Studia Universitatis "Vasile Goldiş", Seria Ştiinţele Vieţii
Vol. 23, issue 1, 2013, pp. 21-27

TISSUE REGENERATION OF CARTILAGE: A COMBINED COMPUTATIONAL AND EXPERIMENTAL STUDY

Iris MATEESCU, Andrei BANCILA, Laura PAUN, Stefan POPESCU, George ROATA, Andreea CIOTEC, Larisa CALU, Daniela BRATOSIN, Manuela SIDOROFF*
National Institute of Research and Development for Biological Sciences, Bucharest, Romania

ABSTRACT. Computational modeling of whole biological systems from cells to organs is gaining momentum in cell biology, disease studies and tissue-engineered constructs. This computational modeling is a framework for obtaining an integrated understanding of key processes, which include: nutrient transport and utilization, matrix formation, cell population dynamics, cell attachment and migration, and local cell–cell interactions. Theoretical modeling has an enormous potential in applications ranging from the interpretation of experimental results and the identification of the main governing processes, to the optimization of practical tissue engineering protocol. This article introduces a series of mathematical and computational models that we have used to understand the mechanisms involved in obtaining biomaterials and optimizing practical tissue engineering protocols.

Keywords: computational methods, modelling, P systems, tissue engineering, chondrocytes, cell proliferation, extracellular matrix, cellular division

INTRODUCTION

Tissue engineering is a promising methodology for the repair of musculoskeletal tissue (Fraj & Roc, 2002; Chung & Burdick, 2008). However, the tissue engineered constructs needs to possess the appropriate biological and mechanical functionality, which is currently insufficient. To improve these functional properties and control their development, every stage in the tissue engineering process is the subject of a complex/elaborated research, starting with the choice of cell source, cell selection, in vitro cell expansion, scaffold design, cell seeding and bioreactor cultivation and conditioning (Hunziker EB, 2001; El-Ghannam A, 2005; Mauney et al., 2005; Mistry AS and Mikos AG, 2005; ROC et al., 2005; Patrachari et al., 2012; Chawla et al., 2012).

The complexity of this research requires a multidisciplinary approach: cellular biology, biochemistry, integrating biology, clinical medicine as well as material science, physics and engineering disciplines.

In vitro culture of a cartilage construct involves numerous internal and external regulatory mechanisms. However, the intrinsic complexity of biological systems means that in many cases it is not possible to separate these mechanisms experimentally. Quantification by means of computational modelling is therefore essential in order to interpret experimental results and to identify the dominating mechanisms (MacArthur et al., 2005). Furthermore, predictive modelling offers huge potential in the optimization of culture conditions and the design of experimental protocols.

The key processes and parameters that determine the success of tissue engineering protocols includes: choosing of nutrient, cell proliferation and population dynamic - including cell growth, attachment, migration and interaction with biochemical factors, matrix formation and local cell-cell interaction. The key issue is how these different aspects need to be combined using computational and experimental methods in order to contribute to an integrated approach to tissue engineering. In the final analysis, an appreciation of the complexity involved in these systems and a need to recognize the limitations of focusing on single parameters is an obvious first step.

One of the well-known problems that appear is homogeneous spatial distribution of cells and/or extracellular matrix (ECM). Typically, the matrix accumulates in the periphery of the scaffold, while in the center we have lower matrix content (may be even empty). The causes for an eventually inadequate growth of the tissue can be associated with: transport restrictions of the nutrients and soluble factors, the removal of waste products, or inhomogeneous cell seeding as well as migration. The main control parameter that determines whether solute gradients will occur is the required cell density in combination with the cellular nutrient utilization rate, under given culture conditions. The transport of nutrients will become a problem to take in consideration during tissue formation, since augmented cell proliferation and matrix formation in the periphery of the scaffold will pose ever increasing transport restrictions for cells in the center of the scaffold (Sengers et al., 2007).

Theoretical models are useful in relating global measurements to local cellular utilization by taking into account the spatial distribution of solutes (i.e.: by modelling the oxygen uptake and diffusive transport in chondrocyte pellets, we can estimate the cellular utilization). This can also help to account for the time dependency that is important when using finite medium volumes, which are most convenient for measuring changes in concentration (Sengers et al., 2007).

