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Stem Cell

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Stem Cell Physics Research Literatures

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Abstract: Stem cells are derived from embryonic and non-embryonic tissues. Most stem cell studies are for animal stem cells and plants have also stem cell. Stem cells were discovered in 1981 from early mouse embryos. Stem cells have the potential to develop into all different cell types in the living body. Stem cell is a body repair system. When a stem cell divides it can be still a stem cell or become adult cell, such as a brain cell. Stem cells are unspecialized cells and can renew themselves by cell division, and stem cells can also differentiate to adult cells with special functions. Stem cells replace the old cells and repair the damaged tissues. Embryonic stem cells can become all cell types of the body because they are pluripotent. Adult stem cells are thought to be limited to differentiating into different cell types of their tissue of origin. This article introduces recent research reports as references in the related studies.
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Key words: stem cell; physics; life; research; literature

Introduction

The stem cell is the origin of an organism's life that has the potential to develop into many different types of cells in life bodies. In many tissues stem cells serve as a sort of internal repair system, dividing essentially without limit to replenish other cells as long as the person or animal is still alive. When a stem cell divides, each new cell has the potential either to remain a stem cell or become another type of cell with a more specialized function, such as a red blood cell or a brain cell. This article introduces recent research reports as references in the related studies.

The following introduces recent reports as references in the related studies.

Accardo, A., et al. (2019). "Interfacing cells with microengineered scaffolds for neural tissue reconstruction." Brain Res Bull **152**: 202-211.

The development of cellular microenvironments suitable for neural tissue engineering purposes involves a plethora of research fields ranging from cell biology to biochemistry, neurosciences, physics, nanotechnology, mechanobiology. In the last two decades, this multi-disciplinary activity has led to the emergence of numerous strategies to create architectures capable of reproducing the topological, biochemical and mechanical properties of the extracellular matrix present in the central (CNS) and peripheral nervous system (PNS). Some of these approaches have succeeded in inducing the functional recovery of damaged areas in the CNS and the PNS to address the current lack of effective medical treatments for this type of injury. In this review, we analyze recent developments in the realization of two-dimensional and three-dimensional neuronal scaffolds following either top-down or bottom-up approaches. After providing an overview of the different fabrication techniques employed for tailoring the biomaterials, we draw on specific examples to describe the major features of the developed approaches. We then conclude with prospective proof of concept studies on guiding scaffolds and regenerative models on macro-scale brain implants targeting neural regeneration.

Akamatsu, M., et al. (2020). "Principles of self-organization and load adaptation by the actin cytoskeleton during clathrin-mediated endocytosis." Elife **9**.

Force generation by actin assembly shapes cellular membranes. An experimentally constrained multiscale model shows that a minimal branched actin network is sufficient to internalize endocytic pits against membrane tension. Around 200 activated Arp2/3 complexes are required for robust internalization. A newly developed molecule-counting method determined that ~200 Arp2/3 complexes assemble at sites of clathrin-mediated endocytosis in human cells. Simulations predict that actin self-organizes into a radial branched array with growing ends oriented toward the base of the pit. Long actin filaments bend between attachment sites in the coat and the base of the pit. Elastic energy stored in bent filaments, whose presence was confirmed by cryo-

electron tomography, contributes to endocytic internalization. Elevated membrane tension directs more growing filaments toward the base of the pit, increasing actin nucleation and bending for increased force production. Thus, spatially constrained actin filament assembly utilizes an adaptive mechanism enabling endocytosis under varying physical constraints.

The outer membrane of a cell is a tight but elastic barrier that controls what enters or leaves the cell. Large molecules typically cannot cross this membrane unaided. Instead, to enter the cell, they must be packaged into a pocket of the membrane that is then pulled inside. This process, called endocytosis, shuttles material into a cell hundreds of times a minute. Endocytosis relies on molecular machines that assemble and disassemble at the membrane as required. One component, a protein called actin, self-assembles near the membrane into long filaments with many repeated subunits. These filaments grow against the membrane, pulling it inwards. But it was not clear how actin filaments organize in such a way that allows them to pull on the membrane with enough force - and without a template to follow. Akamatsu et al. set about identifying how actin operates during endocytosis by using computer simulations that were informed by measurements made in living cells. The simulations included information about the location of actin and other essential molecules, along with the details of how these molecules work individually and together. Akamatsu et al. also developed a method to count the numbers of molecules of a key protein at individual sites of endocytosis. High-resolution imaging was then used to create 3D pictures of actin and endocytosis in action in human cells grown in the laboratory. The analysis showed the way actin filaments arrange themselves depends on the starting positions of a few key molecules that connect to actin. Imaging confirmed that, like a pole-vaulting pole, the flexible actin filaments bend to store energy and then release it to pull the membrane inwards during endocytosis. Finally, the simulations predicted that the collection of filaments adapts its shape and size in response to the resistance of the elastic membrane. This makes the system opportunistic and adaptable to the unpredictable environment within cells.

Ambriz, X., et al. (2018). "The Mechanobiology of the Actin Cytoskeleton in Stem Cells during Differentiation and Interaction with Biomaterials." Stem Cells Int **2018**: 2891957.

An understanding of the cytoskeleton's importance in stem cells is essential for their manipulation and further clinical application. The cytoskeleton is crucial in stem cell biology and depends on physical and chemicals signals to define its structure. Additionally, cell culture conditions will be important in the proper maintenance of stemness, lineage commitment, and differentiation. This review focuses on the following areas: the role of the actin cytoskeleton of stem cells during differentiation, the significance of cellular morphology, signaling pathways involved in cytoskeletal rearrangement in stem cells, and the mechanobiology and mechanotransduction processes implicated in the interactions of stem cells with different surfaces of biomaterials, such as nanotopography, which is a physical cue influencing the differentiation of stem cells. Also, cancer stem cells are included since it is necessary to understand the role of their mechanical properties to develop new strategies to treat cancer. In this context, to study the stem cells requires integrated disciplines, including molecular and cellular biology, chemistry, physics, and immunology, as well as mechanobiology. Finally, since one of the purposes of studying stem cells is for their application in regenerative medicine, the deepest understanding is necessary in order to establish safety protocols and effective cell-based therapies.

Askenasy, N. (2006). "From the atom to the cell: Is the cat alive? Quantum mechanics and stem cell plasticity as deja vu." Stem Cells Dev **15**(4): 488-491.

The concepts submitted by quantum mechanics fascinated the scientific community during the first half of the 20(th) century. The second half was dominated by biology, culminating in the sequencing of the human genome and the study of stem cells. Although the anticipated revolution of cellular therapies in medicine is in its infancy, the conceptual debate over stem cell plasticity shares similarities with evolution of the quantum theory. Are there notions and modes of thinking that stem cell biologists should adopt from the evolution in the interpretation of the laws of physics?

Bakhman, A., et al. (2019). "Residue-level determinants of angiopoietin-2 interactions with its receptor Tie2." Proteins **87**(3): 185-197.

We combined computational and experimental methods to interrogate the binding determinants of angiopoietin-2 (Ang2) to its receptor tyrosine kinase (RTK) Tie2-a central signaling system in angiogenesis, inflammation, and tumorigenesis. We used physics-based electrostatic and surface-area calculations to identify the subset of interfacial Ang2 and Tie2 residues that can affect binding directly. Using random and site-directed mutagenesis and yeast surface display (YSD), we validated these predictions and identified additional Ang2 positions that affected receptor binding. We then used burial-based calculations to classify the larger set of Ang2 residues that are buried in the Ang2 core, whose mutations can perturb the Ang2 structure and thereby affect interactions with Tie2 indirectly. Our analysis showed that the Ang2-

Tie2 interface is dominated by nonpolar contributions, with only three Ang2 and two Tie2 residues that contribute electrostatically to intermolecular interactions. Individual interfacial residues contributed only moderately to binding, suggesting that engineering of this interface will require multiple mutations to reach major effects. Conversely, substitutions in substantially buried Ang2 residues were more prevalent in our experimental screen, reduced binding substantially, and are therefore more likely to have a deleterious effect that might contribute to oncogenesis. Computational analysis of additional RTK-ligand complexes, c-Kit-SCF and M-CSF-c-FMS, and comparison to previous YSD results, further show the utility of our combined methodology.

Bhuiyan, N. U. and J. W. Poston, Sr. (2005). "A revised model for electron dosimetry in the human small intestine." Health Phys **88**(1): 23-36.

In this study, the absorbed dose was calculated to the small intestine (SI) wall of an adult human from electrons in its lumen contents. The effects on dose due to variations in the lumen radius and wall-thickness also were studied. The SI model was based on values gleaned from anatomic and histologic reviews of the adult human SI. Histologic and radiological analyses of the SI suggested the microscopic intricacy of this walled organ could be avoided for dosimetric purposes and a set of concentric cylinders could be used to model the SI. The model was input into the Monte Carlo N-Particle (MCNP) version 4A computational package, which was used to simulate energy deposition in the SI by electrons of fifty discrete energies ranging 10-500 keV. The source electrons as well as all resulting particles, such as knock-on electrons, bremsstrahlung, and electrons created from bremsstrahlung interactions, were transported until the particle energies fell below the 1 keV low-energy cutoff. Detailed physics treatments for secondary photons were made. With a reasonable number of histories, appropriate variance reduction techniques were used to improve the precision of the Monte Carlo calculations. The model used very small tally regions, which ranged in thickness from 0.5 microm to 200 microm depending on the electron energy studied and tally location in the wall. Relative errors associated with these calculations were maintained at less than 5%. The large number of tally results across the wall for each of the energies studied enabled the construction of the energy-specific depth dose curves in the wall. Each of these curves was consistent with the anticipated energy deposition pattern. These curves showed that only a small fraction of the energy absorbed at the contents-mucus interface reaches the stem cell layers because the cells are located deep in the mucosa. This fraction was found to vary from 1.66 x 10(-6) to 1.21 x 10(-1) over the energy range 10-500 keV. These results demonstrated the interface dose, which has been routinely reported as the "wall" dose, is a significant overestimate of the actual dose to the stem cells. The dose uncertainties associated with variations of the critical cell depth were shown to be very high for electrons whose CSDA ranges in the soft tissue exceeded the depth of the critical cells. This study showed that the uncertainty in the wall-thickness had no effect on depth doses while variation in the lumen radius significantly changes depth doses. The results suggest that these changes could be approximated by the inverse square of the lumen radius.

Bianco, S., et al. (2019). "Modeling Single-Molecule Conformations of the HoxD Region in Mouse Embryonic Stem and Cortical Neuronal Cells." Cell Rep **28**(6): 1574-1583 e1574.

Complex architectural rearrangements are associated to the control of the HoxD genes in different cell types; yet, how they are implemented in single cells remains unknown. By use of polymer models, we dissect the locus 3D structure at the single DNA molecule level in mouse embryonic stem and cortical neuronal cells, as the HoxD cluster changes from a poised to a silent state. Our model describes published Hi-C, 3-way 4C, and FISH data with high accuracy and is validated against independent 4C data on the Nsi-SB 0.5-Mb duplication and on triple contacts. It reveals the mode of action of compartmentalization on the regulation of the HoxD genes that have gene- and cell-type-specific multi-way interactions with their regulatory elements and high cell-to-cell variability. It shows that TADs and higher-order 3D structures, such as metaTADs, associate with distinct combinations of epigenetic factors, including but not limited to CCCTC-binding factor (CTCF) and histone marks.

Biava, P. M., et al. (2019). "Stem Cell Differentiation Stage Factors and their Role in Triggering Symmetry Breaking Processes during Cancer Development: A Quantum Field Theory Model for Reprogramming Cancer Cells to Healthy Phenotypes." Curr Med Chem **26**(6): 988-1001.

A long history of research has pursued the use of embryonic factors isolated during cell differentiation processes for the express purpose of transforming cancer cells back to healthy phenotypes. Recent results have clarified that the substances present at different stages of cell differentiation-which we call stem cell differentiation stage factors (SCDSFs)-are proteins with low molecular weight and nucleic acids that regulate genomic expression. The present review summarizes how these substances, taken at different stages of cellular maturation, are able to retard proliferation of many human tumor cell lines and thereby reprogram cancer cells to healthy phenotypes. The model presented here is a quantum field theory (QFT) model in which SCDSFs are able to trigger

symmetry breaking processes during cancer development. These symmetry breaking processes, which lie at the root of many phenomena in elementary particle physics and condensed matter physics, govern the phase transitions of totipotent cells to higher degrees of diversity and order, resulting in cell differentiation. In cancers, which share many genomic and metabolic similarities with embryonic stem cells, stimulated redifferentiation often signifies the phenotypic reversion back to health and nonproliferation. In addition to acting on key components of the cellular cycle, SCDSFs are able to reprogram cancer cells by delicately influencing the cancer microenvironment, modulating the electrochemistry and thus the collective electrodynamic behaviors between dipole networks in biomacromolecules and the interstitial water field. Coherent effects in biological water, which are derived from a dissipative QFT framework, may offer new diagnostic and therapeutic targets at a systemic level, before tumor instantiation occurs in specific tissues or organs. Thus, by including the environment as an essential component of our model, we may push the prevailing paradigm of mutation-driven oncogenesis toward a closer description of reality.

Binhi, V. (2008). "Do naturally occurring magnetic nanoparticles in the human body mediate increased risk of childhood leukaemia with EMF exposure?" Int J Radiat Biol **84**(7): 569-579.

PURPOSE: To develop the hypothesis that magnetic nanoparticles, found in many organisms and often involved in biological reactions to weak electromagnetic fields (EMF), mediate EMF-induced DNA damage which could result in increased risk of childhood leukaemia and other cancers. MATERIALS AND METHODS: An analysis of current research into magnetic nanoparticles. Physics estimates and the development of the hypothesis that intracellular magnetic nanoparticles chronically change the free radical concentration and can mediate the enhanced rate of DNA damage in hematopoietic stem cells. RESULTS: The properties of magnetic nanoparticles are considered and the naturally occurring magnetic field generated by a magnetic nanoparticle within a cell is calculated to be in the range of about 1-200 millitesla, which exceeds the level of the natural geomagnetic field by orders of magnitude. Experiments are summarized on the biological effects of static magnetic field in this range. It is shown that magnetic nanoparticles can increase the rate of free radical formation by a few percent, in the course of an idealized radical-pair reaction in a cell. A mechanism is discussed that explains how weak alternating magnetic fields, of the order of 0.4 muT, could cause an increase in the rate of leukaemia via millitesla fields produced around superparamagnetic nanoparticles in hematopoietic stem cells. CONCLUSIONS: The postulated presence of magnetic nanoparticles located in hematopoietic stem cells could constitute a cancer risk factor. Superparamagnetic nanoparticles can possibly mediate increased level of leukaemia caused by background exposure to low-frequency weak EMF.

Bond, V. P. (1995). "Dose, effect severity, and imparted energy in assessing biological effects." Stem Cells **13 Suppl 1**: 21-29.

Because of the widespread efforts in cancer radioepidemiological studies to attach a value of absorbed dose to each exposed individual, the notion seems to have become prevalent that dose plays an essential role in the medical determination of the diagnosis and prognosis of the individual. This view is enhanced by the fact that, while the present quantities and units for radiological physics were developed in the context of the acute effects of large exposures to radiation, e.g., in radiotherapy where they still apply well, these same quantities and units have been used, without modification, to apply to cancer radioepidemiology in the context of low level irradiation. A principle purpose of the present communication is to show that, in medicine, dose plays a limited role even in the deterministic application of therapeutic agents, and that diagnosis and estimates of prognosis in medicine are based, not on dose, but on the severity of effect on, or damage to the organ or organs involved in a particular medical condition. Thus it is "going backward" to view estimates of the severity of effect, e.g., the fraction of cells with abnormalities, or killed, as a "biological dosimeter," rather than as a quantitative estimate of the severity of effect. The use of biological indicators is of maximum value in noncancerous disease or injury in which the severity of an effect causative for organ failure and a consequent quantal, e.g., a lethal response in the individual, can be measured with increasing accuracy by modern medical techniques.(ABSTRACT TRUNCATED AT 250 WORDS)

Booth, C., et al. (2012). "Christopher S. Potten, 1940-2012." Health Phys **103**(4): 339.

