Introduction to Nanotechnology

- Textbook：
  Nanophysics and Nanotechnology
  by:
  Edward L. Wolf

Instructor: ***H. Hosseinkhani***
E-mail: **hosseinkhani@yahoo.com**

Classroom: A209
Time: Thursday; 13:40-16:30 PM
Office hour: Thur., 10:00-11:30 AM or by appointment

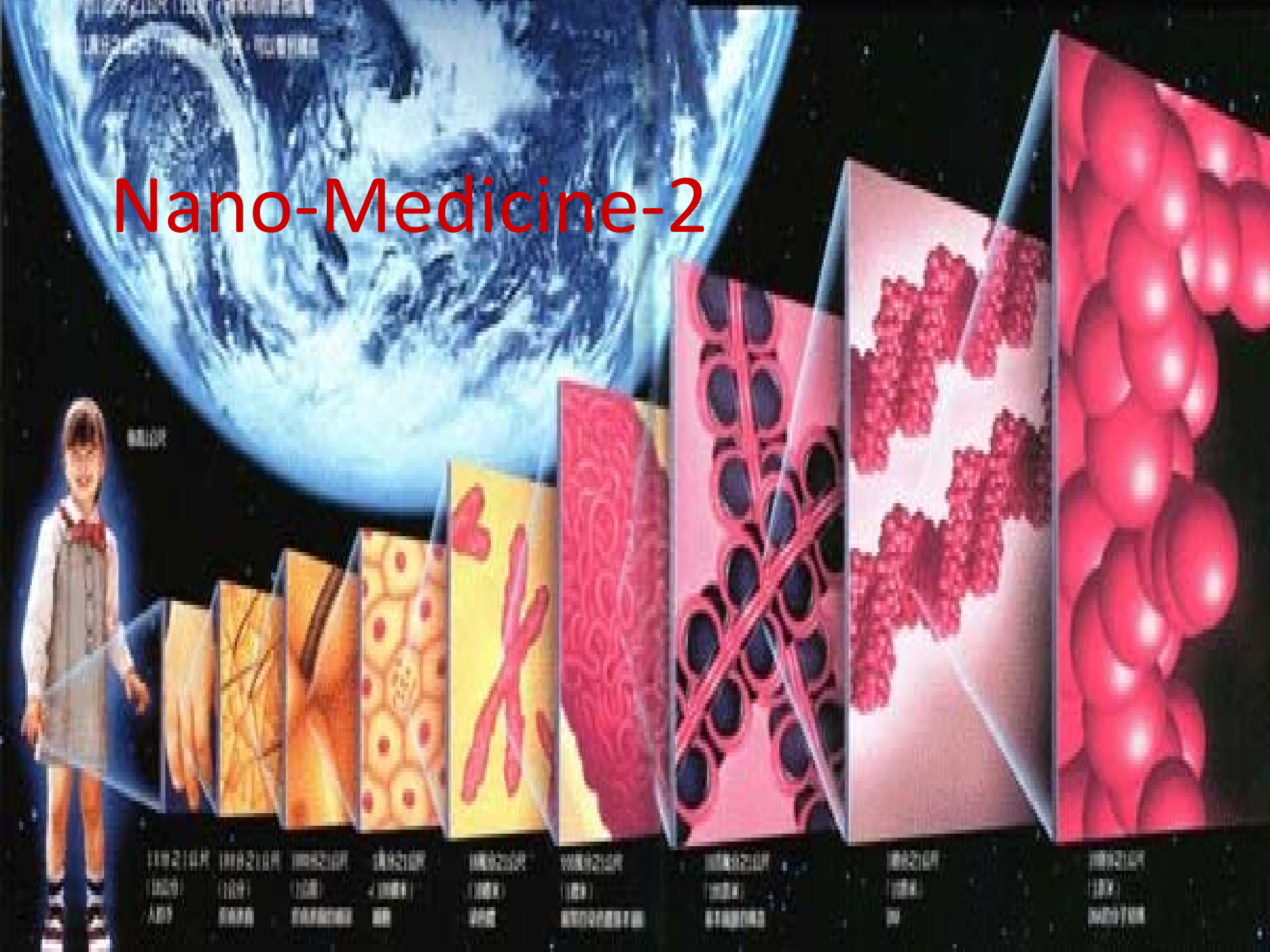
Objective of the course

The course, Introduction to Nanotechnology (IN), will focus on understanding of the basic molecular structure principals of Nano-materials. It will address the molecular structures of various materials. The long term goal of this course is to teach molecular design of materials for a broad range of applications. A brief history of biological materials and its future perspective as well as its impact to the society will be also discussed.

Evaluation; Score: 100%:

Mid-term Exam: 30%
Final Exam: 30%
Scientific Activity: 40 % (Home work, Innovation Design)

Nano-Medicine-2



Subjects:

Biodegradable and Biocompatible Nanoparticles, Nanofibers

1. Drug Delivery
2. Tissue Engineering
3. Diagnostic Tools

Motivation

- Since 1970s, organ transplantation has become a common therapeutic approach for end-stage organ failure patients.
- Demand >> Supply (UNOS National Patient Waiting List)
  - 19,095 patients (1989)
  - 80,766 patients (December 2002)
- Cost of organ replacement therapy: $305 billion (US, 2000)

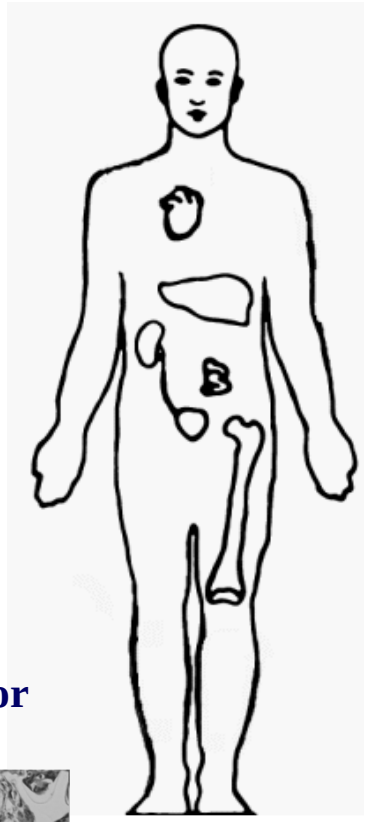
Potential of Tissue Engineering

People in the world affected by diseases that may be helped by regenerative medicine and tissue engineering.

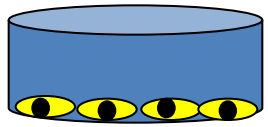
Condition	Number of persons affected (just in USA)
Cardiovascular diseases	58 million
Autoimmune diseases	30 million
Diabetes	16 million
Osteoporosis	10 million
Cancer	8.2 million
Alzheimer's disease	4 million
Parkinson's disease	1.5 million
Burns (severe)	0.3 million
Spinal cord injuries	0.25 million
Birth defects	150,000 (per year)
Total	128.4 million

Tissue Engineering

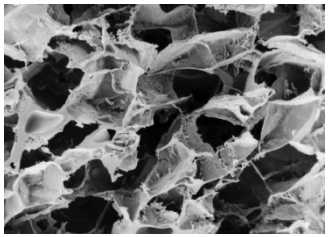
"Tissue engineering is an interdisciplinary field that applies principles & methods of engineering toward the development of biological substitutes to improve the function of damaged tissue & organs."
(Langer & Vacanti, 1998)



Cell Isolation Stem cells

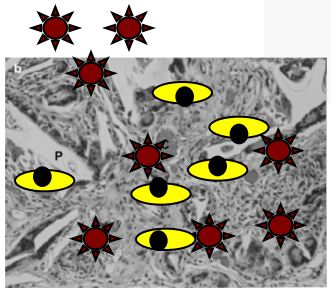


Expansion



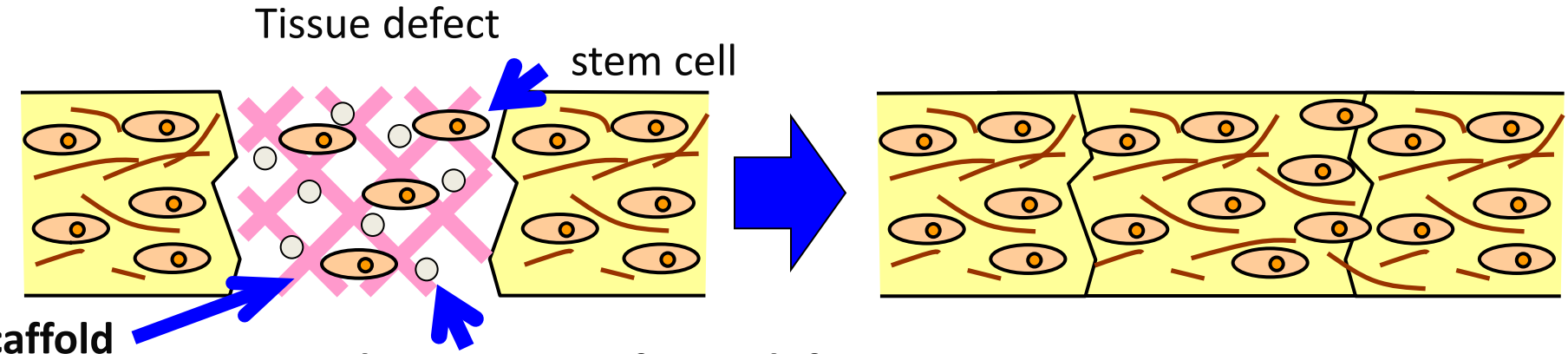
Biodegradable Polymer Scaffold fabrication

