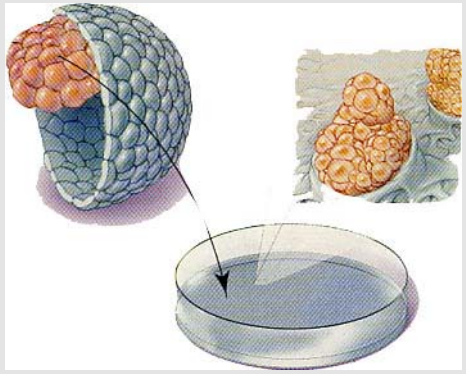
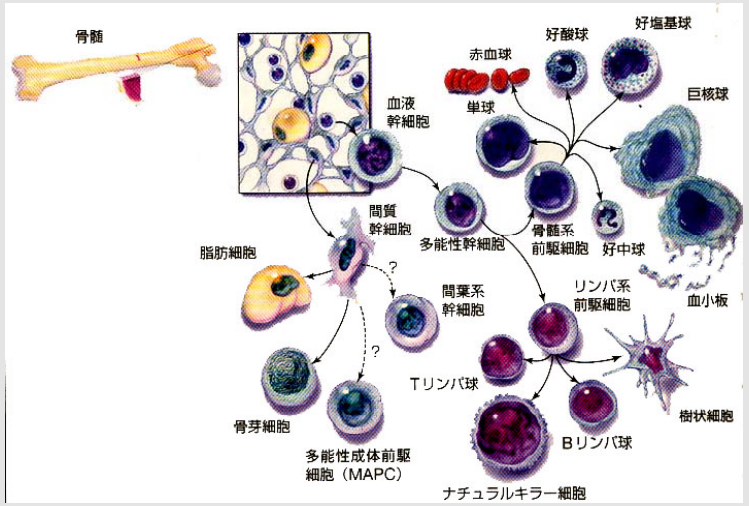
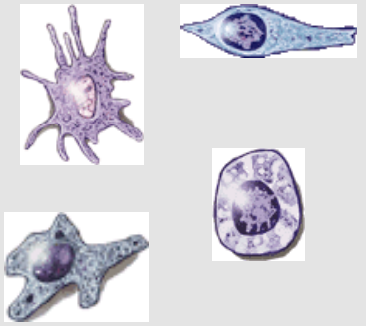
“Tissue Engineering

- Cartilage Regeneration –“

Takashi USHIDA
(Mechano-bioengineering)



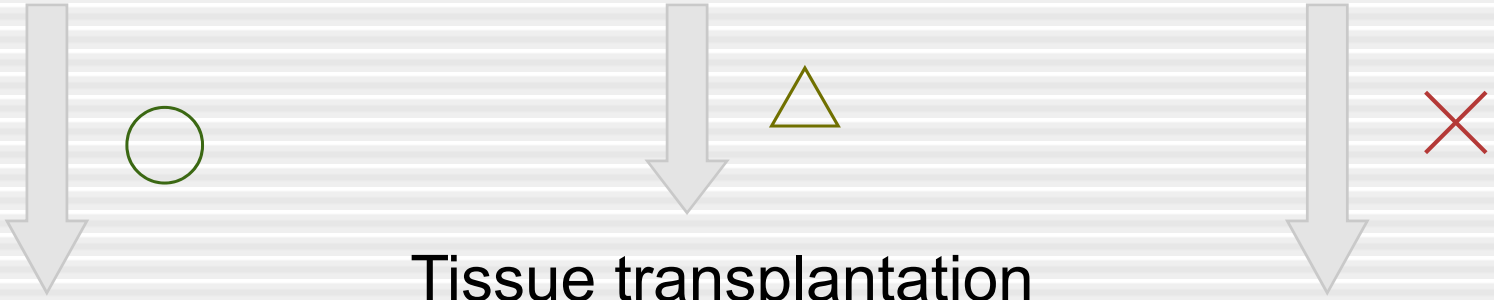




ES Cell

Bone Marrow Stem Cell

Matured Cell



Cell transplantation

Tissue transplantation

Organ transplantation

1. Cell sources

2. 3 dimensional scaffolds

Cell source

From who ?

1. Autologous

2. Allogeneic

3. Heterologous

1. Autologous

merit

- No immuno-rejection
- No problem concerning virus

demerit

- Limitation of cell sources
- Low activity of cells

2. Allogeneic

merit

- Higher activity of cells
- Capable of virus check

demerit

- Immuno-rejection
- Ethical problems in some cases

3. Heterologous

merit

- No limitation of cell sources
- Higher activity of cells

demerit

- Immuno-rejection
- Risk of unknown virus

Cell source

From where ?

