REVIEW

Substitution for *In Vitro* and *In Vivo* Tests: Computational Models from Cell Attachment to Tissue Regeneration

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Abstract To get an optimal product of orthopaedic implant or regenerative medicine needs to follow trial-and-error analyses to investigate suitable product's material, structure, mechanical properties etc. The whole process from *in vivo* tests to clinical trials is expensive and time-consuming. Computational model is seen as a useful analysis tool to make the product development. A series of models for simulating tissue engineering process from cell attachment to tissue regeneration are reviewed. The challenging is that models for simulating tissue engineering processes are developed separately. From cell to tissue regeneration, it would go through blood injection after moving out the defect; to cell disperse and attach on the scaffold; to proliferation, migration and differentiation; and to the final part—becoming mature tissues. This paper reviewed models that related to tissue engineering process, aiming to provide an opportunity for researchers to develop a mature model for whole tissue engineering process. This article focuses on the model analysis methods of cell adhesion, nutrient transport and cell proliferation, differentiation and migration in tissue engineering. In cell adhesion model, one of the most accurate method is to use discrete phase model to govern cell movement and use Stanton-Rutland

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model for simulating cell attachment. As for nutrient transport model, numerical model coupling with volume of fluid model and species transport model together is suitable for predicting nutrient transport process. For cell proliferation, differentiation and migration, finite element method with random-walk algorithm is one the most advanced way to simulate these processes. Most of the model analysis methods require further experiments to verify the accuracy and effectiveness. Due to the lack of technology to detect the rate of nutrient diffusion, there are especially few researches on model analysis methods in the area of blood coagulation. Therefore, there is still a lot of work to be done in the research of the whole process model method of tissue engineering. In the future, the numerical model would be seen as an optimal way to investigate tissue engineering products bioperformance and also enable to optimize the parameters and material types of the tissue engineering products.

OSTEoarthritis is a long-term chronic disease which results in creating stiffness, pain, and impaired movement. Joint replacements are major surgical procedures, which are only undertaken where there is no alternative. Patients with osteoarthritis often suffer pain, loss of mobility and the loss of cartilage and bone at the joint, which significantly reduces their quality of life. As a major category in regenerative medicine, tissue engineering provides a new approach with broad application prospects for clinical repair and reconstruction of injured tissues.

Tissue engineering have emerged as an alternative treatment for osteoarthritis and have shown promise in the repair of damaged bone and cartilage. Kang *et al.*^[1] utilized a multi-layered scaffold to provide a suitable chondrogenic and osteogenic environment for cartilage and bone regeneration respectively. In osteochondral tissue engineering field, a unique composition scaffold with multiple layers is needed with specific biological and mechanical properties.^[2] Osteochondral scaffolds have been used in the past few decades for cartilage and bone regeneration.^[3] These scaffolds are thought to maintain the biological and mechanical properties of the interfaces between cartilage, subchondral bone plate and trabecular bone after regeneration.^[3, 4] The scaffolds need to be integrated with the surrounding host tissues and to maintain cell viability and proliferation during implantation, and enable cell differentiation and migration post-implantation.^[2] The difficulty for trabecular bone regeneration remains a major obstacle for osteochondral scaffolds. To ensure a high-quality of hyaline cartilage regeneration, the osteochondral scaffold should not only have a similar structure to the trabecular bone with optimal strength and fatigue performance, but also the ability to bond to the soft materials which are used for cartilage regeneration. The scaffold should be a highly porous

structure and not elicit immunological or foreign body reactions in patients.^[2]

Tissue engineering products for repairing damaged bone and cartilage require large investments for *in vitro* tests, *in vivo* tests and four phases of clinical trials before they enter the market. Although the clinical trials could not be substituted, as medical authorities need to scrutinize the risk, safety and efficacy of the product, much of the *in vitro* and *in vivo* tests could be reduced and substituted with numerical models. Based on limited experimental studies, numerical models have been developed by researchers to help understand the tissue engineering process and find optimal tissue engineering products.