Incorporation cells and growth factor



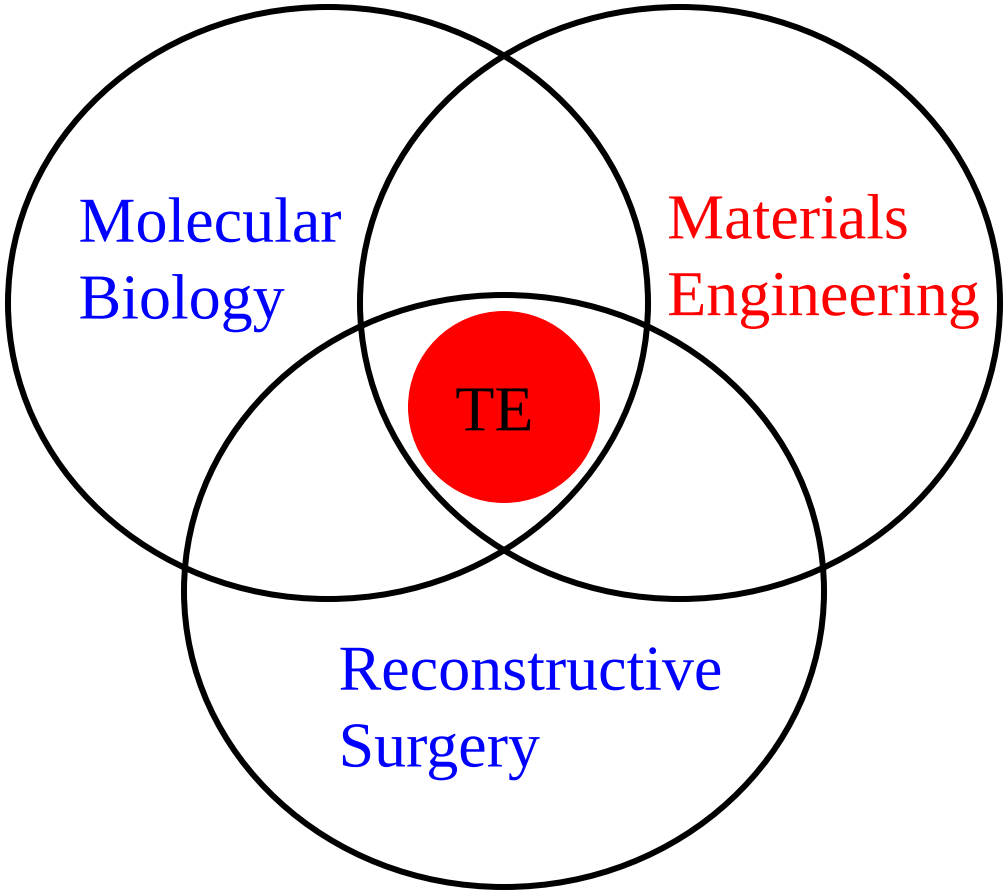
Growth Factor

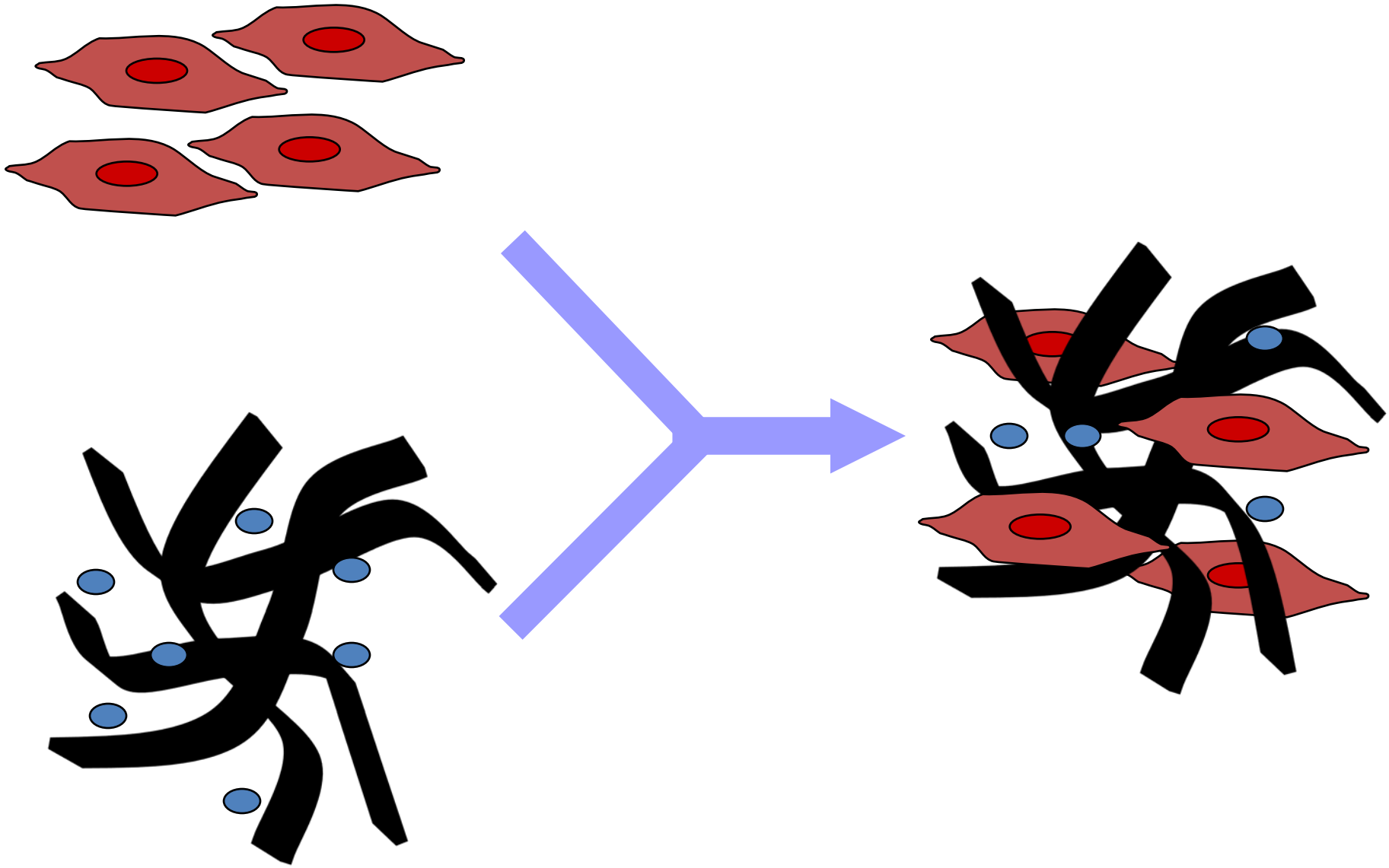
Tissue Engineering



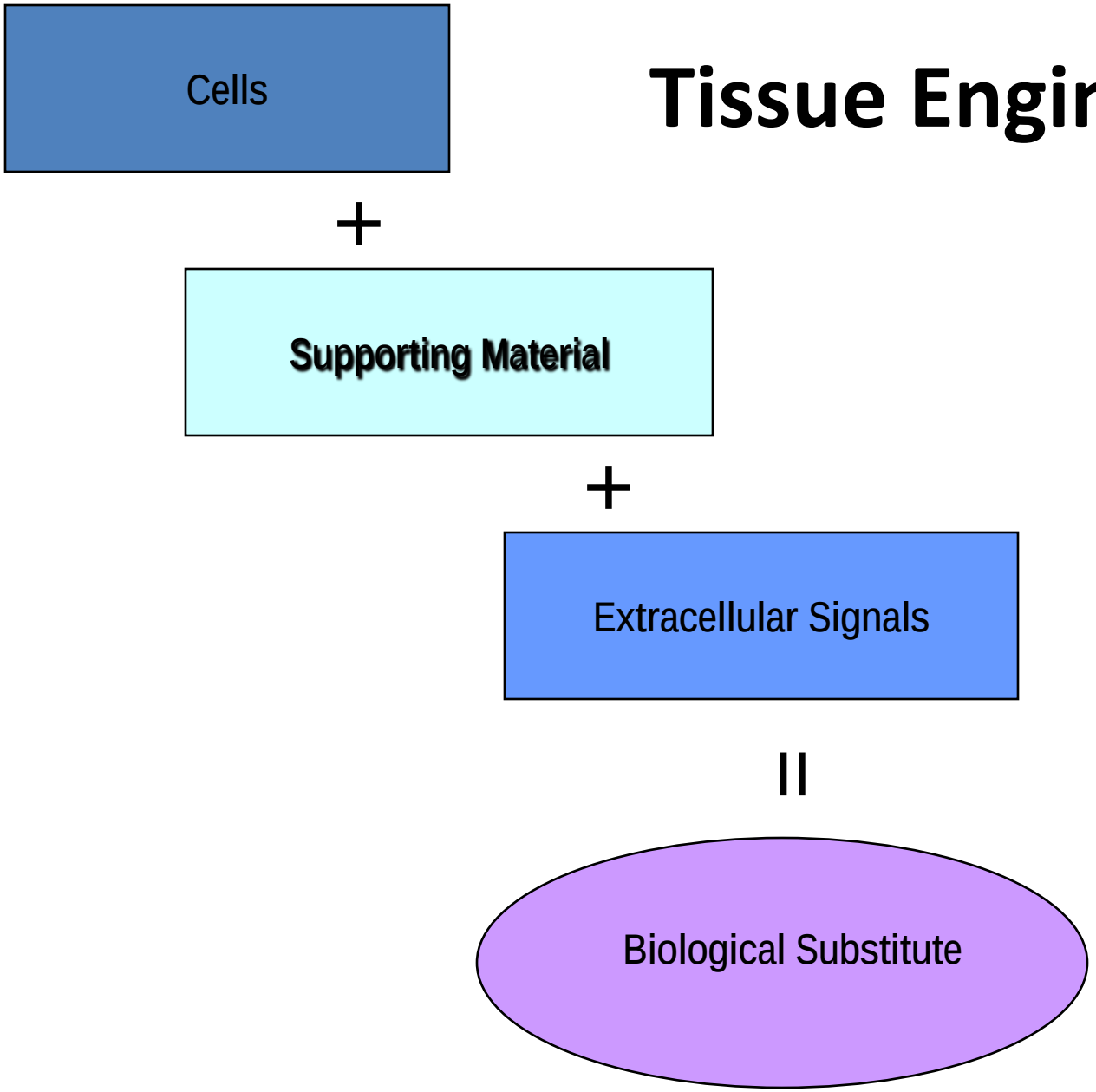
"Protein release from gelatin matrices"
Advanced Drug Delivery Rev. 1998; 31; 287-301
Tabata Y, Ikada Y

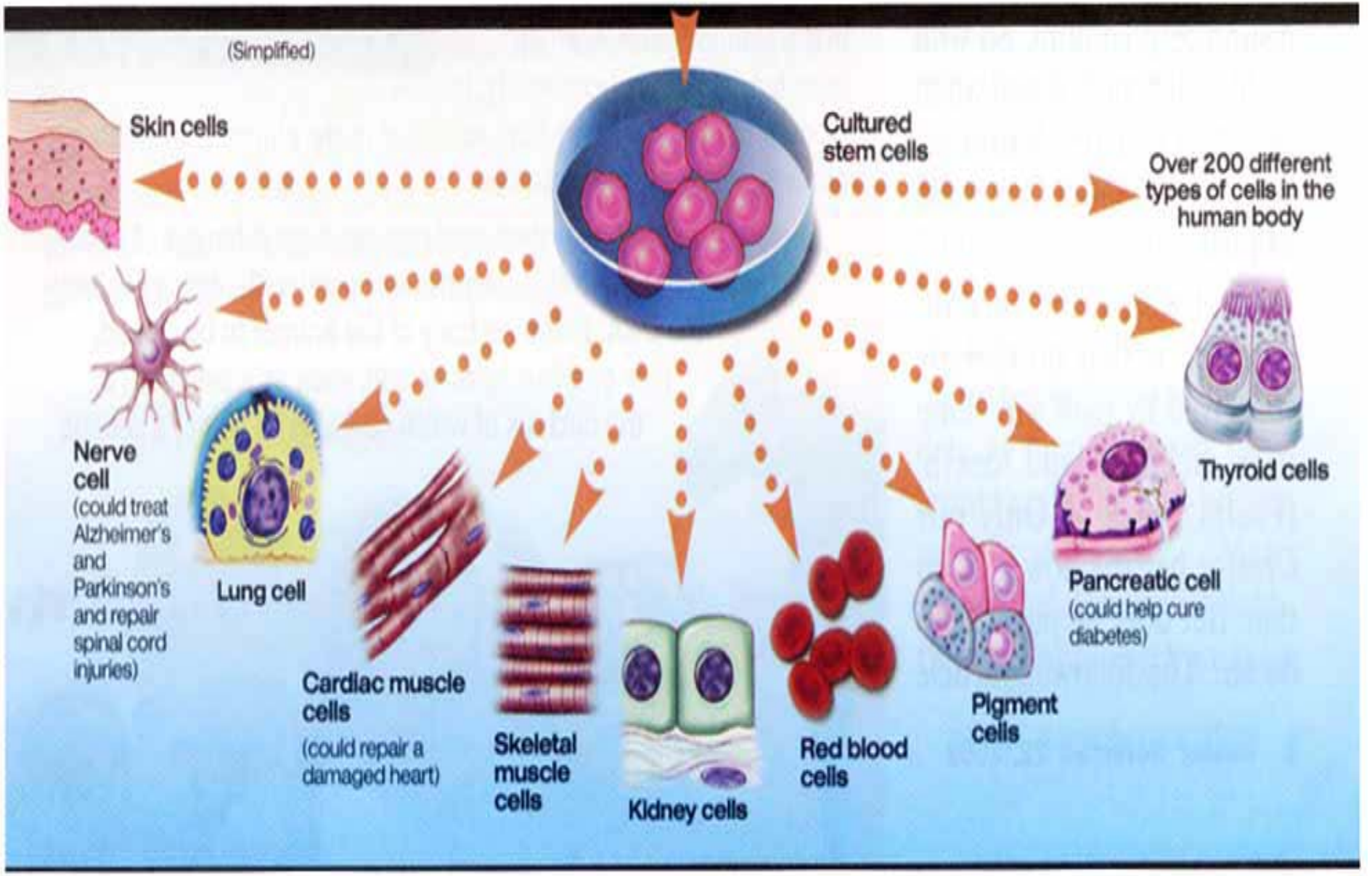
Interdisciplinary Approach





Tissue Engineering




(Simplified)
Skin cells
Cultured stem cells
Over 200 different types of cells in the human body
Nerve cell
(could treat Alzheimer's and Parkinson's and repair spinal cord injuries)
Lung cell
Cardiac muscle cells
(could repair a damaged heart)
Skeletal muscle cells
Kidney cells
Red blood cells
Pigment cells
Pancreatic cell
(could help cure diabetes)
Thyroid cells

Source of Stem Cells

Embryonic Stem Cells

ES cells constitute stem cell mass and give rise to a multiple of cell types and tissues.

Comparison

Compared to ES cells, AS cells are preferable for therapeutic purposes since they are considered safer for implantation, with lesser proliferation capacity and tumorgenecity. They are also easier to differentiate to specific lineages, while ES cells can give a wide range of tissues following local implantation.

Adult Stem Cells

AS cells constitute adult tissues and give rise to differentiated tissue-specialized cells, and are responsible for the regenerative capacities of tissue. AS cells present a more limited range of differentiation lineages.

Mesenchymal Stem Cells (MSCs) are stem cells residing in variety of adult mesenchymal tissues, and can be isolated from bone marrow, or other hematopoietic and non-hematopoietic tissues.

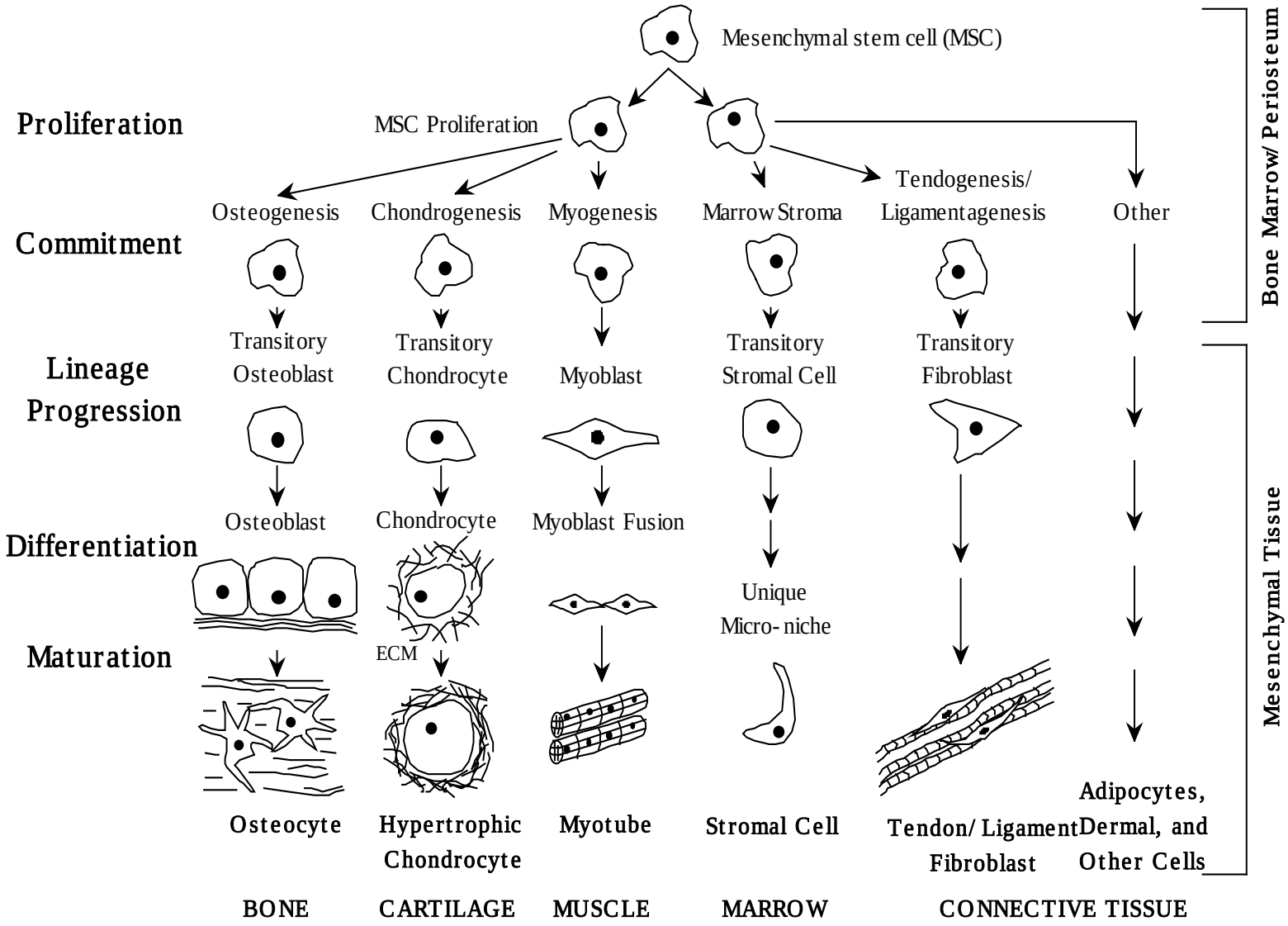
MSCs cells derived from non-hematopoietic tissues, such as adipose tissues are very attractive future area of tissue engineering.

- Bone marrow
- Adipose Tissue

Embryonic Germ Cells

EG cells are derived from the cells in the ridge of an embryo or a fetus, which give rise to eggs or sperm. They are able to rise to virtually all cell types. This potential makes pluripotent cells very attractive candidates for the development of unprecedental medical treatments.

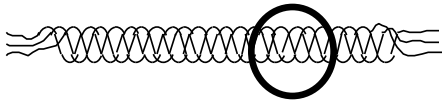
Tissue Regeneration By Use of Cells (mesenchymal stem cells, precursor cells, and blast cells)



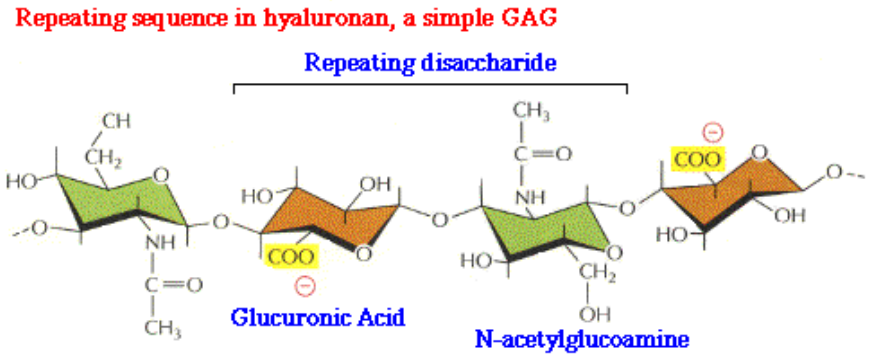
Differentiation tree of mesenchymal stem cells

Cellular Microenvironment:
1. Extracellular Matrix (ECM)

1. Main Components

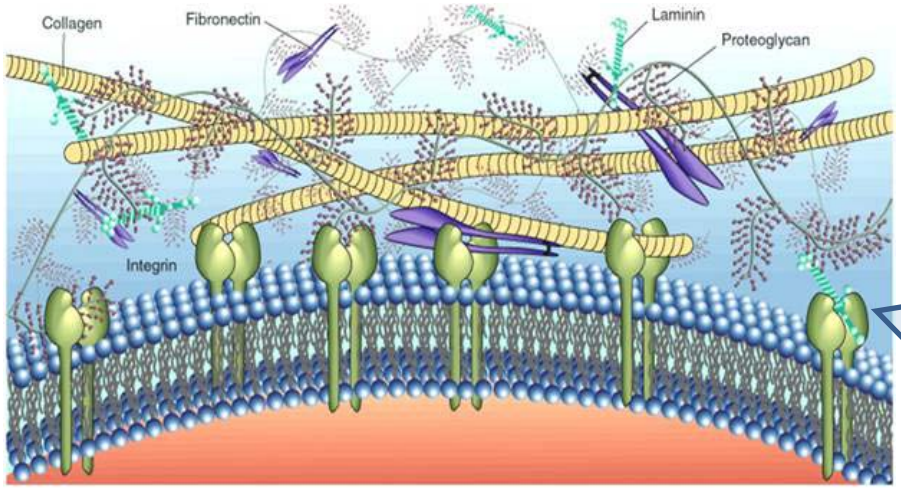


Collagen fibrils



Hyaluronic Acid (HA)

- Natural component of the ECM
- Negatively charged polysaccharide
- Highly water soluble
- Forms absorbed layers on hydrophilic substrates