1. ES cell
2. Bone marrow cell
3. Somatic stem cell
4. Matured cell

1. ES cell

ES (Embryonic Stem) cell

Stem cell
- Self renewal
- Multipotential

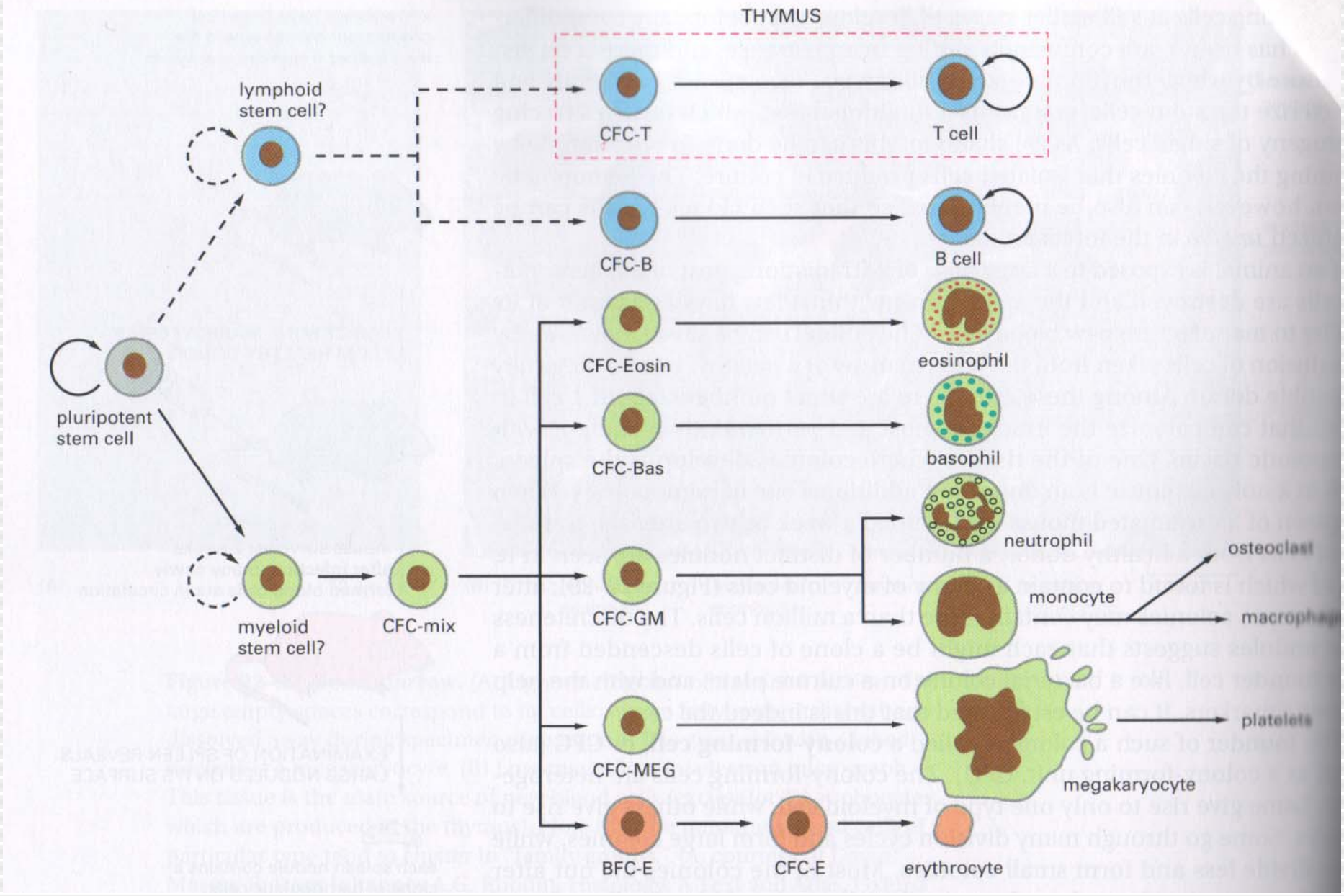
ES cell
- Self renewal
- Pluripotential

- Allogenic
- Ethical problem by broking zygocyte

Bone Marrow Derived Stem Cells

1. Hematopoetic Stem Cell

2. Mesenchymal Stem Cell



Differentiation of Hematopoetic Stem Cell

3. Somatic Stem Cell

Stem cells located in tissues

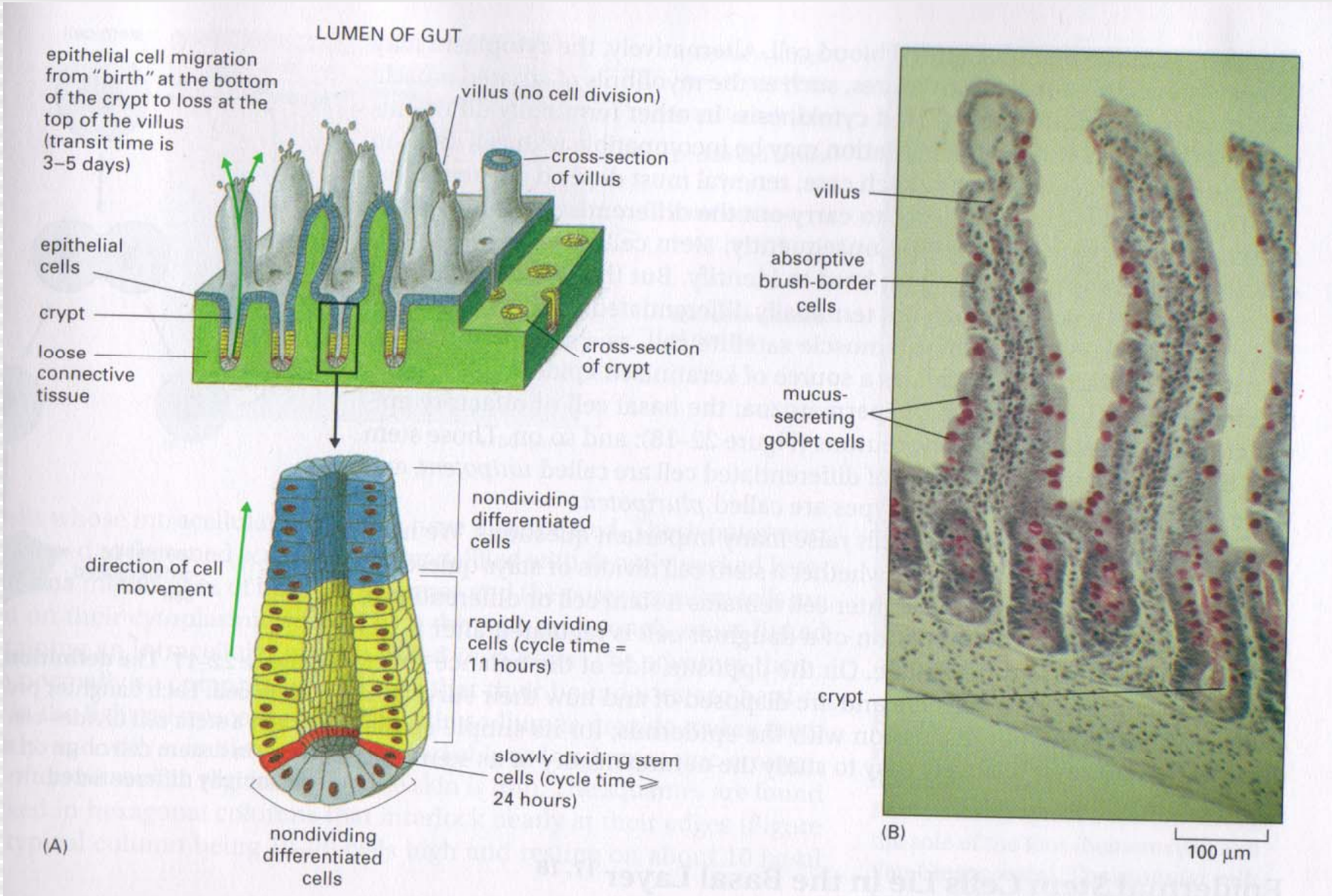
1）Neural Stem Cell

2）Epidermal Stem Cell

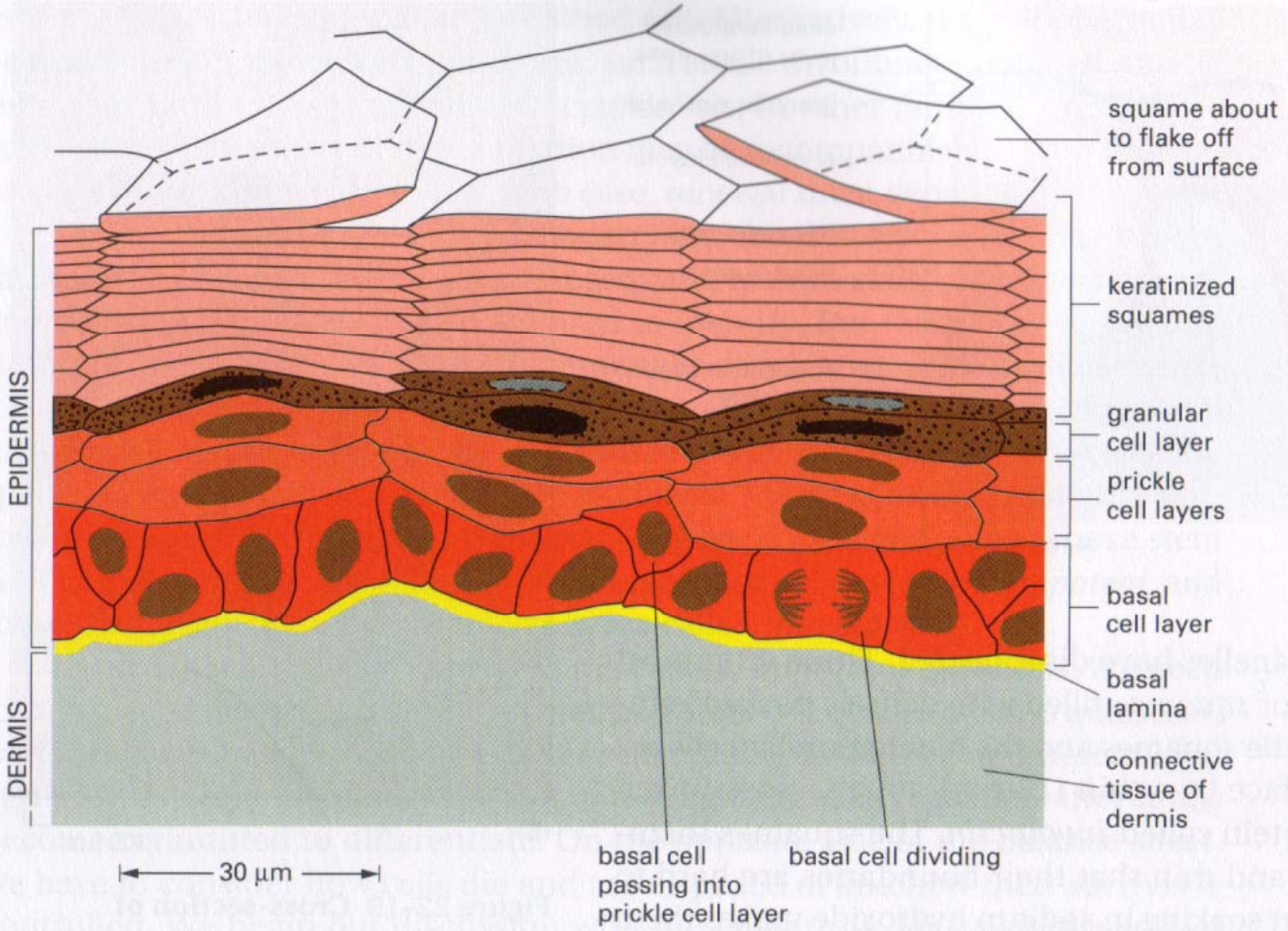
3）Hepatic Stem Cell

4）Small Intestine Epithelial Stem C

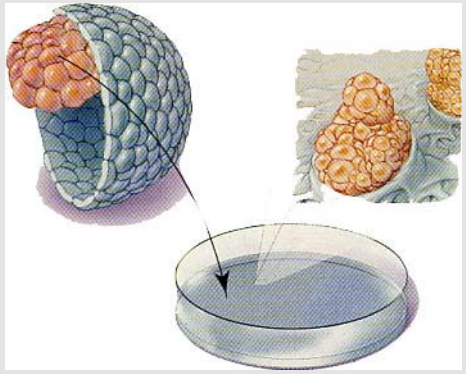