Current modelling approaches for transport and utilization offer a relatively simple approach to obtain a

***Correspondence:** Dr. Manuela Sidoroff, National Institute of Research & Development for Biological Sciences Spl. Independentei nº 296, 060031 Bucharest, Romania Tel/Fax: 40.21.2200881 E-mail: manuelasidoroff@yahoo.com*
Article received: October 2012; published: February 2013

first-order estimate used to identify what nutrients are potentially limiting and how will the model evolve from the nutrient point of view. Such models can be applied to design and test operating conditions of hollow bioreactors for bone tissue growth.

For tissue engineering applications, the culture environment should provide optimal conditions for matrix synthesis and for that it is crucial to identify the underlying mechanism and rate limiting process in matrix formation. The computational modelling can provide a major contribution by enabling the investigation of proposed specific relationships in a quantified manner.

Another important aspect to take into consideration when designing a model is cell proliferation and nutrient limitations. For producing rapidly large quantities of ECM, significant cell numbers may be required - but this will imply a high overall utilization rate, leading to nutrient depletion and tissue abnormal (inhomogeneous) development. A good example that can be used to understand cell proliferation of cell growth in cartilage tissue engineering is proposed by Galban and Locke (Galban and Locke et al., 1999). In this model, extensive kinetic relationships are employed, including nutrient saturation, product inhibition and the effects of cell density and cell death. The experimental trends could be represented only quantitatively and not qualitatively (Sengers et al., 2007).

It is important that the model used to provide evidence for a proposed mechanism to be able to capture different experimental conditions. For the matrix synthesis, most studies attribute cell growth to a single limiting factor, without excluding other possibilities. This can be a useful indicator for additional research, but not to be taken as a fact.

Mechanical interactions between different components (cells, water and scaffold material) can determine whether cells form aggregates or disperse throughout the scaffold; also volumetric growth of the different matrix constituents (proteoglycans, collagen) and their mechanical interaction determine the tissue development. This can be described as growth-mixture theory and also multiphase mixture theory that can be used to describe tissue mechanics and swelling in response to changes in solute concentration by including a fluid phase, a charged solid phase and positive and negative ions (Huyghe and Janssen et al., 1997; Mauney et al., 2005). This type of modelling is used to predict and control the shape stability of tissue engineered construct.

Many strategies rely on rapid cell expansion and maintaining the functionality of the tissue. The cell populations derived from marrow are not homogeneous. Often heterogeneous populations are harvested, containing only a small fraction of stem and progenitor cells, which has to be directed towards the relevant tissue type. Studying the behavior of a few cells, one can predict the eventual outcome of the tissue engineering process. In the system formed by these cell populations, stem cell activation, proliferation and differentiation are supervised and controlled with environment factors, like inhibition by available space, cell-cell interactions, biochemical factors or nutrient availability (Andreson et al., 2010).

By integrating both biochemical and mechanical factors, theoretical models can be used to investigate hypotheses for actual mechanisms that govern tissue regeneration.

MATERIALS AND METHODS

A simulation of spreading and tissue regeneration

It is well known that both discrete and continuous models can be used to describe the forming of tissues and the choice between them is depending on the length scale of interest. We can also derive different global parameters from them, like cell diffusion coefficient (from the behaviour of discrete cells). However, the continuous models are more suited to represent the behaviour of large populations of cells, organization patterns are generated by the behaviour of discrete, individual, cells. These types of models are used in regenerative medicine and their purpose is to simulate (predict) tissue regeneration (bone tissue or the repair of the musculoskeletal tissue we can use a simple model to simulate (interpret) the base (simple) evolution of a cell population by starting with the following assumptions:

1) the cell division (growth) is following a known distribution (usually exponential);

2) the cell transformation into the type of tissue needed is happening with a certain probability during a time interval;

3) the cells used to create the tissue (stem cells) multiply, divide and die;

4) the cells forming the tissue also multiply, divide and die;

We say that the system state at time t, X(t) represents the number of each type (the original type A (usually stem cells, bone marrow cells, bone cells) and the needed type B (depending on the type of tissue that is required));

Consider the following cell evolution model, starting at Lotka-Volterra predator-prey model, to simulate the evolution of the cell population (Andreson et al., 2010):

1) $X(t) = (X_A(t), X_B(t))$ is the model state at time t.