One of the main purpose for numerical models is to find optimal design of various structural parameters such as pore size, porosity, and permeability. The above-mentioned multiple parameters affect the cell-scaffold interaction effect and thus the efficiency of bone regeneration. Therefore, it is very important to further analyze the influence of the various parameters of the scaffold on the bioperformance response. Simulation models for tissue engineering process mainly focus on two processes. The first one is cell attachment process.^[5, 6] The cells dispersed in blood would be carried into the void space of the defect area when the disease area is removed and replaced by scaffold. The second process that are well explored is cell proliferation, differentiation and migration. Damien and Byrne *et al.* made a new three-dimensional approach for model tissue regeneration within scaffolds which differentiation is modelled based on mechano-regulation algorithm models and proliferation, and migration are modelled based on random work theory.^[7, 8]

In reality, tissue engineering contains a lot processes. From cell to tissue regeneration, it would go through blood injection after moving out the defect; to cell dispersion and attachment on the scaffold; to

proliferation, migration and differentiation; and to the final part—becoming mature tissues shown in **Fig. 1**.^[9-13] This paper reviews the models from 1997 until 2021 that related to tissue engineering process for simulating cell attachment until tissue regeneration. The models that are reviewed in the paper are selected based on the accurate physical model chosen and contains more biological processes. It aims to provide an opportunity for researchers to develop a mature model for whole tissue engineering process. The model would not only help researchers to fully understand the entire tissue engineering process, but also enable optimization of the parameters and material types of the tissue engineering products.

CELL ADHESION

Tissue regeneration is strongly related to initial cell attachment.^[14, 15] Preceding all other steps, a better cell

adhesion and an even spatial distribution are associated with good culture results.^[6] Although this is one of the determinants of the final bioperformance of a scaffold, it is difficult to observe the cells seeding process and cells distribution within the scaffold during the seeding process. In general, the cell adhesion could be separated to two processes—blood filling and cell attachment.

Blood filling

Bone tissue contains blood vessels. When it is broken, the blood vessels at the fracture site will be destroyed. Subsequently, after the scaffold is implanted in the bone, blood carried with bone mesenchymal stem cells (BMSCs) will flow into the voids of the scaffold.

When analyzing the blood flow in tissue engineering process, the blood characteristics need to be well considered. Researchers usually assume that blood as a Newtonian fluid with a constant viscosity to simplify the model. In fact, the tiny particles in the blood such

Figure 1. Schematic diagram of the whole process of bone tissue engineering. A. Bone tissue engineering scaffold implantation;^[9] B. Blood filling after implantation;^[9, 10] C. Cells adhesion;^[11, 12] D. Blood coagulation;^[10] E. Cell proliferation, differentiation and migration;^[13] F. Formation of an organization.^[9]

as cells, proteins and other components will invalidate this assumption. Because blood contains tiny particles and cannot be completely used as a Newtonian fluid, it is widely regarded as a shear thinning fluid.^[16]

In order to accurately build the model, researchers began to treat blood as a non-Newtonian fluid. For example, Reymond *et al.*^[17] used standard computational fluid dynamics (CFD) model to simulate the steady flow in a patient-specific model of the entire system circulation considered blood as Newtonian and non-Newtonian. The results showed that using Newtonian and Non-Newtonian models for blood rheology is not very significant. Truscello *et al.*^[18] regarded the culture medium as an incompressible homogeneous Newtonian fluid, which has the characteristics of water, and based on this assumption, they established a computational fluid dynamics model to predict the permeability of the scaffold. Subsequently, the pressure drop between the different units of all models is obtained through experiments, which is used to calculate the permeability coefficient of the scaffold and compare it with the predicted value of the model. The relative difference between the measured value and the predicted value in the final result is between 2% to 27%.

However, in tissue engineering field, cells are dispersed in blood which could not be simplified as Newtonian fluid. Assuming blood as Newtonian fluid could not get accurate prediction results. In order to accurately build the model, researchers began to treat blood as a non-Newtonian fluid. Xiang *et al.*^[19] used the Newtonian viscosity model to compare two non-Newtonian models (Casson and Herschel-Bulkley) and found that the Newtonian fluid hypothesis may underestimate the viscosity and overestimate the shear rate in the stagnant or slowly recirculating secondary vortex region.