5）Hair Root Stem Cell

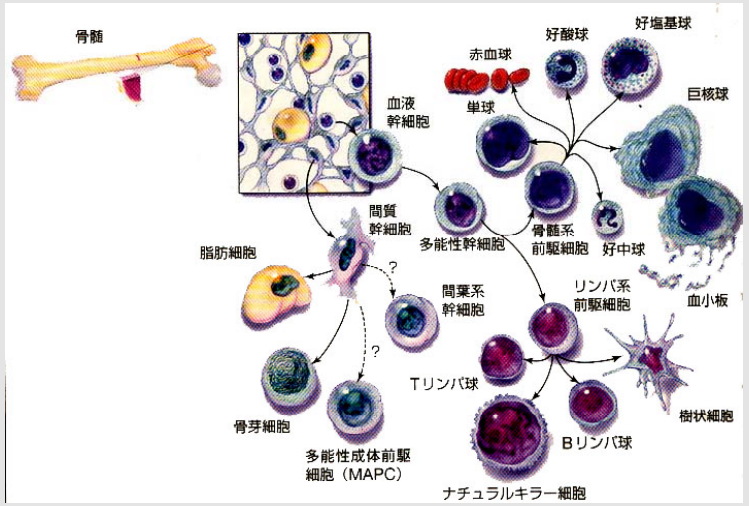


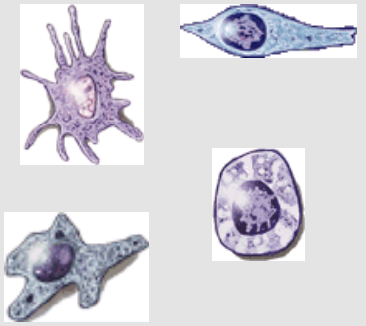
Small Intestine Epithelial Stem Cell



Epidermal Stem Cell



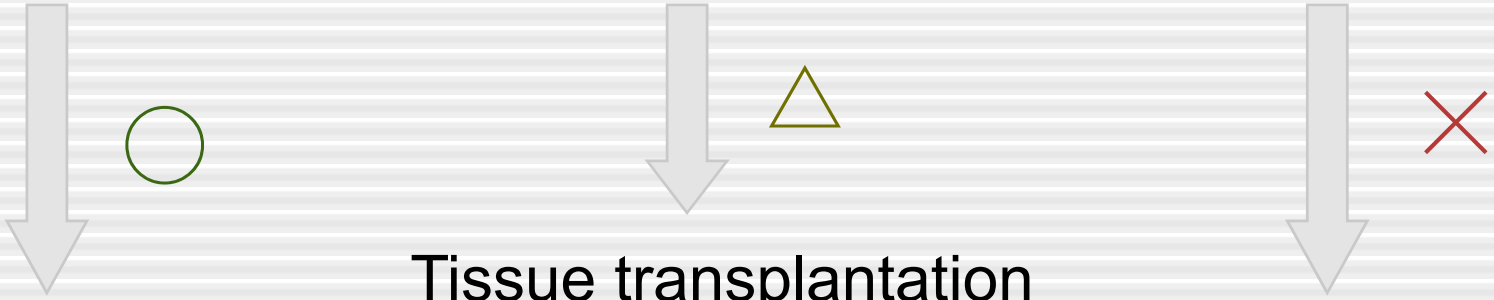




ES Cell

Bone Marrow Stem Cell

Matured Cell



Cell transplantation

Tissue transplantation

Organ transplantation

1. Cell sources
2. 3 dimensional scaffolds

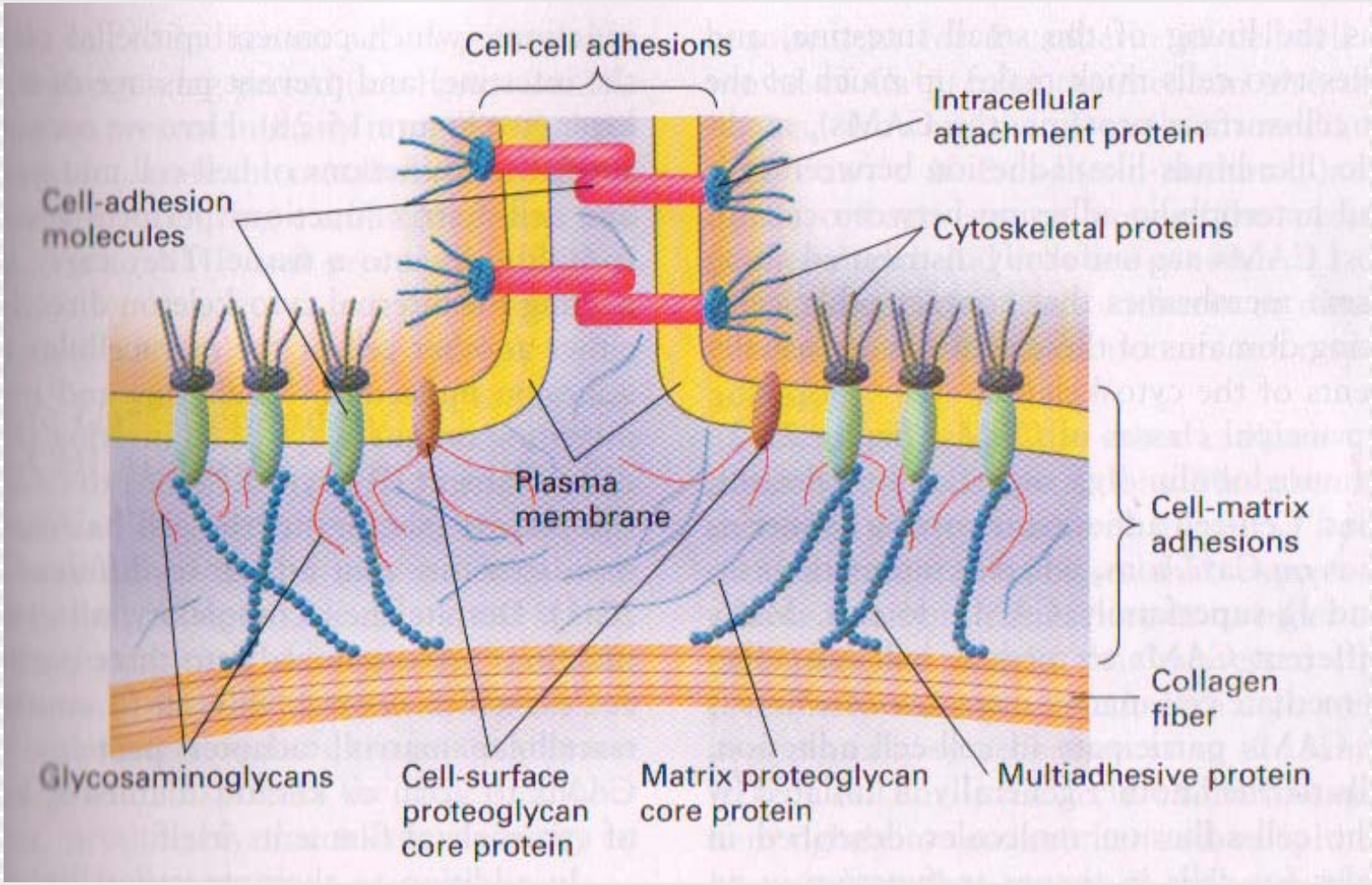
Biomaterial

Collagen, Hyaluronan, Laminin, Elastin

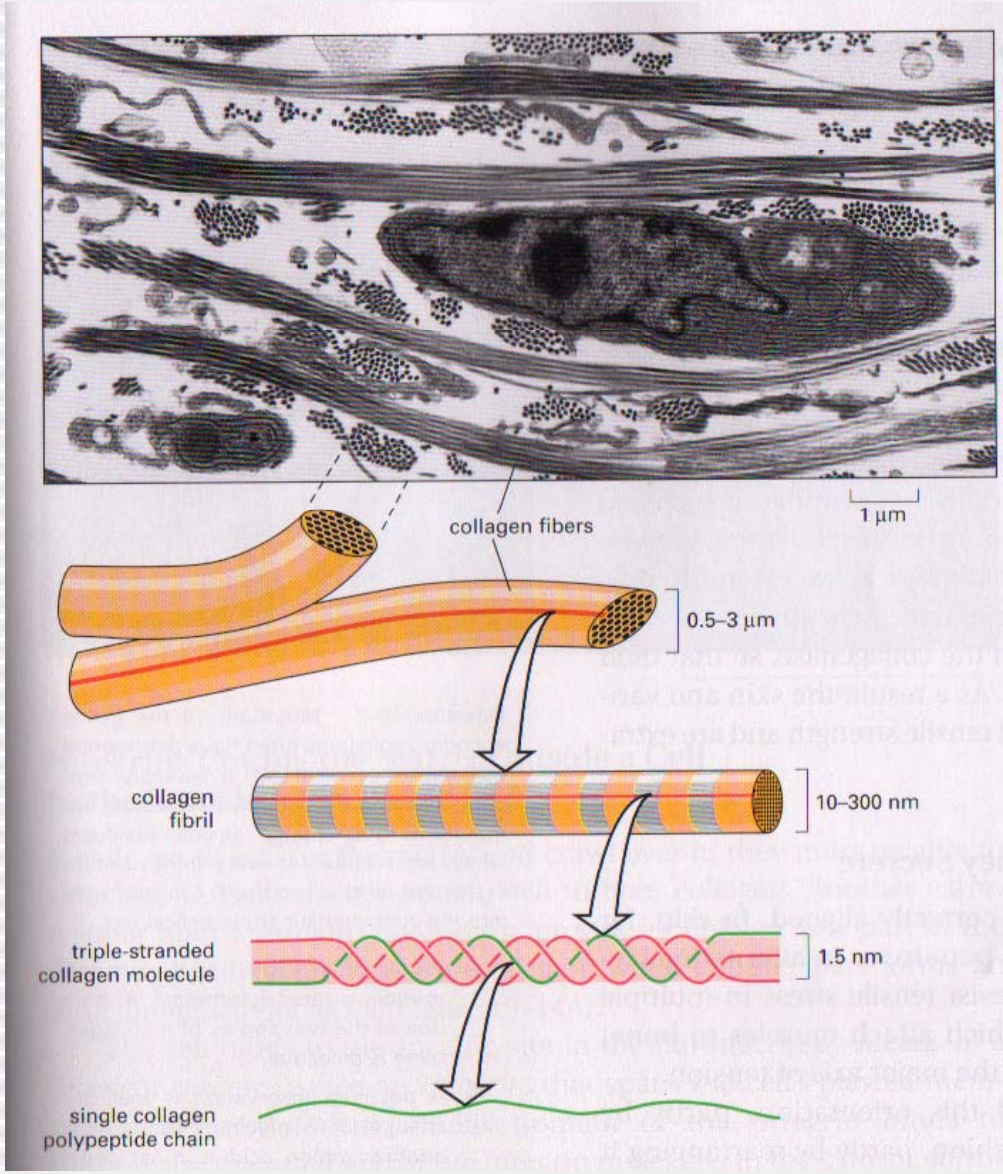
Biodegradable polymer

PLLA, PGA, PLGA

Biopolymers Used in the Field of Tissue Engineeri



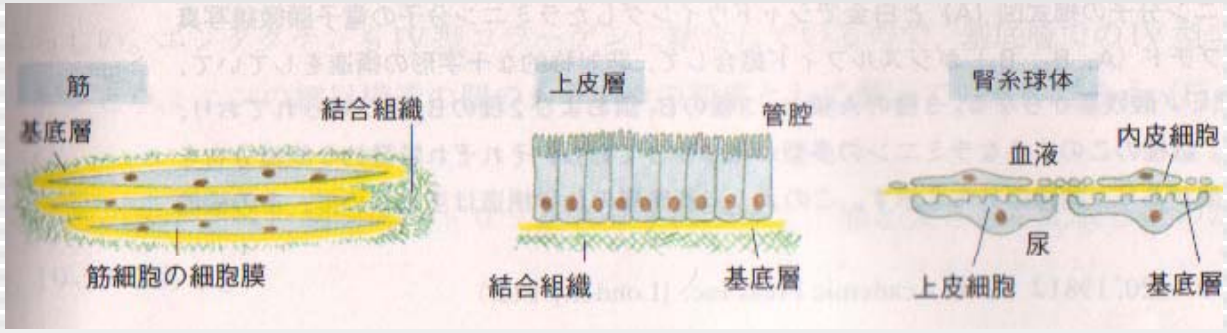
Cell - cell interaction
Cell - matrix interaction