2) we use a Poisson process to model a series of observations. The rate of the process is λ (it can be constant or not) (Andreson, 2010).

Thus, we will have:

$$P\{Y_A(t+\Delta t) - Y_A(t) > 0\} \approx 1 - e^{\lambda(t)\Delta t} \approx \lambda(t)\Delta t$$

so we will have:

$R(t) = Y\left(\int_0^t kX_A(s)X_B(s)ds\right)$ where Y is the Poisson process.

Studia Universitatis "Vasile Goldiş", Seria Ştiinţele Vieţii
Vol. 23, issue 1, 2013, pp. 21-27

3) we establish the following rules:

$A \xrightarrow{p1} 2A$, $A \xrightarrow{p2} B$, $A \xrightarrow{p3} \emptyset$, $B \xrightarrow{p4} 2B$, $B \xrightarrow{p5} \emptyset$

We can start with a value for A (optional for B) and the values (definitions) for simulate the model:

a) the deterministic version:

$x(t) = [A(t), B(t)]^T$

$$x(t) = x(0) + p1\int_0^t x_1(s)ds\begin{bmatrix}1\\0\end{bmatrix} + p2\int_0^t x_1(s)x_2 ds\begin{bmatrix}-1\\1\end{bmatrix} + p3\int_0^t x_1(s)ds\begin{bmatrix}-1\\0\end{bmatrix} + p4\int_0^t x_2(s)ds\begin{bmatrix}1\\0\end{bmatrix} + p5\int_0^t x_2(s)ds\begin{bmatrix}0\\-1\end{bmatrix}$$

b) the stochastic version:

$$X(t) = X(0) + Y_1\left(p1\int_0^t x_1(s)ds\right)\begin{bmatrix}1\\0\end{bmatrix} + Y_2\left(p2\int_0^t x_1(s)x_2 ds\right)\begin{bmatrix}-1\\1\end{bmatrix}$$

$X(t) - [A(t), B(t)]^T$

$$+Y_3\left(p3\int_0^t x_1(s)ds\right)\begin{bmatrix}-1\\0\end{bmatrix} + Y_4\left(p4\int_0^t x_2(s)ds\right)\begin{bmatrix}1\\0\end{bmatrix} + Y_5\left(p5\int_0^t x_2(s)ds\right)\begin{bmatrix}0\\-1\end{bmatrix}$$

This describes the evolution of the system, considering the conditions named above, in the stochastic version, using the Poisson process model from 2). Also, this model is not influenced by any other variable (like the ones mentioned above). We can say that it is a theoretical and untroubled model. In a real environment, this will not happen. In real modelling situations, you have to take into account biosynthesis, diffusion, binding and degradation, all depending on the oxygen transport and utilization or the nutrients used in mechanical interactions. Thus a model that will simulate a real situation is really complex and requires lots of time for succeeding to take into account all the variables and the factor that will influence the model. Also for the mathematical modelling part to be precise and give valuable information it has to be done in close relation with the biochemical one that gives the support and which) can verify the theoretical data revealed by the model.

The normal evolution does not follow a static evolution because the tissue evolution is subject to environmental variables and most of them influence of tissue growth in time. For dealing with this problem different techniques were tried in order to maintain the growth constant and also reach the desired shape and size of the tissue.

Fast Computations by Using Division

Cellular division is an important process at the level of the membrane, which can be modelled as well in this area.

In this section we recall results obtained for P systems with proteins on membranes and membrane division.

The P systems with proteins on membranes can be viewed as a model combining membrane systems (Paun et al., 2002) and brane calculi (Cardelli et al., 2005) as introduced by (Paun and Popa et al., 2006). The paper is in the major area of P systems started by Gh. Paun (Paun et al., 2002).

What is remarkable is the fact that the aforementioned author was active in many areas, with significant results in formal language theory and natural computing. The interested reader is referred to (Ciobanu, Pan and Paun et al., 2007), (Freund, Kari and Paun et al., 2004), (Freund, Martin-Vide and Paun et al., 2004), (Martin-Vide, Paun and Rozenberg et al., 2002), (Mateescu, Paun and Rozenberg et al., 1998) and (Paun and Rozenberg et al., 1998) for further details. The novelty of the framework of the current paper is the fact that we are modelling proteins embedded in the membranes which control the application of the rules in the system.

