Field Review

On the Mathematical Representation of Stem Cells

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Himanshu obtained his DPhil in Engineering Science at St Anne's College. His research focused on the development of a modeling platform that can quantify the dynamic relationship between cells and their microenvironment. As part of his research, Himanshu extended the principle of Dynamic Reciprocity as well as introduced the computational principle of Dynamic Assimilation. This article discusses the various methods that could be employed to represent stem cells mathematically. The computational ontologies thereby generated can be employed to develop mathematical models that can provide fundamental insights into the mechanics governing cellular behaviour, which for practical or conceptual reasons have, thus far, eluded observation.

Introduction

Stem cells are the less specialised, precursor cells that are capable of forming any type of more specialised cells in the body of multicellular organisms. *Stemness* can, therefore, be described as the cell's capacity to participate in this specialisation process. In other words, the richer the variety of resulting specialised cells, the more "*potent*" the stemness. Stem Cells are classified based on their source: *embryonic* stem cells are derived from the epiblast tissue of the ICM of a blastocyst[1]; *adult* stem cells are found in adult tissue and include hematopoietic (bone marrow), mesenchymal (connective as well as non-marrow tissue), and neural stem cells[2]; and *amniotic* stem cells come from amniotic fluid[3]. In terms of functionality, stem cells are either unspecialised (Embryonic Stem Cells) or slightly specialised (Mesenchymal Stem Cells, Osteoblasts, etc.). This is to say that while embryonic stem cells can form any

kind of cells (retinal, cardiac, or neural cells, for example) and thus relevant structures in the body, osteoblasts can only lead to bone formation. Beyond their ability to acquire any/a specialised phenotype, stem cells possess limitless proliferative potential[4] (i.e. they can produce a large number of similar cell types) as well as the ability to differentiate into different (specialised) cell types beyond the tissues in which they normally reside[5] ("stem cell plasticity"). This means that a fat cell (adipocyte) can be transformed into a nerve cell. The two features collectively make stem cells appealing from a therapeutic perspective, and form the cornerstone of the technique of cell therapy, which involves reinforcing the compromised tissue or cell population with relevant stem cells. This is usually achieved by delivering the cells via a catheter or syringe, though other alternatives also exist.

Stemness and the Stem Cell Niche

While the exact causes of *stemness* are not yet quite clear, two schools of thought dominate the scene. The first attributes stemness to the cell itself whereas the other claims it to be a part of the stem cell *niche* (Figure 1). Although in French, the term *niche* refers to a dog-house[6], the term *stem cell niche* refers to a dynamic environment replete with anatomical, functional and physiological cues that can promote self-renewal, reproduction, or differentiation[6,7] (the latter being the process by which a less specialised cell turns into a more specialised one). The limited functionality of adult stem cells and hematopoietic stem cells without their niche seems to support the niche concept[6], which Schofield is credited to have first[6,8] proposed in 1978[9].

This instructional relationship, whereby information acting as "cues" flow dynamically, between stem cells and their microenvironment is best captured by the principle of *dynamic*

reciprocity[13], which further extends the instructive behaviour to cells in addition to their extra-cellular matrix (ECM): "*the ECM affects the cell which in turn responds by synthetic and degradative processes causing the composition and the structure of ECM to change which in turn influences the cell and so forth.*"[13] The general inability of cells to form functional structures when cultured as monolayers or on two-dimensional substrates (with certain exceptions) also testifies to the importance of the cells' microenvironment to tissue development. The dependence of tissue micro-architecture on the ECM forming ability of cells further validates this principle[14,15]. In 2013 Kaul et al[16] extended dynamic reciprocity to include the impact of local transport processes on tissue development.