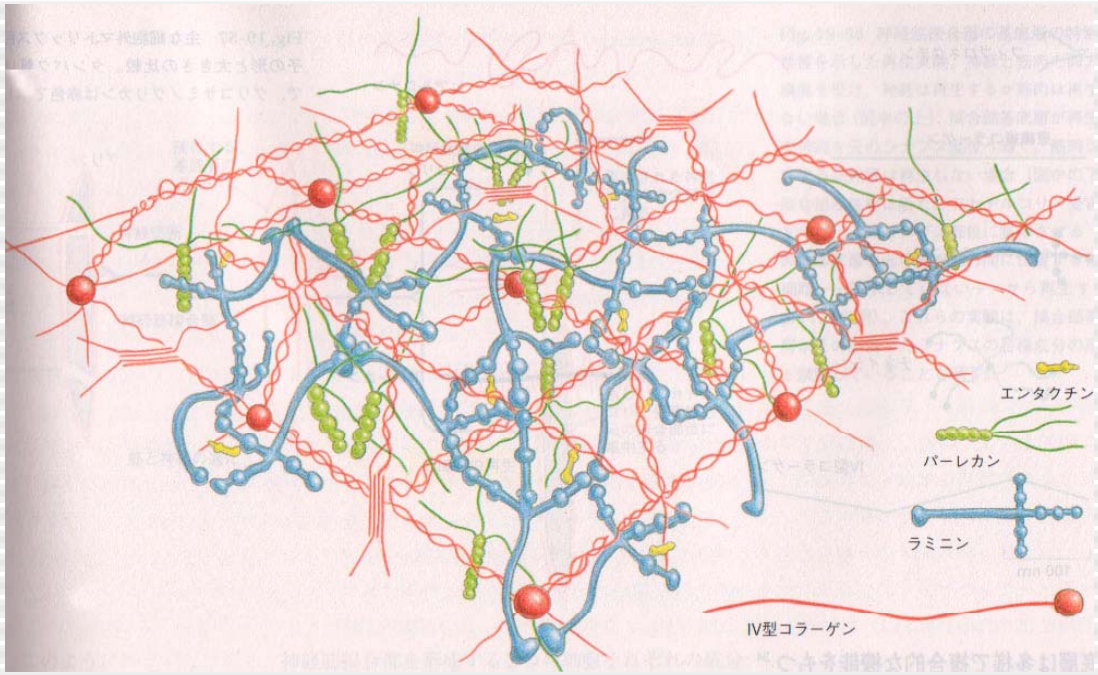
Collagen Type I

	Type	Molecular Formula	Polymerized Form	Tissue Distribution
FIBRIL-FORMING (FIBRILLAR)	I	$[\alpha1(I)]_2\alpha2(I)$	fibril	bone, skin, tendon, ligaments, cornea, internal organs (accounts for 90% of body collagen)
	II	$[\alpha1(II)]_3$	fibril	cartilage, intervertebral disc, notochord, vitreous humor of the eye
	III	$[\alpha1(III)]_3$	fibril	skin, blood vessels, internal organs
	V	$[\alpha1(V)]_2\alpha2(V)$	fibril (with type I)	as for type I
	XI	$\alpha1(XI)\alpha2(XI)\alpha3(XI)$	fibril (with type II)	as for type II
FIBRIL-ASSOCIATED	IX	$\alpha1(IX)\alpha2(IX)\alpha3(IX)$ with type II fibrils	lateral association	cartilage
	XII	$[\alpha1(XII)]_3$ with some type I fibrils	lateral association	tendon, ligaments, some other tissues
NETWORK-FORMING	IV	$[\alpha1(IV)_2\alpha2(IV)$	sheetlike network	basal laminae
	VII	$[\alpha1(VII)]_3$	anchoring fibrils	beneath stratified squamous epithelia

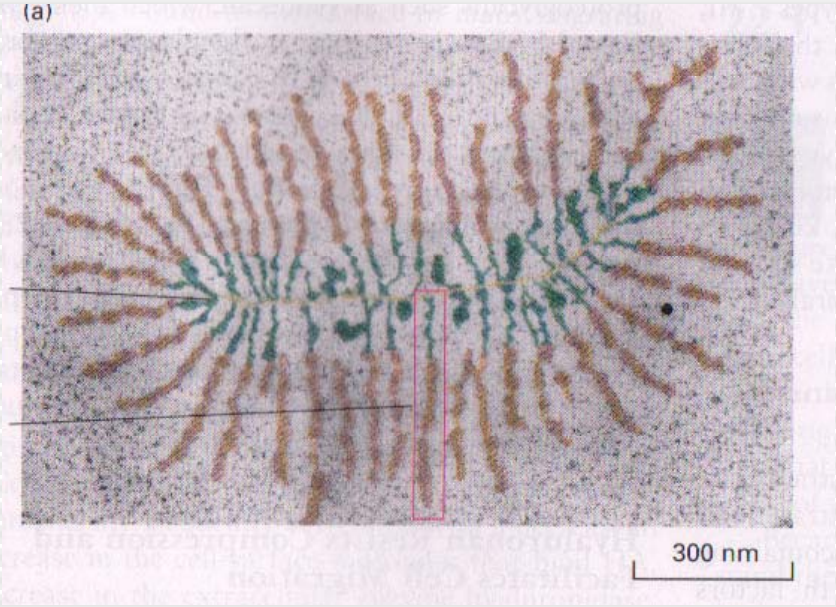
Types of Collagens

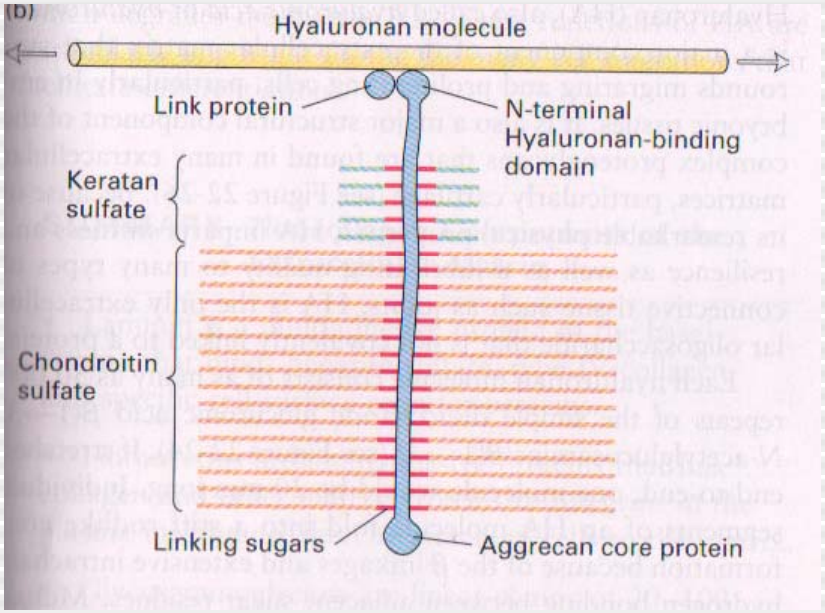


Basement membrane



Structure of basement membrane





Proteoglycan

Structure of proteoglycan

Biodegradable polymer

- ＰＬＬＡ （Polylactic acid） year
- ＰＬＧＡ （Polylactic-acid-polyglycolic-acid-copo
  month
- ＰＧＡ （polyglycolic acid） week