Buxboim, A., et al. (2010). "Matrix elasticity, cytoskeletal forces and physics of the nucleus: how deeply do cells 'feel' outside and in?" J Cell Sci **123**(Pt 3): 297-308.

Cellular organization within a multicellular organism requires that a cell assess its relative location, taking in multiple cues from its microenvironment. Given that the extracellular matrix (ECM) consists of the most abundant proteins in animals and contributes both structure and elasticity to tissues, ECM probably provides key physical cues to cells. In vivo, in the vicinity of many tissue cell types, fibrous characteristics of the ECM are less discernible than the measurably distinct elasticity that characterizes

different tissue microenvironments. As a cell engages matrix and actively probes, it senses the local elastic resistance of the ECM and nearby cells via their deformation, and--similar to the proverbial princess who feels a pea placed many mattresses below--the cell seems to possess feedback and recognition mechanisms that establish how far it can feel. Recent experimental findings and computational modeling of cell and matrix mechanics lend insight into the subcellular range of sensitivity. Continuity of deformation from the matrix into the cell and further into the cytoskeleton-caged and -linked nucleus also supports the existence of mechanisms that direct processes such as gene expression in the differentiation of stem cells. Ultimately, cells feel the difference between stiff or soft and thick or thin surroundings, regardless of whether or not they are of royal descent.

Chalut, K. J., et al. (2011). "Stem cell differentiation indicated by noninvasive photonic characterization and fractal analysis of subcellular architecture." Integr Biol (Camb) **3**(8): 863-867.

We hypothesised that global structural changes in stem cells would manifest with differentiation, and that these changes would be observable with light scattering microscopy. Analysed with a fractal dimension formalism, we observed significant structural changes in differentiating human mesenchymal stem cells within one day after induction, earlier than could be detected by gene expression profiling. Moreover, light scattering microscopy is entirely non-perturbative, so the same sample could be monitored throughout the differentiation process. We explored one possible mechanism, chromatin remodelling, to account for the changes we observed. Correlating with the staining of HP1alpha, a heterochromatin protein, we applied novel microscopy methods and fractal analysis to monitor the plastic dynamics of chromatin within stem cell nuclei. We showed that the level of chromatin condensation changed during differentiation, and provide one possible explanation for the changes seen with the light scattering method. These results lend physical insight into stem cell differentiation while providing physics-based methods for non-invasive detection of the differentiation process.

Chen, G. Y., et al. (2012). "A graphene-based platform for induced pluripotent stem cells culture and differentiation." Biomaterials **33**(2): 418-427.

Induced pluripotent stem cells (iPSCs) hold great promise as a cell source for regenerative medicine yet its culture, maintenance of pluripotency and induction of differentiation remain challenging. Conversely, graphene (G) and graphene oxide (GO) have captured tremendous interests in the fields of materials science, physics, chemistry and nanotechnology. Here we report on that G and GO can support the mouse iPSCs culture and allow for spontaneous differentiation. Intriguingly, G and GO surfaces led to distinct cell proliferation and differentiation characteristics. In comparison with the glass surface, iPSCs cultured on the G surface exhibited similar degrees of cell adhesion and proliferation while iPSCs on the GO surface adhered and proliferated at a faster rate. Moreover, G favorably maintained the iPSCs in the undifferentiated state while GO expedited the differentiation. The iPSCs cultured on both G and GO surfaces spontaneously differentiated into ectodermal and mesodermal lineages without significant disparity, but G suppressed the iPSCs differentiation towards the endodermal lineage whereas GO augmented the endodermal differentiation. These data collectively demonstrated that the different surface properties of G and GO governed the iPSCs behavior and implicate the potentials of graphene-based materials as a platform for iPSCs culture and diverse applications.

Chen, T. H., et al. (2012). "Patterns of periodic holes created by increased cell motility." Interface Focus **2**(4): 457-464.

The reaction and diffusion of morphogens is a mechanism widely used to explain many spatial patterns in physics, chemistry and developmental biology. However, because experimental control is limited in most biological systems, it is often unclear what mechanisms account for the biological patterns that arise. Here, we study a biological model of cultured vascular mesenchymal cells (VMCs), which normally self-organize into aggregates that form into labyrinthine configurations. We use an experimental control and a mathematical model that includes reacting and diffusing morphogens and a third variable reflecting local cell density. With direct measurements showing that cell motility was increased ninefold and threefold by inhibiting either Rho kinase or non-muscle myosin-II, respectively, our experimental results and mathematical modelling demonstrate that increased motility alters the multicellular pattern of the VMC cultures, from labyrinthine to a pattern of periodic holes. These results suggest implications for the tissue engineering of functional replacements for trabecular or spongy tissue such as endocardium and bone.

Cipitria, A. and M. Salmeron-Sanchez (2017). "Mechanotransduction and Growth Factor Signalling to Engineer Cellular Microenvironments." Adv Healthc Mater **6**(15).

Engineering cellular microenvironments involves biochemical factors, the extracellular matrix (ECM) and the interaction with neighbouring cells. This progress report provides a critical overview of key studies that incorporate growth factor (GF) signalling and mechanotransduction into the design of advanced microenvironments. Materials systems have been

developed for surface-bound presentation of GFs, either covalently tethered or sequestered through physico-chemical affinity to the matrix, as an alternative to soluble GFs. Furthermore, some materials contain both GF and integrin binding regions and thereby enable synergistic signalling between the two. Mechanotransduction refers to the ability of the cells to sense physical properties of the ECM and to transduce them into biochemical signals. Various aspects of the physics of the ECM, i.e. stiffness, geometry and ligand spacing, as well as time-dependent properties, such as matrix stiffening, degradability, viscoelasticity, surface mobility as well as spatial patterns and gradients of physical cues are discussed. To conclude, various examples illustrate the potential for cooperative signalling of growth factors and the physical properties of the microenvironment for potential applications in regenerative medicine, cancer research and drug testing.

Ciucci, S., et al. (2017). "Enlightening discriminative network functional modules behind Principal Component Analysis separation in differential-omic science studies." Sci Rep **7**: 43946.

Omic science is rapidly growing and one of the most employed techniques to explore differential patterns in omic datasets is principal component analysis (PCA). However, a method to enlighten the network of omic features that mostly contribute to the sample separation obtained by PCA is missing. An alternative is to build correlation networks between univariately-selected significant omic features, but this neglects the multivariate unsupervised feature compression responsible for the PCA sample segregation. Biologists and medical researchers often prefer effective methods that offer an immediate interpretation to complicated algorithms that in principle promise an improvement but in practice are difficult to be applied and interpreted. Here we present PC-corr: a simple algorithm that associates to any PCA segregation a discriminative network of features. Such network can be inspected in search of functional modules useful in the definition of combinatorial and multiscale biomarkers from multifaceted omic data in systems and precision biomedicine. We offer proofs of PC-corr efficacy on lipidomic, metagenomic, developmental genomic, population genetic, cancer promoteromic and cancer stem-cell mechanomic data. Finally, PC-corr is a general functional network inference approach that can be easily adopted for big data exploration in computer science and analysis of complex systems in physics.

Constine, L. S., et al. (2019). "Pediatric Normal Tissue Effects in the Clinic (PENTEC): An International Collaboration to Analyse Normal Tissue Radiation Dose-Volume Response Relationships for Paediatric Cancer Patients." Clin Oncol (R Coll Radiol) **31**(3): 199-207.

With advances in multimodality therapy, childhood cancer cure rates approach 80%. However, both radiotherapy and chemotherapy can cause debilitating or even fatal late adverse events that are critical to understand, mitigate or prevent. QUANTEC (Quantitative Analysis of Normal Tissue Effects in the Clinic) identified radiation dose constraints for normal tissues in adults and pointed out the uncertainties in those constraints. The range of adverse events seen in children is different from that in adults, in part due to the vulnerability/characteristics of radiation damage to developing tissues, and in part due to the typical body sites affected by childhood cancer that lead to collateral irradiation of somewhat different normal tissues and organs compared with adults. Many childhood cancer survivors have a long life expectancy and may develop treatment-induced secondary cancers and severe organ/tissue injury 10, 20 or more years after treatment. Collaborative long-term observational studies and clinical research programmes for survivors of paediatric and adolescent cancer provide adverse event data for follow-up periods exceeding 40 years. Data analysis is challenging due to the interaction between therapeutic and developmental variables, the lack of radiation dose-volume data and the fact that most childhood malignancies are managed with combined modality therapy. PENTEC (Pediatric Normal Tissue Effects in the Clinic) is a volunteer research collaboration of more than 150 physicians, medical physicists, mathematical modellers and epidemiologists organised into 18 organ-specific working groups conducting a critical review and synthesis of quantitative data from existing studies aiming to: (1) establish quantitative, evidence-based dose/volume/risk guidelines to inform radiation treatment planning and, in turn, improve outcomes after radiation therapy for childhood cancers; (2) explore the most relevant risk factors for toxicity, including developmental status; (3) describe specific physics and dosimetric issues relevant to paediatric radiotherapy; and (4) propose dose-volume outcome reporting standards for publications on childhood cancer therapy outcomes. The impact of other critical modifiers of normal tissue radiation damage, including chemotherapy, surgery, stem cell transplantation and underlying genetic predispositions are also considered. The aims of the PENTEC reports are to provide clinicians with an analysis of the best available data to make informed decisions regarding radiation therapy normal organ dose constraints for planning childhood cancer treatment, and to define future research priorities.

Cotlet, G. and T. E. Blue (2000). "A stochastic model of radiation-induced bone marrow damage." Health Phys **78**(3): 289-294.

A stochastic model, based on consensus principles from radiation biology, is used to estimate bone-marrow stem cell pool survival (CFU-S and stroma cells) after irradiation. The dose response model consists of three coupled first order linear differential equations which quantitatively describe time dependent cellular damage, repair, and killing of red bone marrow cells. This system of differential equations is solved analytically through the use of a matrix approach for continuous and fractionated irradiations. The analytic solutions are confirmed through the dynamical solution of the model equations using SIMULINK. Rate coefficients describing the cellular processes of radiation damage and repair, extrapolated to humans from animal data sets and adjusted for neutron-gamma mixed fields, are employed in a SIMULINK analysis of criticality accidents. The results show that, for the time structures which may occur in criticality accidents, cell survival is established mainly by the average dose and dose rate.

Dahl-Jensen, S. and A. Grapin-Botton (2017). "The physics of organoids: a biophysical approach to understanding organogenesis." Development **144**(6): 946-951.

Organoids representing a diversity of tissues have recently been created, bridging the gap between cell culture and experiments performed in vivo Being small and amenable to continuous monitoring, they offer the opportunity to scrutinize the dynamics of organ development, including the exciting prospect of observing aspects of human embryo development live. From a physicist's perspective, their ability to self-organize - to differentiate and organize cells in space - calls for the identification of the simple rules that underlie this capacity. Organoids provide tractable conditions to investigate the effects of the growth environment, including its molecular composition and mechanical properties, along with the initial conditions such as cell number and type(s). From a theoretical standpoint, different types of in silico modeling can complement the measurements performed in organoids to understand the role of chemical diffusion, contact signaling, differential cell adhesion and mechanical controls. Here, we discuss what it means to take a biophysical approach to understanding organogenesis in vitro and how we might expect such approaches to develop in the future.

Delsanto, P. P., et al. (2008). "A multilevel approach to cancer growth modeling." J Theor Biol **250**(1): 16-24.

Cancer growth models may be divided into macroscopic models, which describe the tumor as a single entity, and microscopic ones, which consider the tumor as a complex system whose behavior emerges from the local dynamics of its basic components, the neoplastic cells. Mesoscopic models (e.g. as based on the Local Interaction Simulation Approach [Delsanto, P.P., Mignogna, R., Scalerandi, M., Schechter, R., 1998. In: Delsanto, P.P. Saenz, A.W. (Eds.), New Perspectives on Problems in Classical and Quantum Physics, vol. 2. Gordon & Breach, New Delhi, p. 5174]), which explicitly consider the behavior of cell clusters and their interactions, may be used instead of the microscopic ones, in order to study the properties of cancer biology that strongly depend on the interactions of small groups of cells at intermediate spatial and temporal scales. All these approaches have been developed independently, which limits their usefulness, since they all include relevant features and information that should be cross-correlated for a deeper understanding of the mechanisms involved. In this contribution we consider multicellular tumor spheroids as biological reference systems and propose an intermediate model to bridge the gap between a macroscopic formulation of tumor growth and a mesoscopic one. Thus we are able to establish, as an important result of our formalism, a direct correspondence between parameters characterizing processes occurring at different scales. In particular, we analyze their dependence on an important limiting factor to tumor growth, i.e. the extra-cellular matrix pressure. Since the macro and meso-models stem from totally different roots (energy conservation and clinical observations vs. cell groups dynamics), their consistency may be used to validate both approaches. It may also be interesting to note that the proposed formalism fits well into a recently proposed conjecture of growth laws universality.

Diaz, J. A. and M. F. Murillo (2012). "Phenotype characterization of embryoid body structures generated by a crystal comet effect tail in an intercellular cancer collision scenario." Cancer Manag Res **4**: 9-21.

Cancer is, by definition, the uncontrolled growth of autonomous cells that eventually destroy adjacent tissues and generate architectural disorder. However, this concept cannot be totally true. In three well documented studies, we have demonstrated that cancer tissues produce order zones that evolve over time and generate embryoid body structures in a space-time interval. The authors decided to revise the macroscopic and microscopic material in well-developed malignant tumors in which embryoid bodies were identified to determine the phenotype characterization that serves as a guideline for easy recognition. The factors responsible for this morphogenesis are physical, bioelectric, and magnetic susceptibilities produced by crystals that act as molecular designers for the topographic gradients that guide the surrounding silhouette and establish tissue head-tail positional identities. The structures are located in amniotic-like cavities and show characteristic somite-like embryologic segmentation.

Immunophenotypic study has demonstrated exclusion factor positional identity in relation to enolase-immunopositive expression of embryoid body and human chorionic gonadotropin immunopositivity exclusion factor expression in the surrounding tissues. The significance of these observations is that they can also be predicted by experimental image data collected by the Large Hadron Collider (LHC) accelerator at the European Organization for Nuclear Research, in which two-beam subatomic collision particles in the resulting debris show hyperorder domains similar to those identified by us in intercellular cancer collisions. Our findings suggest that we are dealing with true reverse biologic system information in an activated collective cancer stem cell memory, in which physics participates in the elaboration of geometric complexes and chiral biomolecules that serve to build bodies with embryoid print as it develops during gestation. Reversal mechanisms in biology are intimately linked with DNA repair. Further genotype studies must be carried out to determine whether the subproducts of these structures can be used in novel strategies to treat cancer.

Discher, D. E., et al. (2017). "Matrix Mechanosensing: From Scaling Concepts in 'Omics Data to Mechanisms in the Nucleus, Regeneration, and Cancer." Annu Rev Biophys **46**: 295-315.