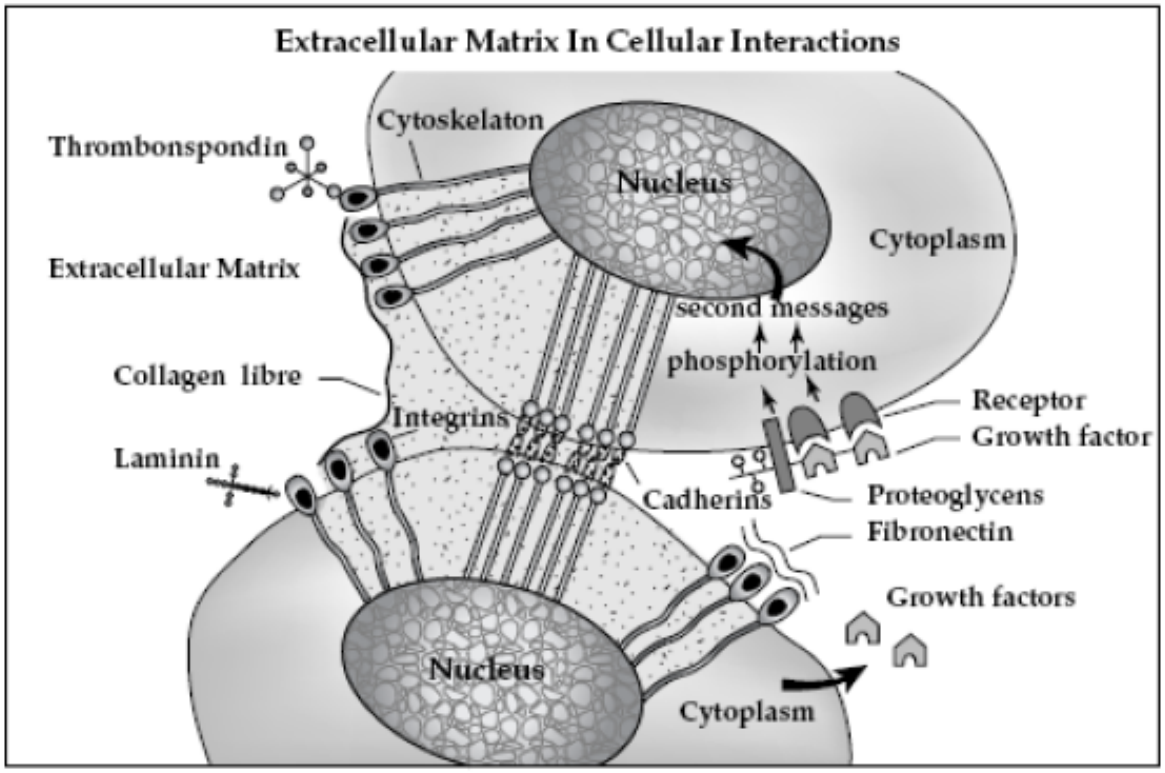
2. Specific Components

- Fibronectin
- Vitronectin
- **Laminin**
- Elastin
- Adhesion Proteins

GENERAL ECM CHARACTERISTICS

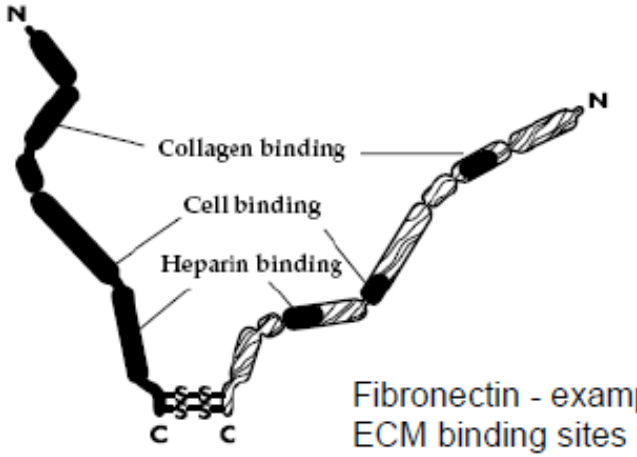
- They are very large molecules (molecular weight: 10^5–10^6) and therefore have very low diffusivities.
- They are multimodal in that they often have cell-binding domains as well as domains to bind other ECM molecules.
- They can regulate cell fates, including differentiation, apoptosis, and migration.
- They can change conformation under mechanical load, thereby exposing active binding sites.
- Many of the critical binding domains that have been identified can be mimicked by small peptides.
- These molecules often remain *bound* to isolated mammalian cells, occupying a subpopulation of integrin receptors.

Network of Communication: Cells ←→ ECM



Component	Function	Location
Collagens	Tissue architecture, tensile strength Cell-matrix interactions Matrix-matrix interactions	Ubiquitously distributed
Elastin	Tissue architecture and elasticity	Tissues requiring elasticity, eg. lung, blood vessels, heart, skin
Proteoglycans	Cell-matrix interactions Matrix-matrix interactions Cell proliferation Cell migration	Ubiquitously distributed
Hyaluronan	Cell-matrix interactions Matrix-matrix interactions Cell proliferation Cell migration	Ubiquitously distributed
Laminin	Basement membrane component Cell migration	Basement membranes
Epiligrin	Basement membrane component (epithelium)	Basement membranes
Entactin (nidogen)	Basement membrane component	Basement membranes
Fibronectin	Tissue architecture Cell-matrix interactions Matrix-matrix interactions Cell proliferation Cell migration Opsonin	Ubiquitously distributed
Vitronectin	Cell-matrix interactions Matrix-matrix interactions Hemostasis	Blood Sites of wound formation
Fibrinogen	Cell proliferation Cell migration Hemostasis	Blood Sites of wound formation
Fibrillin	Microfibrillar component of elastic fibers	Tissues requiring elasticity, eg. blood vessels, heart, skin
Tenascin Anti-adhesive	Modulates cell-matrix interactions remodeling matrix	Transiently expressed associated with anti-proliferative
SPARC (osteonectin)	Modulates cell-matrix interactions Anti-adhesive	Transiently expressed associated with remodeling matrix anti-proliferative
Thrombospodin	Modulates cell-matrix interactions	Platelet & granules
Adhesion molecules	Cell surface proteins mediating cell adhesion to matrix or adjacent cells Mediators of transmembrane signals	Ubiquitously distributed
von Willebrand factor	Mediates platelet adhesion Carrier for procoagulant factor VIII	Plasma protein Subendothelium

Extracellular Matrix – complexity



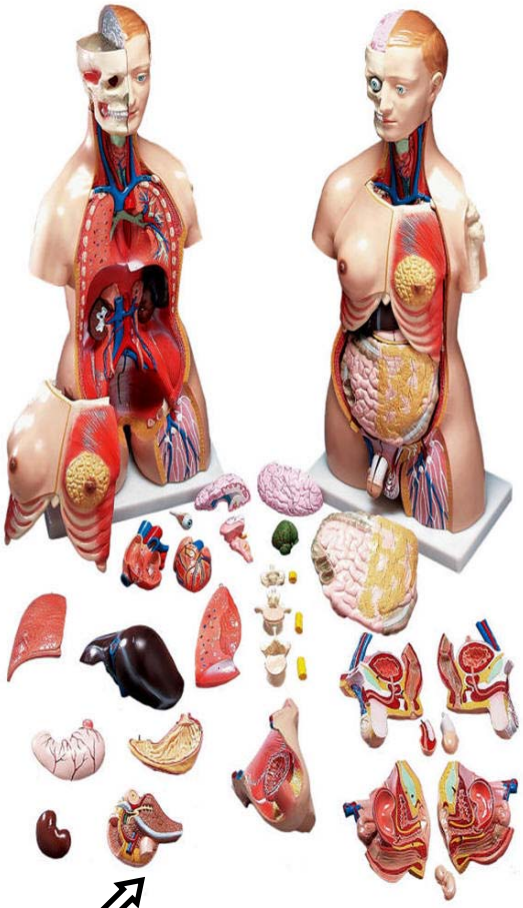
(B)

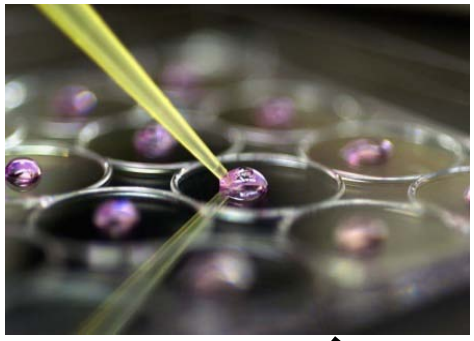
Fibronectin - example
ECM binding sites

Nanotechnology

in

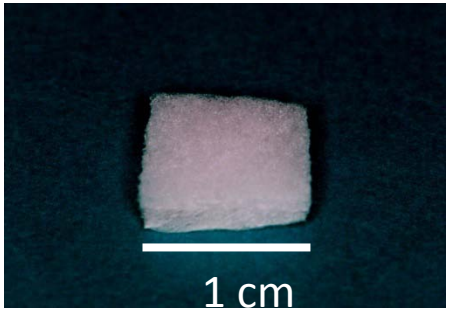
Tissue Engineering



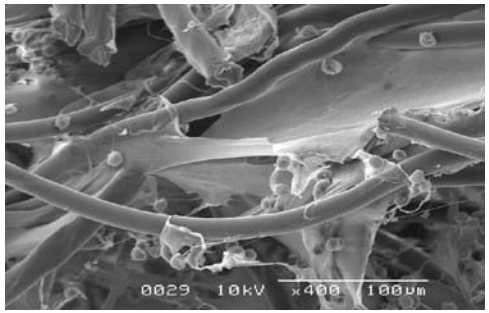


Cell isolation

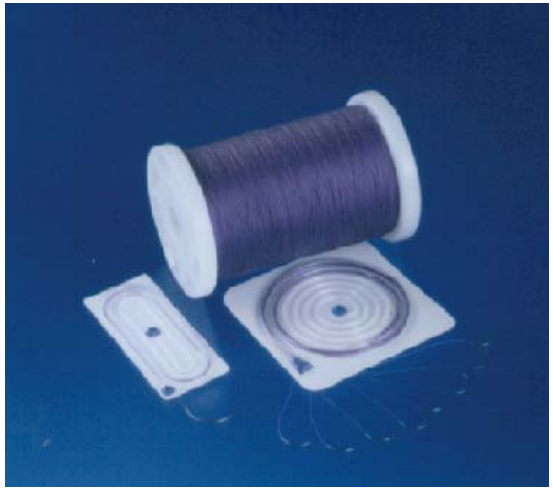
2D *in vitro* culture



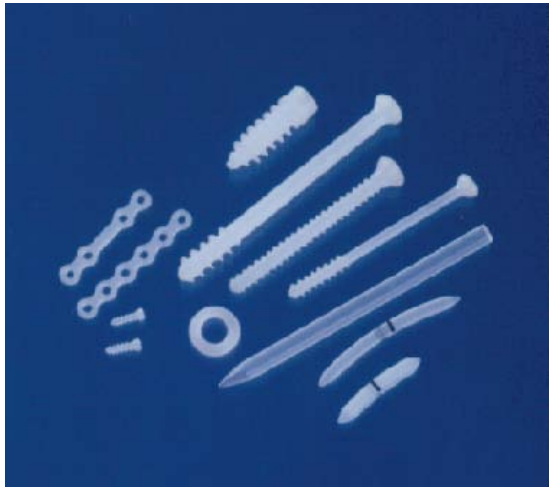
3D *in vitro* scaffold



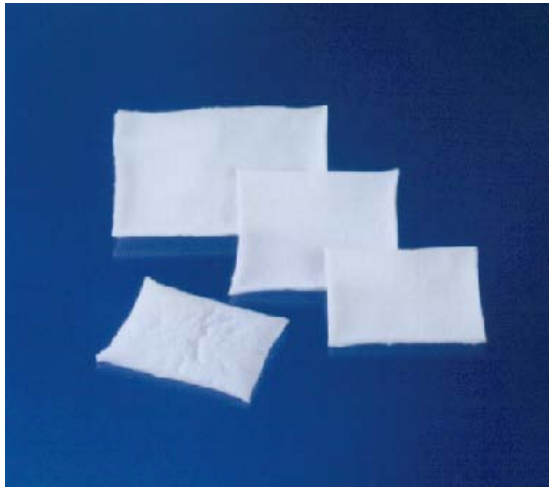
Incorporation of cells and growth factor



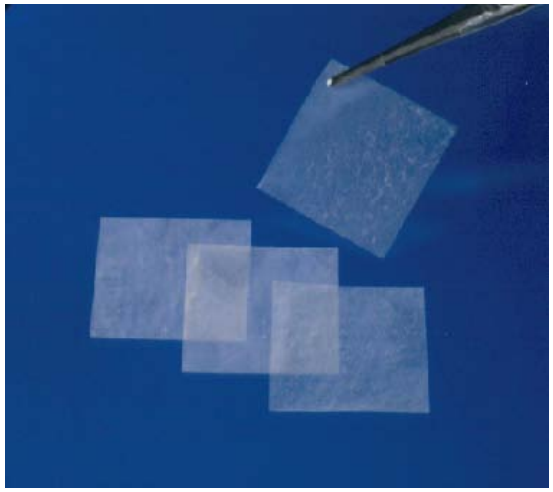
Abxorbable suture (PGA)



Bone fixation device (PLLA)

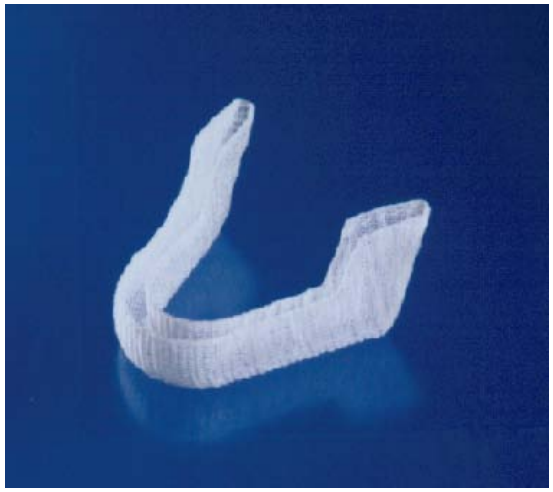


Artificial dermis (Collagen)



Dura substitute (PLA-co-εCL)

Artificial ligament (PLLA)



Mandibular mesh tray（PLLA）

techniques are involved in the fabrication of 3D scaffolding materials:

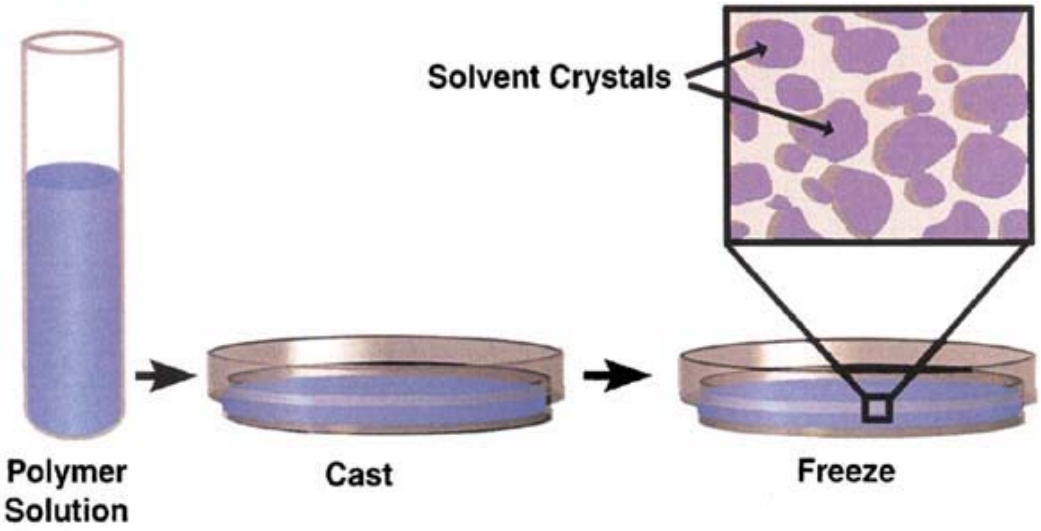
Freeze Drying
Phase Separation
Gas Foaming
Fiber Bonding
Photolithography
Solid Free Form (SFF)
Solvent Casting in Combination with Particle Leaching

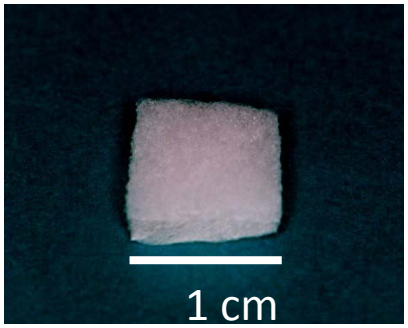
Freeze Drying: principles of freeze drying are based on the lyophilization process. Lyophilization is a process which extracts the water from products so that the products remain stable and are easier to store at room temperature (ambient air temperature). Lyophilization is carried out using a simple principle of physics called sublimation. Sublimation is the transition of a substance from the solid to the vapor state, without first passing through an intermediate liquid phase. A schematic view of freeze drying for the fabrication of 3D scaffolding materials is shown in Figure 1

Phase Separation: In this technique, a porous structure can be easily obtained by adjusting thermodynamic and kinetic parameters. However, because of the complexity of the processing variables involved in phase-separation technique the pore structure cannot be easily controlled. Moreover, it is difficult to obtain large pores and may exhibit a lack of interconnectivity

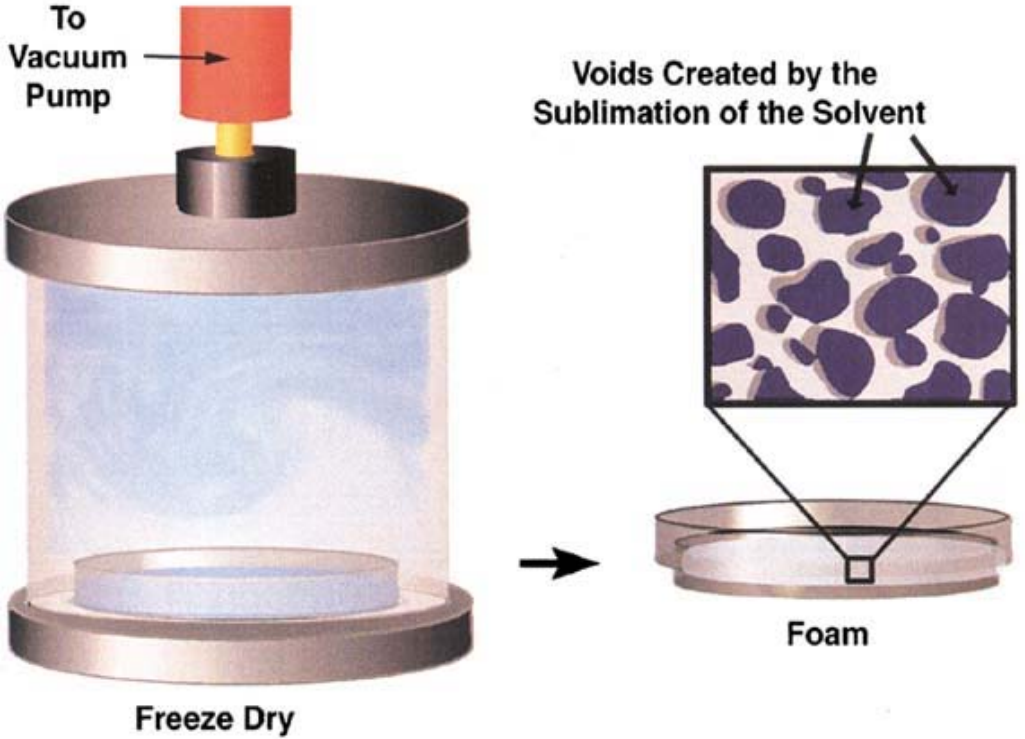
Gas Foaming: This technology has the advantage of room temperature processing but produces a largely non-porous outer skin layer and a mixture of open and closed pores within the center leaving incomplete interconnectivity. The main disadvantage of the gas foaming method is that it often result in a non-connected cellular structure within the scaffold .