Biomaterial

Merit

- good biocompatibility
- having cell adhesion sites

Demerit

- difficult for forming
- weak mechanical properties

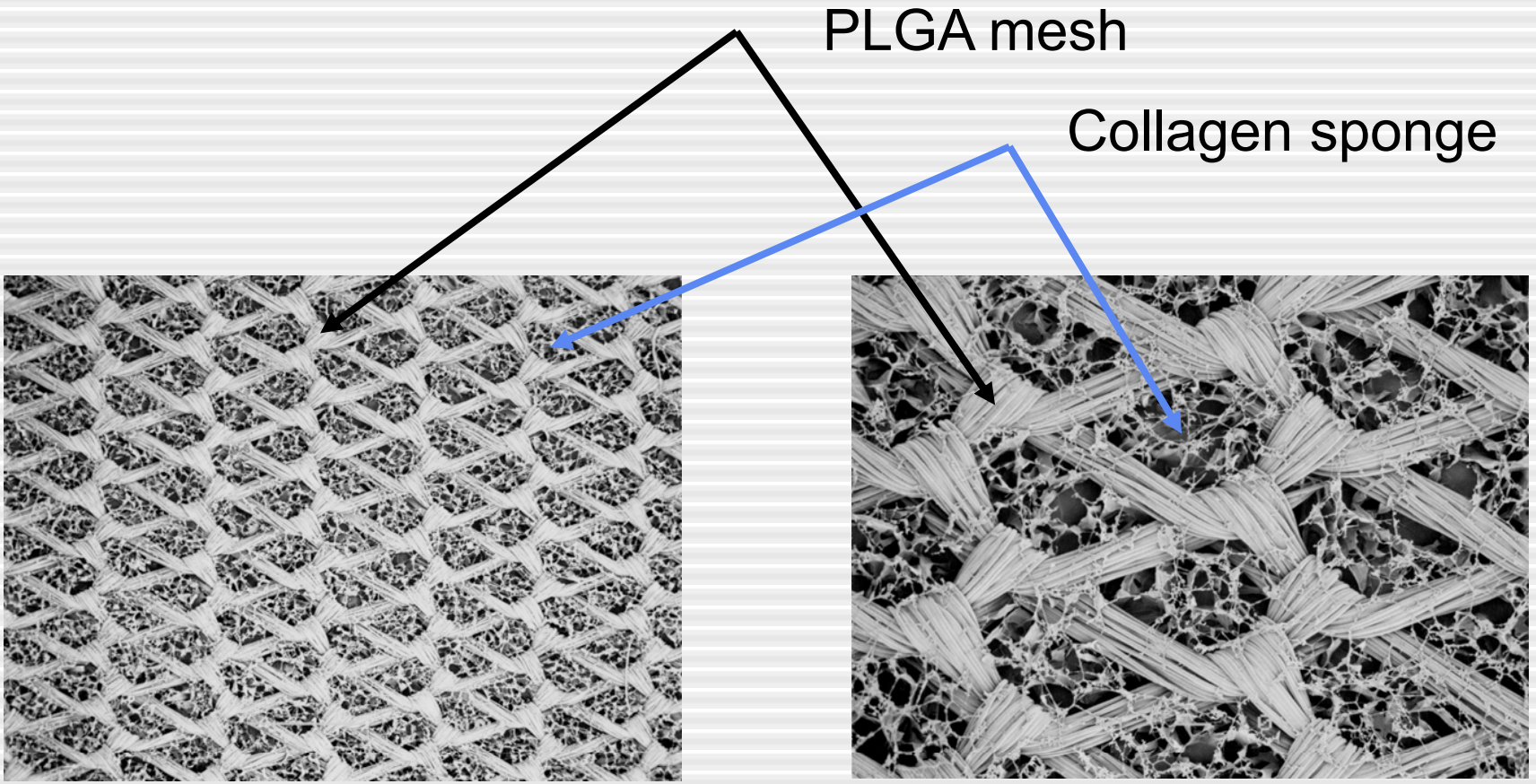
Biodegradable polymer

Merit

- easy for forming
- enough mechanical properties

Demerit

- hydrophobic
- no cell adhesion site



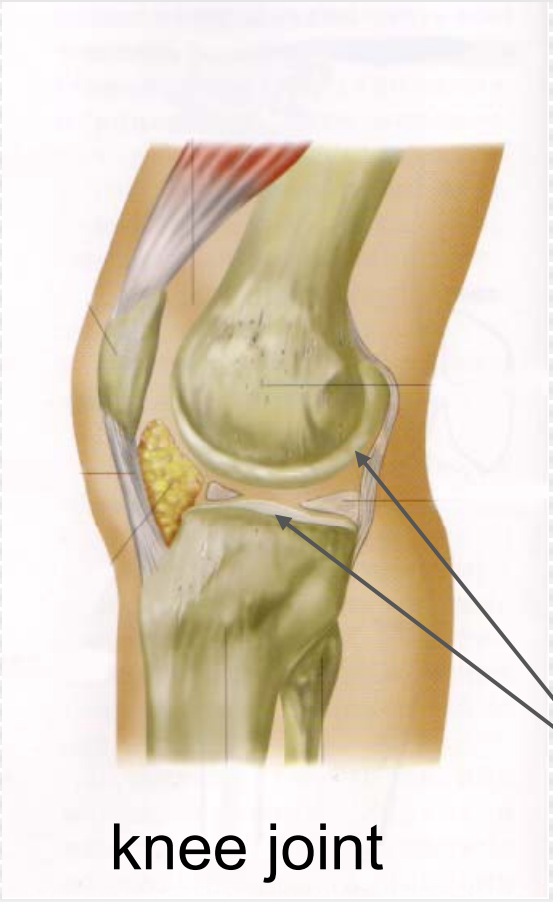
PLGA-collagen mesh at original magnification × 20

PLGA-collagen mesh at original magnification × 60

SEM photomicrographs of PLGA knitted mesh & its composite with collagen

Calcium Phosphates

Ca/P		formula	
0.5	Calcium diphosphate	$Ca(H_2PO_4)_2 \cdot H_2O$ α, β, γ -$Ca(PO_3)_2$	MCPM MTCP
1.0	Calcium monophosphate	$CaHPO_4 \cdot 2H_2O$ $CaHPO_4$ α, β, γ -$Ca_2P_2O_7$	DCPD DCP α, β, γ -CPP
1.34	Octa-calcium phosphate	$Ca_8H_2(PO_4)_6 \cdot 5H_2O$	OCP
1.5	Caicium triphosphate	α, β, γ -$Ca_3(PO_4)_2$	α, β, γ -TCP
1.67	Hydroxyapatite	$Ca_{10}(PO_4)_6 \cdot (OH)_2$	HAp,HA
2.0	Tetra-calciumphosphate	$Ca_4P_2O_9$	TTCP



knee joint

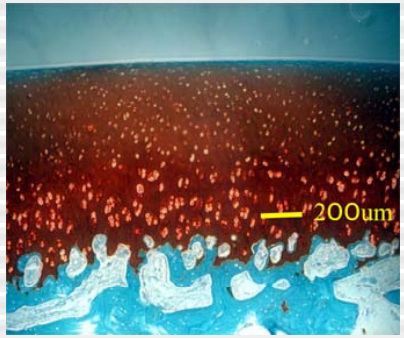
3 major diseases of articular cartilage

sport injuries

osteoarthritis (OA)

rheumatoid arthritis (RA)

Articular cartilage



Lateral femoral groove stained with S-O

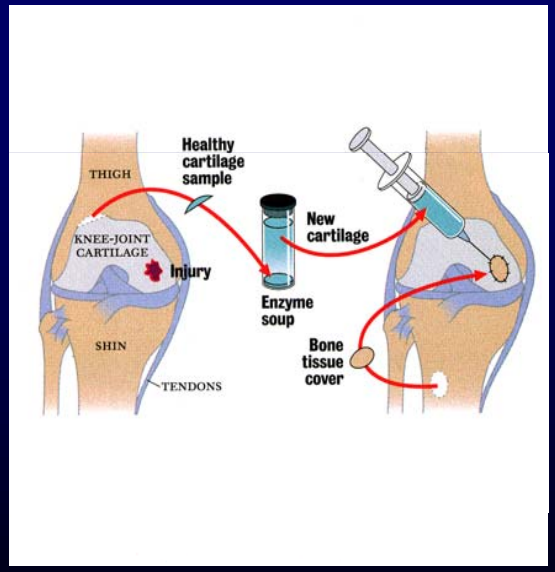
Type II collagen

(Type I collagen fibrous cartilage)