Many of the most important molecules of life are polymers. In animals, the most abundant of the proteinaceous polymers are the collagens, which constitute the fibrous matrix outside cells and which can also self-assemble into gels. The physically measurable stiffness of gels, as well as tissues, increases with the amount of collagen, and cells seem to sense this stiffness. An understanding of this mechanosensing process in complex tissues, including fibrotic disease states with high collagen, is now utilizing 'omics data sets and is revealing polymer physics-type, nonlinear scaling relationships between concentrations of seemingly unrelated biopolymers. The nuclear structure protein lamin A provides one example, with protein and transcript levels increasing with collagen 1 and tissue stiffness, and with mechanisms rooted in protein stabilization induced by cytoskeletal stress. Physics-based models of fibrous matrix, cytoskeletal force dipoles, and the lamin A gene circuit illustrate the wide range of testable predictions emerging for tissues, cell cultures, and even stem cell-based tissue regeneration. Beyond the epigenetics of mechanosensing, the scaling in cancer of chromosome copy number variations and other mutations with tissue stiffness suggests that genomic changes are occurring by mechanogenomic processes that now require elucidation.

Dritschilo, A. and D. S. Sherman (1981). "Radiation and chemical injury in the bone marrow." Environ Health Perspect **39**: 59-64.

Hematopoietic system toxicity is a major limiting factor in the use of aggressive combined modality therapy in the treatment of malignant disease. In this review, the known drug-x-ray interactions using tissue culture systems are extended to the bone marrow compartment. Two hypotheses prevail for late bone marrow failure: (1) stromal damage to the vasculature with subsequent fibrosis and (2) irreversible stem cell depletion in the irradiated site. Clinical extensions of the experimental data for bone marrow kinetics in the animal model have not proven successful to date. The future strategies for therapy of malignancies in which both radiation and chemotherapy are employed may require dose modification or treatment planning to limit bone marrow toxicity.

English, D. (2004). "PR vs. PR: will press release top peer review in stem cell research?" Stem Cells Dev **13**(2): 157-159.

Esdaille, C. J., et al. (2021). "Regenerative engineering: a review of recent advances and future directions." Regen Med **16**(5): 495-512.

Regenerative engineering is defined as the convergence of the disciplines of advanced material science, stem cell science, physics, developmental biology and clinical translation for the regeneration of complex tissues and organ systems. It is an expansion of tissue engineering, which was first developed as a method of repair and restoration of human tissue. In the past three decades, advances in regenerative engineering have made it possible to treat a variety of clinical challenges by utilizing cutting-edge technology currently available to harness the body's healing and regenerative abilities. The emergence of new information in developmental biology, stem cell science, advanced material science and nanotechnology have provided promising concepts and approaches to regenerate complex tissues and structures.

Font-Clos, F., et al. (2018). "Topography of epithelial-mesenchymal plasticity." Proc Natl Acad Sci U S A **115**(23): 5902-5907.

The transition between epithelial and mesenchymal states has fundamental importance for embryonic development, stem cell reprogramming, and cancer progression. Here, we construct a topographic map underlying epithelial-mesenchymal transitions using a combination of numerical simulations of a Boolean network model and the analysis of bulk and single-cell gene expression data. The map reveals a multitude of metastable hybrid phenotypic states, separating stable epithelial and mesenchymal states, and is reminiscent of the free energy measured in glassy materials and disordered solids. Our work not only elucidates the nature of hybrid mesenchymal/epithelial states but also provides a general strategy to construct a topographic

representation of phenotypic plasticity from gene expression data using statistical physics methods.

Fredj, A., et al. (2012). "The single T65S mutation generates brighter cyan fluorescent proteins with increased photostability and pH insensitivity." PLoS One **7**(11): e49149.

Cyan fluorescent proteins (CFP) derived from Aequorea victoria GFP, carrying a tryptophan-based chromophore, are widely used as FRET donors in live cell fluorescence imaging experiments. Recently, several CFP variants with near-ultimate photophysical performances were obtained through a mix of site-directed and large scale random mutagenesis. To understand the structural bases of these improvements, we have studied more specifically the consequences of the single-site T65S mutation. We find that all CFP variants carrying the T65S mutation not only display an increased fluorescence quantum yield and a simpler fluorescence emission decay, but also show an improved pH stability and strongly reduced reversible photoswitching reactions. Most prominently, the Cerulean-T65S variant reaches performances nearly equivalent to those of mTurquoise, with QY = 0.84, an almost pure single exponential fluorescence decay and an outstanding stability in the acid pH range (pK(1/2) = 3.6). From the detailed examination of crystallographic structures of different CFPs and GFPs, we conclude that these improvements stem from a shift in the thermodynamic balance between two well defined configurations of the residue 65 hydroxyl. These two configurations differ in their relative stabilization of a rigid chromophore, as well as in relaying the effects of Glu222 protonation at acid pHs. Our results suggest a simple method to greatly improve numerous FRET reporters used in cell imaging, and bring novel insights into the general structure-photophysics relationships of fluorescent proteins.

Garcia, A., et al. (2016). "Modeling of the mechano-chemical behaviour of the nuclear pore complex: current research and perspectives." Integr Biol (Camb) **8**(10): 1011-1021.

Recent evidence suggests that mechanical deformation of the cell nucleus regulates the nuclear import of the transcriptional activators of genes involved in primary physiological cell responses such as stem cell differentiation. In addition, this nuclear mechanosensing response is de-regulated in pathological states, such as cancer and neurodegeneration. One hypothesis that could greatly advance the field is that the deformation of the nuclear envelope activates nuclear pore complexes through a direct mechanical link. The understanding of this possible mechanism for nuclear pore complex stretch-activation entails studying the mechanical connection of this complex to the nuclear envelope at the nanoscale. The nanomechanics of the nuclear pore complex is thus emerging as a novel research field, bridging nanoscience with nanotechnology. This review examines the frontier of research methodologies that are potentially useful for building a computational model of this interaction. This includes, for example, electron tomography to assess the geometrical features of the nuclear pore complex and nanoindentation to estimate its mechanical properties and that of the nuclear envelope. In order to summarize the state-of-the-art and perspectives in the field of NPC nanomechanics, this review covers highly interdisciplinary experimental and theoretical research methodologies pertaining to the fields of physics, chemistry, biology, materials and mechanics.

Gazquez, J., et al. (2016). "Emerging Diluted Ferromagnetism in High-Tc Superconductors Driven by Point Defect Clusters." Adv Sci (Weinh) **3**(6): 1500295.

Defects in ceramic materials are generally seen as detrimental to their functionality and applicability. Yet, in some complex oxides, defects present an opportunity to enhance some of their properties or even lead to the discovery of exciting physics, particularly in the presence of strong correlations. A paradigmatic case is the high-temperature superconductor YBa2Cu3O7-delta (Y123), in which nanoscale defects play an important role as they can immobilize quantized magnetic flux vortices. Here previously unforeseen point defects buried in Y123 thin films that lead to the formation of ferromagnetic clusters embedded within the superconductor are unveiled. Aberration-corrected scanning transmission microscopy has been used for exploring, on a single unit-cell level, the structure and chemistry resulting from these complex point defects, along with density functional theory calculations, for providing new insights about their nature including an unexpected defect-driven ferromagnetism, and X-ray magnetic circular dichroism for bearing evidence of Cu magnetic moments that align ferromagnetically even below the superconducting critical temperature to form a dilute system of magnetic clusters associated with the point defects.

Germain, L., et al. (2006). "[Canadian founders of haematopoietic stem cell research receive the Lasker Award]." Med Sci (Paris) **22**(2): 212-213.

Glauche, I., et al. (2013). "Stem cell clonality -- theoretical concepts, experimental techniques, and clinical challenges." Blood Cells Mol Dis **50**(4): 232-240.

Here we report highlights of discussions and results presented at an International Workshop on Concepts and Models of Stem Cell Organization held on July 16th and 17th, 2012 in Dresden, Germany. The goal of the workshop was to undertake a systematic survey of state-of-the-art methods and results of

clonality studies of tissue regeneration and maintenance with a particular emphasis on the hematopoietic system. The meeting was the 6th in a series of similar conceptual workshops, termed StemCellMathLab,(2) all of which have had the general objective of using an interdisciplinary approach to discuss specific aspects of stem cell biology. The StemCellMathLab 2012, which was jointly organized by the Institute for Medical Informatics and Biometry, Medical Faculty Carl Gustav Carus, Dresden University of Technology and the Institute for Medical Informatics, Statistics and Epidemiology, Medical Faculty, University of Leipzig, brought together 32 scientists from 8 countries, with scientific backgrounds in medicine, cell biology, virology, physics, computer sciences, bioinformatics and mathematics. The workshop focused on the following questions: (1) How heterogeneous are stem cells and their progeny? and (2) What are the characteristic differences in the clonal dynamics between physiological and pathophysiological situations? In discussing these questions, particular emphasis was placed on (a) the methods for quantifying clones and their dynamics in experimental and clinical settings and (b) general concepts and models for their description. In this workshop summary we start with an introduction to the current state of clonality research and a proposal for clearly defined terminology. Major topics of discussion include clonal heterogeneity in unperturbed tissues, clonal dynamics due to physiological and pathophysiological pressures and conceptual and technical issues of clone quantification. We conclude that an interactive cross-disciplinary approach to research in this field will continue to promote a conceptual understanding of tissue organization.

Gleicher, N., et al. (2017). "A single trophectoderm biopsy at blastocyst stage is mathematically unable to determine embryo ploidy accurately enough for clinical use." Reprod Biol Endocrinol **15**(1): 33.

BACKGROUND: It has become increasingly apparent that the trophectoderm (TE) at blastocyst stage is much more mosaic than has been appreciated. Whether preimplantation genetic screening (PGS), utilizing a single TE biopsy (TEB), can reliably determine embryo ploidy has, therefore, increasingly been questioned in parallel. METHODS: We for that reason here established 2 mathematical models to assess probabilities of false-negative and false-positive results of an on average 6-cell biopsy from an approximately 300-cell TE. This study was a collaborative effort between investigators at The Center for Human Reproduction in New York City and the Center for Studies in Physics and Biology and the Brivanlou Laboratory of Stem Cell Biology and Molecular Embryology, the latter two both at Rockefeller University in New York City. RESULTS: Both models revealed that even under best case scenario, assuming even distribution of mosaicism in TE (since mosaicism is usually clonal, a highly unlikely scenario), a biopsy of at least 27 TE cells would be required to reach minimal diagnostic predictability from a single TEB. CONCLUSIONS: As currently performed, a single TEB is, therefore, mathematically incapable of reliably determining whether an embryo can be transferred or should be discarded. Since a single TEB, as currently performed, apparently is not representative of the complete TE, this study, thus, raises additional concern about the clinical utilization of PGS.

Guillemin, A. and H. S. MP (2020). "Non-equilibrium statistical physics, transitory epigenetic landscapes, and cell fate decision dynamics." Math Biosci Eng **17**(6): 7916-7930.

Statistical physics provides a useful perspective for the analysis of many complex systems; it allows us to relate microscopic fluctuations to macroscopic observations. Developmental biology, but also cell biology more generally, are examples where apparently robust behaviour emerges from highly complex and stochastic sub-cellular processes. Here we attempt to make connections between different theoretical perspectives to gain qualitative insights into the types of cell-fate decision making processes that are at the heart of stem cell and developmental biology. We discuss both dynamical systems as well as statistical mechanics perspectives on the classical Waddington or epigenetic landscape. We find that non-equilibrium approaches are required to overcome some of the shortcomings of classical equilibrium statistical thermodynamics or statistical mechanics in order to shed light on biological processes, which, almost by definition, are typically far from equilibrium.

Higgins, A. Z., et al. (2011). "Effects of freezing profile parameters on the survival of cryopreserved rat embryonic neural cells." J Neurosci Methods **201**(1): 9-16.

The ability to successfully cryopreserve neural cells would represent an important advance with benefits to neural tissue engineering, neural transplantation, and neuroscience research. We have examined key factors responsible for damage to rat embryonic neural cells during cryopreservation using a two-step temperature profile, with an emphasis on the effects of cooling rate and plunge temperature. Our results indicate that the initial addition of 8% dimethyl sulfoxide (DMSO) and seeding of extracellular ice do not significantly decrease viable cell yield. However, subsequent freezing resulted in significant cell losses for all profile parameter combinations examined. A maximum post-thaw survival of 56% (compared to unfrozen controls) was observed after cooling at 2 degrees C/min to -80 degrees C followed by direct

immersion in liquid nitrogen. Single-step removal of DMSO after thawing was associated with an additional 40-70% loss of viable cells, and the number of viable cells was further reduced by approximately 70% after 2 days of cell culture (resulting in a net viable cell yield of 9.6+/-0.4%). Nonetheless, the cryopreserved neurons that did survive displayed a normal morphology, including formation of neurites. Trends in neuronal viability conformed with predictions of existing theoretical models of cell freezing, with reduced survival for rapid cooling rates or high plunge temperatures (attributable to intracellular ice formation), and decreasing viability with increasing profile duration (consistent with the known effects of cell dehydration at suboptimal cooling rates). These observations suggest that neural cells are good candidates for further refinement of freezing profile design using a physics-based approach to parameter optimization.

Higuera, G. A., et al. (2012). "The physics of tissue formation with mesenchymal stem cells." Trends Biotechnol **30**(11): 583-590.

Cells react to various forms of physical phenomena that promote and maintain the formation of tissues. The best example of this are cells of musculoskeletal origin, such as mesenchymal stem cells (MSCs), which consistently proliferate or differentiate under cues from hydrostatic pressure, diffusive mass transport, shear stress, surface chemistry, mechanotransduction, and molecular kinetics. To date, no other cell type shows greater receptiveness to macroscopic and microscopic cues, highlighting the acute sensitivity of MSCs and the importance of physical principles in tissue homeostasis. In this review, we describe the literature that has shown how physical phenomena govern MSCs biology and provide insight into the mechanisms and strategies that can spur new biotechnological applications with tissue biology.

Hsiao, W. W., et al. (2016). "Fluorescent Nanodiamond: A Versatile Tool for Long-Term Cell Tracking, Super-Resolution Imaging, and Nanoscale Temperature Sensing." Acc Chem Res **49**(3): 400-407.

Fluorescent nanodiamond (FND) has recently played a central role in fueling new discoveries in interdisciplinary fields spanning biology, chemistry, physics, and materials sciences. The nanoparticle is unique in that it contains a high density ensemble of negatively charged nitrogen-vacancy (NV(-)) centers as built-in fluorophores. The center possesses a number of outstanding optical and magnetic properties. First, NV(-) has an absorption maximum at approximately 550 nm, and when exposed to green-orange light, it emits bright fluorescence at approximately 700 nm with a lifetime of longer than 10 ns. These spectroscopic properties are little affected by surface modification but are distinctly different from those of cell autofluorescence and thus enable background-free imaging of FNDs in tissue sections. Such characteristics together with its excellent biocompatibility render FND ideal for long-term cell tracking applications, particularly in stem cell research. Next, as an artificial atom in the solid state, the NV(-) center is perfectly photostable, without photobleaching and blinking. Therefore, the NV-containing FND is suitable as a contrast agent for super-resolution imaging by stimulated emission depletion (STED). An improvement of the spatial resolution by 20-fold is readily achievable by using a high-power STED laser to deplete the NV(-) fluorescence. Such improvement is crucial in revealing the detailed structures of biological complexes and assemblies, including cellular organelles and subcellular compartments. Further enhancement of the resolution for live cell imaging is possible by manipulating the charge states of the NV centers. As the "brightest" member of the nanocarbon family, FND holds great promise and potential for bioimaging with unprecedented resolution and precision. Lastly, the NV(-) center in diamond is an atom-like quantum system with a total electron spin of 1. The ground states of the spins show a crystal field splitting of 2.87 GHz, separating the ms = 0 and +/-1 sublevels. Interestingly, the transitions between the spin sublevels can be optically detected and manipulated by microwave radiation, a technique known as optically detected magnetic resonance (ODMR). In addition, the electron spins have an exceptionally long coherence time, making FND useful for ultrasensitive detection of temperature at the nanoscale. Pump-probe-type nanothermometry with a temporal resolution of better than 10 mus has been achieved with a three-point sampling method. Gold/diamond nanohybrids have also been developed for highly localized hyperthermia applications. This Account provides a summary of the recent advances in FND-enabled technologies with a special focus on long-term cell tracking, super-resolution imaging, and nanoscale temperature sensing. These emerging and multifaceted technologies are in synchronicity with modern imaging modalities.