In addition, Jiang *et al.*^[20] used CFD models based on the finite volume method to perform numerical studies on the hemodynamics of blood models with Newtonian and non-Newtonian characteristics, and found that the area of low wall shear stress (WSS) of blood models with non-Newtonian characteristics was smaller, which shows that the blood model is very important for accurately predicting hemodynamics.

As for tissue engineering process, Liu *et al.* started to use volume of fluid (VOF) model to calculate the blood filling process and set blood as non-Newtonian fluid carrying cells.^[9] The filling process is well considered because the blood filling and air coming out is fully governed by VOF model. This model gives research-

ers an opportunity to well investigate following tissue engineering process which no one has done before.

Cell attachment

Cell attachment is a significant step for achieving successful tissue engineering. Cell seeding efficiency and cell distribution can affect the final biological performance of the tissue engineering. As *in vitro*, *in vivo* and clinical tests usually require a large investment, numerical models offer a more ethical and economical alternative and controllable choice to investigate cells attachment to scaffolds. Although several researchers investigated this issue for the past few decades, the accuracy of prediction of cell seeding still needs to be well improved. As Lagrangian method to describe cell movement requires a long time for calculation, researchers prefer to use Eulerian method to govern fluid movement. To simplify the cell attachment model, only fluid properties like velocity and shear stress are calculated to compare with the experiment results. The velocity and shear stress value at the area that has much cell attachment would be seen as the optimal value for cell attachment. Culture medium is mostly used to investigate cell seeding efficiency rather blood because inner body bioperformance is hard to be observed. Numerical models for prediction of cell attachment and cell distribution should consider cell adhesion and fluid properties simultaneously to get a more accurate results.^[21, 22]

Normal CFD model (see cells as pure water) In the early model analysis, in order to simplify the model to the greatest extent, the researchers regarded the medium (including cells) in the scaffold as pure water, although this caused a certain deviation in the analysis results. Xu *et al.*^[5] concluded that fluid velocities and shear stresses influence cell seeding density on a scaffold with a random architecture. However, only considering the culture medium's velocity and shear stress to investigate cell attachment process is insufficient. Each cell has an individual movement, which cannot be neglected and is as important as the fluid flow. To improve the accuracy of predicting the result of cell seeding, an understanding of the type of cell-material interaction is crucial. Euler-Lagrange numerical approach, using the Eulerian method to describe mediums and Lagrangian method to describe cells movement, would be a good solution.

Trap model Olivares *et al.*^[6] used Eulerian wall film model followed by Euler-Lagrange approach to

simulate cell seeding process assuming that only one situation occurs when a cell impinges a wall, with each cell being seen as sphere and trapped by the scaffold after it touches the material surface (**Fig. 2.**)^[6] Olivares *et al.*^[6] innovatively introduced the theory of wall film used to simulate the deposition of fuel particles in the combustion state to simulate the adhesion state of cells when they hit the scaffold wall. The simulation results are quantitatively compared with the experimental results. The cell density values of the two show a consistent distribution trend in the radial direction of the scaffold. The cell density of one type of scaffold

is 11%-31% in the experiment and 6.5%-14% in the model. There is a big difference in absolute value between the two.

In the above model, the interaction between cells and materials is simplified. For example, the wall of the bioreactor adopts non-slip and non-adhesive conditions, without considering some impinge regimes like splashing (cells breakup) and rebound. The above simplifications may cause the deviation between the predicted value and the experimental value, but in general, considering the movement of the cell itself is a big improvement.