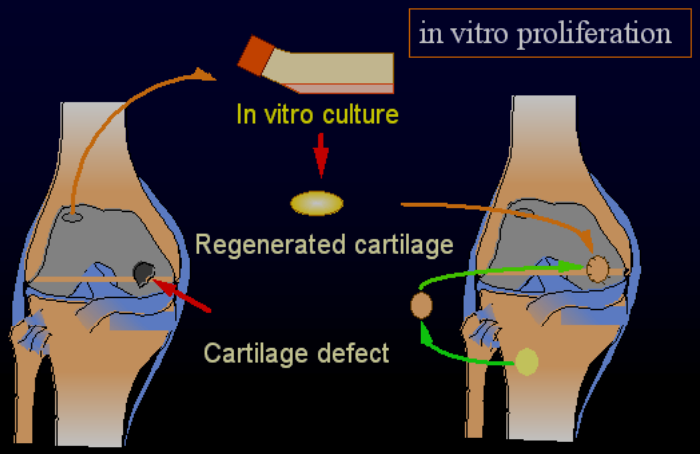
no blood vessel no nerve

↓

low regeneration activity



Clinical application of autologous chondrocyte implantation



Tissue engineered therapy for cartilage defect

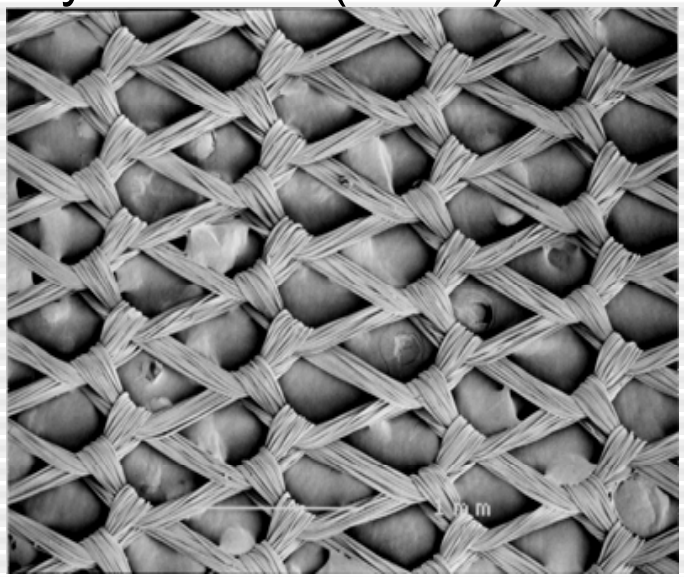
Cell Transplantation

↓

Regenerated Tissue Transplantation

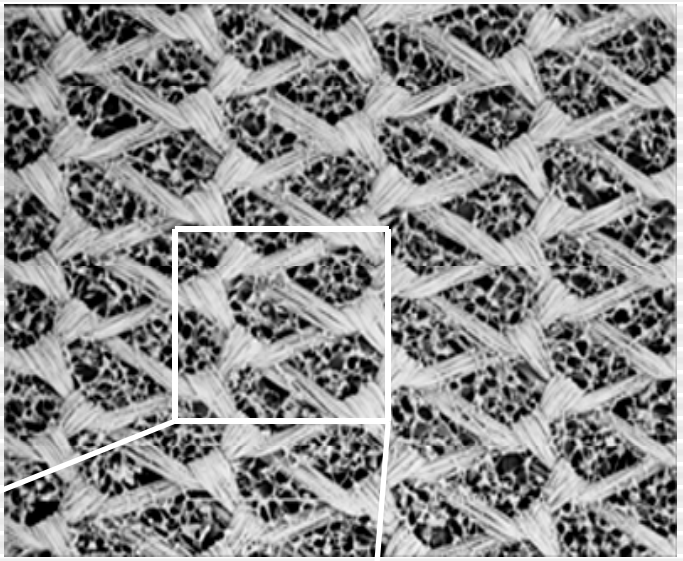
SEM Photomicrographs

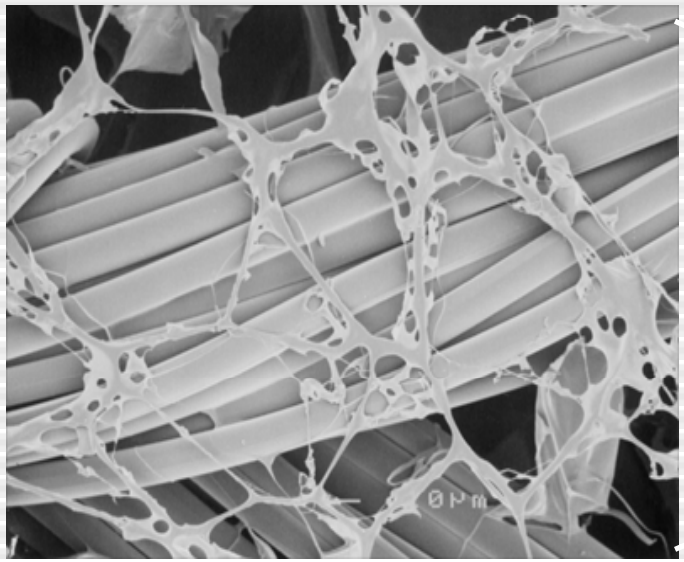
Polylactin 910 (PLGA) kitted mesh

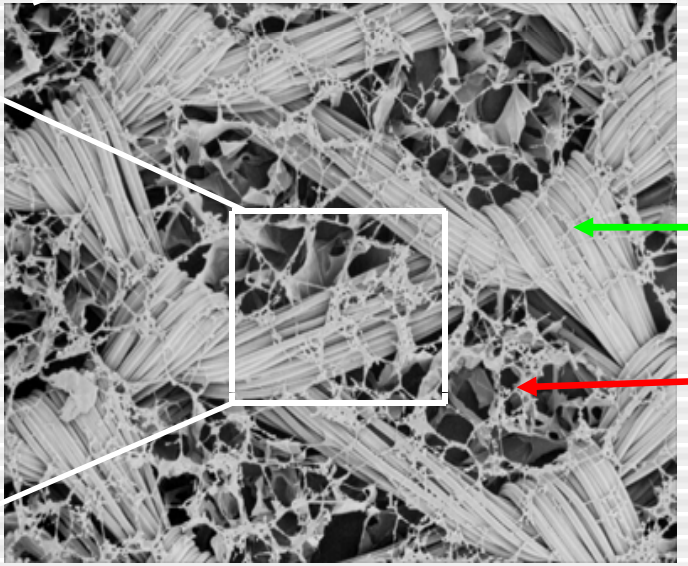


Hybridization

PLGA-collagen hybrid mesh





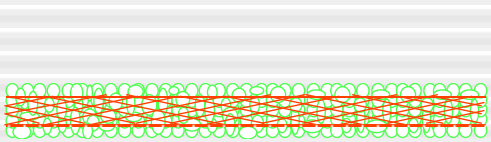


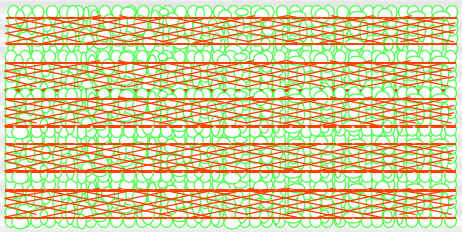
PLGA

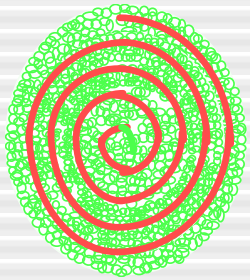
Collagen microsponges

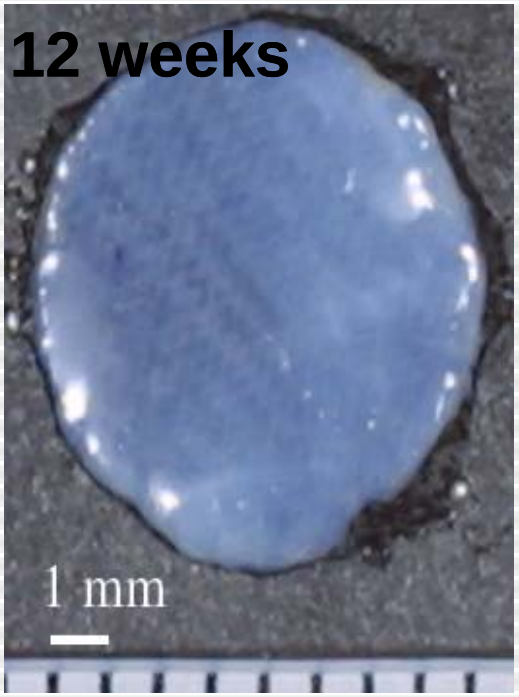
Gross Appearance

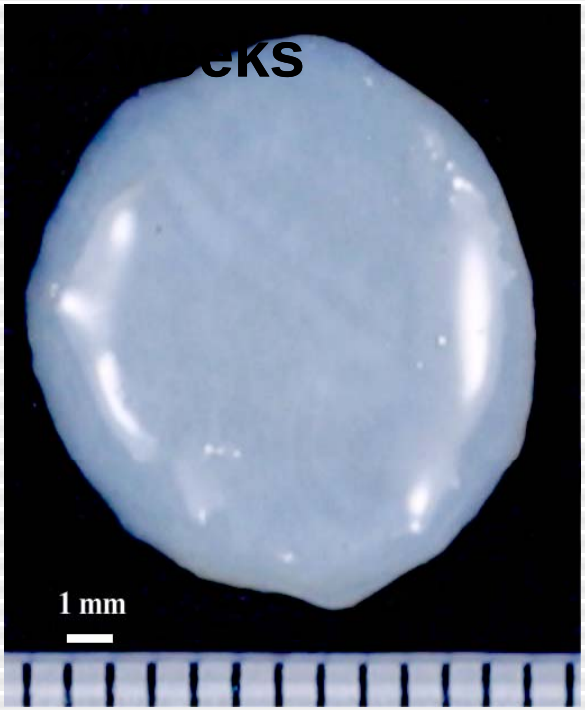
Single Sheet **Laminated (5-sheet)** **Rolled**