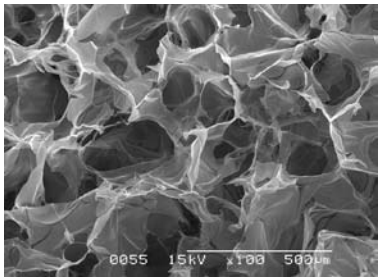
Figure 1





collagen sponge after cutting by a razor





SEM image of highly porous collagen sponge

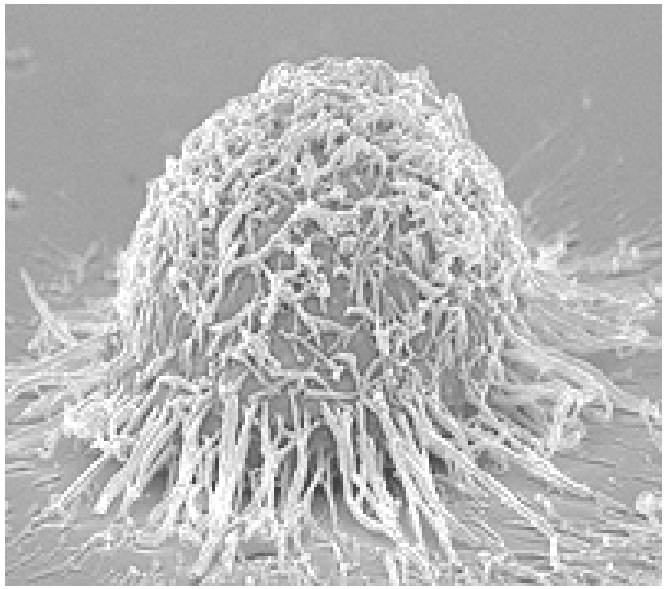
Fiber Bonding: This technique provides a large surface area for cell attachment and a rapid diffusion of nutrients in favor of cell survival and growth. However, these scaffolds, as the ones used to construct a network of bonded polyglycolic acid (PGA), lacked the structural stability necessary for in vivo use. In addition, the technique does not lend itself to easy and independent control of porosity and pore size.

Photolitography: This technology has been employed for patterning, obtaining structures with high resolution, although this resolution may be unnecessary for many applications of patterning in cell biology. In any case, the disadvantage of this technique is the high cost of the equipment need limits their applicability.

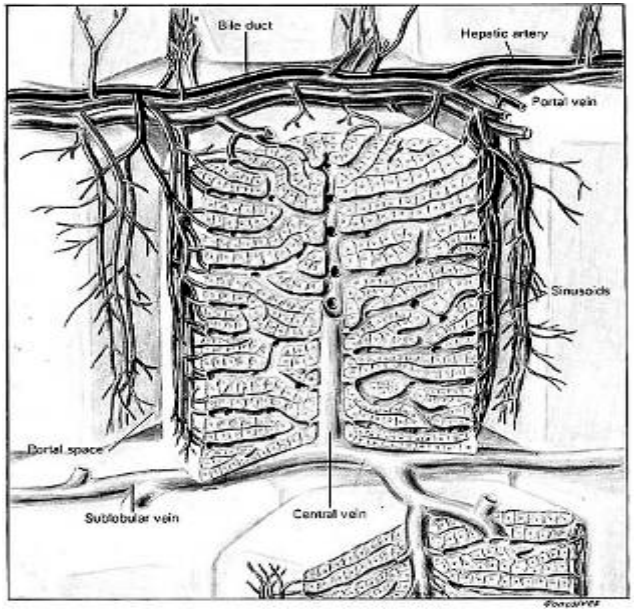
Solid Free Form (SFF): This manufacturing method provide excellent control over scaffold external shape, and internal pore interconnectivity and geometry, but offer limited micro-scale resolution. Moreover, the minimum size of global-pores is 100 µm. Additionally; SFF requires complex correction of scaffold design for anisotropic shrinkage during fabrication. Moreover, it needs high cost equipments

Solvent Casting in Combination with Particle Leaching: This technology involves the casting of a mixture of monomers and initiator solution and a porogen in a mold, polymerization, followed by leaching-out of the porogen with the proper solvent to generate the pores, is inexpensive but still has to overcome some disadvantages in order to find engineering applications, namely the problem of residual porogen remains, irregular shaped pores, and insufficient interconnectivity .The proposed scaffolds may find applications as structures that facilitate either tissue regeneration or repair during reconstructive operations . Intended application of these structures is nerve surgery. The new structure could also find applications in other areas in which the pore morphology may play an essential role, such as membranes and filters

Microenvironmental Interactions



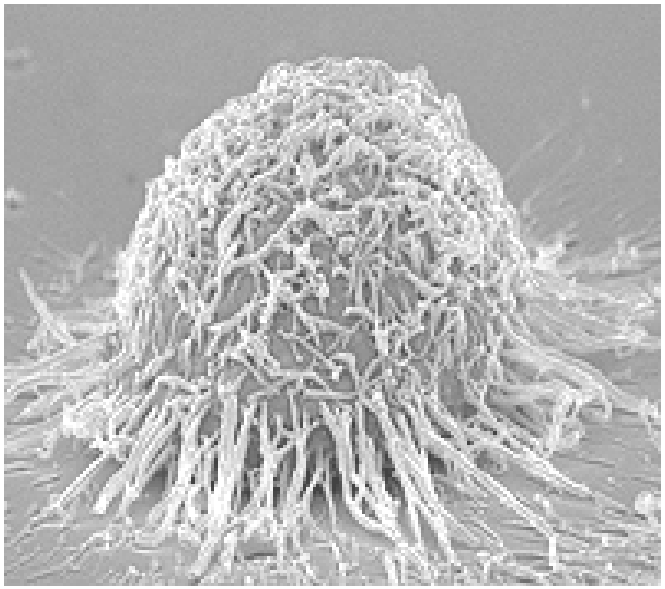
in vitro



in vivo

Challenges:

Microenvironmental Interactions



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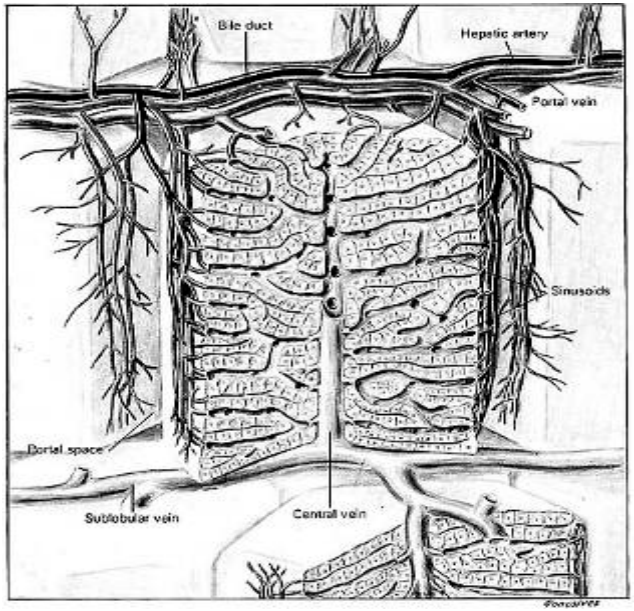
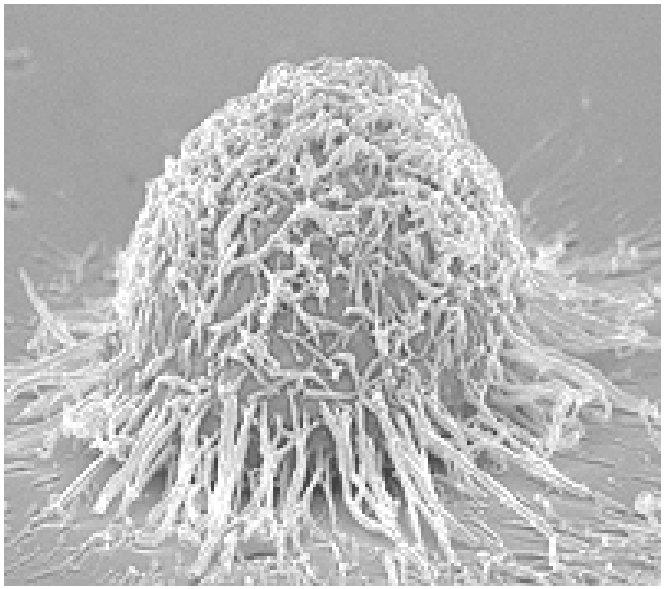


in vivo

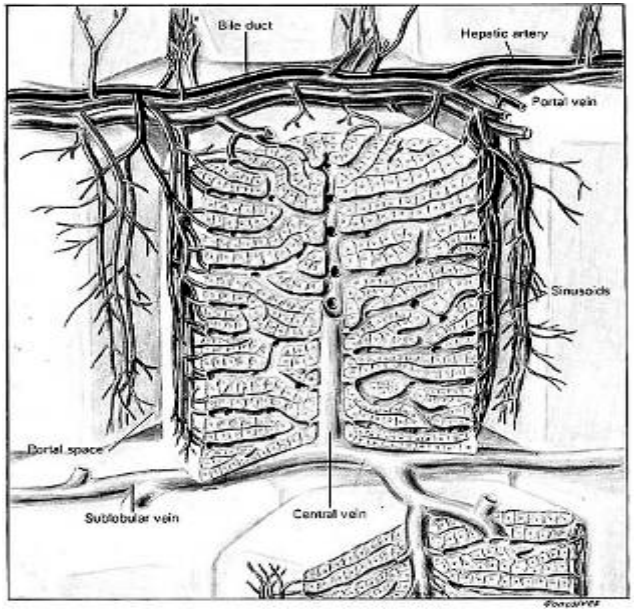
Challenges:

- *In vitro* systems have the inability to mimic the complex cell-microenvironmental interactions.

Microenvironmental Interactions



in vitro

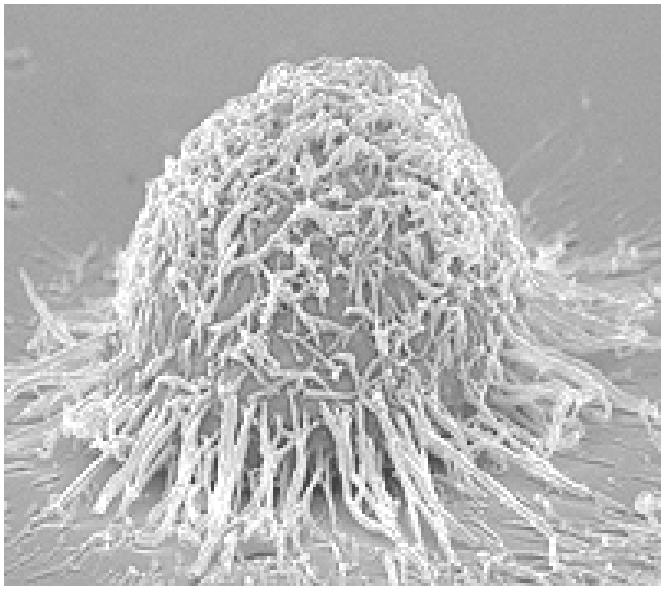


in vivo

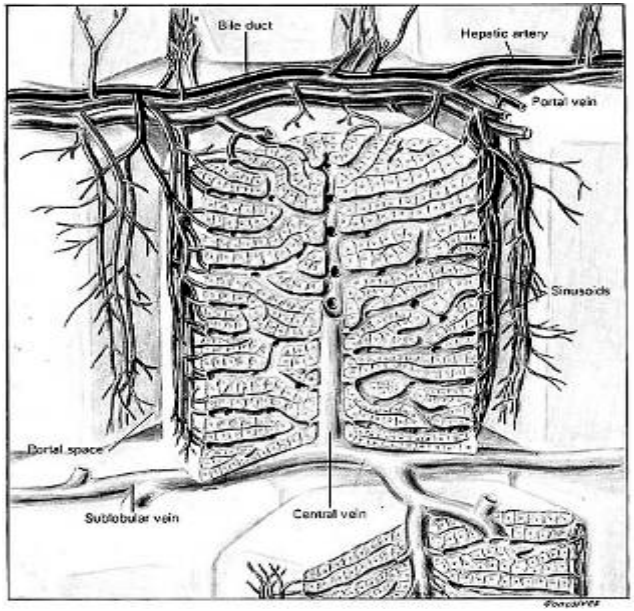
Challenges:

- *In vitro* systems have the inability to mimic the complex cell-microenvironmental interactions.
- *In vitro* systems fails to mimic *in vivo* topography such as three dimensional (3D) orientation and architecture of cells.

Microenvironmental Interactions



in vitro



in vivo

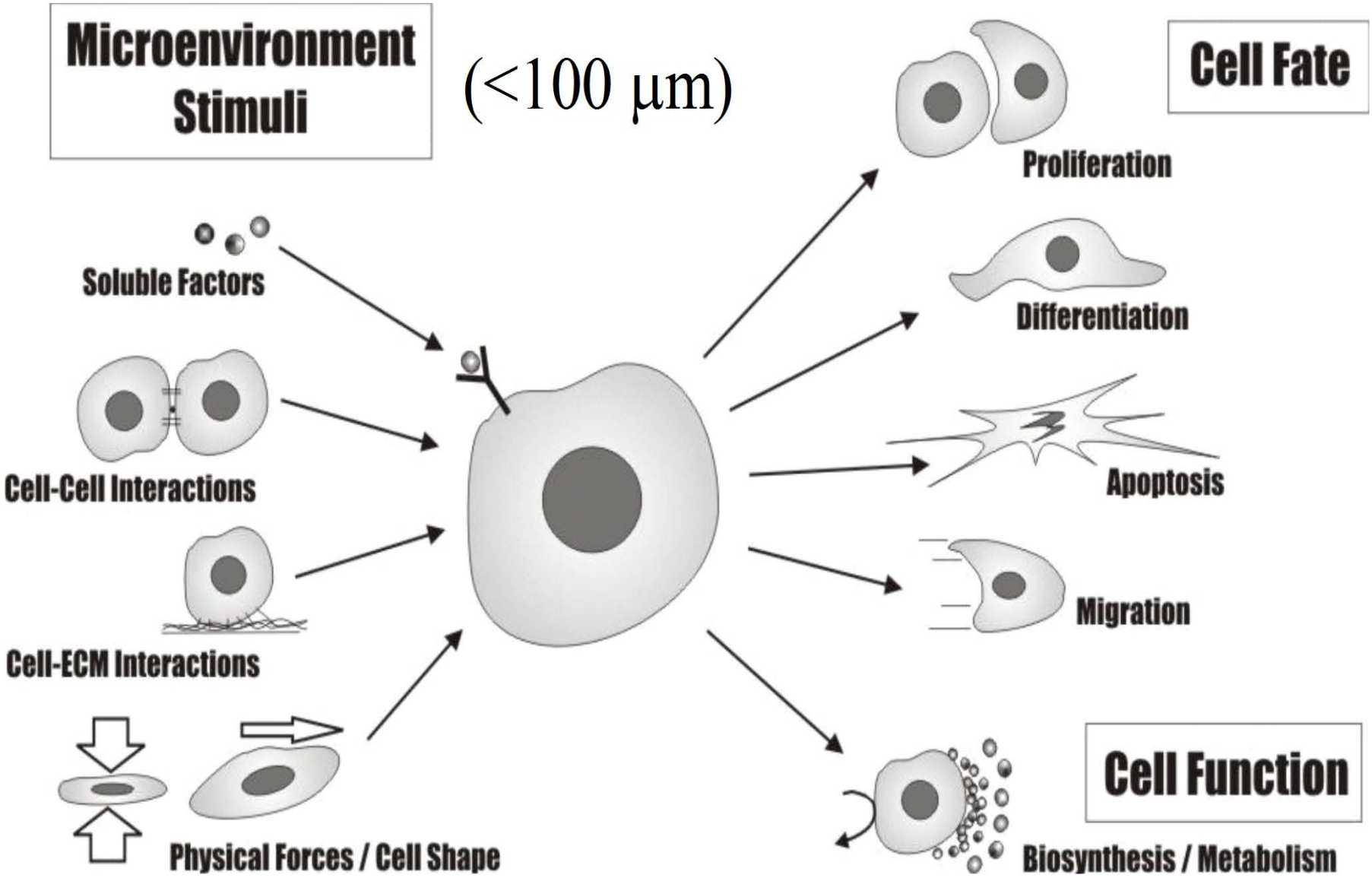
Challenges:

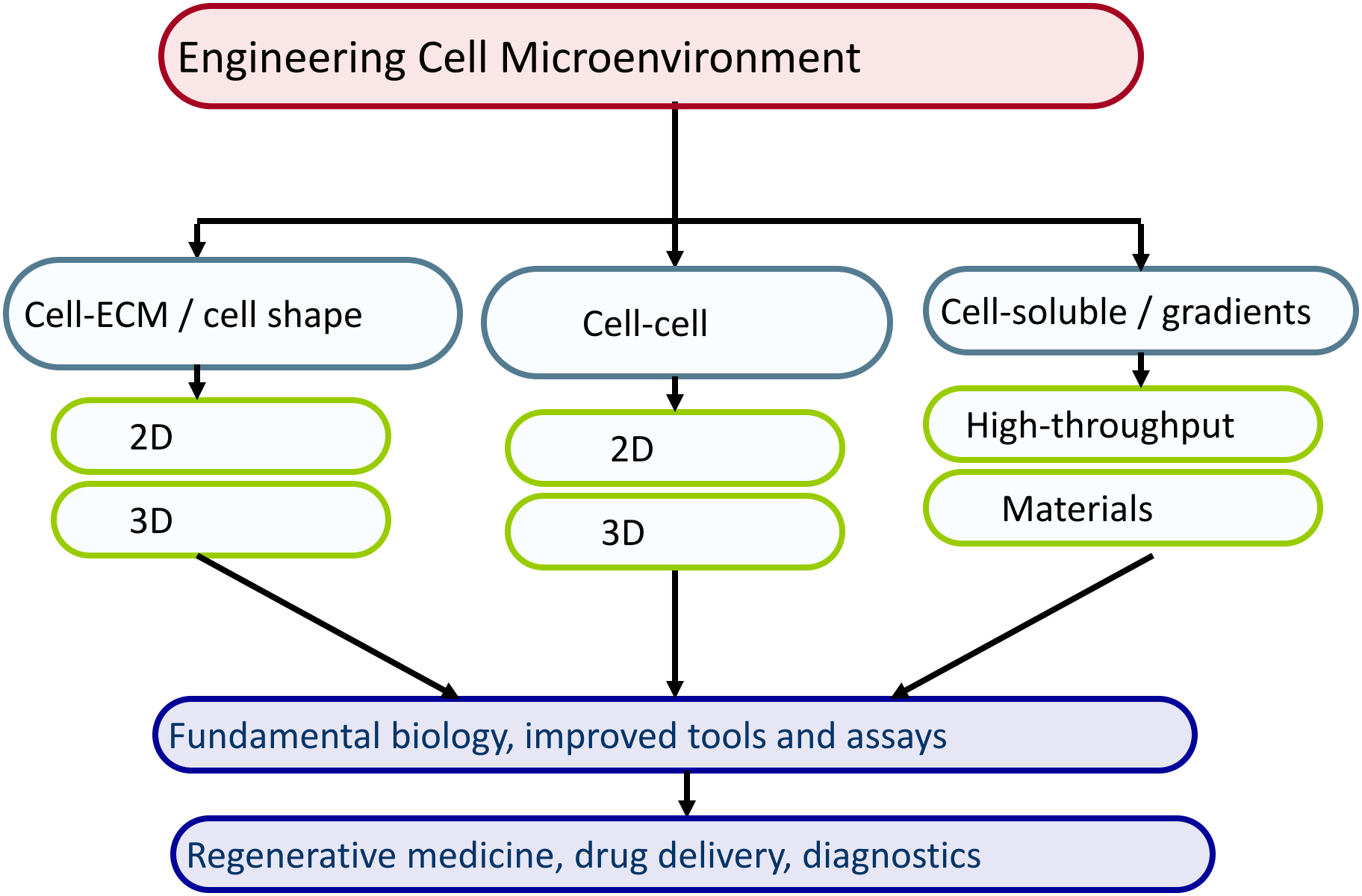
- *In vitro* systems have the inability to mimic the complex cell-microenvironmental interactions.
- *In vitro* systems fails to mimic *in vivo* topography such as three dimensional (3D) orientation and architecture of cells.
- Failure in homotypic and heterotypic cell-cell contact with microscale resolution.

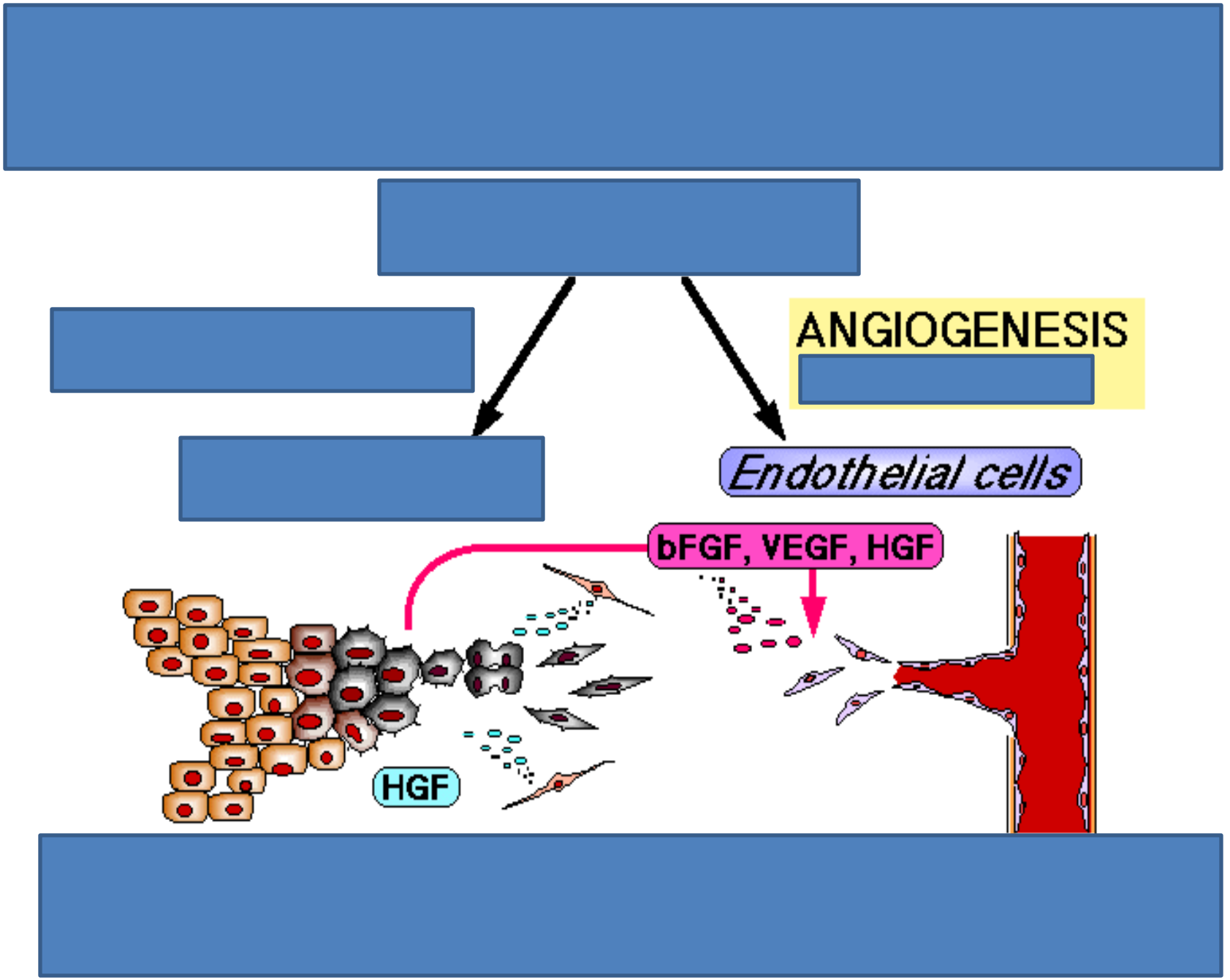
Organ function depends on organ microstructure

- Nanoscale support structures (< 1 micron) to control individual cell behavior
  - adhesion, migration, proliferation
- Microscale support structures (1-100 microns) to control cell-cell interactions and cell-substrate interactions
- Macroscale structures (> 100 microns) for structural support

The cell microenvironment (niche) directs cellular fate and function

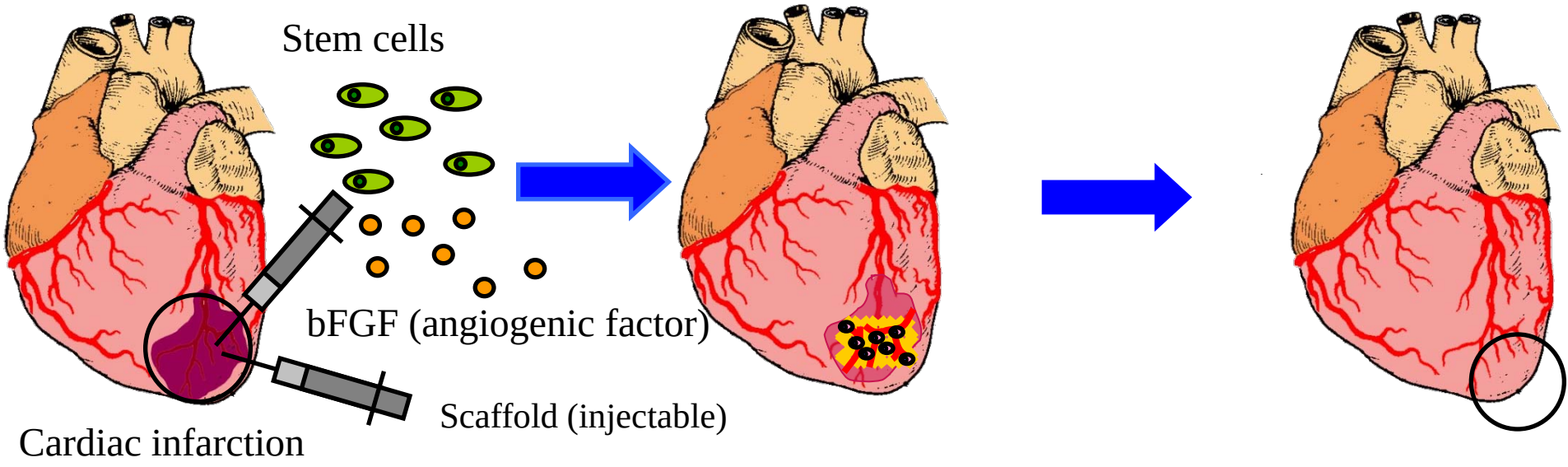



Engineering Cell Microenvironment
Cell-ECM / cell shape
Cell-cell
Cell-soluble / gradients
2D
3D
2D
3D
High-throughput
Materials
Fundamental biology, improved tools and assays
Regenerative medicine, drug delivery, diagnostics


ANGIOGENESIS
Endothelial cells
bFGF, VEGF, HGF
HGF

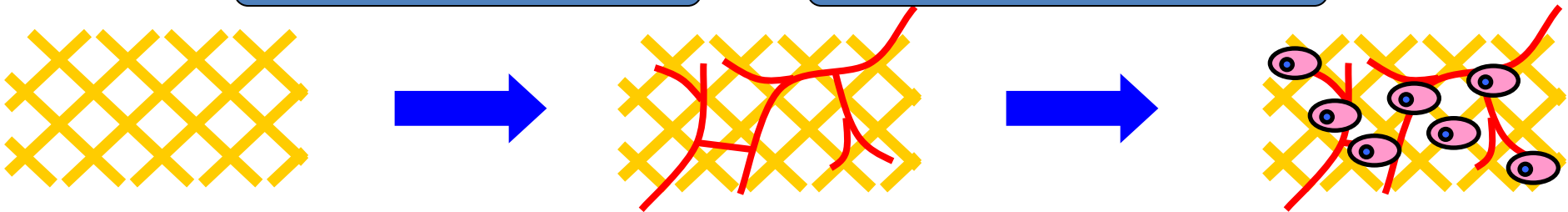
Tissue Engineering

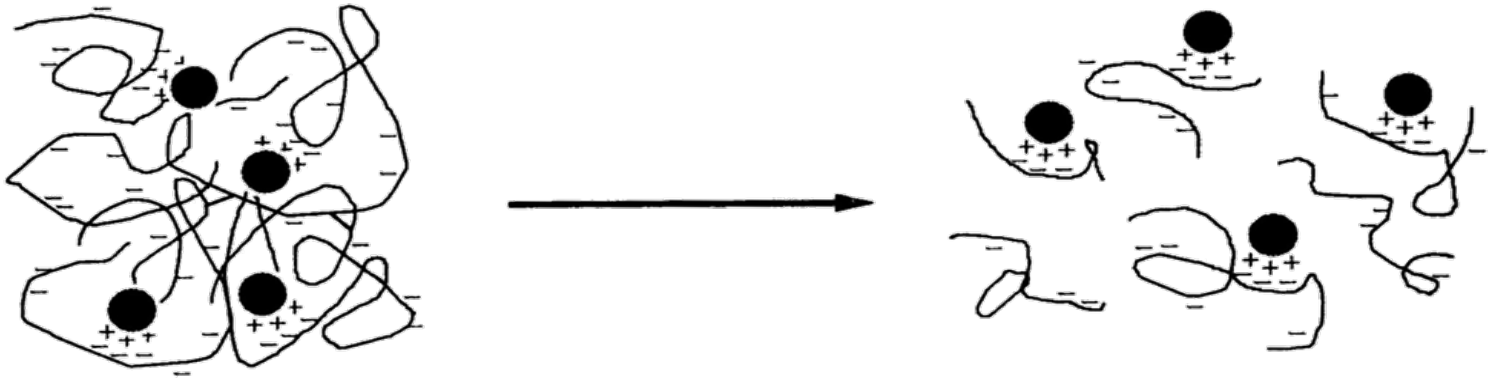
Challenge: Clinical application



Prevascularization

Cell transplantation





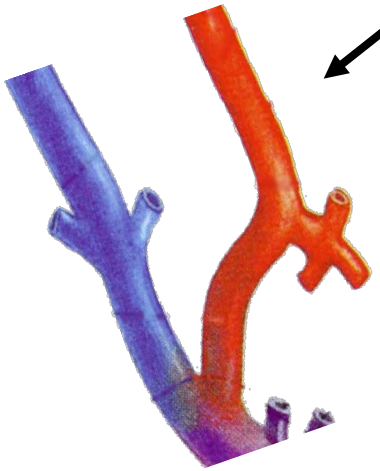
Hydrogel: Electrostatic, Hydrogen bond, Hydrophobic

Controlled release of drug

Protein (growth factor)