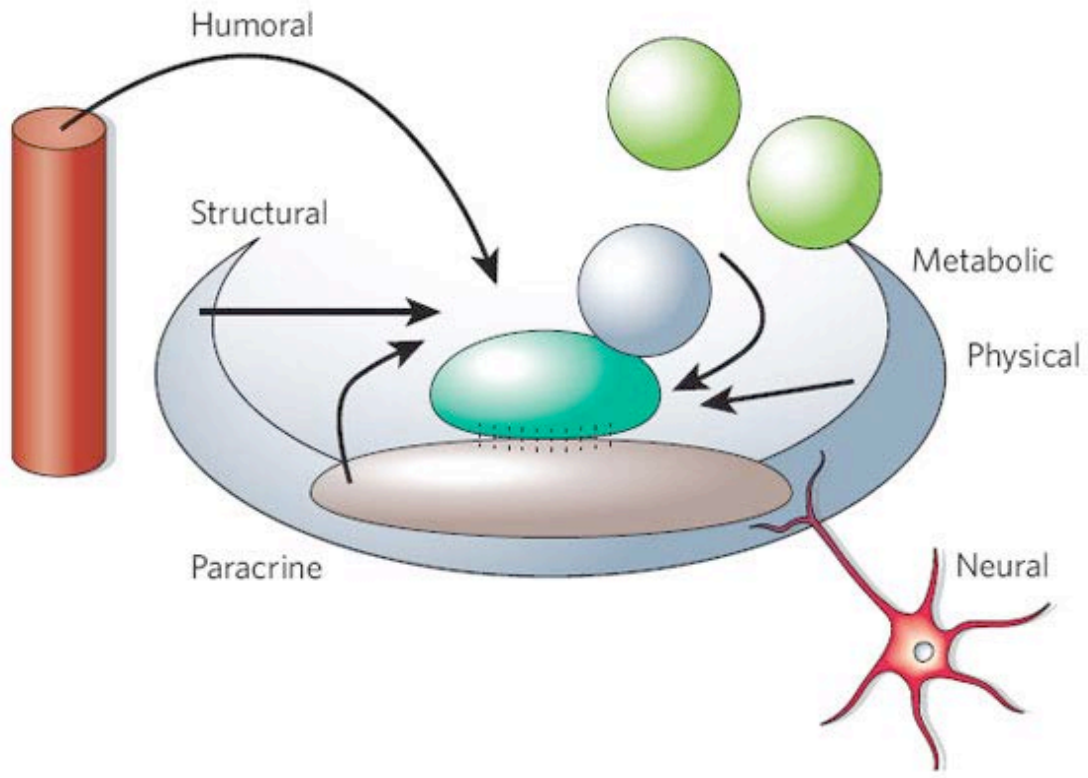

Figure 1: The stem cell niche. The figure is a schematic representation of the stem cell niche and shows the various elements in a cell's microenvironment that can influence the expression of its genotype. The figure also shows the variety of

regulatory signals a cell has to consider in the course of its lifetime. In order to capture the dynamism exhibited by the cellular system, a computational framework must be able to offer suitable ontologies to not only the dynamic structure that is the niche but also capture the broad array of regulatory cues (such as electrical, chemical, mechanical, and architectural) that a cell relies on for growth as well as instructions. *Reproduced with kind permission from Ref.* 6 *© (2006) Nature Publishing Group.*

Computational Modelling of Stem Cell Behaviour

The dynamic reciprocity principle provides a special perspective into stem cell dynamics. Stemness as a property is often considered an *endogenous* attribute: Either a cell has stemness, and will fulfil its role as one, or not. If that is indeed the case, then stem cells divide either:

- *asymmetrically*, producing two daughter cells of different phenotype, one stem and the other more specialised, or
- *symmetrically*, producing two daughter stem cells, thereby preserving the stemness.