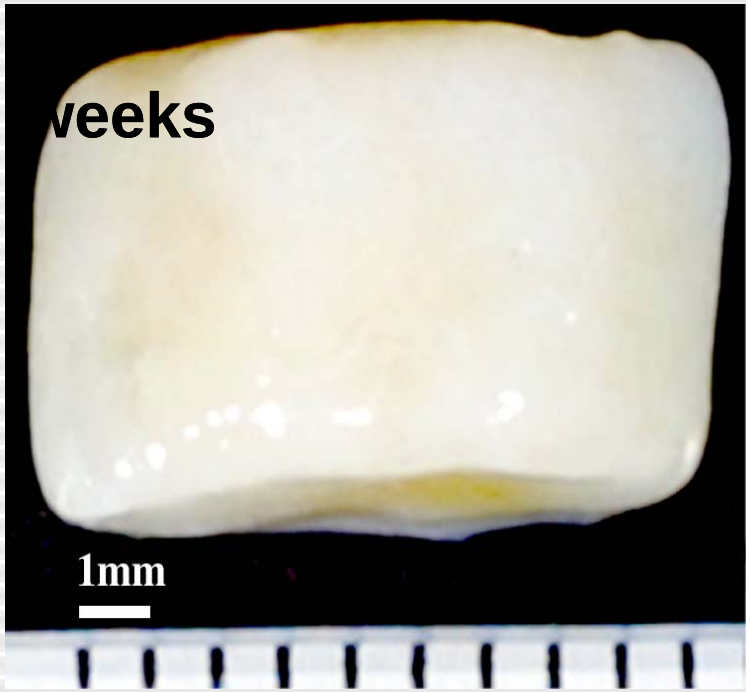






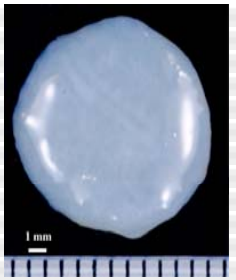






Thickness: 200 μm **1 mm** **8 mm**

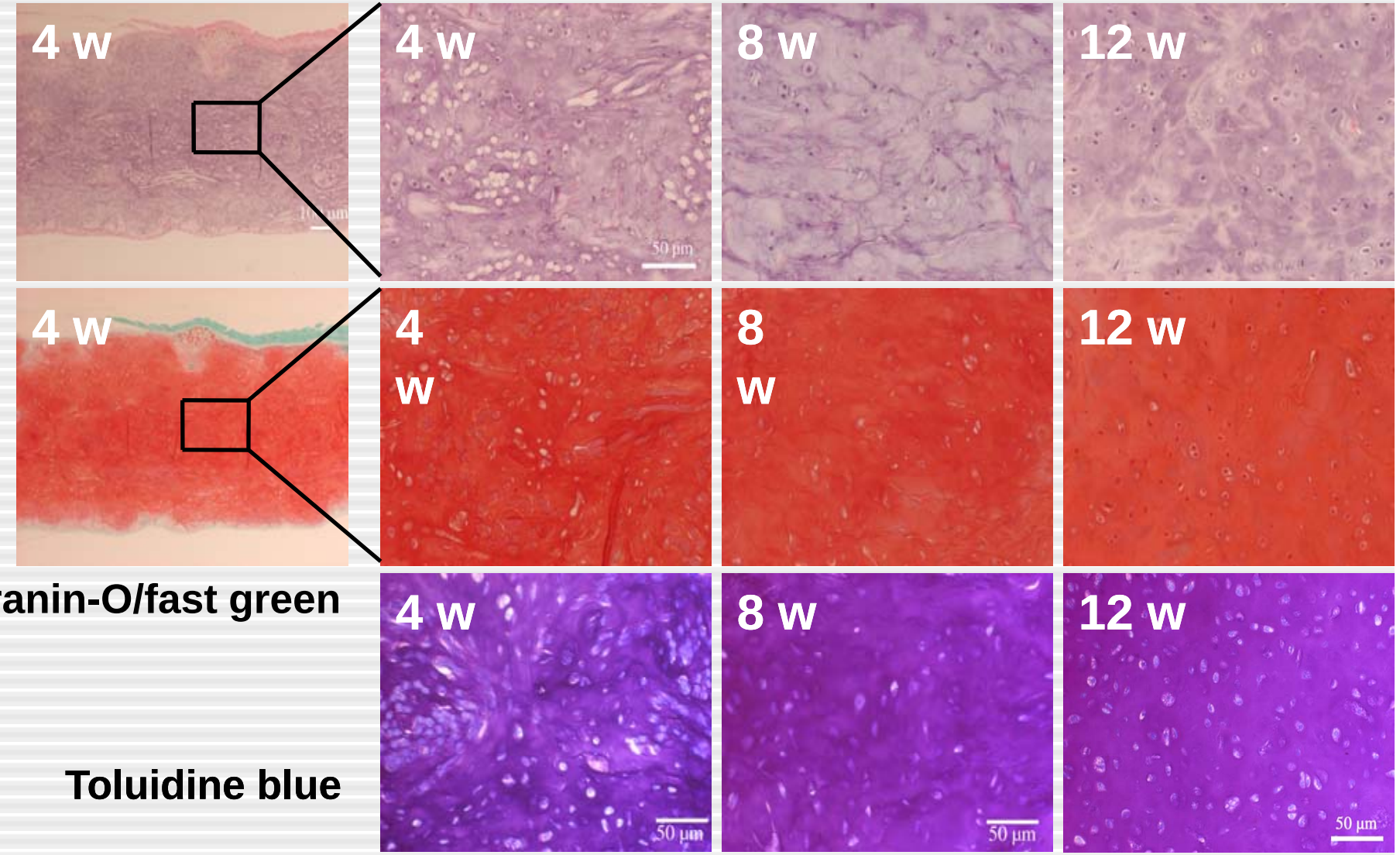
Histological Results of 5-Sheet Implant

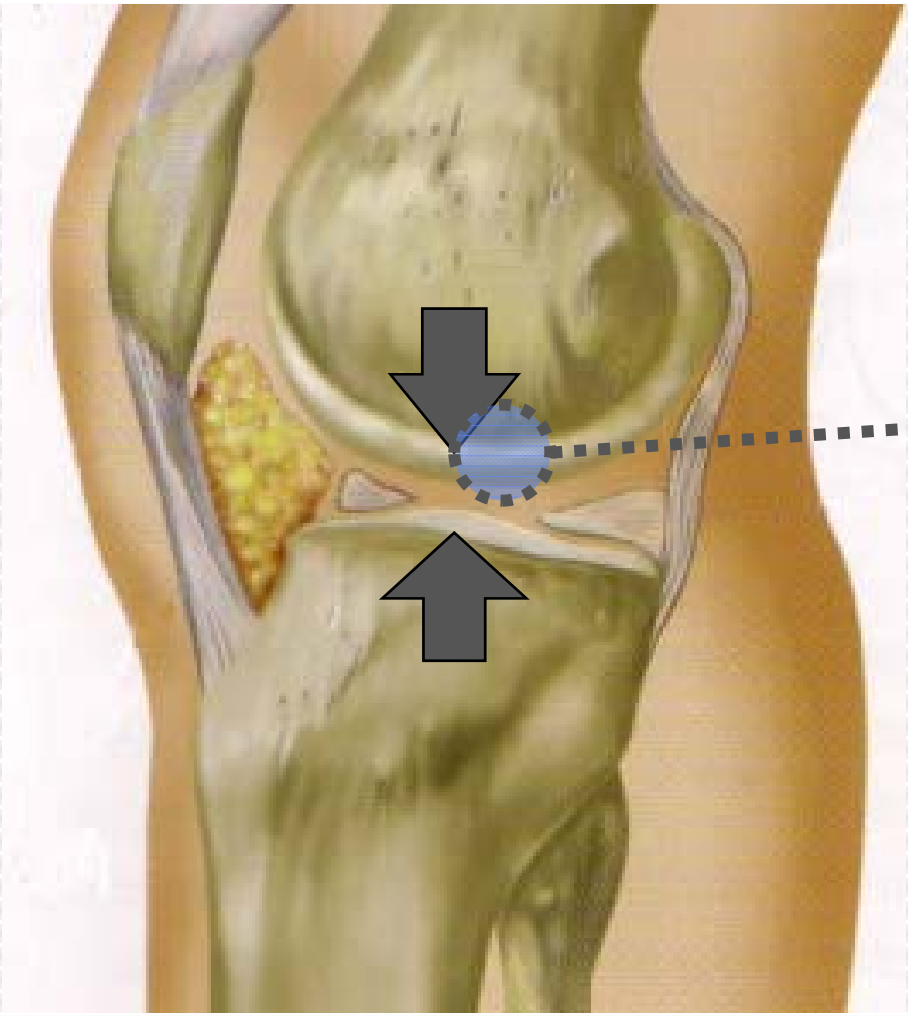


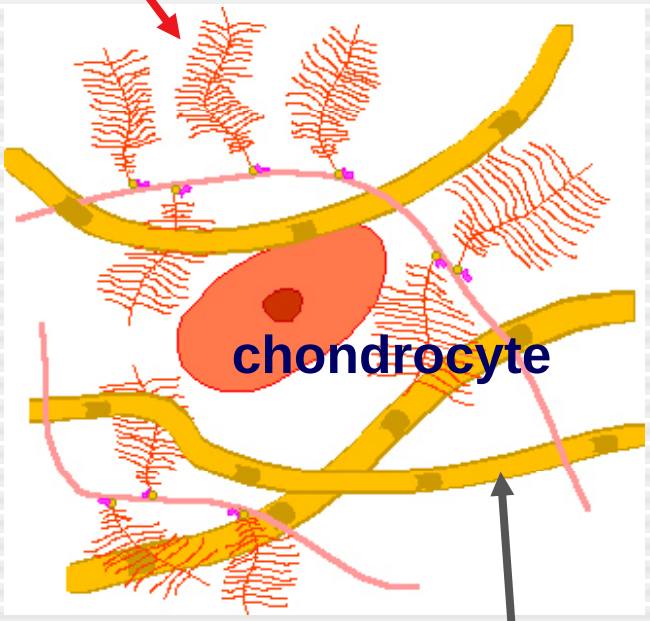
HE

Safranin-O/fast green

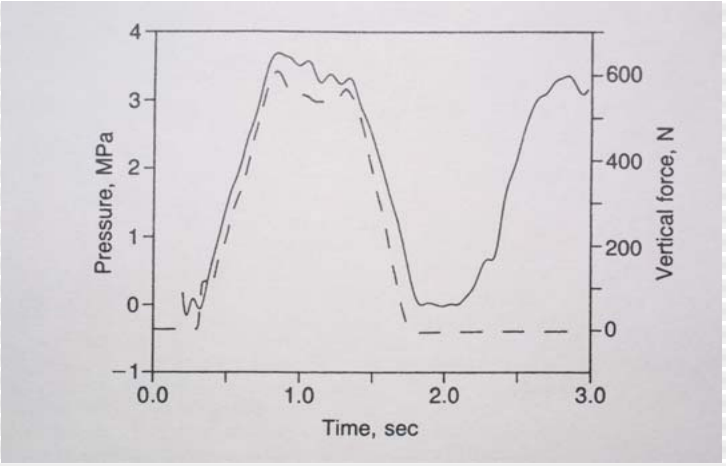
Toluidine blue





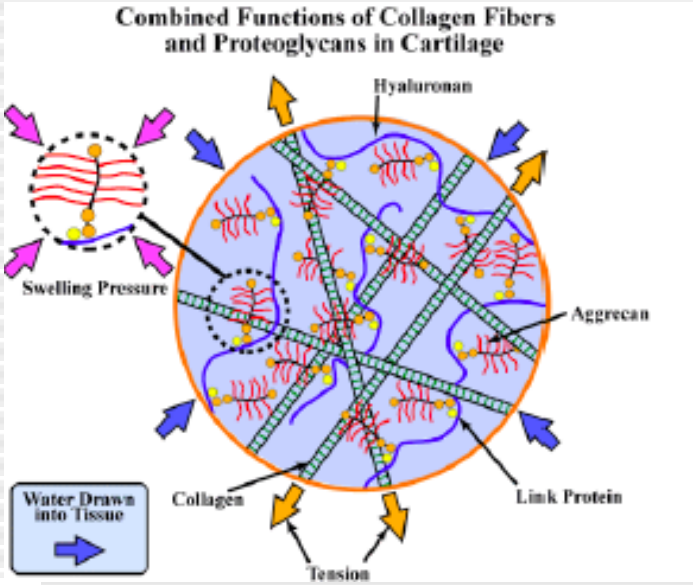


Hydrostatic pressure loading to chondrocytes by weight or moving



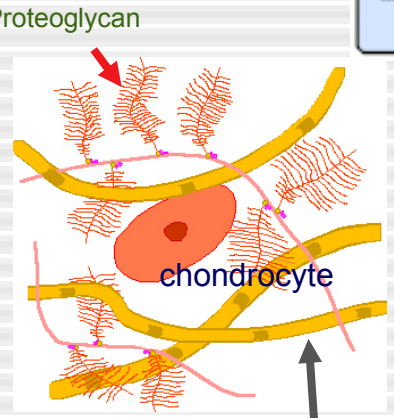
Hodge et al. 1985

Direct measurement of loaded stress to hip joint



Hardingham, Glycoforum 1998