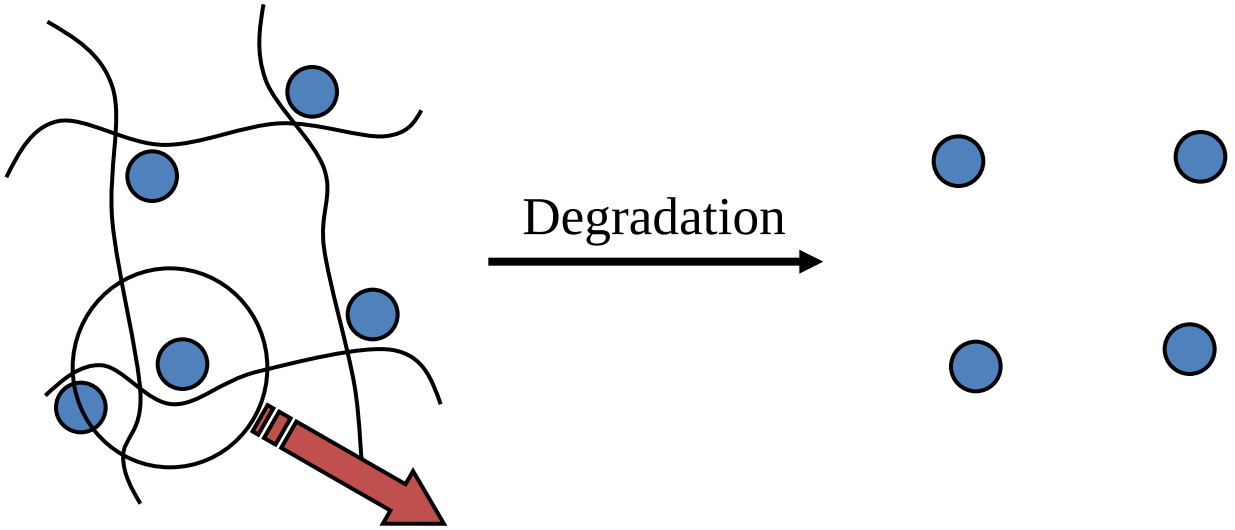
Carrier

bFGF
VEGF

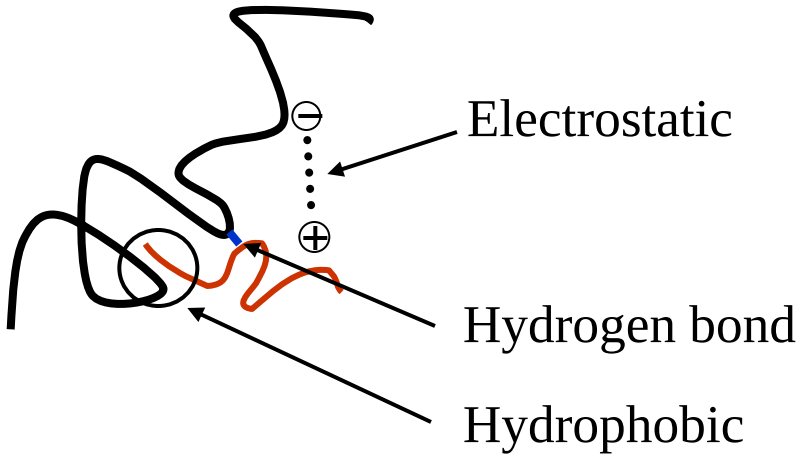


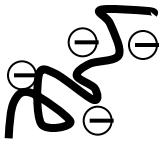
Angiogenesis

Mechanism on the controlled release of Drug



Possible intermolecular interaction between carrier matrix and growth factor





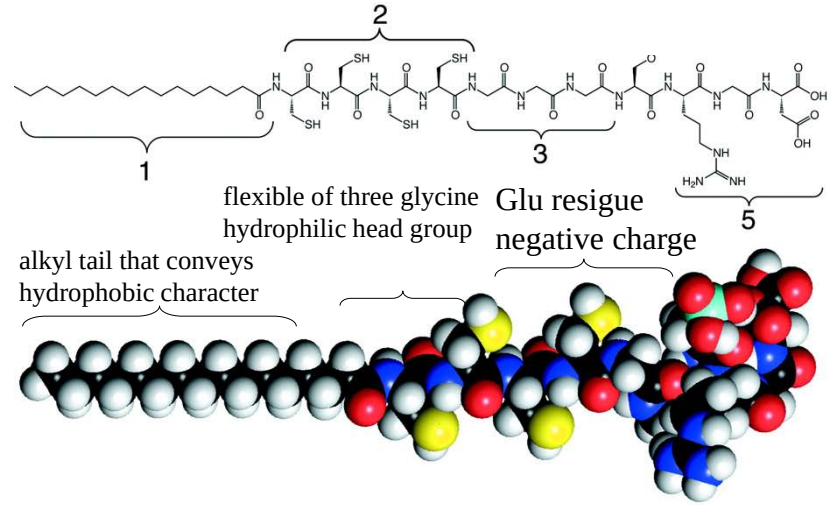
: Carrier matrix

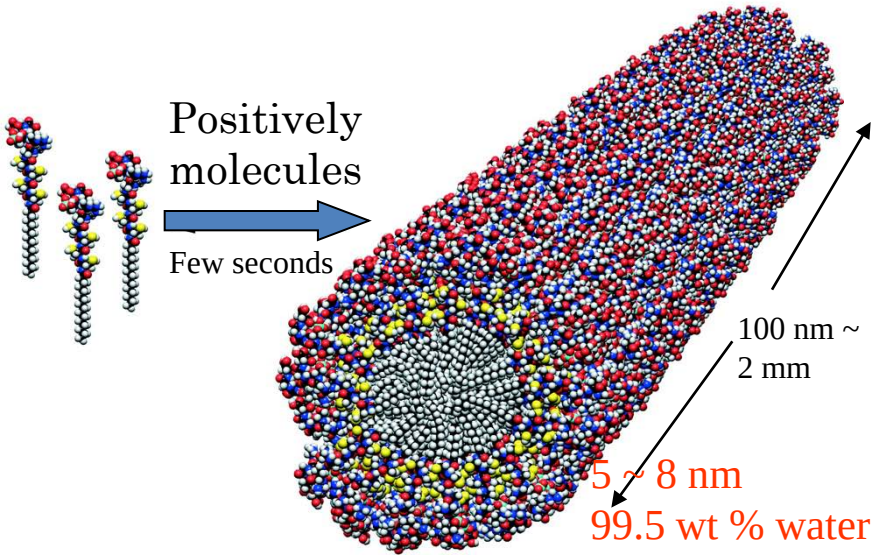
: Growth factor

Table 1 Characteristics of the growth factors used in this study

Growth factor	Isoelectric point (IEP)	Molecular weight (kDa)	Biological substances for growth factor binding	Functions of growth factor
Basic fibroblast growth factor (bFGF)	**9.6**	16	Heparin or heparan sulfate	Stimulating the cells involved in the healing process (bone, cartilage, nerve, etc). Angiogenesis
Transforming growth factor β1 (TGFβ1)	**9.5**	25	Heparin or heparan sulfate Collagen type IV Latency associated protein Latent TGFβ1 binding protein	Enhancing the wound healing, Stimulating the osteoblast proliferation to enhance bone formation
Bone morphogenetic protein-2 (BMP-2)	**8.5**	32	Collagen type IV	Stimulating the mesenchymal stem cells to osteoblast lineage and inducing the bone formation both at bone and ectopic sites.
Vascular endothelial growth factor (VEGF)	**8.5**	38	Heparin or heparan sulfate	Stimulating the endothelial cell growth, angiogenesis, and capillary permeability
Hepatocyte growth factor (HGF)	**5.5**	100	Heparin or heparan sulfate	Stimulating of matrix remodeling and epithelial regeneration (liver, spleen, kidney, etc)

ECM-mimicking Hydrogel





High aspect ratio
High surface area

Samuel I. Stupp of Northwestern University in Chicago has discovered and considered to Peptide Amphiphile (PA) as its great capacity to form nanofibers (Science, Vol 303, 2004).

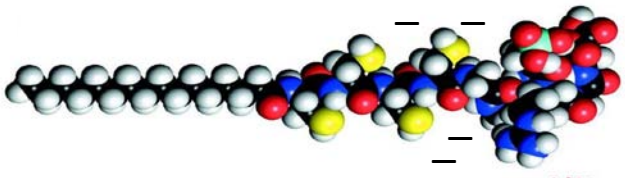
PA can self assemble into sheets, spheres, rods, disks, or
channels depending on the shape, charge, and environment

Amphiphiles with a conical shape in which the hydrophilic head group is somewhat bulkier than its narrow hydrophobic tail have been shown to form cylindrical micelles.

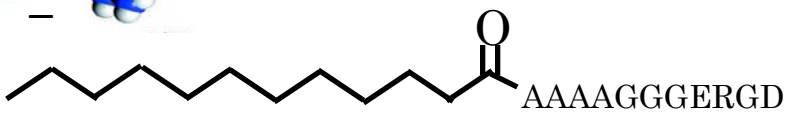
PA with mono- or di-alkyl tails were found to associate in conformations such as triple helical structures found in collagen.

Cations in human body can screen electrostatic repulsion among peptide, and 3-D monofiber are stabilized by ions such as Ca2+ through hydrogen and electrostatic interactions, nonspecific van der Waals interactions, hydrophobic forces, and repulsive steric forces.

Fabrication of Hydrogel that mimic ECM nano-fibers



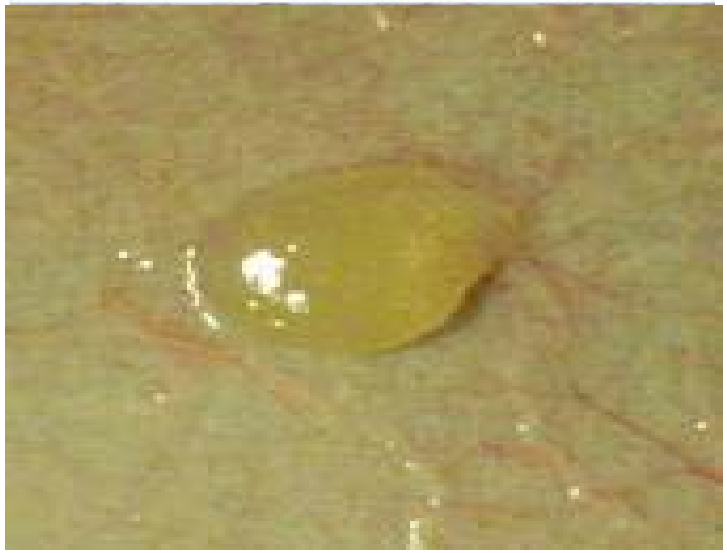
Peptide

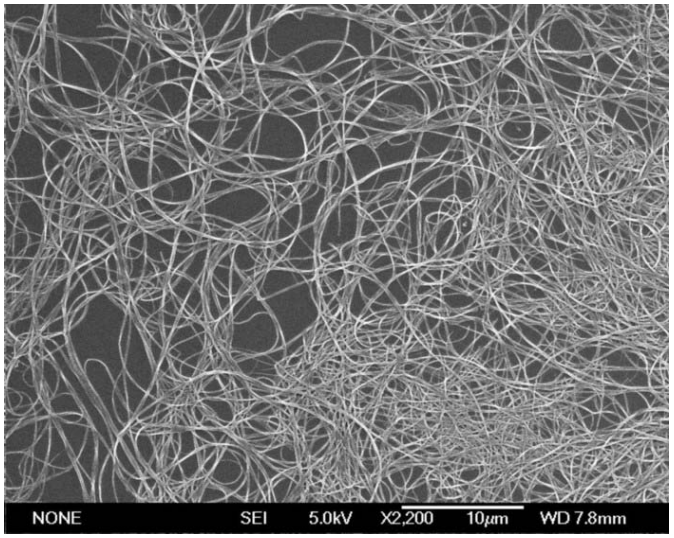


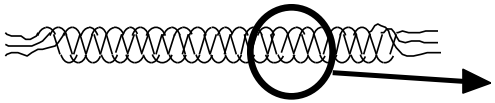
C-16

bFGF

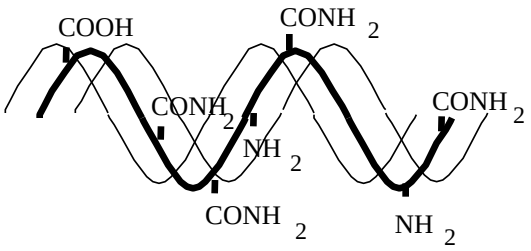
Gel formation

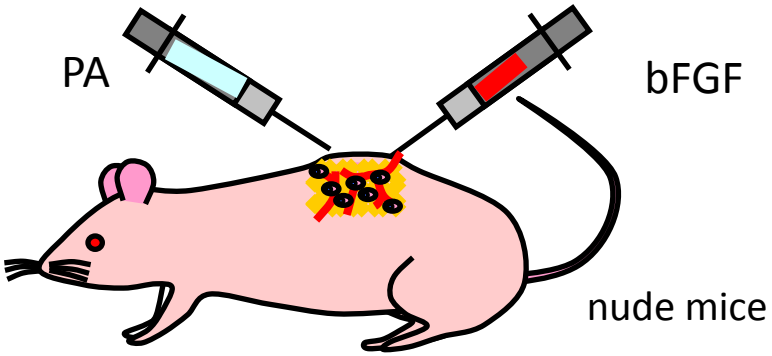


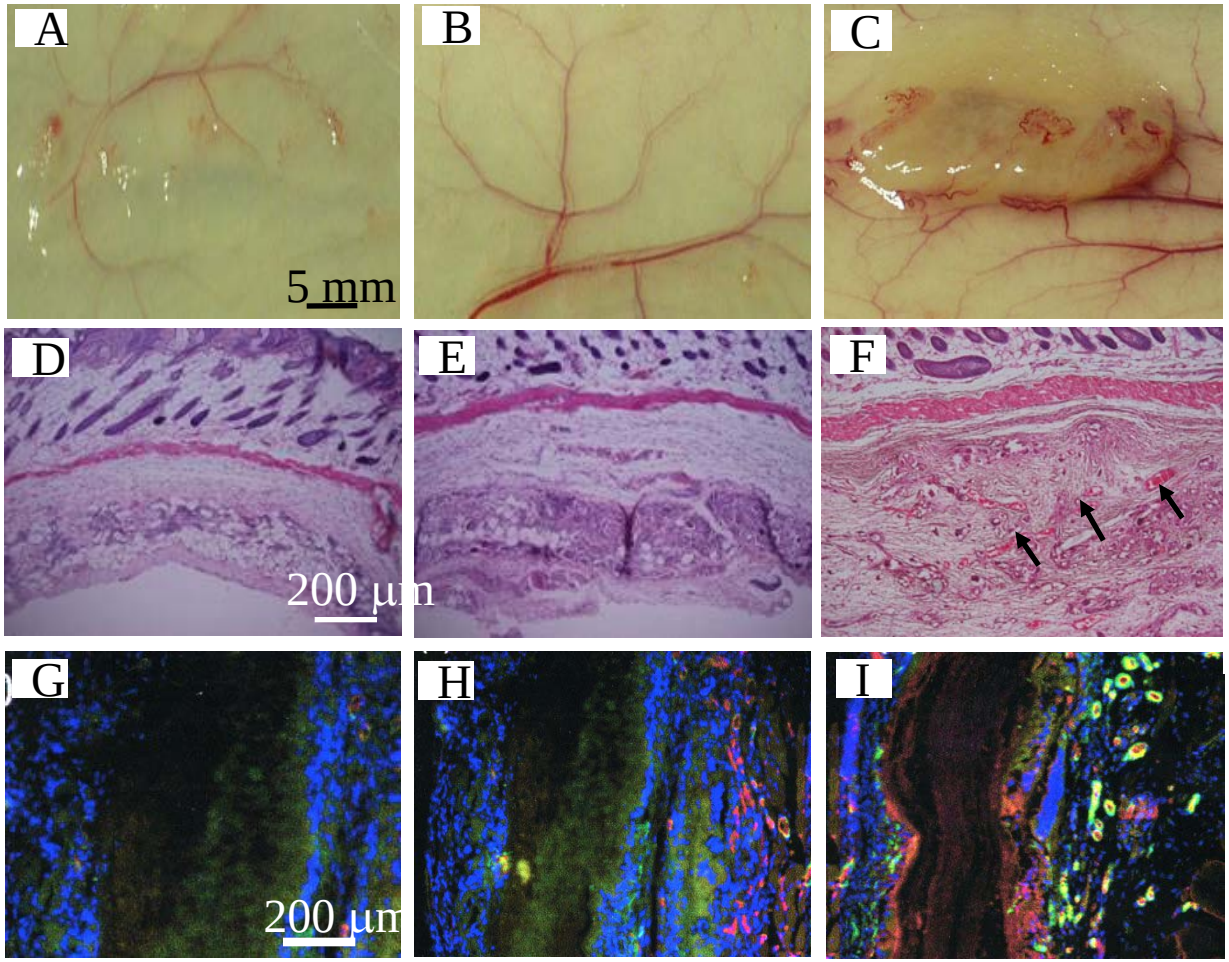




collagen







green: SMC α-actin
blue: nuclei
red: CD31

Rationale: Hydrogels are remarkably similar to human tissues.

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…hydrogels lack the desired mechanical and biological properties that are associated with tissues in the body.

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Mechanically Bio-mimicking

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Mechanically Bio-mimicking

Chemically Bio-mimicking

Rationale: Hydrogels are remarkably similar to human tissues.

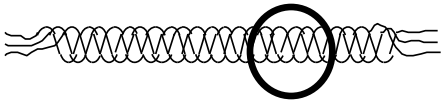
…hydrogels lack the desired mechanical and biological properties that are associated with tissues in the body.

Mechanically Bio-mimicking

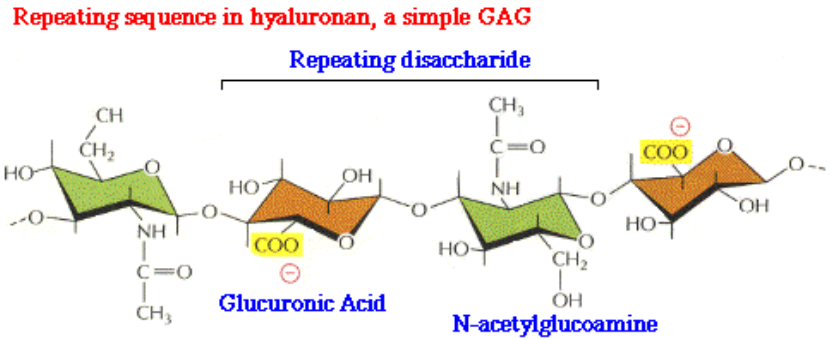
Chemically Bio-mimicking

Biologically Bio-mimicking

Natural Cellular Microenvironment
ECM:



Collagen fibrils + growth factors (bFGF, HGF BMP-2,4..IGF…)



Hyaluronic Acid (HA)

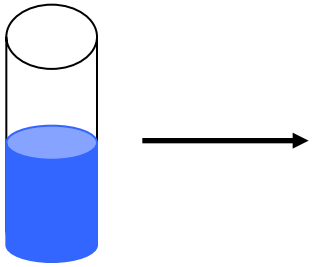
- Natural component of the ECM
- Negatively charged polysaccharide
- Highly water soluble
- Forms absorbed layers on hydrophilic substrates!