Bone marrow transplantation, first performed in 1968, relies on the ability of hematopoietic stem cells to divide symmetrically and in large numbers[17] so as to reconstitute the entire hematopoietic system of the recipient. However, the stem cell *niche* theory challenges that notion and proposes that stemness is an exogenous property that is dependent on the entire microenvironment, not just the cell[6]. The failure of cells to differentiate into desired lineages *ex vivo*, due to lack of relevant chemical, mechanical, or even microenvironmental spatial cues adds credence to this theory. Accordingly, advances in material science and fabrication techniques[18-20] are being used to recreate

the architecture as well as spatiotemporal cues native to the developing tissue.

Stem cell behaviour can be simulated by *population-based*, *cell-based*, *attractor-state*, or *statistical* mathematical approaches. The cell-based approach is a form of the discrete approach, the statistical-based approach a form of the empirical approach, and the rest form the continuum method of modelling stem cell behaviour where a population of stem cells is assumed to behave in an identical manner. In assuming absence of heterogeneous behaviour within cell population these models treat cell population as a continuum. Statistical methods, such as *Bayesian networks*, are employed to mine data sets to identify principal variables impacting the overall behaviour of the population. Such methods can potentially highlight unintuitive network behaviours. However, this approach entails fitting models to experimental data that do not track individual cell responses and, therefore, do not account very well for the heterogeneity of a stem cell niche.

Population-based models rely on ordinary or partial differential equations to capture the global behaviour of stem cell populations. They involve representing stem cell behaviour with mathematical equations that may either be algebraic, or capture variability in cell behaviour temporally or spatiotemporally[21]. Both *statistical* and *population* based models usually ignore the microscopic details of the cellular microenvironment, even cellular heterogeneity, and capture average responses of a cell population[22]. As such, they exhibit large cellular variability[22] and rarely offer much beyond qualitative similarity.

The attractors approach[23], relying on differential equations, better represents the stem cell niche. In the niche context, a set of ordinary or partial differential equations describing a dynamic system will converge towards attractors as stable phenotypes.

Attractors are the equilibrium states to which dynamical systems phase space converges on[8,23]. Equations representing the system can have multiple attractors[23].

To visualise an attractor, imagine a ball rolling around in a landscape with several depressions (Figure 2): the ball represents the cell, and the depressions attractors. The ball settled in the depression indicates that based on the initial conditions, represented by the basin of the attractor[23], the equations have converged to a particular solution.