Huang, A., et al. (2018). "BioBits Explorer: A modular synthetic biology education kit." Sci Adv **4**(8): eaat5105.

Hands-on demonstrations greatly enhance the teaching of science, technology, engineering, and mathematics (STEM) concepts and foster engagement and exploration in the sciences. While numerous chemistry and physics classroom demonstrations exist, few biology demonstrations are practical and accessible due to the challenges and concerns of growing living cells in classrooms. We introduce BioBits Explorer, a synthetic biology educational kit based on shelf-stable,

freeze-dried, cell-free (FD-CF) reactions, which are activated by simply adding water. The FD-CF reactions engage the senses of sight, smell, and touch with outputs that produce fluorescence, fragrances, and hydrogels, respectively. We introduce components that can teach tunable protein expression, enzymatic reactions, biomaterial formation, and biosensors using RNA switches, some of which represent original FD-CF outputs that expand the toolbox of cell-free synthetic biology. The BioBits Explorer kit enables hands-on demonstrations of cutting-edge science that are inexpensive and easy to use, circumventing many current barriers for implementing exploratory biology experiments in classrooms.

Huang, S. (2011). "Systems biology of stem cells: three useful perspectives to help overcome the paradigm of linear pathways." Philos Trans R Soc Lond B Biol Sci **366**(1575): 2247-2259.

Stem cell behaviours, such as stabilization of the undecided state of pluripotency or multipotency, the priming towards a prospective fate, binary fate decisions and irreversible commitment, must all somehow emerge from a genome-wide gene-regulatory network. Its unfathomable complexity defies the standard mode of explanation that is deeply rooted in molecular biology thinking: the reduction of observables to linear deterministic molecular pathways that are tacitly taken as chains of causation. Such culture of proximate explanation that uses qualitative arguments, simple arrow-arrow schemes or metaphors persists despite the ceaseless accumulation of 'omics' data and the rise of systems biology that now offers precise conceptual tools to explain emergent cell behaviours from gene networks. To facilitate the embrace of the principles of physics and mathematics that underlie such systems and help to bridge the gap between the formal description of theorists and the intuition of experimental biologists, we discuss in qualitative terms three perspectives outside the realm of their familiar linear-deterministic view: (i) state space (ii), high-dimensionality and (iii) heterogeneity. These concepts jointly offer a new vista on stem cell regulation that naturally explains many novel, counterintuitive observations and their inherent inevitability, obviating the need for ad hoc explanations of their existence based on natural selection. Hopefully, this expanded view will stimulate novel experimental designs.

Hurtado, J. L., et al. (2012). "Hybrid computational phantoms representing the reference adult male and adult female: construction and applications for retrospective dosimetry." Health Phys **102**(3): 292-304.

Currently, two classes of computational phantoms have been developed for dosimetry calculation: (1) stylized (or mathematical) and (2) voxel (or tomographic) phantoms describing human anatomy through mathematical surface equations and 3D voxel matrices, respectively. Mathematical surface equations in stylized phantoms are flexible, but the resulting anatomy is not as realistic. Voxel phantoms display far better anatomical realism, but they are limited in terms of their ability to alter organ shape, position, and depth, as well as body posture. A new class of computational phantoms called hybrid phantoms takes advantage of the best features of stylized and voxel phantoms-flexibility and anatomical realism, respectively. In the current study, hybrid computational phantoms representing the adult male and female reference anatomy and anthropometry are presented. These phantoms serve as the starting framework for creating patient or worker sculpted whole-body phantoms for retrospective dose reconstruction. Contours of major organs and tissues were converted or segmented from computed tomography images of a 36-y-old Korean volunteer and a 25-y-old U.S. female patient, respectively, with supplemental high-resolution CT images of the cranium. Polygon mesh models for the major organs and tissues were reconstructed and imported into Rhinoceros for non-uniform rational B-spline (NURBS) surface modeling. The resulting NURBS/polygon mesh models representing body contour and internal anatomy were matched to anthropometric data and reference organ mass data provided by Centers for Disease Control and Prevention and International Commission on Radiation Protection, respectively. Finally, two hybrid adult male and female phantoms were completed where a total of eight anthropometric data categories were matched to standard values within 4% and organ volumes matched to ICRP data within 1% with the exception of total skin. The hybrid phantoms were voxelized from the NURBS phantoms at resolutions of 0.158 x 0.158 x 0.158 cm and 0.126 x 0.126 x 0.126 cm for the male and female, respectively. To highlight the flexibility of the hybrid phantoms, graphical displays are given of (1) underweight and overweight adult male phantoms, (2) a sitting position for the adult female phantom, and (3) extraction and higher-resolution voxelization of the small intestine for localized dosimetry of mucosal and stem cell layers. These phantoms are used to model radioactively contaminated individuals and to then assess time-dependent detector count rate thresholds corresponding to 50, 250, and 500 mSv effective dose, as might be needed during in-field radiological triage by first responders or first receivers.

Ivanovska, I., et al. (2010). "Physical plasticity of the nucleus and its manipulation." Methods Cell Biol **98**: 207-220.

The genome is virtually identical in all cells within an organism, with epigenetic changes contributing largely to the plasticity in gene expression

during both development and aging. These changes include covalent modifications of chromatin components and altered chromatin organization as well as changes in other nuclear components, such as nuclear envelope lamins. Given that DNA in each chromosome is centimeters long and dozens of chromosomes are compacted into a microns-diameter nucleus through non-trivial interactions with the bounding envelope, the polymer physics of such a structure under stress can be complex but perhaps systematic. We summarize micromanipulation methods for measuring the physical plasticity of the nucleus, with recent studies documenting the extreme flexibility of human embryonic stem cells and the rigidification in model aging of progerin-type nuclei. Lamin-A/C is a common molecular factor, and methods are presented for its knockdown and measurement.

Iwamuro, M., et al. (2012). "A preliminary study for constructing a bioartificial liver device with induced pluripotent stem cell-derived hepatocytes." Biomed Eng Online **11**: 93.

BACKGROUND: Bioartificial liver systems, designed to support patients with liver failure, are composed of bioreactors and functional hepatocytes. Immunological rejection of the embedded hepatocytes by the host immune system is a serious concern that crucially degrades the performance of the device. Induced pluripotent stem (iPS) cells are considered a desirable source for bioartificial liver systems, because patient-derived iPS cells are free from immunological rejection. The purpose of this paper was to test the feasibility of a bioartificial liver system with iPS cell-derived hepatocyte-like cells. METHODS: Mouse iPS cells were differentiated into hepatocyte-like cells by a multi-step differentiation protocol via embryoid bodies and definitive endoderm. Differentiation of iPS cells was evaluated by morphology, PCR assay, and functional assays. iPS cell-derived hepatocyte-like cells were cultured in a bioreactor module with a pore size of 0.2 mum for 7 days. The amount of albumin secreted into the circulating medium was analyzed by ELISA. Additionally, after a 7-day culture in a bioreactor module, cells were observed by a scanning electron microscope. RESULTS: At the final stage of the differentiation program, iPS cells changed their morphology to a polygonal shape with two nucleoli and enriched cytoplasmic granules. Transmission electron microscope analysis revealed their polygonal shape, glycogen deposition in the cytoplasm, microvilli on their surfaces, and a duct-like arrangement. PCR analysis showed increased expression of albumin mRNA over the course of the differentiation program. Albumin and urea production was also observed. iPS-Heps culture in bioreactor modules showed the accumulation of albumin in the medium for up to 7 days. Scanning electron microscopy revealed the attachment of cell clusters to the hollow fibers of the module. These results indicated that iPS cells were differentiated into hepatocyte-like cells after culture for 7 days in a bioreactor module with a pore size of 0.2 mum. CONCLUSION: We consider the combination of a bioreactor module with a 0.2-mum pore membrane and embedded hepatocytes differentiated from iPS cells to be a promising option for bioartificial liver systems. This paper provides the basic concept and preliminary data for an iPS cell-oriented bioartificial liver system.PACS code: 87. Biological and medical physics, 87.85.-d Biomedical engineering, 87.85.Lf Tissue engineering, 87.85.Tu Modeling biomedical systems.

Jang, J. Y., et al. (2018). "Cold atmospheric plasma (CAP), a novel physicochemical source, induces neural differentiation through cross-talk between the specific RONS cascade and Trk/Ras/ERK signaling pathway." Biomaterials **156**: 258-273.

Plasma, formed by ionization of gas molecules or atoms, is the most abundant form of matter and consists of highly reactive physicochemical species. In the physics and chemistry fields, plasma has been extensively studied; however, the exact action mechanisms of plasma on biological systems, including cells and humans, are not well known. Recent evidence suggests that cold atmospheric plasma (CAP), which refers to plasma used in the biomedical field, may regulate diverse cellular processes, including neural differentiation. However, the mechanism by which these physicochemical signals, elicited by reactive oxygen and nitrogen species (RONS), are transmitted to biological system remains elusive. In this study, we elucidated the physicochemical and biological (PCB) connection between the CAP cascade and Trk/Ras/ERK signaling pathway, which resulted in neural differentiation. Excited atomic oxygen in the plasma phase led to the formation of RONS in the PCB network, which then interacted with reactive atoms in the extracellular liquid phase to form nitric oxide (NO). Production of large amounts of superoxide radical (O2(-)) in the mitochondria of cells exposed to CAP demonstrated that extracellular NO induced the reversible inhibition of mitochondrial complex IV. We also demonstrated that cytosolic hydrogen peroxide, formed by O2(-) dismutation, act as an intracellular messenger to specifically activate the Trk/Ras/ERK signaling pathway. This study is the first to elucidate the mechanism linking physicochemical signals from the CAP cascade to the intracellular neural differentiation signaling pathway, providing physical, chemical and biological insights into the development of therapeutic techniques to treat neurological diseases.

Jansen, K. A., et al. (2015). "A guide to mechanobiology: Where biology and physics meet." Biochim Biophys Acta **1853**(11 Pt B): 3043-3052.

Cells actively sense and process mechanical information that is provided by the extracellular environment to make decisions about growth, motility and differentiation. It is important to understand the underlying mechanisms given that deregulation of the mechanical properties of the extracellular matrix (ECM) is implicated in various diseases, such as cancer and fibrosis. Moreover, matrix mechanics can be exploited to program stem cell differentiation for organ-on-chip and regenerative medicine applications. Mechanobiology is an emerging multidisciplinary field that encompasses cell and developmental biology, bioengineering and biophysics. Here we provide an introductory overview of the key players important to cellular mechanobiology, taking a biophysical perspective and focusing on a comparison between flat versus three dimensional substrates. This article is part of a Special Issue entitled: Mechanobiology.

Jenkinson, G., et al. (2017). "Potential energy landscapes identify the information-theoretic nature of the epigenome." Nat Genet **49**(5): 719-729.

Epigenetics is the study of biochemical modifications carrying information independent of DNA sequence, which are heritable through cell division. In 1940, Waddington coined the term "epigenetic landscape" as a metaphor for pluripotency and differentiation, but methylation landscapes have not yet been rigorously computed. Using principles from statistical physics and information theory, we derive epigenetic energy landscapes from whole-genome bisulfite sequencing (WGBS) data that enable us to quantify methylation stochasticity genome-wide using Shannon's entropy, associating it with chromatin structure. Moreover, we consider the Jensen-Shannon distance between sample-specific energy landscapes as a measure of epigenetic dissimilarity and demonstrate its effectiveness for discerning epigenetic differences. By viewing methylation maintenance as a communications system, we introduce methylation channels and show that higher-order chromatin organization can be predicted from their informational properties. Our results provide a fundamental understanding of the information-theoretic nature of the epigenome that leads to a powerful approach for studying its role in disease and aging.

Kaneko, K. (2011). "The challenges facing systemic approaches in biology: an interview with kunihiko kaneko." Front Physiol **2**: 93.

Interviewed by Kumar Selvarajoo and Masa Tsuchiya at Institute for Advanced Biosciences, Keio University, Japan, I discuss my approach to biology, what I call complex systems biology. The approach is constructive in nature, and is based on dynamical systems theory and statistical physics. It is intended to understand universal characteristics of life systems; generic adaptation under noise, differentiation from stem cells in interacting cells, robustness and plasticity in evolution, and so forth. Current status and future directions in systems biology in Japan are also discussed.

Kirby, N., et al. (2011). "Physics strategies for sparing neural stem cells during whole-brain radiation treatments." Med Phys **38**(10): 5338-5344.

PURPOSE: Currently, there are no successful long-term treatments or preventive strategies for radiation-induced cognitive impairments, and only a few possibilities have been suggested. One such approach involves reducing the dose to neural stem cell compartments (within and outside of the hippocampus) during whole-brain radiation treatments for brain metastases. This study investigates the fundamental physics issues associated with the sparing of neural stem cells during photon radiotherapy for brain metastases. METHODS: Several factors influence the stem cell dose: intracranial scattering, collimator leakage, beam energy, and total number of beams. The relative importance of these factors is investigated through a set of radiation therapy plans, which are all variations of an initial 6 MV intensity-modulated radiation therapy (IMRT) plan designed to simultaneously deliver a whole-brain dose of 30 Gy and maximally reduce stem cell compartment dose. Additionally, an in-house leaf segmentation algorithm was developed that utilizes jaw motion to minimize the collimator leakage. RESULTS: The plans are all normalized such that 50% of the PTV receives 30 Gy. For the initial 6 MV IMRT plan, 50% of the stem cells receive a dose greater than 6.3 Gy. Calculations indicate that 3.6 Gy of this dose originates from intracranial scattering. The jaw-tracking segmentation algorithm, used in conjunction with direct machine parameter optimization, reduces the 50% stem cell dose to 4.3 and 3.7 Gy for 6 and 10 MV treatment beams, respectively. CONCLUSIONS: Intracranial scattering alone is responsible for a large dose contribution to the stem cell compartment. It is, therefore, important to minimize other contributing factors, particularly the collimator leakage, to maximally reduce dose to these critical structures. The use of collimator jaw tracking in conjunction with modern collimators can minimize this leakage.

La Porta, C. A. (2012). "Thoughts about cancer stem cells in solid tumors." World J Stem Cells **4**(3): 17-20.

Cancer chemotherapy efficacy is frequently impaired by either intrinsic or acquired tumor resistance. A fundamental problem in cancer research is identifying the cell type that is capable of sustaining neoplastic growth and its origin from normal tissue cells. In recent years, the cancer stem cell (CSC) theory has changed the classical view of tumor growth and therefore the therapeutic perspective. Overcoming intrinsic and acquired resistance of cancer

stem/progenitor cells to current clinical treatments represents a major challenge in treating and curing the most aggressive and metastatic cancers. On the other hand, the identification of CSCs in vivo and in vitro relies on specific surface markers that should allow the sorting cancer cells into phenotypically distinct subpopulations. In the present review, recent papers published on CSCs in solid tumors (breast, prostate, brain and melanoma) are discussed, highlighting critical points such as the choice of markers to sort CSCs and mouse models to demonstrate that CSCs are able to replicate the original tumor. A discussion of the possible role of aldehyde dehydrogenase and CXCR6 biomarkers as signaling molecules in CSCs and normal stem cells is also discussed. The author believes that efforts have to be made to investigate the functional and biological properties of putative CSCs in cancer. Developing diagnostic/prognostic tools to follow cancer development is also a challenge. In this connection it would be useful to develop a multidisciplinary approach combining mathematics, physics and biology which merges experimental approaches and theory. Biological models alone are probably unable to resolve the problem completely.