Figure 2. Dynamic conditions and results of Olivares *et al.*'s research.^[6] A. Suspension process at a constant velocity of 1 mm/s, representation of area covered by the cells after last injection; the cells do not touch the scaffold wall. B. Dispersion of cells after oscillating fluid flow. C. Graphical representation of initial injection conditions and cycles repeating during 2 h of experimentation. Maximum mass flux corresponds to maximum fluid normal velocity of $u_n=1$ mm/s. D. Superior view of cells attached in the type I (top) and type G (bottom) scaffolds. The evolution of increment of cell numbers deposited in each time selected.

Stanton-Rutland model (cell impinge model) In reality, cells can bounce back from the wall or sometimes split to several smaller particles when they approach the wall. The impinge types are similar to spray droplets interaction with the internal walls of a combustion engine. The Stanton-Rutland model is mainly used to simulate internal combustion engines, but it also has potential to simulate cell seeding process, in which cells can be considered as particles.^[23, 24] The great advantage of this model is that when cells impinge a scaffold wall, four regimes (stick, rebound, spread, and splash) are considered.^[25] This novel method can interact with both discrete phase model (DPM) and Eulerian multiphase model. In the Eulerian multiphase interaction, the first phase represents the fluid (medium solution) and the secondary phase rep-

resents cells that can be captured by wall surfaces.

Compared with the model of Olivares *et al.*,^[6] Liu *et al.*^[12] proposed a further model for simulating cell attachment based on Stanton-Rutland model (**Fig. 3**).^[12] In addition to the "absorption/capture" situation of cell impact, their model also includes more impact types, such as bounce and splash. Liu *et al.* assumed that cells are able to bounce back from the scaffold wall, and split into several smaller particles which means cells would die.

Of course, this CFD model also has certain limitations. In the model assumption, the cells that are stuck or spread on the scaffold are all seen as alive. However, not all the cells would alive even they attached on the scaffold. Further investigations is needed to find a suitable equation to solve this issue.

Figure 3. Stanton-Rutland model proposed by Liu *et al.*^[12] A. Impinge regimes definition. B. Cells growth on porous titanium matrix was examined by confocal microscopy, and the confocal image was further processed to determine the cells distribution. C. Predicted cell distribution during cell seeding process. From left to right represent the cells distribution at second one, two, three, four, five and six, respectively.

Blood coagulation

After cell adhesion, the next step in tissue engineering is blood coagulation. Obviously, blood coagulation process control has a greater impact on the biomechanical properties of the final biological tissue formed by tissue engineering. However, we found that there are only a handful of literatures on modeling research of blood coagulation in this field. This process is really important for cell proliferation and tissue growth because nutrients transport mainly occurs during this process. Before coagulation, the nutrient transport way is convection and diffusion. After blood coagulation, only diffusion would be occurred for nutrient transport. Although this process could affect final bioperformance, few researchers investigated this process as there is no facilities at the moment to measure the speed of the nutrient diffusion speed after blood coagulation.

Nutrient transport

Nutrient supply is a critical factor for tissue engineering success.^[26] Cells require nutrients and oxygen to survive. *In vitro* studies have shown that nutrient concentration in the scaffold is decreasing toward its center due to the consumption of nutrients by the cells and the relatively slow supply by diffusion. Only cells up to a distance of approximately 200 μm have access to sufficient nutrients.^[27, 28] This results in nutrient limitations for cells that are further away from the engineered tissue surface, and thus reduce cell proliferation or non-optimal conditions for new matrix production. There is an urgent need to understand the nutrient distribution for both *in vitro* and *in vivo* studies. However, no technology is currently available for researchers to observe the nutrient transport during these dynamic processes.

Two of the main mechanisms governing nutrient transportation are convection and diffusion.^[6, 29] Convection is a faster mass transport method due to bulk fluid motion and diffusion is a significantly slower transport mechanism due to its reliance upon a concentration gradient. In a bioreactor system, the solution transportation is usually generated by external sources called forced convection. In most bioreactor systems, the flow speed caused by external sources is far higher than the diffusion speed, meaning that the transportation type in most bioreactor systems mostly relies upon convection rather than diffusion. Unlike normal solutions for cultivating cells, bone marrow is a semi-solid tissue^[30] which is hard to mimic experi-

mentally. During *in vivo* and *in vitro* tests, there are no techniques which are able to monitor the entire biosystem including temperature, cell density, bone marrow speed, nutrients transportation and density. Computational analysis provides a promising method to understand the tissue regeneration process from cell seeding to tissue regeneration.^[12]