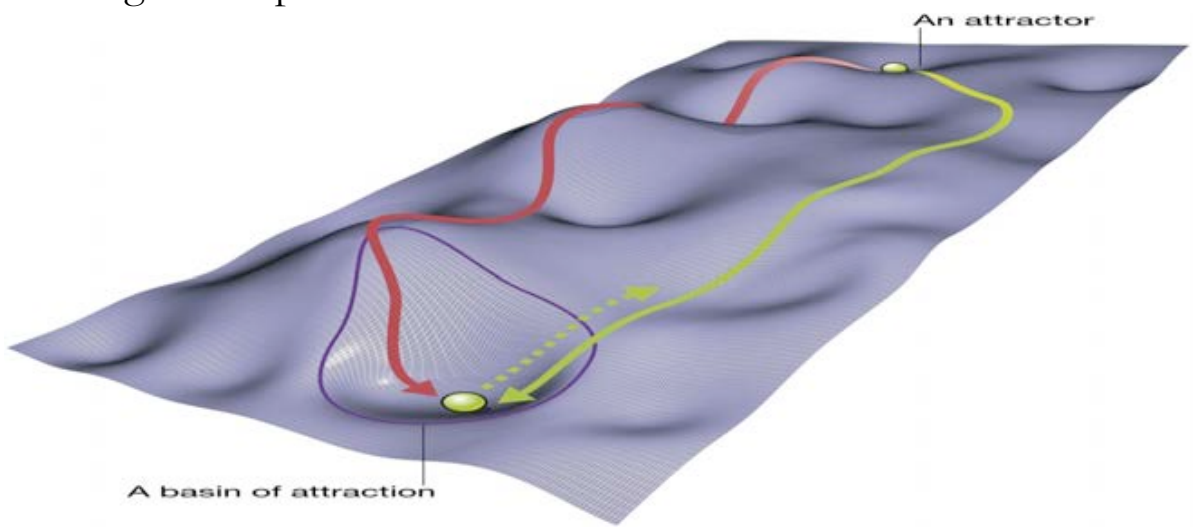

Figure 2: Landscape of Cell Phenotype. The basic idea is to project cellular phenotype or behaviour on a 2D plane, with the third dimension representing the energy of a cell displaying relevant behaviours (such as differentiation, dedifferentiation, proliferation, apoptosis, etc.), which are depicted by the depressions in the landscape. The hollows represent the most stable states, which the cells are likeliest to adopt. Mathematically, the hollows are 'attractors' signifying stable solutions to the equations capturing the dynamics of the concerned system. *Reproduced with kind permission from Ref.* 23 *© (2009) Elsevier.*

In the cellular context this means that the cell, based on endogenous or exogenous variables, has acquired a particular phenotype. There may have been multiple phenotypes (attractors)

that the cell could have acquired, but that is dependent upon its genotype and/or exogenous signals. Although quite promising, Complexity science, which considers dynamic systems in the light of attractors, is still developing.

Single-cell models form a part of the discrete mathematical approach that traces the evolution of a cellular system arising from a combination of cellular interactions and exogenous stimuli. Consequently, they are a better ontological choice (in other words a better form of representation) for the systems being modelled. Cellular automata and agent-based modelling are two of the most common frameworks employed to simulate tissue development based on interactions between (stem) cells and their microenvironment. The problem, however, with the single-cell based approach is that the models tend to be computationally expensive. Other approaches include Chemical equilibrium dynamics[24-26], Activation energy[27], Boolean networks[28], and Kinetic models[29,30].

While there exist a plethora of computational techniques that can be employed to model stem cell behaviour, the accuracy of model-based predictions is heavily reliant on the choice of approach as well as the system being modelled. For example, if the modeller is trying to understand the proliferative rate of stem cells, the statistical approach might seem the best alternative – these models are empirical and computationally fast. However, stem cell proliferation is not an isolated event and, based on the system under consideration, depend upon a variety of parameters: nutrient concentration *in vitro*, presence of growth factors or other relevant protein molecules *in vivo*, etc. Here, the population-based approach, which would consider, and model, the surrounding environment (perfusion rate of a nutrient medium, for example) might capture the system more accurately and provide more reliable predictions.

But what if stem cell proliferation and differentiation additionally depended upon the location of these cells? In this case, the cell-based approach will fare better in predicting system outcomes (and answer the modeller's questions) more accurately as the approach considers each cell individually in determining their response to their immediate environment.

It may appear from the discussion above that statistical models may not be very useful. This is not the case, for while the population- and cell-based approaches capture the underlying physics more accurately than their statistical counterpart, the statistical approach may be utilised if the system being modelled is highly complex (while modelling multiple scales, for example) and/or speed of computation is important. A hybrid model integrating either of the aforementioned approaches can also be employed if accuracy is of the utmost importance. Combining the continuum approach with cell-based approach, as an example, where the cell-based approach accounts for cellular behaviour and continuum approach captures the variations observed in the environment, is arguably a most suitable ontology for simulating stemness, as emerging from the stem cell niche. This hybrid approach can also be utilised if stemness is considered as an endogenous trait. The last few years have witnessed a rapid increase in the application of hybrid models towards modelling biological systems.

In conclusion, there seems to be no single right approach to model stem cell behaviour and stemness. As such, the choice of modelling method(s) depends upon (among other desirables) the complexity of the system being modelled, desired accuracy, speed of outcome, and the questions being asked of the model. However, one needs to be mindful that each of the aforementioned techniques have merits and drawbacks. Yet, each of these

techniques can yield valuable information into the dynamics of stem cell behaviour that may not be apparent from direct observation.

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