Considering that only recently the membrane proteins were modelled in the field of membrane computing, we are now focusing on studying their properties as related to the membrane computing area.

In the P systems, which we consider below, we use two types of objects, proteins and usual objects; the former are placed on the membranes, the latter are placed in the regions delimited by membranes. The fact that a protein p is on a membrane (with label) i is written in the form. Both the regions of a membrane structure and the membranes can contain multisets of objects and of proteins, respectively.

We consider the following types of rules for handling the objects and the proteins; in all of them a, *b, c, d* are objects, *p, p'* are two proteins (possibly equal; if *p = p'*, then the rules of the type *cp* become rules of the type res; i.e. restricted), and *i* is a label ("*cp*" stands for "change protein"):

Type	Rule	Effect (besides changing also the protein)
1cp	$[ip\|a \to [ip^{Tr}\|b$ $a[ip\| \to b[ip^{Tr}\|$	modify an object, but not move
2cp	$[ip\|a \to a[ip^{Tr}\|$ $a[ip\| \to [ip^{Tr}\|a$	move one object unmodified
3cp	$[ip\|a \to b[ip'\|$ $a[ip\| \to [ip^{Tr}\|b$	modify and move one object
4cp	a $a[ip\|b \to b[ip^{Tr}\|$	a interchange two objects
5cp	d $a[ip\|b \to c[ip^{Tr}\|$ d	interchange and modify two objects

As a note on the types of rules considered above, when we need to model cellular processes involving membranes such as signal transduction, we will need to add "dummy" objects for the modelling to work. A few examples follow. For signal binding to a receptor $s\,[ir| \rightarrow [irs|$ would be modelled with 1cp rules such as $s[[ir| \rightarrow s^{Tr}\,[irs|$: cytoplasmic protein recruitment (ir|s (irs| would be done for example through 2cp rules: $[ir|s \rightarrow s^{Tr}\,[irs|$ etc. For signal binding to a receptor $s[ir| \rightarrow [irs|$ would be modelled with 1cp rules such as $s[ir \rightarrow s^{Tr}\,[irs|$: cytoplasmic protein recruitment would be done for example through 2cp rules: etc.

There are processes that change the number of proteins at the plasma membrane; such as receptor dimerization $[ip + p| \rightarrow [ip2|$ that could not be easily modelled using the rules above since our rules do not change the number of proteins in the system.

One could model though the dimerization at the moment when the ribosome produces the second copy of the receptor (through a rule of type 1cp for example)

Results for Fast Computations by Using Division

In this section we recall results obtained for P systems with proteins on membranes and membrane division.

Cellular division is an important process at the level of the membrane which can be modelled as well in this area.

Accepting P systems can be used for solving hard decidability problems.

To this aim (to trade space for time), we need tools for producing an exponential workspace in polynomial (if possible, linear) time, and the usual way to do it is by considering rules for membrane division. This is particularly natural in the case of P systems with objects on membranes, because we can consider such rules with the division of a membrane controlled by proteins placed on the membrane itself (not inside the delimited region, as in P systems with active membranes).

Specifically, we can consider rules of the form $[ip+|]i \rightarrow [ip^{Tr}\,]i[ip''|\,]i$, where p, p', p" are proteins. Under the influence of protein p, membrane i is divided into two copies with the same label, with protein p replaced by p' and p", respectively.

Using such rules and rules as in Subsection 1, solutions to SAT can be obtained in a time which is polynomial with respect to the number of variables and the number of clauses. We refer to (26) for details of the construction.

We denote by NOPD*m*(pror;list-of-types-of-rules) the family of sets of numbers N(II) accepted by systems II with at most m membranes, using rules as specified in the list-of-types-of-rules, and with at most r proteins present on a membrane, and working with division rules. When parameters *m* or *r* are not bounded, we use * as a subscript.

Solving NP-complete problems in linear time

In the paper (Paun and Popa et al., 2006) it was reported a construction solving SAT in polynomial time. Namely, systems of the form NOPD5(pro5mn+15n+3m+21;3res, 2cp, 5cp) can solve SAT problems with n variables, in conjunctive normal form using m clauses in exactly 2nm+ 5n + 7 steps.