Lang, A. H., et al. (2014). "Epigenetic landscapes explain partially reprogrammed cells and identify key reprogramming genes." PLoS Comput Biol **10**(8): e1003734.

A common metaphor for describing development is a rugged "epigenetic landscape" where cell fates are represented as attracting valleys resulting from a complex regulatory network. Here, we introduce a framework for explicitly constructing epigenetic landscapes that combines genomic data with techniques from spin-glass physics. Each cell fate is a dynamic attractor, yet cells can change fate in response to external signals. Our model suggests that partially reprogrammed cells are a natural consequence of high-dimensional landscapes, and predicts that partially reprogrammed cells should be hybrids that co-express genes from multiple cell fates. We verify this prediction by reanalyzing existing datasets. Our model reproduces known reprogramming protocols and identifies candidate transcription factors for reprogramming to novel cell fates, suggesting epigenetic landscapes are a powerful paradigm for understanding cellular identity.

Laurencin, C. T. and L. S. Nair (2016). "The Quest toward limb regeneration: a regenerative engineering approach." Regen Biomater **3**(2): 123-125.

The Holy Grail to address the clinical grand challenge of human limb loss is to develop innovative strategies to regrow the amputated limb. The remarkable advances in the scientific understanding of regeneration, stem cell science, material science and engineering, physics and novel surgical approaches in the past few decades have provided a regenerative tool box to face this grand challenge and address the limitations of human wound healing. Here we discuss the convergence approach put forward by the field of Regenerative Engineering to use the regenerative tool box to design and develop novel translational strategies to limb regeneration.

Leapman, R. D. (2017). "Application of EELS and EFTEM to the life sciences enabled by the contributions of Ondrej Krivanek." Ultramicroscopy **180**: 180-187.

The pioneering contributions of Ondrej Krivanek to the development of electron energy loss spectrometers, energy filters, and detectors for transmission and scanning transmission electron microscopes have provided researchers with indispensible tools across a wide range of disciplines in the physical sciences, ranging from condensed matter physics, to chemistry, mineralogy, materials science, and nanotechnology. In addition, the same instrumentation has extended its reach into the life sciences, and it is this aspect of Ondrej Krivanek's influential contributions that will be surveyed here, together with some personal recollections. Traditionally, electron microscopy has given a purely morphological view of the biological structures that compose cells and tissues. However, the availability of high-performance electron energy loss spectrometers and energy filters offers complementary information about the elemental and chemical composition at the subcellular scale. Such information has proven to be valuable for applications in cell and structural biology, microbiology, histology, pathology, and more generally in the biomedical sciences.

Lee, Y. B., et al. (2018). "Engineering spheroids potentiating cell-cell and cell-ECM interactions by self-assembly of stem cell microlayer." Biomaterials **165**: 105-120.

Numerous methods have been reported for the fabrication of 3D multi-cellular spheroids and their use in stem cell culture. Current methods typically relying on the self-assembly of trypsinized, suspended stem cells, however, show limitations with respect to cell viability, throughput, and accurate recapitulation of the natural microenvironment. In this study, we developed a new system for engineering cell spheroids by self-assembly of micro-scale monolayer of stem cells. We prepared synthetic hydrogels with the surface of chemically formed micropatterns (squares/circles with width/diameter of 200mum) on which mesenchymal stem cells isolated from human nasal turbinate tissue (hTMSCs) were selectively attached and formed a monolayer. The hydrogel is capable of thermally controlled expansion. As the temperature was decreased from 37 to 4 degrees C, the cell layer detached rapidly (<10min) and assembled to form

spheroids with consistent size (approximately 100mum) and high viability (>90%). Spheroidization was significantly delayed and occurred with reduced efficiency on circle patterns compared to square patterns. Multi-physics mapping supported that delamination of the micro-scale monolayer may be affected by stress concentrated at the corners of the square pattern. In contrast, stress was distributed symmetrically along the boundary of the circle pattern. In addition, treatment of the micro-scale monolayer with a ROCK inhibitor significantly retarded spheroidization, highlighting the importance of contraction mediated by actin stress fibers for the stable generation of spheroidal stem cell structures. Spheroids prepared from the assembly of monolayers showed higher expression, both on the mRNA and protein levels, of ECM proteins (fibronectin and laminin) and stemness markers (Oct4, Sox2, and Nanog) compared to spheroids prepared from low-attachment plates, in which trypsinized single cells are assembled. The hTMSC spheroids also presented enhanced expression levels of markers related to tri-lineage (osteogenic, chondrogenic and adipogenic) differentiation. The changes in microcellular environments and functionalities were double-confirmed by using adipose derived mesenchymal stem cells (ADSCs). This spheroid engineering technique may have versatile applications in regenerative medicine for functionally improved 3D culture and therapeutic cell delivery.

Li, Y., et al. (2017). "Engineering-derived approaches for iPSC preparation, expansion, differentiation and applications." Biofabrication **9**(3): 032001.

Remarkable achievements have been made since induced pluripotent stem cells (iPSCs) were first introduced in 2006. Compared with non-pluripotent stem cells, iPSC research faces several additional complexities, such as the choice of extracellular matrix proteins, growth and differentiation factors, as well as technical challenges related to self-renewal and directed differentiation. Overcoming these challenges requires the integration of knowledge and technologies from multiple fields including cell biology, biomaterial science, engineering, physics and medicine. Here, engineering-derived iPSC approaches are reviewed according to three aspects of iPSC studies: preparation, expansion, differentiation and applications. Engineering strategies, such as 3D systems establishment, cell-matrix mechanics and the regulation of biophysical and biochemical cues, together with engineering techniques, such as 3D scaffolds, cell microspheres and bioreactors, have been applied to iPSC studies and have generated insightful results and even mini-organs such as retinas, livers and intestines. Specific results are given to demonstrate how these approaches impact iPSC behavior, and related mechanisms are discussed. In addition, cell printing technologies are presented as an advanced engineering-derived approach since they have been applied in both iPSC studies and the construction of diverse tissues and organs. Further development and possible innovations of cell printing technologies are presented in terms of creating complex and functional iPSC-derived living tissues and organs.

Liu, L., et al. (2019). "Heterogeneous Loop Model to Infer 3D Chromosome Structures from Hi-C." Biophys J **117**(3): 613-625.

Adapting a well-established formalism in polymer physics, we develop a minimalist approach to infer three-dimensional folding of chromatin from Hi-C data. The three-dimensional chromosome structures generated from our heterogeneous loop model (HLM) are used to visualize chromosome organizations that can substantiate the measurements from fluorescence in situ hybridization, chromatin interaction analysis by paired-end tag sequencing, and RNA-seq signals. We demonstrate the utility of the HLM with several case studies. Specifically, the HLM-generated chromosome structures, which reproduce the spatial distribution of topologically associated domains from fluorescence in situ hybridization measurement, show the phase segregation between two types of topologically associated domains explicitly. We discuss the origin of cell-type-dependent gene-expression level by modeling the chromatin globules of alpha-globin and SOX2 gene loci for two different cell lines. We also use the HLM to discuss how the chromatin folding and gene-expression level of Pax6 loci, associated with mouse neural development, are modulated by interactions with two enhancers. Finally, HLM-generated structures of chromosome 19 of mouse embryonic stem cells, based on single-cell Hi-C data collected over each cell-cycle phase, visualize changes in chromosome conformation along the cell-cycle. Given a contact frequency map between chromatic loci supplied from Hi-C, HLM is a computationally efficient and versatile modeling tool to generate chromosome structures that can complement interpreting other experimental data.

Lopez-Garcia, C., et al. (2010). "Intestinal stem cell replacement follows a pattern of neutral drift." Science **330**(6005): 822-825.

With the capacity for rapid self-renewal and regeneration, the intestinal epithelium is stereotypical of stem cell-supported tissues. Yet the pattern of stem cell turnover remains in question. Applying analytical methods from population dynamics and statistical physics to an inducible genetic labeling system, we showed that clone size distributions conform to a distinctive scaling behavior at short times. This result demonstrates that intestinal stem cells form an equipotent population in which the loss of a stem cell is compensated by the multiplication of a neighbor, leading to neutral drift dynamics in which clones

expand and contract at random until they either take over the crypt or they are lost. Combined with long-term clonal fate data, we show that the rate of stem cell replacement is comparable to the cell division rate, implying that neutral drift and symmetrical cell divisions are central to stem cell homeostasis.

Lowell, S. (2013). "Getting the measure of things: the physical biology of stem cells." Development **140**(20): 4125-4128.

In July 2013, the diverse fields of biology, physics and mathematics converged to discuss 'The Physical Biology of Stem Cells', the subject of the third annual symposium of the Cambridge Stem Cell Institute, UK. Two clear themes resonated throughout the meeting: the new insights gained from advances in the acquisition and interpretation of quantitative data; and the importance of 'thinking outside the nucleus' to consider physical influences on cell fate.

MacArthur, B. D., et al. (2008). "Toward stem cell systems biology: from molecules to networks and landscapes." Cold Spring Harb Symp Quant Biol **73**: 211-215.

The last few years have seen significant advances in our understanding of the molecular mechanisms of stem-cell-fate specification. New and emerging high-throughput techniques, as well as increasingly accurate loss-of-function perturbation techniques, are allowing us to dissect the interplay among genetic, epigenetic, proteomic, and signaling mechanisms in stem-cell-fate determination with ever-increasing fidelity (Boyer et al. 2005, 2006; Ivanova et al. 2006; Loh et al. 2006; Cole et al. 2008; Jiang et al. 2008; Johnson et al. 2008; Kim et al. 2008; Liu et al. 2008; Marson et al. 2008; Mathur et al. 2008). Taken together, recent reports using these new techniques demonstrate that stem-cell-fate specification is an extremely complex process, regulated by multiple mutually interacting molecular mechanisms involving multiple regulatory feedback loops. Given this complexity and the sensitive dependence of stem cell differentiation on signaling cues from the extracellular environment, how are we best to develop a coherent quantitative understanding of stem cell fate at the systems level? One approach that we and other researchers have begun to investigate is the application of techniques derived in the computational disciplines (mathematics, physics, computer science, etc.) to problems in stem cell biology. Here, we briefly sketch a few pertinent results from the literature in this area and discuss future potential applications of computational techniques to stem cell systems biology.

Mandal, B. B., et al. (2013). "Laminar silk scaffolds for aligned tissue fabrication." Macromol Biosci **13**(1): 48-58.

3D-biomaterial scaffolds with aligned architecture are of vital importance in tissue regeneration. A generic method is demonstrated to produce aligned biomaterial scaffolds using the physics of directional ice freezing. Homogeneously aligned 3D silk scaffolds with high porosity and alignment are prepared. The method can be adapted to a wide range of polymers and is devoid of any chemical reactions, thus avoiding potential complications associated with by-products. Mechanical properties and cellular responses with chondrocytes and bone-marrow-derived hMSCs are studied, assessing survival, proliferation, and differentiation. In vivo tests suggest biocompatibility of the matrices for future tissue engineering applications, specifically in areas where high cellular alignment is needed.

Marcu, L. G. (2017). "Photons - Radiobiological issues related to the risk of second malignancies." Phys Med **42**: 213-220.

Photons are widely used in radiotherapy and while they are low LET radiation, can still pose a risk in developing second malignant neoplasms (SMN). Due to the physics of photons that allow distribution of energy outside the target volume, out-of-field irradiation is an important component of SMN risk assessment. The epidemiological evidence supporting this risk should be augmented with radiobiological justifications for a better understanding of the underlying processes. There are several factors that impact second cancer risk which can be analysed from a radiobiological perspective: age at irradiation, type of irradiated tissue, irradiated volume, treatment technique, previous irradiation/radiological investigations. Age-dependence has a radiobiological foundation given by the higher radiosensitivity of children as compared to adult patients. However, in its 2013 report, UNSCEAR advises against generalisation of the effects of childhood radiation exposure, given the fact that these effects are strongly organ dependent. Furthermore, the age-dependent radiation sensitivity has a bimodal distribution, since aging cells present an increase in the oxidative stress, which can promote premalignant cells. Non-targeted effects such as radiation-induced genomic instability, bystander or abscopal effects could also impact on the risk of SMN. Recent studies show that beside the known cellular changes, bystander effects can be manifested through increased cell proliferation, which could be a culprit for SMN development. Furthermore, new evidence on the existence of tumour-specific cancer stem cells that are long-lived and more quiescent and radioresistant than non-stem cancer cells can raise questions about their association with SMN risk.

Matai, I., et al. (2020). "Progress in 3D bioprinting technology for tissue/organ regenerative engineering." Biomaterials **226**: 119536.

Escalating cases of organ shortage and donor scarcity worldwide are alarming reminders of the need

for alternatives to allograft tissues. Within the last three decades, research efforts in the field of regenerative medicine and tissue engineering continue to address the unmet need for artificial tissues and organs for transplant. Work in the field has evolved to create what we consider a new field, Regenerative Engineering, defined as the Convergence of advanced materials science, stem cell science, physics, developmental biology and clinical translation towards the regeneration of complex tissues and organ systems. Included in the regenerative engineering paradigm is advanced manufacturing. Three-dimensional (3D) bioprinting is a promising and innovative biofabrication strategy to precisely position biologics, including living cells and extracellular matrix (ECM) components, in the prescribed 3D hierarchal organization to create artificial multi-cellular tissues/organs. In this review, we outline recent progress in several bioprinting technologies used to engineer scaffolds with requisite mechanical, structural, and biological complexity. We examine the process parameters affecting bioprinting and bioink-biomaterials and review notable studies on bioprinted skin, cardiac, bone, cartilage, liver, lung, neural, and pancreatic tissue. We also focus on other 3D bioprinting application areas including cancer research, drug testing, high-throughput screening (HTS), and organ-on-a-chip models. We also highlight the current challenges associated with the clinical translation of 3D bioprinting and conclude with the future perspective of bioprinting technology.

Mengsteab, P. Y., et al. (2016). "The past, present and future of ligament regenerative engineering." Regen Med **11**(8): 871-881.

Regenerative engineering has been defined as the convergence of Advanced Materials Sciences, Stem Cell Sciences, Physics, Developmental Biology and Clinical Translation for the regeneration of complex tissues and organ systems. Anterior cruciate ligament (ACL) reconstruction necessitates the regeneration of bone, ligament and their interface to achieve superior clinical results. In the past, the ACL has been repaired with the use of autologous and allogeneic grafts, which have their respective drawbacks. Currently, investigations on the use of biodegradable matrices to achieve knee stability and permit tissue regeneration are making promising advancements. In the future, utilizing regenerative biology cues to induce an endogenous regenerative response may aid the enhancement of clinical ACL reconstruction outcomes.

Monzel, C., et al. (2018). "Dynamic cellular phynotyping defines specific mobilization mechanisms of human hematopoietic stem and progenitor cells induced by SDF1alpha versus synthetic agents." Sci Rep **8**(1): 1841.