Cartilage model with collagen, proteoglycans & water



weight, movement

↓

articular cartilage

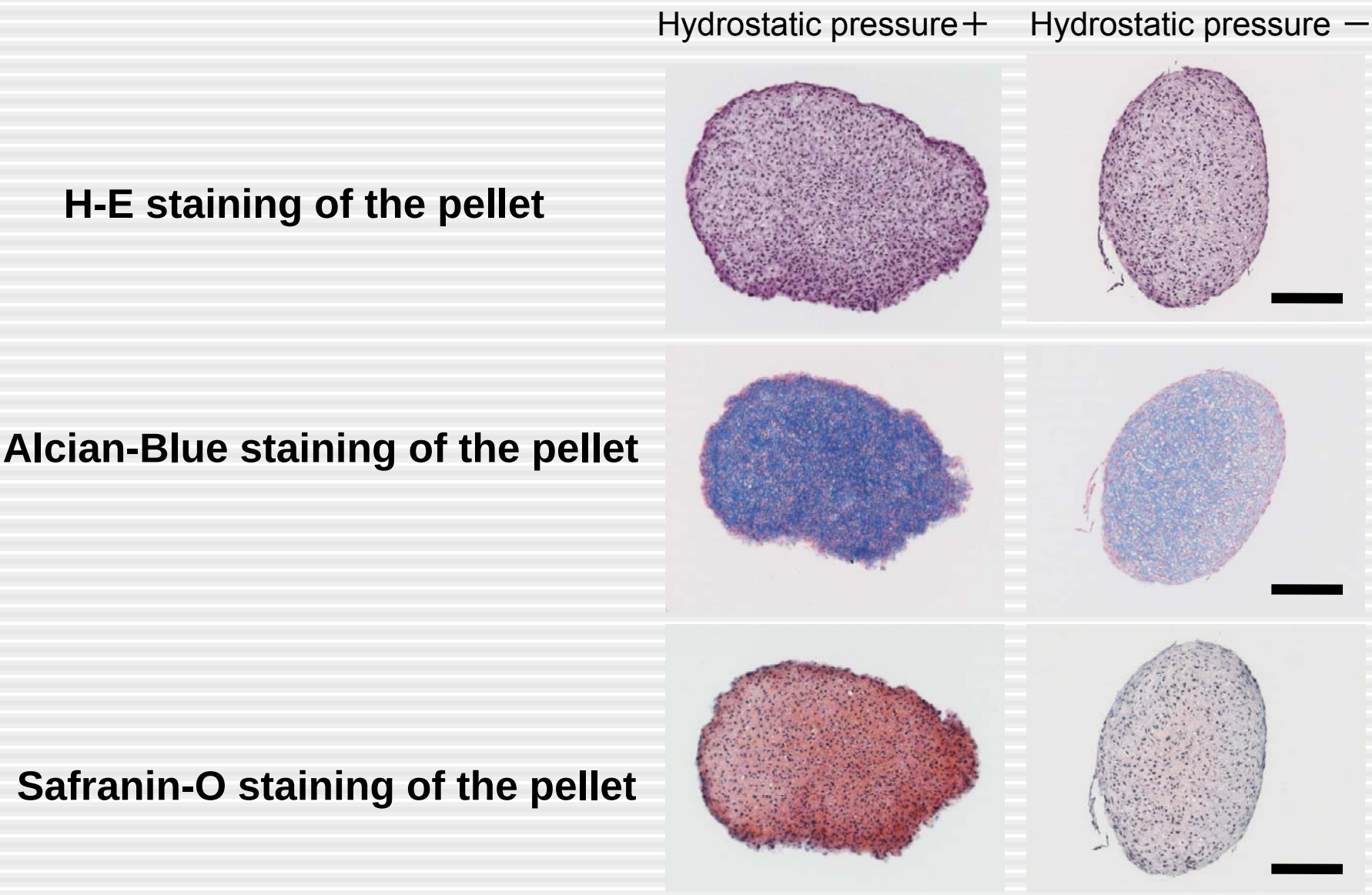
hydrostatic pressure

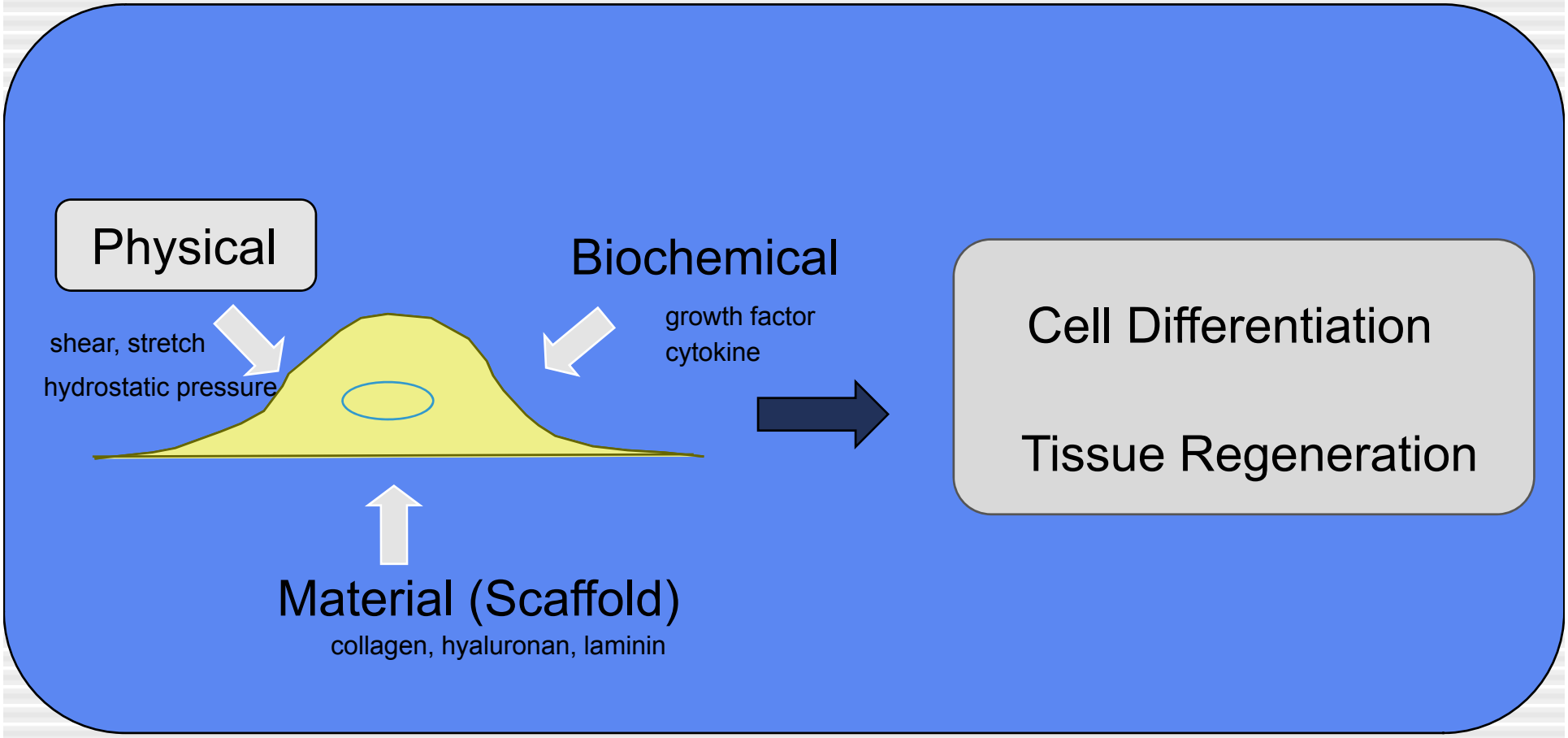
↓

chondrocyte

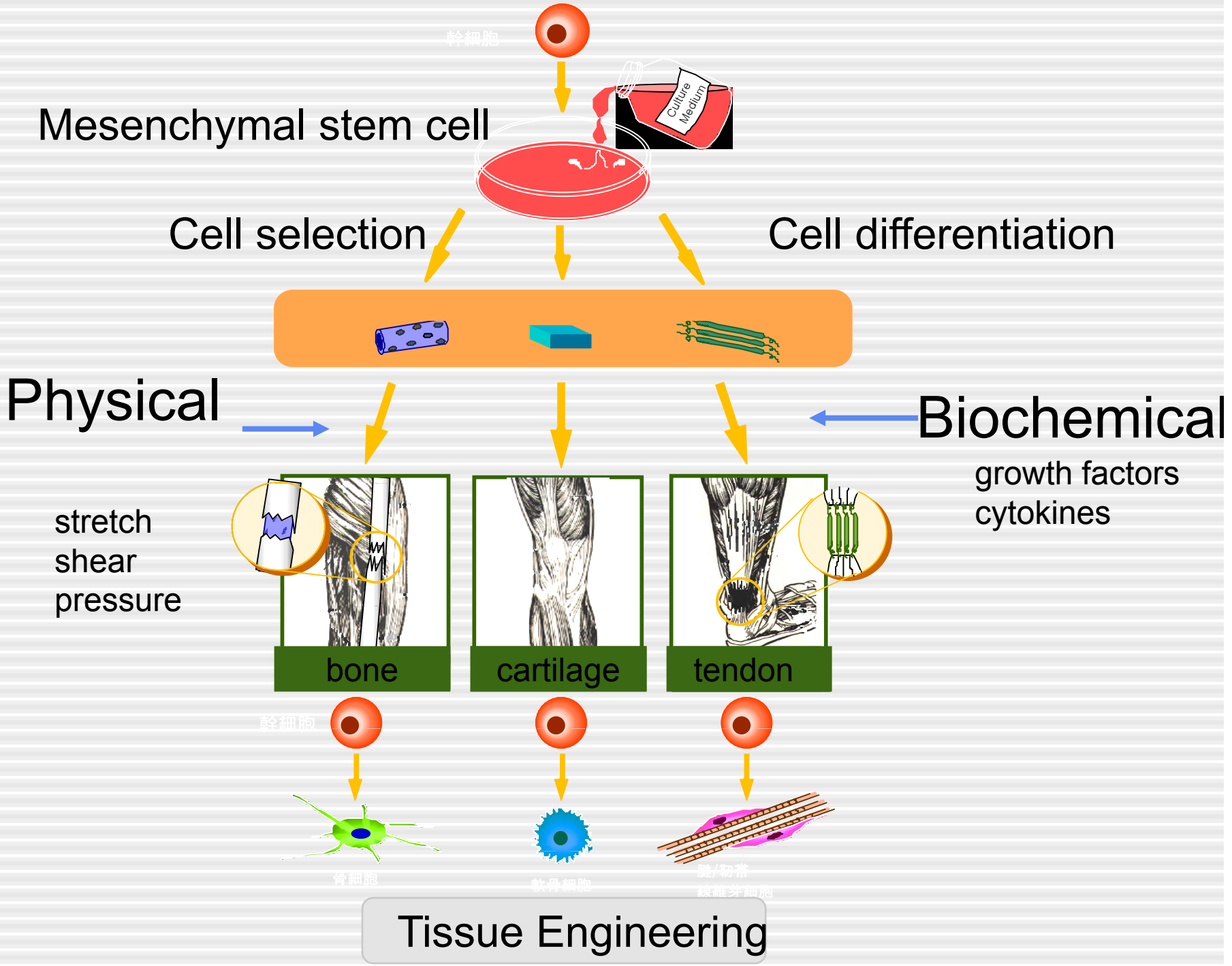
Chondrocytes physiologically loaded with hydrostatic pressure

Effect of Hydrostatic Pressure Loading On Matrices Production in Chondrocyte Pellets





3 Essential Factors for in vitro Tissue Regeneration


幹細胞
Mesenchymal stem cell
Culture Medium
Cell selection
Cell differentiation
Physical
Biochemical
growth factors
cytokines
stretch
shear
pressure
bone
cartilage
tendon
幹細胞
骨細胞
軟骨細胞
腱/靭帯
線維芽細胞
Tissue Engineering