When considering the same framework of such devices able to use division, a recent result obtained in (Sosik and Rodriguez-Paton et al., 2010) generalizes the previous result: the family of such devices is PSPACE-complete. We give the main results in this respect in the following sub-section.

Membrane systems with division are PSPACE-complete

It is shown that P systems with proteins on membranes can compute in polynomial time exactly the class of problems PSPACE. Mathematically, this property can be expressed as

M-PTIME = M-NPTIME = PSPACE,

Where M-(N)PTIME is the class of problems solved in polynomial time by a (non-) deterministic computing model M (In our case, M is a uniform family of P systems with proteins on membranes.) This relation is also known as the Parallel Computation Thesis (Van Emde Boas et al., 1990). Models of computation with this property form the so-called second machine class. Typical representations of this class are the alternating Turing machine, SIMDAG (also known as SIMD PRAM) and other standard parallel computer models (Van Emde Boas et al., 1990).

This result is related to several other studies of computational potential of various natural computing models. For example, in the field of DNA computing, where fragments of DNA molecules are used as an information medium, an analogous result was presented in (Beaver et al., 1995). Later (Dantsin and Wolpert et al., 2003) presented another, more robust DNA computing model capturing **PSPACE** in polynomial time. Also, when abstracting from biochemical aspects of DNA to the operation of genetic crossing-over, (Pudlak et al., 2001) obtained a second-class computer embodied in so-called genetic Turing machine, see also (Arora, Rabani and Vazirani et al., 1994). Finally, in the field of membrane computing, results giving evidence that abstract operations inspired by processes in cellular membranes also wield the power of the second machine class were given by (Alhazov, Martin-Vide and Paun et al., 2003) and (Sosik and Rodriguez-Paton et al., 2007). These studies suggest that the class **PSPACE** provides a characterization of the computational potential of natural computing machinery.

The main result in (33) was that uniform families of recognizer P systems with proteins on membranes can compute in polynomial time exactly the class of problems **PSPACE**. More precisely, the authors

provide a semi-uniform solution to problems in this class (the QSAT problem was solved by P systems in linear time as well as uniform families of P systems have been simulated in polynomial space). Hence, their information processing potential is equivalent to that of a standard parallel computing model as PRAM or alternating Turing machine.

CONCLUSIONS

To establish protocols for tissue engineering, a balance will have to be achieved between the predictive value of the model and the complexity and number of parameters involved. Due to the complexity of cellular behaviour and interactions with numerous cellular environments, it is necessary to study computational and experimental contributions for understanding the culture medium, nutrient transport, cell attachment, cell-cell interactions and population dynamics. It is not advisable to reproduce exactly the same aspects of cell behaviour in a single model and modelling should focus on identifying key mechanisms. These are significant challenges, but they are key to the development of regenerative strategies.

ACKNOWLEDGMENTS

This work was supported by Structural Funds POSCCE-A2-O2.1.2: OPERATIONAL PROGRAMME "INCREASE OF ECONOMIC COMPETITIVENESS" Priority axis 2 –Research, Technological Development and Innovation for Competitiveness, Operation 2.1.2: Complex research projects fostering the participation of high-level international experts, Nr. Project SMIS-CSNR: 12449, Financing contract nr. 204/20.07.2010, "Biotechnological center for cell therapy and regenerative medicine based on stem cells and apoptosis modulators"-BIOREGMED.

REFERENCES

Alhazov A, Martin-Vide C, Pan L, Solving a PSPACE-complete problem by P systems with restricted active membranes. Fundamenta Informaticae, 58 (2), 67–77, 2003

Andreson D F, Stochastic models in biology, CIBM Seminar, (web), December 7th, 2010

Arora S, Rabani Y, Vazirani U, Simulating quadratic dynamical systems is PSPACE-complete (preliminary version). In: STOC 26. pp. 459–467,1994

Beaver D., A universal molecular computer. In: R.J. Lipton, E. B. (Ed.), DNA Based Computers. Vol. 27 of DIMACS. American Mathematical Society, 29–36, 1995.

Ciobanu, G, Pan LQ, Paun G, P systems with minimal parallelism, Theoretical computer Science, Vol. 378 (1), 117-130, 2007.

Besozzi D, Rozenberg G, Extended P systems for the analysis of (trans)membrane proteins, WMC7, 2006, 8-9, 2006.