Convection (blood filling, before coagulation)

Convection is a faster method of mass transport due to bulk fluid motion whilst diffusion is a significantly slower transportation movement due to its reliance on a concentration gradient. In a bioreactor system, the solution transportation is usually generated by external sources which are called forced convection.^[31] In most cases, the flow speed caused by external sources is far higher than the diffusion speed; therefore the major type of transportation in a bioreactor system relies on convection rather than diffusion. For tissue engineering field, nutrient transport before blood coagulation mainly depends on convection (diffusion also occurred during the process).

Diffusion (blood filling and after blood coagulation)

After blood coagulation, nutrient transport is mainly rely on diffusion which is slower than before. During *in vivo* and *in vitro* tests, there are no techniques which are able to monitor the entire biosystem including temperature, cell density, bone marrow speed, nutrients transportation and density. Computational analysis provides a promising method for researchers to understand the tissue regeneration process from the initial cell seeding to tissue regeneration.^[12]

However, most of the studies are mainly focus on bioreactor experiments rather inner body experiments. In consideration of diffusion speed of nutrient (glucose), Botchwey *et al.*^[32] explored the effects of flow-induced nutrient transport and fluid perfusion for cells on scaffolds in a rotational bioreactor during dynamic bioreactor cultivation. As oxygen is also one of the most important ingredients of nutrients, Mofrad *et al.*^[33] developed a numerical model considering both convection and diffusion to understand the relationship between oxygen supplying and cell density in a channeled scaffold.

In this tissue engineering process, after blood coagulation, nutrient transport is mainly depending on diffusion due to the nutrient concentration gradients. Fick's law would be one of the suitable law to use for

calculating diffusion flux which could be written as:

$$\vec{J}_i = -\rho D_{i,m} \nabla Y_i - D_{T,i} \frac{\nabla T}{T}$$

R_i is the rate of production of species; $\vec{J}_i$ represents the diffusion flux of nutrients species due to their concentration gradients; $D_{i,m}$ defined as mass diffusion coefficient for nutrients in the mixture and $D_{T,i}$ represents the thermal diffusion coefficient.

Cell proliferation, differentiation and migration

After MSCs attachment, cells would start to proliferate and differentiate when absorb enough nutrients. A random walk method could be used to predict MSCs migration, proliferation and differentiation to predict tissue engineering bioperformance. It is a mathematical object, known as a stochastic or random, which describes a path that consists of a succession of random steps on some mathematical space such as the integers. According to the Pérez's work, cells disperse with a preferential direction of proliferation.^[34] (Fig. 4.) The highest probability positions are the ones in which one daughter cell tends to appear in a preferred direction (downwards) with p_1 . The corresponding value of p is computed fulfilling $\sum_{i=1}^{n_1} p_1 + \sum_{i=1}^{n_2} p_2 + \sum_{i=1}^{n_3} p_3 = 1$, and the preferred direction is seen as $p_1 = 10, p_2 = 60, p_3 = p = \frac{4}{13}$. MSCs differentiation can be expressed as^[35]

$$S = \frac{\gamma}{a} + \frac{\vartheta}{b}$$

High levels of stimuli ($S > 3$) promote the differentiation of MSCs into fibroblasts, intermediate levels ($1 < S < 3$) stimulate the differentiation into chondrocytes and low levels ($S < 1$) favor the differentiation of osteoblasts from the MSC pool. Cells then synthesize the appropriate extracellular matrix leading to tissue

formation and new mechanical properties for the tissue. The values are $a = 0.0375$ and $b = 3 \mu\text{ms}^{-1}$, based on Søball's experimental results.^[35] The geometry of the scaffold will be optimized by 2 parameters getting from this program. Most of osteoblast should grow on bottom and most of chondrocytes should grow on top.