Aim 1

Mechanically Bio-mimicking Hydrogels

Aim 1

Mechanically Bio-mimicking Hydrogels

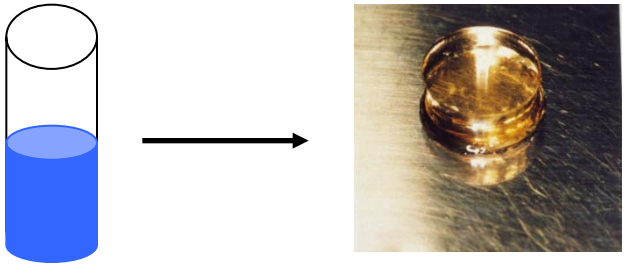


4 C

Heat-crosslinkable hydrogels (Collagen)

Aim 1

Mechanically Bio-mimicking Hydrogels



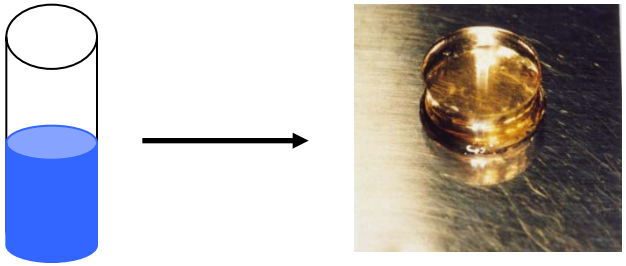
4 C

37 C

Heat-crosslinkable hydrogels (Collagen)

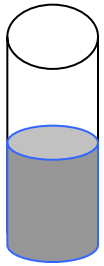
Aim 1

Mechanically Bio-mimicking Hydrogels



4 C

37 C

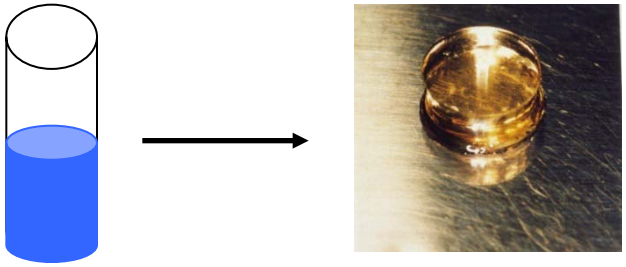


Peptide

Heat-crosslinkable hydrogels (Collagen)

Aim 1

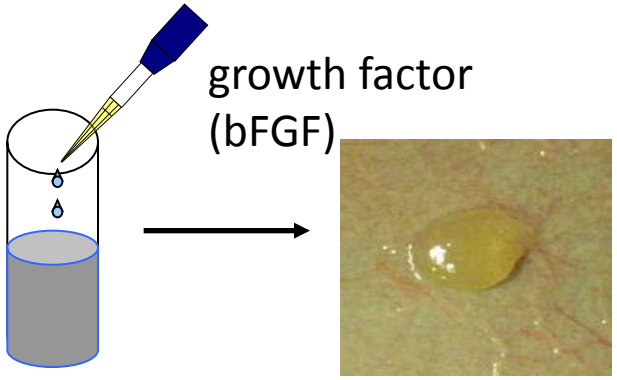
Mechanically Bio-mimicking Hydrogels



4 C

37 C

Heat-crosslinkable hydrogels (Collagen)

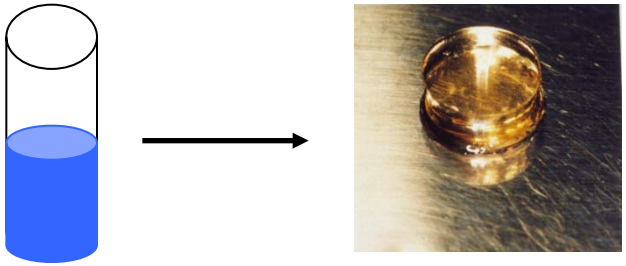


Peptide

Self-assembled Peptide hydrogel

Aim 1

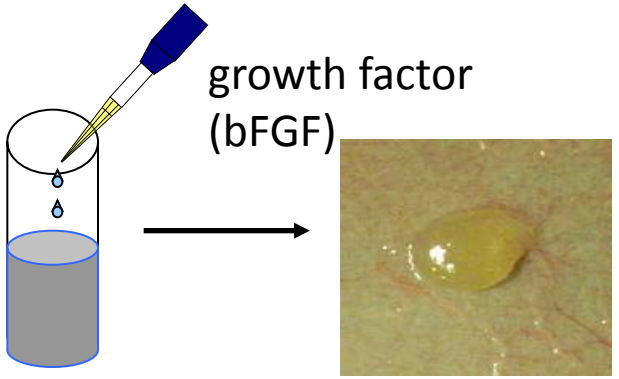
Mechanically Bio-mimicking Hydrogels



4 C

37 C

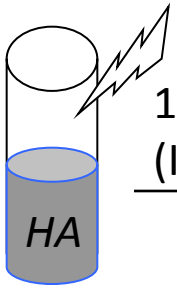
Heat-crosslinkable hydrogels (Collagen)



Peptide

Self-assembled Peptide hydrogel

UV light (1 kW, 80 W/cm)



1% photoinitiator
(Irgacure 2959)

60 sec.

Photocrosslinkable hydrogels (HA)

5% meHA in PBS (methacrylated)

Aim 1

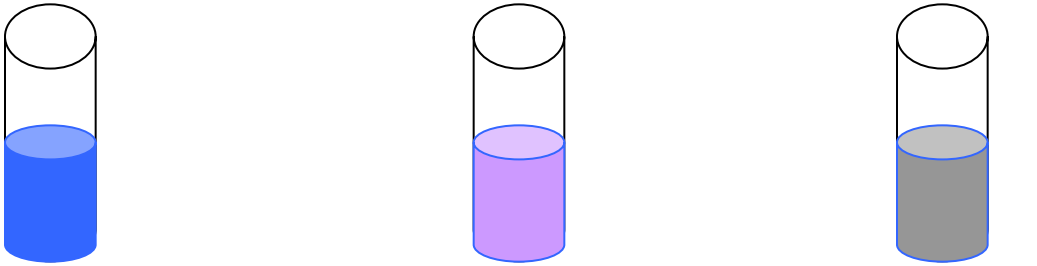
Mechanically and Biologically Bio-mimicking Hydrogels

To mimic the cellular microenvironment (ECM)

Aim 1

Mechanically and Biologically Bio-mimicking Hydrogels

To mimic the cellular microenvironment (ECM)

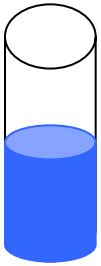


Collagen HA Peptide

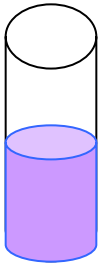
Aim 1

Mechanically and Biologically Bio-mimicking Hydrogels

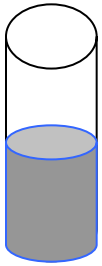
To mimic the cellular microenvironment (ECM)



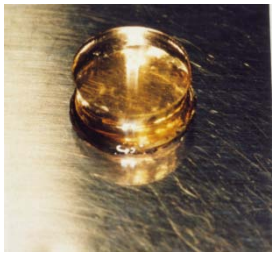
Collagen

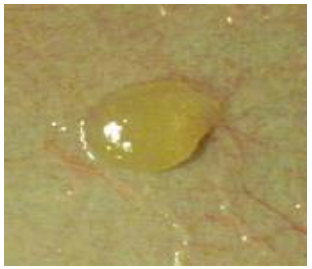


HA



Peptide

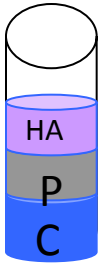




Aim 1

Mechanically and Biologically Bio-mimicking Hydrogels

To mimic the cellular microenvironment (ECM)



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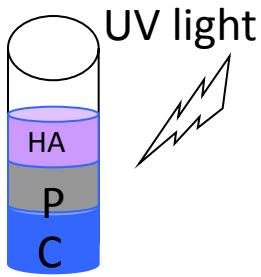
Mechanically and Biologically Bio-mimicking Hydrogels

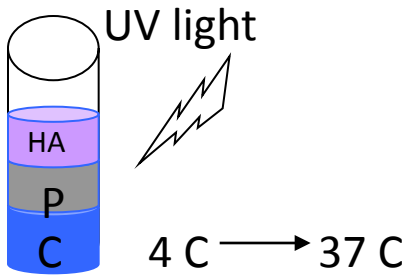
To mimic the cellular microenvironment (ECM)



Aim 1

Mechanically and Biologically Bio-mimicking Hydrogels

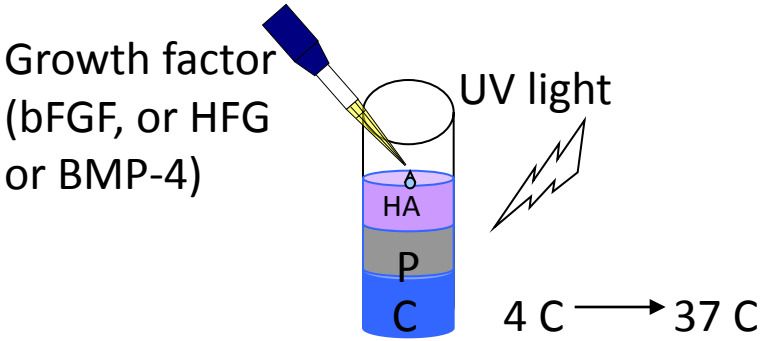
To mimic the cellular microenvironment (ECM)



Aim 1

Mechanically and Biologically Bio-mimicking Hydrogels

To mimic the cellular microenvironment (ECM)

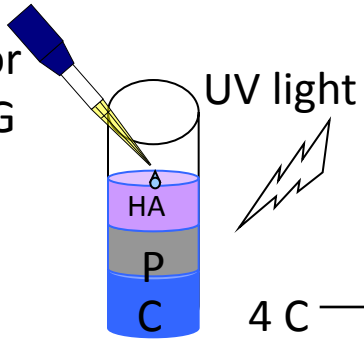


Aim 1

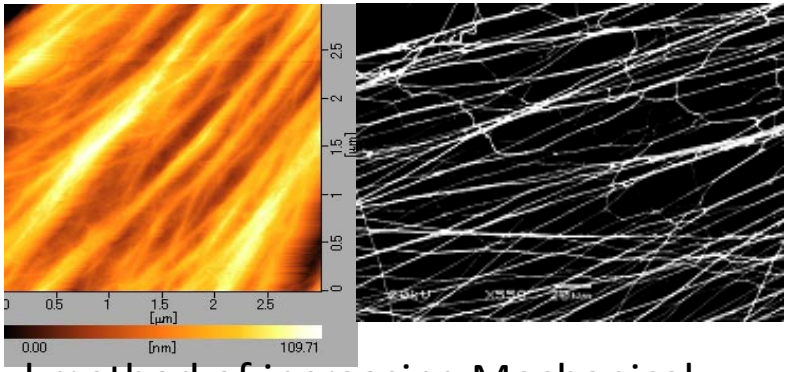
Mechanically and Biologically Bio-mimicking Hydrogels

To mimic the cellular microenvironment (ECM)

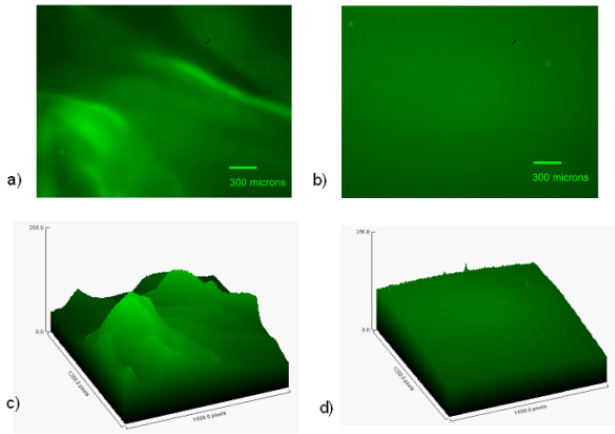
Growth factor (bFGF, or HFG or BMP-4)



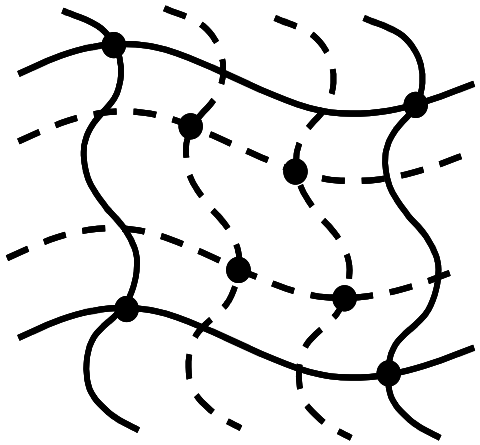
Interpenetrating Networks (IPNs) Hydrogels



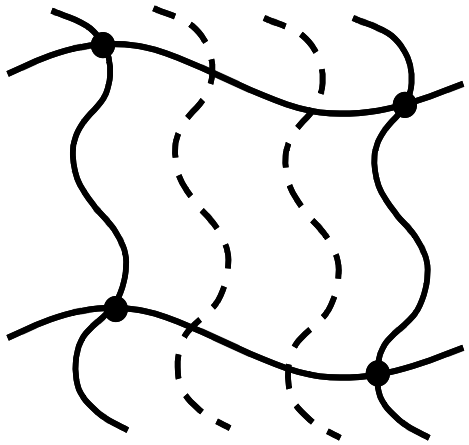
The fluorescent image of FITC-labeled collagen-peptide-HA



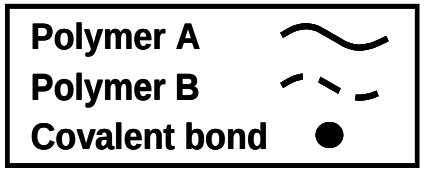
IPNs is a powerful method of increasing Mechanical Properties of Hydrogel, while retaining elasticity.



(a) Interpenetrating network



(b) Semi-interpenetrating network



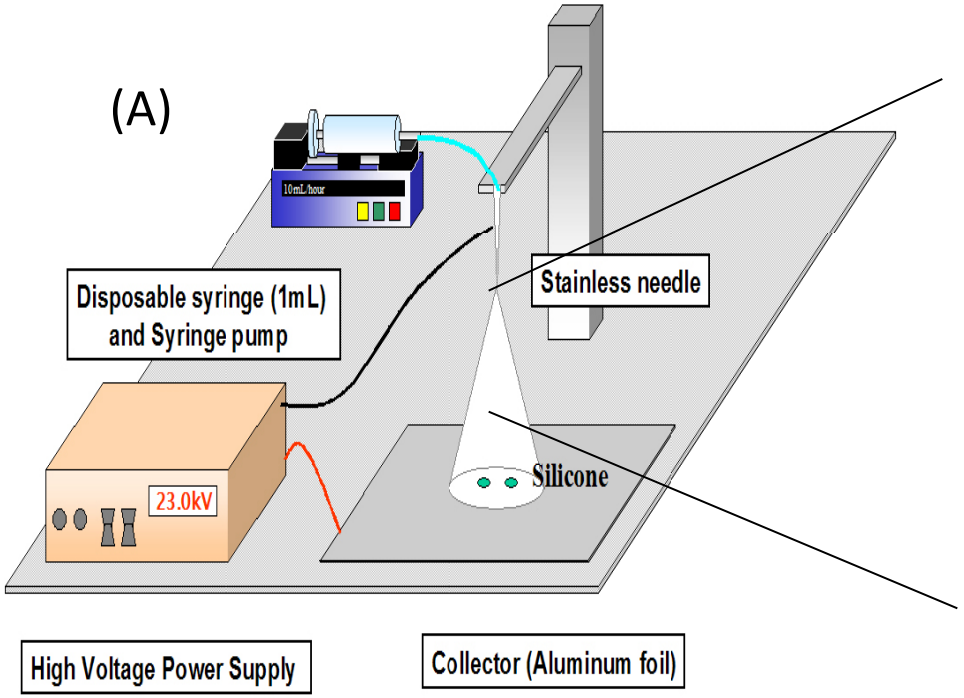
In order to mimic the biological function of ECM proteins, the scaffold materials used in tissue engineering need to be chemically functionalized to promote tissue regeneration as ECM does. Collagen and elastin as ECM proteins are made from fibers in dimension smaller than micrometers. It seems that artificial nanoscaled fibers have great potential application in the field of biomaterials and tissue engineering.