Brijder R, Cavaliere M, Riscos-Nunez A, Rozenberg G, Sburlan D, Membrane systems with marked membranes. Electronic Notes in Theoretical Computer Science, ENTCS, 171, 2 (2007), 25–36, 2007

Brijder R, Cavaliere M, Riscos-Nunez A, Rozenberg G, Sburlan D, Membrane systems with proteins embedded in membranes, Theoretical Computer Science, 404 (1-2), 26–39, 2008

Cardelli L, Brane calculi – interactions of biological membranes. In: Computa52 A. Paun et al. tional Methods in Systems Biology. Vol. 3082, Lecture Notes in Computer Science, Springer-Verlag, Berlin, 257–280, 2005

Cavaliere M, Sedwards S, Membrane systems with peripheral proteins: transport and evolution, CoSBi Technical Report 04/2006, www.cosbi.eu and Electronic Notes in Theoretical Computer Science, ENTCS, 171, 2 (2007), 37–53, 2006

Cavaliere M, Sedwards S, Modelling cellular processes using membrane systems with peripheral and integral proteins, Computational Methods in Systems Biology, Intern. Conference, Trento, (C. Priami, ed.), LNBI 4210, Springer, 108–126, 2006

Chawla K, Yu T, Stutts L, Yen M., Guan, Modulation of chondrocyte behavior through tailoring functional synthetic Z., saccharideepeptide hydrogels, Biomaterials, 33, 6052-6060, 2012

Chung C, Burdick J A, Engineering cartilage tissue, Advanced Drug Delivery, Reviews 60, 243–262, 2008

El-Ghannam A, Bone reconstruction: from bioceramics to tissue engineering, Expert Rev Med Devices, 2(1), 87-101, 2005

Dantsin E, Wolpert A, A robust DNA computation model that captures PSPACE. Int. J. Foundations Comp. Sci. 14 (5), 933–951, 2003

Fraj J R, Roc O, Bone tissue engineering: hope vs. hype, Biochem Biophys Res Commun 292, (1), 1–7, 2002

Freund R, Kari L, Paun G, DNA computing based on splicing: The existence of universal computers, Theory of Computing Systems, Vol. 32 (1), 69–112, 1999

Freund R, Martin-Vide C, Paun G, From regulated rewriting to computing with membranes: collapsing hierarchies, Theoretical Computer Science, Vol. 312 (2-3), 143–188, 2004

Frisco P, Computing with Cells. Advances in Membrane Computing. Oxford University Press, Oxford, 2009

Ibarra OH, Paun A, Counting Time in Computing with Cells, Proceedings of DNA11 conference, June 6-9, London Ontario, Canada, 112–128. Vol. 3892, Lecture Notes in Computer Science. Springer-Verlag, Berlin, pp. 112–128, 2005

Galban CJ, Locke BR, Effects of spatial variation of cells and nutrient and product concentrations

coupled with product inhibition on cell growth in a polymer scaffold, Biotechnol Bioeng, 64, 633-43, 1999

Gu WY, Lai WM, Janssen JD, Mow VC, A mixture theory for charged-hydrated soft tissues containing multi-electrolytes: passive transport and swelling behaviors, J Biomech Eng 120,169-80, 1998

Hunziker EB, Articular cartilage repair: basic science and clinical progress. A review of the current status and prospects, Osteoarthritis Cartilage 10(6), 432–63, 2001

Huyghe JM, Janssen JD, Quadriphasic mechanics of swelling incompressible porous media, Int J Eng Sci 35(8), 793-802, 1997

Krishna SN, Combining Brane Calculus and Membrane Computing, Proc. Bio-Inspired Computing – Theory and Applications Conf., BIC-TA 2006, Wuhan, China, September 2006, Membrane Computing Section and Journal of Automata Languages and Combinatory submitted, 2006

Krishna SN, On the computational power of flip-flop proteins on membranes, Proceedings of CiE'07 Computability and Logic in the Real World. Vol. 4497 of Lecture Notes in Computer Science, Springer-Verlag, Berlin, pp. 695–704, 2007

MacArthur BD, Tare RS, Please CP, Prescott P, Roc O, A non-invasive method for in situ quantification of subpopulation behavior in mixed cell culture, J R Soc Interface, 3(6), 63–9, 2005