In addition, Byrne *et al.*^[8] used a three-dimensional finite element model method to simulate tissue differentiation and bone regeneration (Fig. 5). The model predicts that porosity, Young's modulus, and solubility have a greater impact on bone regeneration.

Figure 4. The research of Pérez *et al.*^[34] A. Possible position that daughter cells can occupy after mitosis. The distance between sites is only schematic; adjacent sites in the algorithm are considered to be exactly the diameter of a mesenchymal stem cell.^[36] B. Possible position that daughter cells can occupy after mitosis with anisotropic proliferation direction (preferred direction is given by the arrow).

Figure 5. The research of Byrne *et al.*^[8] A. Finite element model of a 50% porous scaffold with regular porosity (green). Only one-eighth needs to be modelled because of symmetry (yellow box). The cavity is initially occupied by granulation tissue (in red). B. Lattice generated for each granulation element to model cellular activity. C. Increasing lattice points to account for dissolution of scaffold material.

CONCLUSIONS

The objective of this paper is to sum up all the tissue engineering process models from cell attachment until tissue regeneration. To develop a tissue regeneration model for tissue engineering, all the above models need to be considered. And most of them still need further experiments to improve the accuracy and efficiency. These models provide platforms for researchers to predict and optimize geometry and material design of tissue engineering products.

Conflicts of Interest Statement

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

REFERENCES

- Kang H, Zeng Y, Varghese S. Functionally graded multilayer scaffolds for *in vivo* osteochondral tissue engineering. *Acta Biomater* 2018; 78:365-77. doi: 10.1016/j.actbio.2018.07.039.
- Nooeaid P, Salih V, Beier JP, et al. Osteochondral tissue engineering: scaffolds, stem cells and applications. *J Cell Mol Med* 2012; 16(10):2247-70. doi: 10.1111/j.1582-4934.2012.01571.x.
- Yang PJ, Temenoff JS. Engineering orthopedic tissue interfaces. *Tissue Eng Part B Rev* 2009; 15(2):127-41. doi: 10.1089/ten.teb.2008.0371.
- Hunziker EB. Articular cartilage repair: basic science and clinical progress. A review of the current status and prospects. *Osteoarthritis Cartilage* 2002; 10(6):432-63. doi: 10.1053/joca.2002.0801.
- Xu S, Du P, Xie Y, et al. Cell distribution in a scaffold with random architectures under the influence of fluid dynamics. *J Biomater Appl* 2008; 23(3):229-45. doi: 10.1177/0885328207086322.
- Olivares AL, Lacroix D. Simulation of cell seeding within a three-dimensional porous scaffold: a fluid-particle analysis. *Tissue Eng Part C Methods* 2012; 18(8):624-31. doi: 10.1089/ten.TEC.2011.0660.
- Prendergast PJ, Huiskes R, Søballe K. ESB Research Award 1996. Biophysical stimuli on cells during tissue differentiation at implant interfaces. *J Biomech* 1997; 30(6):539-48. doi: 10.1016/s0021-9290(96)00140-6.
- Byrne DP, Lacroix D, Planell JA, et al. Simulation of tissue differentiation in a scaffold as a function of porosity, Young's modulus and dissolution rate: application of mechanobiological models in tissue engineering. *Biomaterials* 2007; 28(36):5544-54. doi: 10.1016/j.biomaterials.2007.09.003.
- Liu Z, Tamaddon M, Chen SM, et al. Determination of an initial stage of the bone tissue ingrowth into titanium matrix by cell adhesion model. *Front Bioeng Biotechnol* 2021; 9:736063. doi: 10.3389/fbioe.2021.736063.
- Liu J, Chen G, Xu H, et al. Pre-vascularization in fibrin Gel/PLGA microsphere scaffolds designed for bone regeneration. *NPG Asia Materials* 2018; 10(8):827-39. doi: 10.1038/s41427-018-0076-8.
- Daley WP, Peters SB, Larsen M. Extracellular matrix dynamics in development and regenerative medicine. *J Cell Sci* 2008; 121(Pt 3):255-64. doi: 10.1242/jcs.006064.
- Liu Z, Tamaddon M, Gu Y, et al. Cell seeding process experiment and simulation on three-dimensional polyhedron and cross-link design scaffolds. *Front Bioeng Biotechnol* 2020; 8:104. doi: 10.3389/fbioe.2020.00104.
- Ali F, Tareh S, Al-Nuzaily M, et al. Stem cells differentiation and probing their therapeutic applications in hematological disorders: a critical review. *Eur Rev Med Pharmacol Sci* 2016; 20(20):4390-400. doi: 10.1155/2016/7653489.
- Santoro R, Olivares AL, Brans G, et al. Bioreactor based engineering of large-scale human cartilage grafts for joint resurfacing. *Biomaterials* 2010; 31(34):8946-52. doi: 10.1016/j.biomaterials.2010.08.009.
- Wendt D, Stroebel S, Jakob M, et al. Uniform tissues engineered by seeding and culturing cells in 3D scaffolds under perfusion at defined oxygen tensions. *Biorheology* 2006; 43(3, 4):481-8.
- Cherry EM, JK Eaton. Shear thinning effects on blood flow in straight and curved tubes. *Phys FLUIDS* 2013; 25:0733104. doi: 10.1063/1.4816369.
- Reymond P, Perren F, Lazeyras F, et al. Patient-specific mean pressure drop in the systemic arterial tree, a comparison between 1-D and 3-D models. *J Biomech* 2012; 45(15):2499-505. doi: 10.1016/j.jbiomech.2012.07.020.
- Truscetto S, Kerckhofs G, Van Bael S, et al. Prediction of permeability of regular scaffolds for skeletal tissue engineering: a combined computational and experimental study. *Acta Biomater* 2012; 8(4):1648-58. doi: 10.1016/j.actbio.2011.12.021.
- Xiang J, Tremmel M, Kolega J, et al. Newtonian