Efficient mobilization of hematopoietic stem and progenitor cells (HSPC) is one of the most crucial issues for harvesting an adequate amount of peripheral HSPC for successful clinical transplantation. Applying well-defined surrogate models for the bone marrow niche, live cell imaging techniques, and novel tools in statistical physics, we have quantified the functionality of two mobilization agents that have been applied in the clinic, NOX-A12 and AMD3100 (plerixafor), as compared to a naturally occurring chemokine in the bone marrow, SDF1alpha. We found that NOX-A12, an L-enantiomeric RNA oligonucleotide to SDF1, significantly reduced the adhesion of HSPC to the niche surface mediated via the CXCR4-SDF1alpha axis, and stretched the migration trajectories of the HSPC. We found that the stretching of trajectories by NOX-A12 was more prominent than that by SDF1alpha. In contrast, plerixafor exhibited no detectable interference with adhesion and migration. We also found that the deformation of HSPC induced by SDF1alpha or plerixafor was also drastically suppressed in the presence of NOX-A12. This novel technology of quantitative assessment of "dynamic phenotypes" by physical tools has therefore enabled us to define different mechanisms of function for various extrinsic factors compared to naturally occurring chemokines.

Muschler, G. F., et al. (2004). "Engineering principles of clinical cell-based tissue engineering." J Bone Joint Surg Am **86**(7): 1541-1558.

Tissue engineering is a rapidly evolving discipline that seeks to repair, replace, or regenerate specific tissues or organs by translating fundamental knowledge in physics, chemistry, and biology into practical and effective materials, devices, systems, and clinical strategies. Stem cells and progenitors that are capable of forming new tissue with one or more connective tissue phenotypes are available from many adult tissues and are defined as connective tissue progenitors. There are four major cell-based tissue-engineering strategies: (1) targeting local connective tissue progenitors where new tissue is desired, (2) transplanting autogenous connective tissue progenitors, (3) transplanting culture-expanded or modified connective tissue progenitors, and (4) transplanting fully formed tissue generated in vitro or in vivo. Stem cell function is controlled by changes in stem cell activation and self-renewal or by changes in the proliferation, migration, differentiation, or survival of the progeny of stem cell activation, the downstream progenitor cells. Three-dimensional porous scaffolds promote new tissue formation by providing a surface and void volume that promotes the attachment, migration, proliferation, and desired differentiation of connective tissue progenitors throughout the region where new tissue is needed. Critical variables in

scaffold design and function include the bulk material or materials from which it is made, the three-dimensional architecture, the surface chemistry, the mechanical properties, the initial environment in the area of the scaffold, and the late scaffold environment, which is often determined by degradation characteristics. Local presentation or delivery of bioactive molecules can change the function of connective tissue progenitors (activation, proliferation, migration, differentiation, or survival) in a manner that results in new or enhanced local tissue formation. All cells require access to substrate molecules (oxygen, glucose, and amino acids). A balance between consumption and local delivery of these substrates is needed if cells are to survive. Transplanted cells are particularly vulnerable. Theoretical calculations can be used to explore the relationships among cell density, diffusion distance, and cell viability within a graft and to design improved strategies for transplantation of connective tissue progenitors. Rational strategies for tissue engineering seek to optimize new tissue formation through the logical selection of conditions that modulate the performance of connective tissue progenitors in a graft site to produce a desired tissue. This increasingly involves strategies that combine cells, matrices, inductive stimuli, and techniques that enhance the survival and performance of local or transplanted connective tissue progenitors.

Mutlu, B. R., et al. (2018). "Oscillatory inertial focusing in infinite microchannels." Proc Natl Acad Sci U S A **115**(30): 7682-7687.

Inertial microfluidics (i.e., migration and focusing of particles in finite Reynolds number microchannel flows) is a passive, precise, and high-throughput method for microparticle manipulation and sorting. Therefore, it has been utilized in numerous biomedical applications including phenotypic cell screening, blood fractionation, and rare-cell isolation. Nonetheless, the applications of this technology have been limited to larger bioparticles such as blood cells, circulating tumor cells, and stem cells, because smaller particles require drastically longer channels for inertial focusing, which increases the pressure requirement and the footprint of the device to the extent that the system becomes unfeasible. Inertial manipulation of smaller bioparticles such as fungi, bacteria, viruses, and other pathogens or blood components such as platelets and exosomes is of significant interest. Here, we show that using oscillatory microfluidics, inertial focusing in practically "infinite channels" can be achieved, allowing for focusing of micron-scale (i.e. hundreds of nanometers) particles. This method enables manipulation of particles at extremely low particle Reynolds number (Rep < 0.005) flows that are otherwise unattainable by steady-flow inertial microfluidics (which has been limited to Rep > approximately 10(-1)). Using this technique, we demonstrated that synthetic particles as small as 500 nm and a submicron bacterium, Staphylococcus aureus, can be inertially focused. Furthermore, we characterized the physics of inertial microfluidics in this newly enabled particle size and Rep range using a Peclet-like dimensionless number (alpha). We experimentally observed that alpha >> 1 is required to overcome diffusion and be able to inertially manipulate particles.

Nelson, C., et al. (2018). "Nanofiber/Microsphere Hybrid Matrices In Vivo for Bone Regenerative Engineering: A Preliminary Report." Regen Eng Transl Med **4**(3): 133-141.

The demand for bone grafts has led to advances in regenerative engineering, a field at the intersection of advanced biomaterials, stem cell science, physics, developmental biology, and clinical translation. In this work, the authors evaluated a hybrid nanofiber/microsphere matrices both in vitro and in vivo for its ability to promote bone regeneration. Quantitative measures of cellular characteristics in vitro showed a higher fraction of marrow stromal cells with collagen promoter activity on hybrid matrices compared to control matrices (41% vs. 24%, p = 0.02). Control and hybrid matrices were then implanted for 6 weeks in calvarial defects of mice, and the animals received a single injection of calcein 1 day prior to sacrifice to visualize bone formation. Cryohistology of the undecalfied implants were evaluated for markers of bone mineralization, which revealed evidence of higher levels of bone tissue formation in hybrid matrices compared to controls. These data provide support that nanofiber-permeated, sintered, composite microsphere matrices may be a particularly useful matrix for the regenerative engineering of bone.

Ogueri, K. S., et al. (2019). "Generational Biodegradable and Regenerative Polyphosphazene Polymers and their Blends with Poly (lactic-co-glycolic acid)." Prog Polym Sci **98**.

New fields such as regenerative engineering have driven the design of advanced biomaterials with a wide range of properties. Regenerative engineering is a multidisciplinary approach that integrates the fields of advanced materials science and engineering, stem cell science, physics, developmental biology, and clinical translation for the regeneration of complex tissues. The complexity and demands of this innovative approach have motivated the synthesis of new polymeric materials that can be customized to meet application-specific needs. Polyphosphazene polymers represent this fundamental change and are gaining renewed interest as biomaterials due to their outstanding synthetic flexibility, neutral bioactivity (buffering degradation products), and tunable properties across the range. Polyphosphazenes are a unique class of

polymers composed of an inorganic backbone with alternating phosphorus and nitrogen atoms. Each phosphorus atom bears two substituents, with a wide variety of side groups available for property optimization. Polyphosphazenes have been investigated as potential biomaterials for regenerative engineering. Polyphosphazenes for use in regenerative applications have evolved as a class to include different generations of degradable polymers. The first generation of polyphosphazenes for tissue regeneration entailed the use of hydrolytically active side groups such as imidazole, lactate, glycolate, glucosyl, or glyceryl groups. These side groups were selected based on their ability to sensitize the polymer backbone to hydrolysis, which allowed them to break down into non-toxic small molecules that could be metabolized or excreted. The second generation of degradable polyphosphazenes developed consisted of polymers with amino acid ester side groups. When blended with poly (lactic acid-co-glycolic acid) (PLGA), the feasibility of neutralizing acidic degradation products of PLGA was demonstrated. The blends formed were mostly partially miscible. The desire to improve miscibility led to the design of the third generation of degradable polyphosphazenes by incorporating dipeptide side groups which impart significant hydrogen bonding capability to the polymer for the formation of completely miscible polyphosphazene-PLGA blends. Blend system of the dipeptide-based polyphosphazene and PLGA exhibit a unique degradation behavior that allows the formation of interconnected porous structures upon degradation.

Ottolenghi, A., et al. (2015). "The ANDANTE project: a multidisciplinary approach to neutron RBE." Radiat Prot Dosimetry **166**(1-4): 311-315.

UNLABELLED: The usual method for estimating the risk from exposure to neutrons uses the concept of relative biological effectiveness (RBE) compared with the risk from photons, which is better known. RBE has been evaluated using cellular and animal models. But this causes difficulties in applying the concept to humans. The ANDANTE project takes a new approach using three different disciplines in parallel: Physics: a track structure model is used to contrast the patterns of damage to cellular macro-molecules from neutrons compared with photons. The simulations reproduce the same energy spectra as are used in the other two approaches. Stem cell radiobiology: stem cells from thyroid, salivary gland and breast tissue are given well characterised exposures to neutrons and photons. A number of endpoints are used to estimate the relative risk of damage from neutrons compared with photons. Irradiated cells will also be transplanted into mice to investigate the progression of the initial radiation effects in stem cells into tumours in a physiological environment. EPIDEMIOLOGY: the relative incidence rates of second cancers of the thyroid, salivary gland and breast following paediatric radiotherapy (conventional radiotherapy for photons and proton therapy for neutrons) are investigated in a pilot single-institution study, exploring the possible design of a multi-institution prospective study comparing the long-term out-of-field and in-field effects of scanned and scattered protons. The results will be used to validate an RBE-based risk model developed by the project, and validate the corresponding RBE values.

Park, J. Y., et al. (2009). "Simultaneous generation of chemical concentration and mechanical shear stress gradients using microfluidic osmotic flow comparable to interstitial flow." Lab Chip **9**(15): 2194-2202.

Cells are very sensitive to various microenvironmental cues, including mechanical stress and chemical gradients. Therefore, physiologically relevant models of cells should consider how cells sense and respond to microenvironmental cues. This can be accomplished by using microfluidic systems, in which fluid physics can be realized at a nanoliter scale. Here we describe a simple and versatile method to study the generation of chemical concentration and mechanical shear stress gradients in a single microfluidic chip. Our system uses an osmotic pump that produces very slow (<a few microm/s) and controlled flow, allowing a wide and stable diffusion of specific chemical concentration. We also established a shear stress gradient passively via a circular channel in the interstitial level. For evaluation of the system, we used L929 mouse fibroblast cells and simultaneously exposed them to a mechanical stress gradient and a chemical nutrient gradient. The interstitial shear stress level clearly affected cell alignment, mobility velocity, and attachment. At the same time, cell proliferation reflected nutrient concentration level. Our system, which enables continuous and long-term culture of cells in a combined chemical and mechanical gradient, provides physiologically realistic conditions and will be applicable to studies of cancer metastasis and stem cell differentiation.

Pena-Duarte, A., et al. (2021). "Iron Quantum Dots Electro-Assembling on Vulcan XC-72R: Hydrogen Peroxide Generation for Space Applications." ACS Appl Mater Interfaces **13**(25): 29585-29601.

Highly dispersed iron-based quantum dots (QDs) onto powdered Vulcan XC-72R substrate were successfully electrodeposited by the rotating disk slurry electrodeposition (RoDSE) technique. Our findings through chemical physics characterization revealed that the continuous electron pathway interaction between the interface metal-carbon is controlled. The rotating ring-disk electrode (RRDE) and the prototype generation unit (PGU) of in-situ H2O2 generation in fuel cell experiments revealed a high activity for the

oxygen reduction reaction (ORR) via two-electron pathway. These results establish the Fe/Vulcan catalyst at a competitive level for space and terrestrial new materials carriers, specifically for the in-situ H2O2 production. Transmission electron microscopy (TEM) analysis reveals the well-dispersed Fe-based quantum dots with a particle size of 4 nm. The structural and chemical-physical characterization through induced coupled plasma-optical emission spectroscopy (ICP-OES), transmission scanning electron microscopy (STEM), X-ray diffraction (XRD), Raman spectroscopy, X-ray photoelectron spectroscopy (XPS), and X-ray absorption spectroscopy (XAS); reveals that, under atmospheric conditions, our quantum dots system is a Fe(2+/3+)/Fe(3+) combination. The QDs oxidation state tunability was showed by the applied potential. The obtention of H2O2 under the compatibility conditions of the drinking water resources available in the International Space Station (ISS) enhances the applicability of this iron- and carbon-based materials for in-situ H2O2 production in future space scenarios.

Potten, C. S. and M. Loeffler (1990). "Stem cells: attributes, cycles, spirals, pitfalls and uncertainties. Lessons for and from the crypt." Development **110**(4): 1001-1020.

We consider some of the problems involved in current discussions on stem cells in adult mammalian tissues. The present concepts involve a number of pitfalls, weaknesses and logical, semantic and classification problems. This indicates the necessity for new and well-defined concepts that are amenable to experimental analysis. One of the major difficulties in considering stem cells is that they are defined in terms of their functional capabilities which can only be assessed by testing the abilities of the cells, which itself may alter their characteristics during the assay procedure: a situation similar to the uncertainty principle in physics. The terms that describe stem cell functions are often not well defined and are used loosely, which can lead to confusion. If such context-dependent interactions exist between the manipulation and measurement process and the challenged stem cells, the question of, for example, the number of stem cells, in a tissue has to be posed in a new way. Rather than obtaining a single number one might end up with various different numbers under different circumstances, all being complementary. This might suggest that stemness is not a property but a spectrum of capabilities from which to choose. This concept might facilitate a reconciliation between the different and sometimes opposing experimental results. Given certain experimental evidence, we have attempted to provide a novel concept to describe structured cell populations in tissues involving stem cells, transit cells and mature cells. It is based on the primary assumption that the proliferation and differentiation/maturation processes are in principle independent entities in the sense that each may proceed without necessarily affecting the other. Stem cells may divide without maturation while cells approaching functional competence may mature but do not divide. In contrast, transit cells divide and mature showing intermediate properties between stem cells and mature functional cells. The need to describe this transition process and the variable coupling between proliferation and maturation leads us to formulate a spiral model of cell and tissue organisation. This concept is illustrated for the intestinal epithelium. It is concluded that the small intestinal crypts contain 4-16 actual stem cells in steady state but up to 30-40 potential stem cells (clonogenic cells) which may take over stem cell properties following perturbations. This implies that transit cells can under certain circumstances behave like actual stem cells while they undergo maturation under other conditions. There is also evidence that the proliferation and differentiation/maturation processes are subject to controls that ultimately lead to a change in the spiral trajectories.(ABSTRACT TRUNCATED AT 400 WORDS)

Reichenbach, T. and A. J. Hudspeth (2014). "The physics of hearing: fluid mechanics and the active process of the inner ear." Rep Prog Phys **77**(7): 076601.

Most sounds of interest consist of complex, time-dependent admixtures of tones of diverse frequencies and variable amplitudes. To detect and process these signals, the ear employs a highly nonlinear, adaptive, real-time spectral analyzer: the cochlea. Sound excites vibration of the eardrum and the three miniscule bones of the middle ear, the last of which acts as a piston to initiate oscillatory pressure changes within the liquid-filled chambers of the cochlea. The basilar membrane, an elastic band spiraling along the cochlea between two of these chambers, responds to these pressures by conducting a largely independent traveling wave for each frequency component of the input. Because the basilar membrane is graded in mass and stiffness along its length, however, each traveling wave grows in magnitude and decreases in wavelength until it peaks at a specific, frequency-dependent position: low frequencies propagate to the cochlear apex, whereas high frequencies culminate at the base. The oscillations of the basilar membrane deflect hair bundles, the mechanically sensitive organelles of the ear's sensory receptors, the hair cells. As mechanically sensitive ion channels open and close, each hair cell responds with an electrical signal that is chemically transmitted to an afferent nerve fiber and thence into the brain. In addition to transducing mechanical inputs, hair cells amplify them by two means. Channel gating endows a

hair bundle with negative stiffness, an instability that interacts with the motor protein myosin-1c to produce a mechanical amplifier and oscillator. Acting through the piezoelectric membrane protein prestin, electrical responses also cause outer hair cells to elongate and shorten, thus pumping energy into the basilar membrane's movements. The two forms of motility constitute an active process that amplifies mechanical inputs, sharpens frequency discrimination, and confers a compressive nonlinearity on responsiveness. These features arise because the active process operates near a Hopf bifurcation, the generic properties of which explain several key features of hearing. Moreover, when the gain of the active process rises sufficiently in ultraquiet circumstances, the system traverses the bifurcation and even a normal ear actually emits sound. The remarkable properties of hearing thus stem from the propagation of traveling waves on a nonlinear and excitable medium.