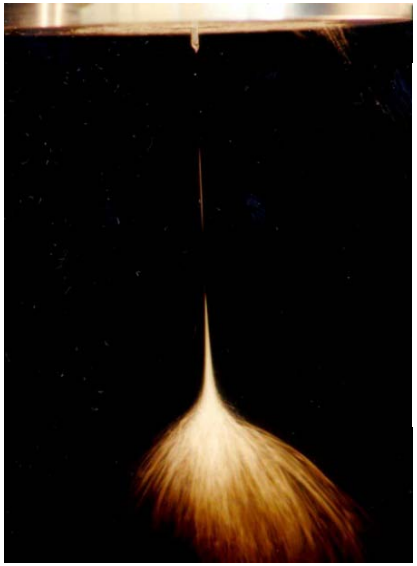
There are three different approaches toward the formation of nanofibrous materials:

1. Phase separation
2. Electrospinning
3. Self-assembly

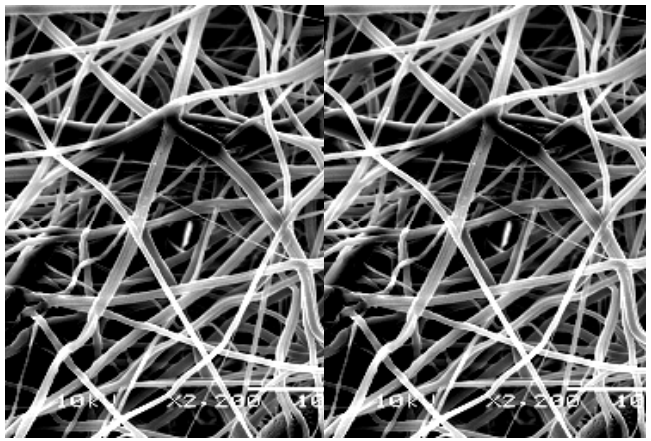
Phase separation and self assembling of biomolecules can generate smaller diameter nanofibers in the same range of natural ECM, while electrospinning generate large diameter nanofibers on the upper end of the range of natural ECM

(A)

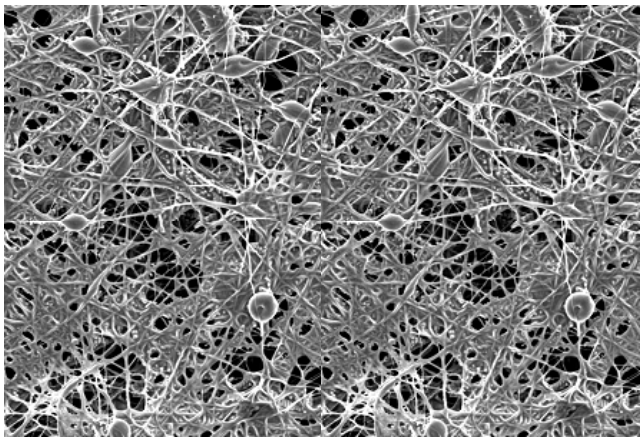

10mL/hour
Disposable syringe (1mL) and Syringe pump
Stainless needle
23.0kV
Silicone
High Voltage Power Supply
Collector (Aluminum foil)



(B)



(C)



Applications of electrospun nanofibres

Filtration

Filtration is necessary in many engineering fields. Fibrous materials used for filter media provide advantages of high filtration efficiency and low air resistance. Filtration efficiency, which is closely associated with the fibre fineness, is one of the most important concerns for the filter performance.

Protective clothing

The protective clothing in military is mostly expected to help maximize the survivability, sustainability, and combat effectiveness of the individual soldier system against extreme weather conditions, ballistics, and NBC (nuclear, biological, and chemical) warfare. In peace ages, breathing apparatus and protective clothing with the particular function of against chemical warfare agents such as sarin, soman, tabun and mustard gas from inhalation and absorption through the skin become special concern for combatants in conflicts and civilian populations in terrorist attacks.

Catalytic nanofibres

Chemical reactions employing enzyme catalysts are important in chemical processes due to their high selectivity and mild reaction conditions. Immobilised enzymes are used largely due to easiness of catalyst separation, enzyme stability, and their availability for continuous operations. The efficiency of these immobilised enzymes depends mainly on the pore structure and diffusion limitations of the substrate material. Nanomaterials are of recent interest as catalyst substrates due to their large surface area per unit mass and the feasibility for high catalyst loading.

Composite application

One of the most important applications of traditional (micro-size) fibres, especially engineering fibres such as carbon, glass, and Kevlar fibres, is to be used as reinforcements in composite developments. With these reinforcements, the composite materials can provide superior structural properties such as high modulus and strength to weight ratios, which generally cannot be achieved by other engineered monolithic materials alone. Needless to say, nanofibres will also eventually find important applications in making nanocomposites. This is because nanofibres can have even better mechanical properties than micro fibres of the same materials, and hence the superior structural properties of nanocomposites can be anticipated.

Electrical and optical application

Conductive nanofibres are expected to be used in the fabrication of tiny electronic devices or machines such as schottky junctions, sensors and actuators. Due to the well-known fact that the rate of electrochemical reactions is proportional to the surface area of the electrode, conductive nanofibrous membranes are also quite suitable for using as porous electrode in developing high performance battery. Conductive (in terms of electrical, ionic and photoelectric) membranes also have potential for applications including electrostatic dissipation, corrosion protection, electromagnetic interference shielding, photovoltaic device, etc.

Medical application

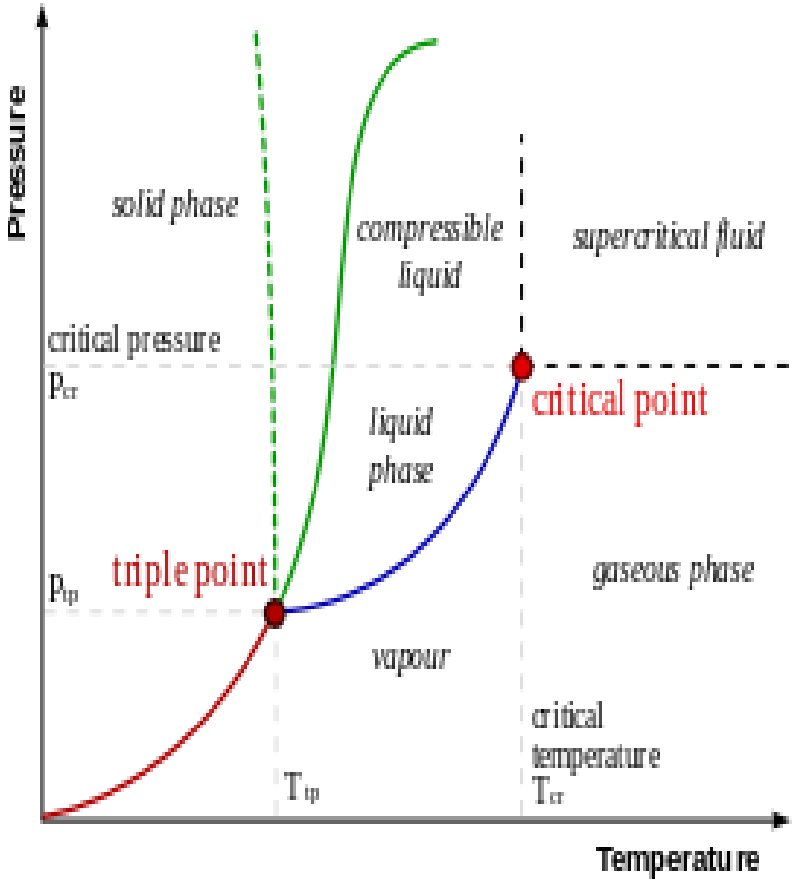
Nanofibres are also used in the medical applications which include, drug and gene delivery, artificial blood vessels, artificial organs, and medical facemasks. Electrospun biocompatible polymer nanofibres can be deposited as a thin porous film onto a hard tissue prosthetic device designed to be implanted into the human body. This coating film is expected to efficiently reduce the stiffness mismatch at the tissue interphase and hence prevent the device failure after the implantation. Nanofibres and webs are capable of delivering medicines directly to internal tissues. Anti-adhesion materials may be used for such applications. Nanofibre, spun from compounds naturally present in blood, can be used as bandages or sutures that ultimately dissolve into body. This nanofibre minimize infection rate, blood loss and is also absorbed by the body.

Phase separation

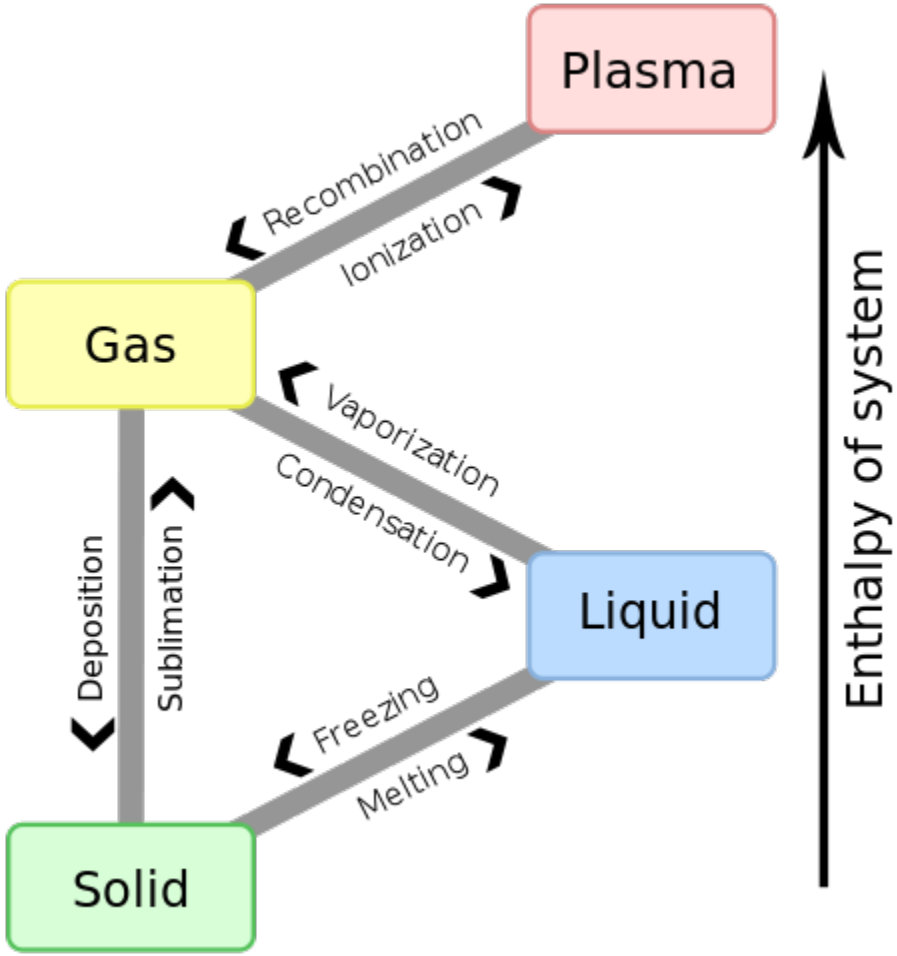
In the physical sciences, a **phase** is a region of space (a thermodynamic system), throughout which all physical properties of a material are essentially uniform.

Examples of physical properties include density, index of refraction, magnetization and chemical composition.

A simple description is that a phase is a region of material that is chemically uniform, physically distinct, and (often) mechanically separable. In a system consisting of ice and water in a glass jar, the ice cubes are one phase, the water is a second phase, and the humid air over the water is a third phase. The glass of the jar is another separate phase



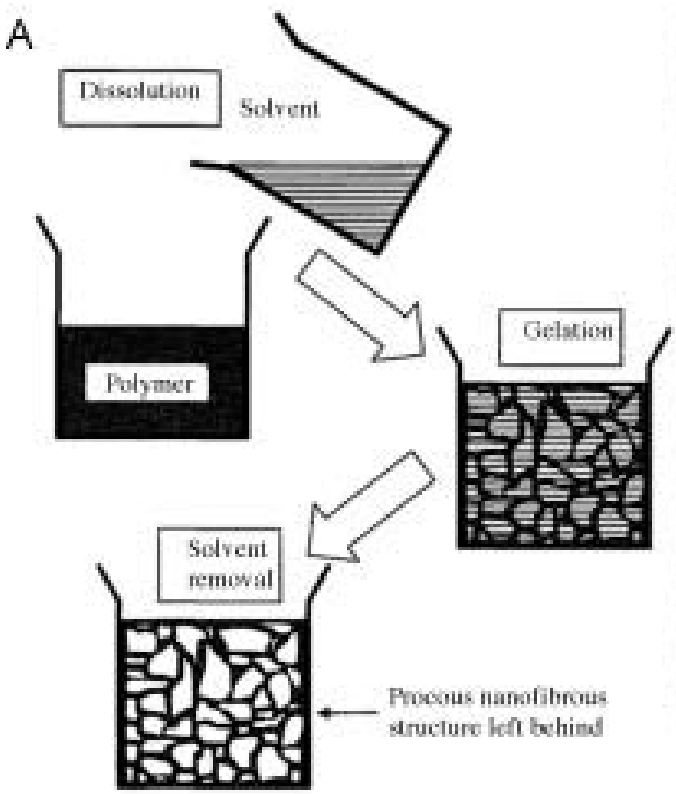
A typical phase diagram for a single-component material, exhibiting solid, liquid and gaseous phases. The solid green line shows the usual shape of the liquid–solid phase line. The dotted green line shows the anomalous behavior of water.

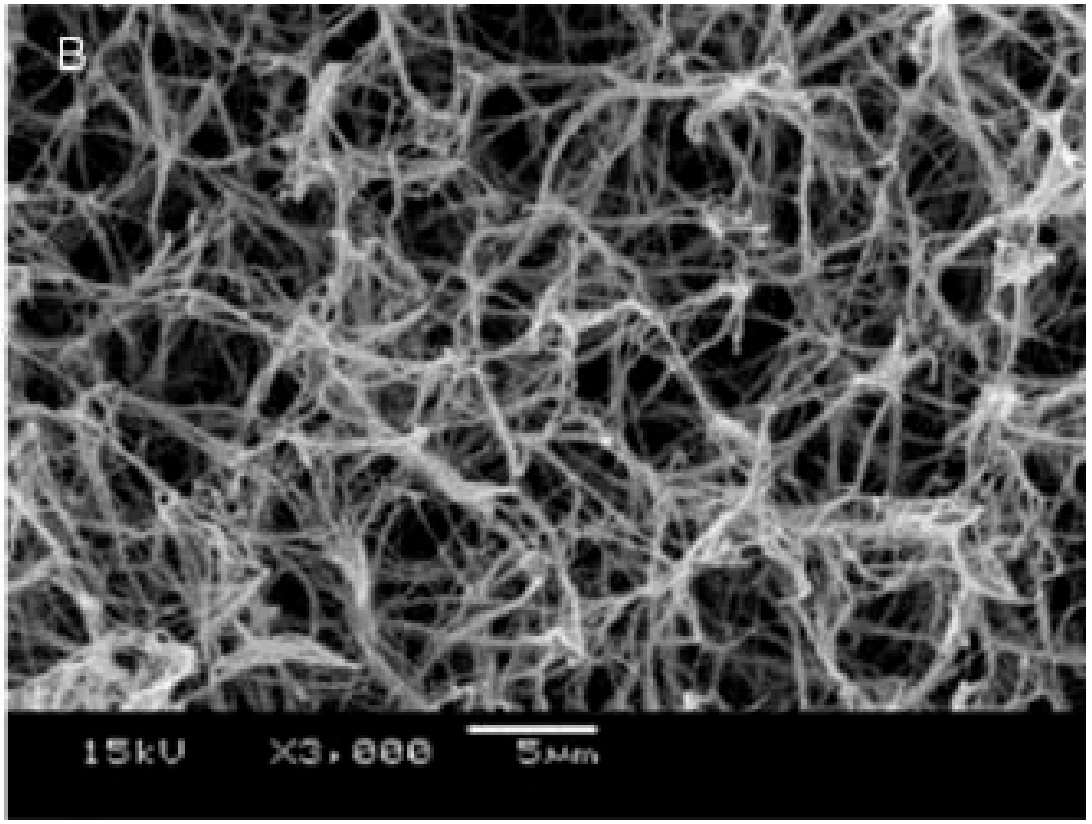

Plasma
Recombination
Ionization
Gas
Vaporization
Condensation
Deposition
Sublimation
Liquid
Freezing
Melting
Solid
Enthalpy of system

The nanofibrous foams produced using the phase separation technique are very similar in size to the natural collagen present in the ECM of tissue in terms of their size (50–500 nm)

This technique involves five basic steps :

1. Dissolution of polymer.
2. Liquid–liquid phase separation process.
3. Polymer gelation (controls the porosity of nanoscale scaffolds at low temperature).
4. Extraction of solvent from the gel with water.
5. Freezing and freeze-drying under vacuum.


A
Dissolution
Solvent
Polymer
Gelation
Solvent removal
Porous nanofibrous structure left behind


B
15kV
X3,000
5μm