Martin-Vide C, Paun G, Rozenberg G,Membrane systems with carriers, Theoretical Computer Science, Vol. 270 (1-2) 779–796, 2002

Mateescu A, Paun G, Rozenberg G, Simple splicing systems, Discrete Applied Mathematics, Vol. 84 (1-3), 145–163, 1998

Mauney JR, Volloch V, Kaplan DL, Role of adult mesenchymal stem cells in bone tissue engineering applications: current status and future prospects, Tissue Eng, 11(5–6),787–802, 2005

Mistry AS, Mikos AG, Tissue engineering strategies for bone regeneration, Adv Biochem Eng Biotechnol 94, 1–22, 2005

Nagda H, Paun A, Rodriguez-Paton A, P Systems with Symport/Antiport and Time, Pre-proceedings of Membrane Computing, International Workshop, WMC7, Leiden, The Netherlands, 429–442, 2006

Patrachari AR, Podichetty JT, Madihally SV, Application of computational fluid dynamics in tissue engineering, J. of Bioscience and Bioengineering, 114 (2), 123-132, 2012

Paun A, Paun M, Rodriguez-Paton A, Sidoroff M, "P Systems with Proteins on Membranes: A Survey", Journal of Foundations of Computer Science, Volume No. 22, Issue No. 01, 39–53, 2011

Paun A, Paun G, The power of communication: P systems with symport/ antiport, New Generation Comput. 20 (3), 295–306, 2002

Paun A, Popa B, P systems with proteins on membranes, Fundamenta Informatica, 72, (4), 467-483, 2006.

Paun A, Popa B, P systems with proteins on membranes and membrane division. In: Ibarra, O., Dang, Z. (Eds.), DLT Vol. 4036 of Lecture Notes in Computer Science. Springer-Verlag, Berlin, pp. 292–303, P Systems with Proteins on Membranes: A Survey 53, 2006

Paun G, Rozenberg G, Salomaa A (Eds.),The Oxford Handbook of Membrane Computing. Oxford University Press, Oxford, 2009

Paun G, Membrane Computing – An Introduction. Springer-Verlag, Berlin

Paun G, Rozenberg G, Sticker systems, Theoretical Computer Science, Vol.204 (1-2), 183–203, 1998

Perez-Jimenez M, A computational complexity theory in membrane computing. In: Paun G, Perez-Jimenez M, Riscos-Nunez A (Eds.), Tenth Workshop on Membrane Computing (WMC10). Vol. 5957 of Lecture Notes in Computer Science.Springer-Verlag, Berlin, pp. 125–148, 2009

Pudlak P, Complexity theory and genetics: The computational power of crossing-over. Information and Computation 171, 201–223, 2001

Roc O, Cooper C, Mason C, Clements M Mesenchymal stem cells—lineage, plasticity, and skeletal therapeutic potential, Stem Cell Rev, 1(2),169–78, 2005

Romero-Campero FJ, Perez-Jimenez M, Modelling gene expression control using P systems: The Lac Operon, a case study. Biosystems 91(3), 438–457, 2008

Sengers BG, Taylor M, Please C P, Roc O, Computational modelling of cell spreading and tissue regeneration in porous scaffolds, Biomaterials 28, 1926-1940, 2007

Sosık P, The computational power of cell division in P systems: Beating down parallel computers? Natural Computing 2 (3), 287–298, 2003

Sosık P, Rodrıguez-Paton A, Membrane computing and complexity theory: A characterization of PSPACE. J. Comput. System Sci. 73 (1), 137–152, 2007

Sosık P, Rodrıguez-Paton A, On the Power of Computing with Proteins on Membranes, Preproceedings of Workshop on Membrane Computing 2009 and Vol. 5957 of Lecture Notes in Computer Science, Springer-Verlag, Berlin, pp. 448–460, 2010

Theise ND, d'Inverno M Understanding cell lineages as complex adaptive systems. Blood Cells Mol Dis 32, 17-20, 2004

van Emde Boas P, Machine models and simulations. In: van Leeuwen, J. (Ed.), Handbook of Theoretical Computer Science. Vol. A. Elsevier, Amsterdam, 1990
The P System Website: http://psystems.disco.unimib.it