*.

Such seemingly paradoxical findings between the predictable population-level behavior and the highly variable individual-level lineage tracings warrant further investigation. Here we examine an intermediate level where we keep track of the number of divisions (generation number) that cells undergo within an invasion wave. At a cell division event, a cell is replaced by two daughter cells in the next generation. The spatial and temporal dynamics of the cell density in each generation i ($i=0,1,2,\ldots$,) will be determined in two ways: using agent lineage tracings in the CA approach and a multi-species generalized Fisher equation in the PDE approach. Both modelling paradigms exhibit spatially well-organized cell generation distributions, in contrast to the individual agent tracings.

If all the initial cells/agents could be labelled and their lineage traced, then it is a straightforward process to determine the generation distribution profiles. Here we investigate the much more difficult inverse problem. We construct a method for determining the lineage tracings of all agents at any given time without labelling and tracking the agents, but through a knowledge of the generation distributions at that time. This methodology involves the construction of a generation-dependent Galton–Watson (GDGW) process. The technique may provide a framework for gaining further information from experiments. For example the green fluorophore KikumeGR in cells of a colonizing population can be converted to red fluorescence in a single cell, which is diluted in intensity by each subsequent cell division (Nishiyama et al., 2012; Stark et al., 2008). When the KikumeGR methods are further refined, it is foreseeable that an index of cycle number may be estimated. Such experimental methods in the future may provide the spatio-temporal distribution of cell generations within the ENC cell invasion process. The GDGW process developed here would then provide an estimate of the full ENC cell lineage tracing, when it is not possible to image every cell.

2. Models

2.1. Agent-based CA model

We model the invasion of ENC cells moving and undergoing division, as a two-dimensional process since the ENC cells are restricted to a cylindrical surface within the intestinal tissue. A discrete-time agent-based CA model on a lattice is used, based on previous work (Binder and Landman, 2009; Simpson et al., 2007b). We use a regular square lattice, with unit spacing, length L and width Y. A lattice site can be occupied by at most one agent (representing an ENC cell) at any time t. This describes an exclusion process (Chowdhury et al., 2005). Such a framework is used to simulate ENS colonization by invasion in an *in vitro* setting, where there is no intestinal tissue domain growth. Agent death is not included here, as there is little evidence of ENC cell death during the colonization process (Chalazonitis et al., 2012).

Suppose that the domain contains $m(t)$ agents at time t. During a single time step from t to $t+1$, an asynchronous updating scheme is used. The $m(t)$ ENC agents are selected uniformly at random and first given the opportunity to move and then given the opportunity to proliferate. For a motility event, a chosen agent at (x,y) attempts to move with probability P_m to one of the four nearest neighbour sites $(x\pm1, y\pm1)$ each with probability $1/4$. For a proliferation event, a mother agent at (x,y) attempts to divide with probability P_p, and one daughter remains at (x,y) while the second daughter is placed at either $(x\pm1, y)$ or $(x, y\pm1)$ each with probability $1/4$. (Note that this proliferation rule places daughters adjacent to each other, Binder et al., 2012, rather than being separated by a single site, Binder and Landman, 2009. However, both rules give qualitatively similar results.) If for any motility or proliferation event the target site is occupied by another agent, then that event is aborted, since we are considering an exclusion process. To represent the cylindrical geometry of the intestine, periodic boundary conditions along the horizontal boundaries, no-flux boundary conditions along the left-hand vertical boundary ($x=0$) and the far vertical boundary are imposed (illustrated in results in Fig. 2). The initial condition taken is 10 fully occupied