Rigaud, S., et al. (2013). "An analysis-synthesis approach for neurosphere modelisation under phase-contrast microscopy." Annu Int Conf IEEE Eng Med Biol Soc **2013**: 3989-3992.

The study of stem cells is one of the most important biomedical research. Understanding their development could allow multiple applications in regenerative medicine. For this purpose, automated solutions for the observation of stem cell development process are needed. This study introduces an on-line analysis method for the modelling of neurosphere evolution during the early time of their development under phase contrast microscopy. From the corresponding phase contrast time-lapse sequences, we extract information from the neurosphere using a combination of phase contrast physics deconvolution and curve detection for locate the cells inside the neurosphere. Then, based on prior biological knowledge, we generate possible and optimal 3-dimensional configuration using 2D to 3D registration methods and evolutionary optimisation algorithm.

Russell, N. S. and H. Bartelink (1992). "Optimized radiotherapy for head and neck cancer." Curr Opin Oncol **4**(3): 491-498.

Optimization of radiation therapy for head and neck tumors requires the combination of several facets of radiation biology and physics. The aim is to achieve optimum tumor control while reducing normal tissue damage. Techniques have been developed to determine tumor radiosensitivity and growth characteristics. Their use as predictive assays of treatment response is gaining importance. As the range of therapeutic options (particularly altered fractionation regimens) increases, it is hoped that the ability to individually tailor patients' treatment will result in improved rates of tumor control and an improved therapeutic ratio. Optimization of treatment delivery based on three-dimensional treatment planning offers the opportunity for dose escalation studies and limitation of normal tissue morbidity. The combination of chemotherapy and radiotherapy continues to be investigated, although major advances using this strategy are unlikely.

Sabloff, M., et al. (2021). "Total Body Irradiation for Hematopoietic Stem Cell Transplantation: What Can We Agree on?" Curr Oncol **28**(1): 903-917.

Total body irradiation (TBI), used as part of the conditioning regimen prior to allogeneic and autologous hematopoietic cell transplantation, is the delivery of a relatively homogeneous dose of radiation to the entire body. TBI has a dual role, being cytotoxic and immunosuppressive. This allows it to eliminate disease and create "space" in the marrow while also impairing the immune system from rejecting the foreign donor cells being transplanted. Advantages that TBI may have over chemotherapy alone are that it may achieve greater tumour cytotoxicity and better tissue penetration than chemotherapy as its delivery is independent of vascular supply and physiologic barriers such as renal and hepatic function. Therefore, the so-called "sanctuary" sites such as the central nervous system (CNS), testes, and orbits or other sites with limited blood supply are not off-limits to radiation. Nevertheless, TBI is hampered by challenging logistics of administration, coordination between hematology and radiation oncology departments, increased rates of acute treatment-related morbidity and mortality along with late toxicity to other tissues.

Sachinidis, A., et al. (2008). "A chemical genetics approach for specific differentiation of stem cells to somatic cells: a new promising therapeutical approach." Comb Chem High Throughput Screen **11**(1): 70-82.

Cell replacement therapy of severe degenerative diseases such as diabetes, myocardial infarction and Parkinson's disease through transplantation of somatic cells generated from embryonic stem (ES) cells is currently receiving considerable attention for the therapeutic applications. ES cells harvested from the inner cell mass (ICM) of the early embryo, can proliferate indefinitely in vitro while retaining the ability to differentiate into all somatic cells thereby providing an unlimited renewable source of somatic cells. In this context, identifying soluble factors, in particular chemically synthesized small molecules, and signal cascades involved in specific differentiation processes toward a defined tissue specific cell type are crucial for optimizing the generation of somatic cells in vitro for therapeutic approaches. However, experimental models are required allowing rapid and "easy-to-handle" parallel screening of chemical libraries to achieve this goal. Recently, the forward chemical genetic screening

strategy has been postulated to screen small molecules in cellular systems for a specific desired phenotypic effect. The current review is focused on the progress of ES cell research in the context of the chemical genetics to identify small molecules promoting specific differentiation of ES cells to desired cell phenotype. Chemical genetics in the context of the cell ES-based cell replacement therapy remains a challenge for the near future for several scientific fields including chemistry, molecular biology, medicinal physics and robotic technologies.

Saritas, E. U., et al. (2013). "Magnetic particle imaging (MPI) for NMR and MRI researchers." J Magn Reson **229**: 116-126.

Magnetic Particle Imaging (MPI) is a new tracer imaging modality that is gaining significant interest from NMR and MRI researchers. While the physics of MPI differ substantially from MRI, it employs hardware and imaging concepts that are familiar to MRI researchers, such as magnetic excitation and detection, pulse sequences, and relaxation effects. Furthermore, MPI employs the same superparamagnetic iron oxide (SPIO) contrast agents that are sometimes used for MR angiography and are often used for MRI cell tracking studies. These SPIOs are much safer for humans than iodine or gadolinium, especially for Chronic Kidney Disease (CKD) patients. The weak kidneys of CKD patients cannot safely excrete iodine or gadolinium, leading to increased morbidity and mortality after iodinated X-ray or CT angiograms, or after gadolinium-MRA studies. Iron oxides, on the other hand, are processed in the liver, and have been shown to be safe even for CKD patients. Unlike the "black blood" contrast generated by SPIOs in MRI due to increased T2* dephasing, SPIOs in MPI generate positive, "bright blood" contrast. With this ideal contrast, even prototype MPI scanners can already achieve fast, high-sensitivity, and high-contrast angiograms with millimeter-scale resolutions in phantoms and in animals. Moreover, MPI shows great potential for an exciting array of applications, including stem cell tracking in vivo, first-pass contrast studies to diagnose or stage cancer, and inflammation imaging in vivo. So far, only a handful of prototype small-animal MPI scanners have been constructed worldwide. Hence, MPI is open to great advances, especially in hardware, pulse sequence, and nanoparticle improvements, with the potential to revolutionize the biomedical imaging field.

Schiermeier, Q. and M. Leeb (2004). "Critics blast 'premature' paper on adult stem cells." Nature **429**(6992): 590.

Segal-Peretz, T., et al. (2015). "Characterizing the Three-Dimensional Structure of Block Copolymers via Sequential Infiltration Synthesis and Scanning Transmission Electron Tomography." ACS Nano **9**(5): 5333-5347.

Understanding and controlling the three-dimensional structure of block copolymer (BCP) thin films is critical for utilizing these materials for sub-20 nm nanopatterning in semiconductor devices, as well as in membranes and solar cell applications. Combining an atomic layer deposition (ALD)-based technique for enhancing the contrast of BCPs in transmission electron microscopy (TEM) together with scanning TEM (STEM) tomography reveals and characterizes the three-dimensional structures of poly(styrene-block-methyl methacrylate) (PS-b-PMMA) thin films with great clarity. Sequential infiltration synthesis (SIS), a block-selective technique for growing inorganic materials in BCPs films in an ALD tool and an emerging technique for enhancing the etch contrast of BCPs, was harnessed to significantly enhance the high-angle scattering from the polar domains of BCP films in the TEM. The power of combining SIS and STEM tomography for three-dimensional (3D) characterization of BCP films was demonstrated with the following cases: self-assembled cylindrical, lamellar, and spherical PS-b-PMMA thin films. In all cases, STEM tomography has revealed 3D structures that were hidden underneath the surface, including (1) the 3D structure of defects in cylindrical and lamellar phases, (2) the nonperpendicular 3D surface of grain boundaries in the cylindrical phase, and (3) the 3D arrangement of spheres in body-centered-cubic (BCC) and hexagonal-closed-pack (HCP) morphologies in the spherical phase. The 3D data of the spherical morphologies was compared to coarse-grained simulations and assisted in validating the simulations' parameters. STEM tomography of SIS-treated BCP films enables the characterization of the exact structure used for pattern transfer and can lead to a better understating of the physics that is utilized in BCP lithography.

Shen, D., et al. (2007). "Coregistration of magnetic resonance and single photon emission computed tomography images for noninvasive localization of stem cells grafted in the infarcted rat myocardium." Mol Imaging Biol **9**(1): 24-31.

This paper demonstrates the application of mutual information based coregistration of radionuclide and magnetic resonance imaging (MRI) in an effort to use multimodality imaging for noninvasive localization of stem cells grafted in the infarcted myocardium in rats. Radionuclide imaging such as single photon emission computed tomography (SPECT) or positron emission tomography (PET) inherently has high sensitivity and is suitable for tracking of labeled stem cells, while high-resolution MRI is able to provide detailed anatomical and functional information of myocardium. Thus,

coregistration of PET or SPECT images with MRI will map the location and distribution of stem cells on detailed myocardium structures. To validate this coregistration method, SPECT data were simulated by using a Monte Carlo-based projector that modeled the pinhole-imaging physics assuming nonzero diameter and photon penetration at the edge. Translational and rotational errors of the coregistration were examined with respect to various SPECT activities, and they are on average about 0.50 mm and 0.82 degrees , respectively. Only the rotational error is dependent on activity of SPECT data. Stem cells were labeled with (111)Indium oxyquinoline and grafted in the ischemic myocardium of a rat model. Dual-tracer small-animal SPECT images were acquired, which allowed simultaneous detection of (111)In-labeled stem cells and of [(99m)Tc]sestamibi to assess myocardial perfusion deficit. The same animals were subjected to cardiac MRI. A mutual-information-based coregistration method was then applied to the SPECT and MRIs. By coregistration, the (111)In signal from labeled cells was mapped into the akinetic region identified on cine MRIs; the regional perfusion deficit on the SPECT images also coincided with the akinetic region on the MR image.

Siregar, P., et al. (2018). "A general framework dedicated to computational morphogenesis Part I - Constitutive equations." Biosystems **173**: 298-313.

In order to understand living organisms, considerable experimental efforts and resources have been devoted to correlate genes and their expressions with cell, tissue, organ and whole organisms' phenotypes. This data driven approach to knowledge discovery has led to many breakthrough in our understanding of healthy and diseased states, and is paving the way to improve the diagnosis and treatment of diseases. Complementary to this data-driven approach, computational models of biological systems based on first principles have been developed in order to deepen our understanding of the multi-scale dynamics that drives normal and pathological biological functions. In this paper we describe the biological, physical and mathematical concepts that led to the design of a Computational Morphogenesis (CM) platform baptized Generic Modeling and Simulating Platform (GMSP). Its role is to generate realistic 3D multi-scale biological tissues from virtual stem cells and the intended target applications include in virtuo studies of normal and abnormal tissue (re)generation as well as the development of complex diseases such as carcinogenesis. At all space-scales of interest, biological agents interact with each other via biochemical, bioelectrical, and mechanical fields that operate in concert during embryogenesis, growth and adult life. The spatio-temporal dependencies of these fields can be modeled by physics-based constitutive equations that we propose to examine in relation to the landmark biological events that occur during embryogenesis.

Strinkovsky, L., et al. (2021). "Spatial correlations constrain cellular lifespan and pattern formation in corneal epithelium homeostasis." Elife **10**.

Homeostasis in adult tissues relies on the replication dynamics of stem cells, their progenitors and the spatial balance between them. This spatial and kinetic coordination is crucial to the successful maintenance of tissue size and its replenishment with new cells. However, our understanding of the role of cellular replicative lifespan and spatial correlation between cells in shaping tissue integrity is still lacking. We developed a mathematical model for the stochastic spatial dynamics that underlie the rejuvenation of corneal epithelium. Our model takes into account different spatial correlations between cell replication and cell removal. We derive the tradeoffs between replicative lifespan, spatial correlation length, and tissue rejuvenation dynamics. We determine the conditions that allow homeostasis and are consistent with biological timescales, pattern formation, and mutants phenotypes. Our results can be extended to any cellular system in which spatial homeostasis is maintained through cell replication.

Su, L., et al. (2019). "Emerging progress on the mechanism and technology in wound repair." Biomed Pharmacother **117**: 109191.

Normal wound repair is a dynamic and complex process involving multiple coordinated interactions between growth factors, cytokines, chemokines, and various cells. Any failure during the repair process may cause chronic wounds or scar formation, which increase the financial burden of patients due to repetitive treatments and considerable medical expenditures, and affect their quality of life. Nowadays, extensive efforts have been made to develop novel therapeutics for wound repair. Genetic engineering technology, tissue engineering technology, stem cell-based therapy, physical and biochemical technology, and vacuum-assisted closure technique have been proposed to be beneficial for wound repair, and shown considerable potential for improving the rate and quality of wound healing and skin regeneration. However, challenges remain as applying these techniques. As the development of cell biology and molecular biology, the understanding of the mechanism under wound repair has gradually deepened. As the growth of interdisciplinary research on physics, chemistry, biology, tissue engineering, and materials, the concept and technique relating wound repair for clinical application have rapidly developed. This article reviews the latest progress on the mechanism and technique in wound repair.

Tamrin, S. H., et al. (2016). "Electromagnetic Fields and Stem Cell Fate: When Physics Meets Biology." Rev Physiol Biochem Pharmacol **171**: 63-97.

Controlling stem cell (SC) fate is an extremely important topic in the realm of SC research. A variety of different external cues mainly mechanical, chemical, or electrical stimulations individually or in combination have been incorporated to control SC fate. Here, we will deconstruct the probable relationship between the functioning of electromagnetic (EMF) and SC fate of a variety of different SCs. The electromagnetic (EM) nature of the cells is discussed with the emphasis on the effects of EMF on the determinant factors that directly and/or indirectly influence cell fate. Based on the EM effects on a variety of cellular processes, it is believed that EMFs can be engineered to provide a controlled signal with the highest impact on the SC fate decision. Considering the novelty and broad applications of applying EMFs to change SC fate, it is necessary to shed light on many unclear mechanisms underlying this phenomenon.

Tanyeri, M. and S. Tay (2018). "Viable cell culture in PDMS-based microfluidic devices." Methods Cell Biol **148**: 3-33.