- viscosity model could overestimate wall shear stress in intracranial aneurysm domes and underestimate rupture risk. *J Neurointerv Surg* 2012; 4(5):351-7. doi: 10.1136/neurintsurg-2011-010089.
20. Jiang Y, Zhang J, Zhao W. Effects of the inlet conditions and blood models on accurate prediction of hemodynamics in the stented coronary arteries. *AIP Adv* 2015; 5(5):057109. doi: 10.1063/1.4919937.
 21. Koch MA, Vrij EJ, Engel E, et al. Perfusion cell seeding on large porous PLA/calcium phosphate composite scaffolds in a perfusion bioreactor system under varying perfusion parameters. *J Biomed Mater Res A* 2010; 95(4):1011-8. doi: 10.1002/jbm.a.32927.
 22. Wendt D, Marsano A, Jakob M, et al. Oscillating perfusion of cell suspensions through three-dimensional scaffolds enhances cell seeding efficiency and uniformity. *Biotechnol Bioeng* 2003; 84(2):205-14. doi: 10.1002/bit.
 23. Stanton DW, CJ Rutland. Modeling fuel film formation and wall interaction in diesel engines. *SAE Trans* 1996; 808-24. doi: 10.4271/960628.
 24. O'Rourke PJ, Amsden AA. A particle numerical model for wall film dynamics in port-injected engines. *SAE Technical Paper Series*. Detroit: SAE; 1996. p.2000-13.
 25. O'Rourke PJ, Amsden AA. A spray/wall interaction submodel for the KIVA-3 wall film model. *SAE Technical Paper Series*. Detroit: SAE; 2000. p.281-98. doi: 10.4271/2000-01-0271.
 26. Karande TS, Ong JL, Agrawal CM. Diffusion in musculoskeletal tissue engineering scaffolds: design issues related to porosity, permeability, architecture, and nutrient mixing. *Ann Biomed Eng* 2004; 32(12):1728-43. doi: 10.1007/s10439-004-7825-2.
 27. Makhaniok A, Haranava Y, Goranov V, et al. In silico prediction of the cell proliferation in porous scaffold using model of effective pore. *Biosystems* 2013; 114(3):227-37. doi: 10.1016/j.biosystems.2013.10.001.
 28. Dunn JC, Chan WY, Cristini V, et al. Analysis of cell growth in three-dimensional scaffolds. *Tissue Eng* 2006; 12(4):705-16. doi: 10.1089/ten.2006.12.705.
 29. Rouwkema J, Koopman B, Blitterswijk C, et al. Supply of nutrients to cells in engineered tissues. *Biotechnol Genet Eng Rev* 2010; 26:163-78. doi: 10.5661/bger-26-163.
 30. Reynolds M, McCann SR. Human marrow stromal cells in short-term semi-solid bone marrow culture in aplastic anaemia. *Scand J Haematol* 1985; 34(2):101-10. doi: 10.1111/j.1600-0609.1985.tb02241.x.
 31. Winterton RHS. Thermal design of nuclear reactors. Elsevier; 1981. Available from: <https://doi.org/10.1016/B978-0-08-024215-6.50003-6>.
 32. Botchwey EA, Dupree MA, Pollack SR, et al. Tissue engineered bone: measurement of nutrient transport in three-dimensional matrices. *J Biomed Mater Res A* 2003; 67(1):357-67. doi: 10.1002/jbm.a.10111.
 33. Mofrad AZ, Mashayekhan S, Bastani D. Simulation of the effects of oxygen carriers and scaffold geometry on oxygen distribution and cell growth in a channeled scaffold for engineering myocardium. *Math Biosci* 2017; 294:160-71. doi: 10.1016/j.mbs.2017.09.003.
 34. Pérez MA, Prendergast PJ. Random-walk models of cell dispersal included in mechanobiological simulations of tissue differentiation. *J Biomech* 2007; 40(10):2244-53. doi: 10.1016/j.jbiomech.2006.10.020.
 35. Søballe K, Hansen ES, B-Rasmussen H, et al. Tissue ingrowth into titanium and hydroxyapatite-coated implants during stable and unstable mechanical conditions. *J Orthop Res* 1992; 10(2):285-99. doi: 10.1002/jor.1100100216.
 36. Lanza R, Gearhart J, Hogan B, et al. Essentials of stem cell biology. Elsevier; 2009. Available from: <https://doi.org/10.1016/C2009-0-00078-6>.