Microfluidics has played a vital role in developing novel methods to investigate biological phenomena at the molecular and cellular level during the last two decades. Microscale engineering of cellular systems is nevertheless a nascent field marked inherently by frequent disruptive advancements in technology such as PDMS-based soft lithography. Viable culture and manipulation of cells in microfluidic devices requires knowledge across multiple disciplines including molecular and cellular biology, chemistry, physics, and engineering. There has been numerous excellent reviews in the past 15 years on applications of microfluidics for molecular and cellular biology including microfluidic cell culture (Berthier et al., 2012; El-Ali, Sorger, & Jensen, 2006; Halldorsson et al., 2015; Kim et al., 2007; Mehling & Tay, 2014; Sackmann et al., 2014; Whitesides, 2006; Young & Beebe, 2010), cell culture models (Gupta et al., 2016; Inamdar & Borenstein, 2011; Meyvantsson & Beebe, 2008), cell secretion (Schrell et al., 2016), chemotaxis (Kim & Wu, 2012; Wu et al., 2013), neuron culture (Millet & Gillette, 2012a, 2012b), drug screening (Dittrich & Manz, 2006; Eribol, Uguz, & Ulgen, 2016; Wu, Huang, & Lee, 2010), cell sorting (Autebert et al., 2012; Bhagat et al., 2010; Gossett et al., 2010; Wyatt Shields Iv, Reyes, & Lopez, 2015), single cell studies (Lecault et al., 2012; Reece et al., 2016; Yin & Marshall, 2012), stem cell biology (Burdick & Vunjak-Novakovic, 2009; Wu et al., 2011; Zhang & Austin, 2012), cell differentiation (Zhang et al., 2017a), systems biology (Breslauer, Lee, & Lee, 2006), 3D cell culture (Huh et al., 2011; Li et al., 2012; van Duinen et al., 2015), spheroids and organoids (Lee et al., 2016; Montanez-Sauri, Beebe, & Sung, 2015; Morimoto & Takeuchi, 2013; Skardal et al., 2016; Young, 2013), organ-on-chip (Bhatia & Ingber, 2014; Esch, Bahinski, & Huh, 2015; Huh et al., 2011; van der Meer & van den Berg, 2012), and tissue engineering (Andersson & Van Den Berg, 2004; Choi et al., 2007; Hasan et al., 2014). In this chapter, we provide an overview of PDMS-based microdevices for microfluidic cell culture. We discuss the advantages and challenges of using PDMS-based soft lithography for microfluidic cell culture and highlight recent progress and future directions in this area.

Teh, B. S., et al. (2008). "[Recent developments in radiation oncology-integrating radiation physics and molecular radiobiology advances into clinical radiotherapy practice and beyond]." Ai Zheng **27**(8): 885-893.

Significant developments in radiation oncology have taken place in recent years as a result of advances in radiation physics and molecular radiobiology. From the conventional 2-dimensional (2D) radiotherapy to 3-dimensional (3D) conformal radiotherapy, we have now entered the era of intensity-modulated radiotherapy (IMRT) and image-guided radiotherapy (IGRT). IMRT/IGRT allows conformal treatment of tumor and conformal avoidance of normal tissues leading to possible improvement of tumor control and decrease in treatment-related toxicity. Frameless stereotactic radiosurgery (SRS) and stereotactic body radiotherapy (SBRT) have now become a reality, offering more treatment options in radiation oncology. With technological advances in image guidance, brachytherapy especially in early stage prostate cancer has progressed and shown excellent long-term outcome data. Charged particle therapy including proton therapy is a promising area for new development. Combining radiotherapy with the more traditional chemotherapy and hormonal therapy to novel targeted therapy and gene therapy is aimed to overcome radio-resistance, improve the radio-therapeutic index and provide better loco-regional and systemic control of cancer. A recent randomized trial in head and neck cancer has shown improved survival data when comparing combined radiotherapy and targeted therapy with radiotherapy alone. Recent advances in functional or molecular imaging offer new opportunity to improve targeting of tumor, for example, hypoxic region, and possibly to perform radiation dose painting with IMRT. Integrating PET/CT in radiotherapy has shown promise in assisting target delineation during treatment planning and assessing radiation treatment response. Cancer stem cell, gene expression profiling and nanotechnology with the implications on radio-resistance are new

exciting areas requiring more research in future as we move toward personalized medicine.

Thacker, V. V., et al. (2020). "A lung-on-chip model of early Mycobacterium tuberculosis infection reveals an essential role for alveolar epithelial cells in controlling bacterial growth." Elife **9**.

We establish a murine lung-on-chip infection model and use time-lapse imaging to reveal the dynamics of host-Mycobacterium tuberculosis interactions at an air-liquid interface with a spatiotemporal resolution unattainable in animal models and to probe the direct role of pulmonary surfactant in early infection. Surfactant deficiency results in rapid and uncontrolled bacterial growth in both macrophages and alveolar epithelial cells. In contrast, under normal surfactant levels, a significant fraction of intracellular bacteria are non-growing. The surfactant-deficient phenotype is rescued by exogenous addition of surfactant replacement formulations, which have no effect on bacterial viability in the absence of host cells. Surfactant partially removes virulence-associated lipids and proteins from the bacterial cell surface. Consistent with this mechanism, the attenuation of bacteria lacking the ESX-1 secretion system is independent of surfactant levels. These findings may partly explain why smokers and elderly persons with compromised surfactant function are at increased risk of developing active tuberculosis.

Tuberculosis is a contagious respiratory disease caused by the bacterium Mycobacterium tuberculosis. Droplets in the air carry these bacteria deep into the lungs, where they cling onto and infect lung cells. Only small droplets, holding one or two bacteria, can reach the right cells, which means that just a couple of bacterial cells can trigger an infection. But people respond differently to the bacteria: some develop active and fatal forms of tuberculosis, while many show no signs of infection. With no effective tuberculosis vaccine for adults, understanding why individuals respond differently to Mycobacterium tuberculosis may help develop treatments. Different responses to Mycobacterium tuberculosis may stem from the earliest stages of infection, but these stages are difficult to study. For one thing, tracking the movements of the few bacterial cells that initiate infection is tricky. For another, studying the molecules, called 'surfactants', that the lungs produce to protect themselves from tuberculosis can prove difficult because these molecules are necessary for the lungs to inflate and deflate normally. Normally, the role of a molecule can be studied by genetically modifying an animal so it does not produce the molecule in question, which provides information as to its potential roles. Unfortunately, due to the role of surfactants in normal breathing, animals lacking them die. Therefore, to reveal the role of some of surfactants in tuberculosis, Thacker et al. used 'lung-on-chip' technology. The 'chip' (a transparent device made of a polymer compatible with biological tissues) is coated with layers of cells and has channels to simulate air and blood flow. To see what effects surfactants have on M. tuberculosis bacteria, Thacker et al. altered the levels of surfactants produced by the cells on the lung-on-chip device. Two types of mouse cells were grown on the chip: lung cells and immune cells. When cells lacked surfactants, bacteria grew rapidly on both lung and immune cells, but when surfactants were present bacteria grew much slower on both cell types, or did not grow at all. Further probing showed that the surfactants pulled out proteins and fats on the surface of M. tuberculosis that help the bacteria to infect their host, highlighting the protective role of surfactants in tuberculosis. These findings lay the foundations for a system to study respiratory infections without using animals. This will allow scientists to study the early stages of Mycobacterium tuberculosis infection, which is crucial for finding ways to manage tuberculosis.

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Tong, W. F., et al. (2002). "Somatic cell nuclear transfer (cloning): implications for the medical practitioner." Singapore Med J **43**(7): 369-376.

The current century will bring tremendous changes to the science and the practice of medicine. This century will be acknowledged as the century of Biology as the fusion of molecular genetics and experimental embryology pushes the barriers of science beyond perimeters that we have thought existed, as much as the past century was the century of Physics, with all the exact scientific calculations and predictions, resulting in electricity, nuclear power and quantum physics. The first major breakthrough has been the pioneering work of Wilmut and Campbell, first with the birth of Megan and Moran in 1995 (1), followed by the birth of Dolly the sheep, the first reported mammalian clone from a fully differentiated adult cell, reported in July 1996 (2). However, current cloning techniques are an extension of over 40 years of research using nuclei derived from non-human embryonic and fetal cells. However, following the birth of Dolly, the prospects of cloning technology have extended to ethically hazier areas of human cloning and embryonic stem cell research. This review hopes to bring the reader closer to the science and the ethics of this new technology, and what the implications are for the medical practitioner.

Urbakh, M., et al. (2004). "The nonlinear nature of friction." Nature **430**(6999): 525-528.

Tribology is the study of adhesion, friction, lubrication and wear of surfaces in relative motion. It remains as important today as it was in ancient times, arising in the fields of physics, chemistry, geology, biology and engineering. The more we learn about

tribology the more complex it appears. Nevertheless, recent experiments coupled to theoretical modelling have made great advances in unifying apparently diverse phenomena and revealed many subtle and often non-intuitive aspects of matter in motion, which stem from the nonlinear nature of the problem.

Wallner, P. E., et al. (2014). "Current status and recommendations for the future of research, teaching, and testing in the biological sciences of radiation oncology: report of the American Society for Radiation Oncology Cancer Biology/Radiation Biology Task Force, executive summary." Int J Radiat Oncol Biol Phys **88**(1): 11-17.

In early 2011, a dialogue was initiated within the Board of Directors (BOD) of the American Society for Radiation Oncology (ASTRO) regarding the future of the basic sciences of the specialty, primarily focused on the current state and potential future direction of basic research within radiation oncology. After consideration of the complexity of the issues involved and the precise nature of the undertaking, in August 2011, the BOD empanelled a Cancer Biology/Radiation Biology Task Force (TF). The TF was charged with developing an accurate snapshot of the current state of basic (preclinical) research in radiation oncology from the perspective of relevance to the modern clinical practice of radiation oncology as well as the education of our trainees and attending physicians in the biological sciences. The TF was further charged with making suggestions as to critical areas of biological basic research investigation that might be most likely to maintain and build further the scientific foundation and vitality of radiation oncology as an independent and vibrant medical specialty. It was not within the scope of service of the TF to consider the quality of ongoing research efforts within the broader radiation oncology space, to presume to consider their future potential, or to discourage in any way the investigators committed to areas of interest other than those targeted. The TF charge specifically precluded consideration of research issues related to technology, physics, or clinical investigations. This document represents an Executive Summary of the Task Force report.

Wang, Q., et al. (2020). "Artificially Engineered Cubic Iron Oxide Nanoparticle as a High-Performance Magnetic Particle Imaging Tracer for Stem Cell Tracking." ACS Nano **14**(2): 2053-2062.

Stem cell therapies are increasingly recognized as the future direction of regenerative medicine, but the biological fate of the administrated stem cells remains a major concern for clinical translation, which calls for an approach to efficiently monitoring the stem cell behaviors in vivo. Magnetic particle imaging (MPI) is an emerging technology for cell tracking; however, its utility has been largely restricted due to the lack of optimal magnetic nanoparticle tracers. Herein, by controlled engineering of the size and shape of magnetic nanoparticles tailored to MPI physics theory, a specialized MPI tracer, based on cubic iron oxide nanoparticles with an edge length of 22 nm (CIONs-22), is developed. Due to the inherent lower proportion of disordered surface spins, CIONs-22 exhibit significantly larger saturation magnetization than that of spherical ones, while they possess similar saturation magnetization but smaller coercivity compared to larger-sized CIONs. These magnetic properties of CIONs-22 warrant high sensitivity and resolution of MPI. With their efficient cellular uptake, CIONs-22 exhibit superior MPI performance for stem cell labeling and tracking compared to the commercialized tracer Vivotrax. By virtue of these advantages, CIONs-22 enable real-time and prolonged monitoring of the spatiotemporal trajectory of stem cells transplanted to hindlimb ischemia mice, which demonstrates the great potential of CIONs-22 as MPI tracers to advance stem cell therapies.

White, D. E., et al. (2013). "Spatial pattern dynamics of 3D stem cell loss of pluripotency via rules-based computational modeling." PLoS Comput Biol **9**(3): e1002952.

Pluripotent embryonic stem cells (ESCs) have the unique ability to differentiate into cells from all germ lineages, making them a potentially robust cell source for regenerative medicine therapies, but difficulties in predicting and controlling ESC differentiation currently limit the development of therapies and applications from such cells. A common approach to induce the differentiation of ESCs in vitro is via the formation of multicellular aggregates known as embryoid bodies (EBs), yet cell fate specification within EBs is generally considered an ill-defined and poorly controlled process. Thus, the objective of this study was to use rules-based cellular modeling to provide insight into which processes influence initial cell fate transitions in 3-dimensional microenvironments. Mouse embryonic stem cells (D3 cell line) were differentiated to examine the temporal and spatial patterns associated with loss of pluripotency as measured through Oct4 expression. Global properties of the multicellular aggregates were accurately recapitulated by a physics-based aggregation simulation when compared to experimentally measured physical parameters of EBs. Oct4 expression patterns were analyzed by confocal microscopy over time and compared to simulated trajectories of EB patterns. The simulations demonstrated that loss of Oct4 can be modeled as a binary process, and that associated patterns can be explained by a set of simple rules that combine baseline stochasticity with intercellular communication. Competing influences between Oct4+

and Oct4- neighbors result in the observed patterns of pluripotency loss within EBs, establishing the utility of rules-based modeling for hypothesis generation of underlying ESC differentiation processes. Importantly, the results indicate that the rules dominate the emergence of patterns independent of EB structure, size, or cell division. In combination with strategies to engineer cellular microenvironments, this type of modeling approach is a powerful tool to predict stem cell behavior under a number of culture conditions that emulate characteristics of 3D stem cell niches.

Wondergem, J. and E. Rosenblatt (2012). "IAEA activities related to radiation biology and health effects of radiation." J Radiol Prot **32**(1): N123-127.

The IAEA is involved in capacity building with regard to the radiobiological sciences in its member states through its technical cooperation programme. Research projects/programmes are normally carried out within the framework of coordinated research projects (CRPs). Under this programme, two CRPs have been approved which are relevant to nuclear/radiation accidents: (1) stem cell therapeutics to modify radiation-induced damage to normal tissue, and (2) strengthening biological dosimetry in IAEA member states.

Yokota, T. (2019). ""Hierarchy" and "Holacracy"; A Paradigm of the Hematopoietic System." Cells **8**(10).

The mammalian hematopoietic system has long been viewed as a hierarchical paradigm in which a small number of hematopoietic stem cells (HSCs) are located at the apex. HSCs were traditionally thought to be homogeneous and quiescent in a homeostatic state. However, recent observations, through extramedullary hematopoiesis and clonal assays, have cast doubt on the validity of the conventional interpretation. A key issue is understanding the characteristics of HSCs from different viewpoints, including dynamic physics and social network theory. The aim of this literature review is to propose a new paradigm of our hematopoietic system, in which individual HSCs are actively involved.

Yoshida, Y. and T. Nakano (2014). "[Topics of radiation biology for cancer treatment]." Igaku Butsuri **34**(2): 48-56.

Recent advances in the field of radiation therapy (RT) have considerably improved treatment outcomes of various cancers. It is related to not only the technological progress in medical physics but also the analytical progress in radiation biological effectiveness. However, the treatment results of RT, especially in advanced cancer, are still insufficient, therefore it is necessary to establish a safety and more effective method for treating cancer. Understanding the radiation biology is essential to appreciate the effect of RT. Hence, we review the controversial point of RT for radiation biology and introduce the results of basic research.

The above contents are the collected information from Internet and public resources to offer to the people for the convenient reading and information disseminating and sharing.

References

[1]. Google. http://www.google.com. 2021.
[2]. Journal of American Science. http://www.jofamericanscience.org. 2021.
[3]. Life Science Journal. http://www.lifesciencesite.com. 2021.
[4]. http://www.sciencepub.net/nature/0501/10-0247-mahongbao-eternal-ns.pdf.
[5]. Ma H. The Nature of Time and Space. Nature and science 2003;1(1):1-11. doi:10.7537/marsnsj010103.01. http://www.sciencepub.net/nature/0101/01-ma.pdf.
[6]. Marsland Press. http://www.sciencepub.net. 2021.
[7]. National Center for Biotechnology Information, U.S. National Library of Medicine. http://www.ncbi.nlm.nih.gov/pubmed. 2021.
[8]. Nature and Science. http://www.sciencepub.net/nature. 2021.
[9]. Wikipedia. The free encyclopedia. http://en.wikipedia.org. 2021.

9/22/2021