中文摘要

组织工程中细胞实验和动物实验的替代计算模型

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摘要

在骨科植入物和再生医学研究领域, 为了得到合适的产品需要经过反复实验分析优化材料、结构、机械性能等指标, 从细胞实验到临床试验的整个过程既昂贵又耗时。计算模拟方法是组织工程产品开发的设计工具, 可以有效降低开发成本和耗时。本文综述了一系列组织工程模型, 他们能模拟从细胞附着到组织再生的全过程。从细胞黏附到组织再生的一体化模型的建立能优化组织工程产品设计和评价过程。然而, 目前模拟组织工程过程的模型均各自独立。本文重点介绍了组织工程中细胞黏附、营养物质运输和细胞增殖、分化、迁移的模型, 试图将所有独立模型串联在一起以供读者使用。在细胞黏附模型中, 离散相模型能最精确地模拟控制细胞运动过程, Stanton-Rutland 模型能模拟细胞黏附过程。营养盐运输模型能是一种能将流体体积模型和组分传输模型耦合的数值模型, 在组织工程中被应用于营养盐运移过程的预测。有限元以及随机游走算法被用于细胞增殖、分化和迁移的模拟。大多数模型的准确性和有效性还有待进一步研究证实。由于尚缺乏检测营养物质扩散速率的技术, 在凝血领域模型分析方法的研究尤其少。因此, 组织工程全过程建模方法的研究还有很多工作要做。在未来, 数值模型将被视为研究组织工程产品生物性能的最佳方法, 并能够优化组织工程产品的参数和材料类型。

关键词: 组织工程; 支架; 计算机辅助设计; 计算流体力学; 有限元模型