Lecture 4-5-6

BT 636

Tissue Engineering and Regenerative Medicine

(3-0-0-6)

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Preamble

- Human tissue and/or organ failure, as a result of disease or trauma, is a major health concern world over.

- Treatment options include donor based transplantation, surgical repair, artificial prostheses etc. Ultimately, however, major damage may never be repaired in a truly satisfactory way.

- For such cases tissue engineering/regenerative medicine has emerged as a potential alternative, whereby tissue and organ failure is addressed by implanting lab grown tissue grafts and organ mimics that are fully functional and compatible.

- A variety of approaches are used to engineer these tissues in combination with stem cells/biomaterial/growth factors etc.

- Stem cells because of their remarkable regenerative potential are a preferred choice.

Syllabus

- **Introduction to tissue engineering & regenerative medicine: principles underlying tissue engineering/regenerative medicine strategies, key concepts of tissue engineering/regenerative medicine, its need and current available technologies.**

- Structure and organization of tissues: various cell and tissue types, its organization, structure-function relationship.

- Stem cells: stem cell types, their characteristics, potency Cell isolation, culture and differentiation: primary cell isolation techniques, cell culture needs, differentiation abilities of stem cells towards specific lineages.

- Biomaterials in tissue engineering & regenerative medicine: knowhow on current biomaterials, natural vs. synthetic, role of a biomaterial in tissue engineering, its properties, biodegradable polymers and 3D scaffold processing techniques; Cell-cell and cell-matrix interactions: knowhow and importance of such interactions, extracellular matrices; tissue repair and angiogenesis.

- Biocompatibility and immune rejection: biomaterial/graft compatibility, host acceptance and rejection Drug, growth factor and gene delivery: knowhow and importance of sustained and controlled delivery, implications and applications.

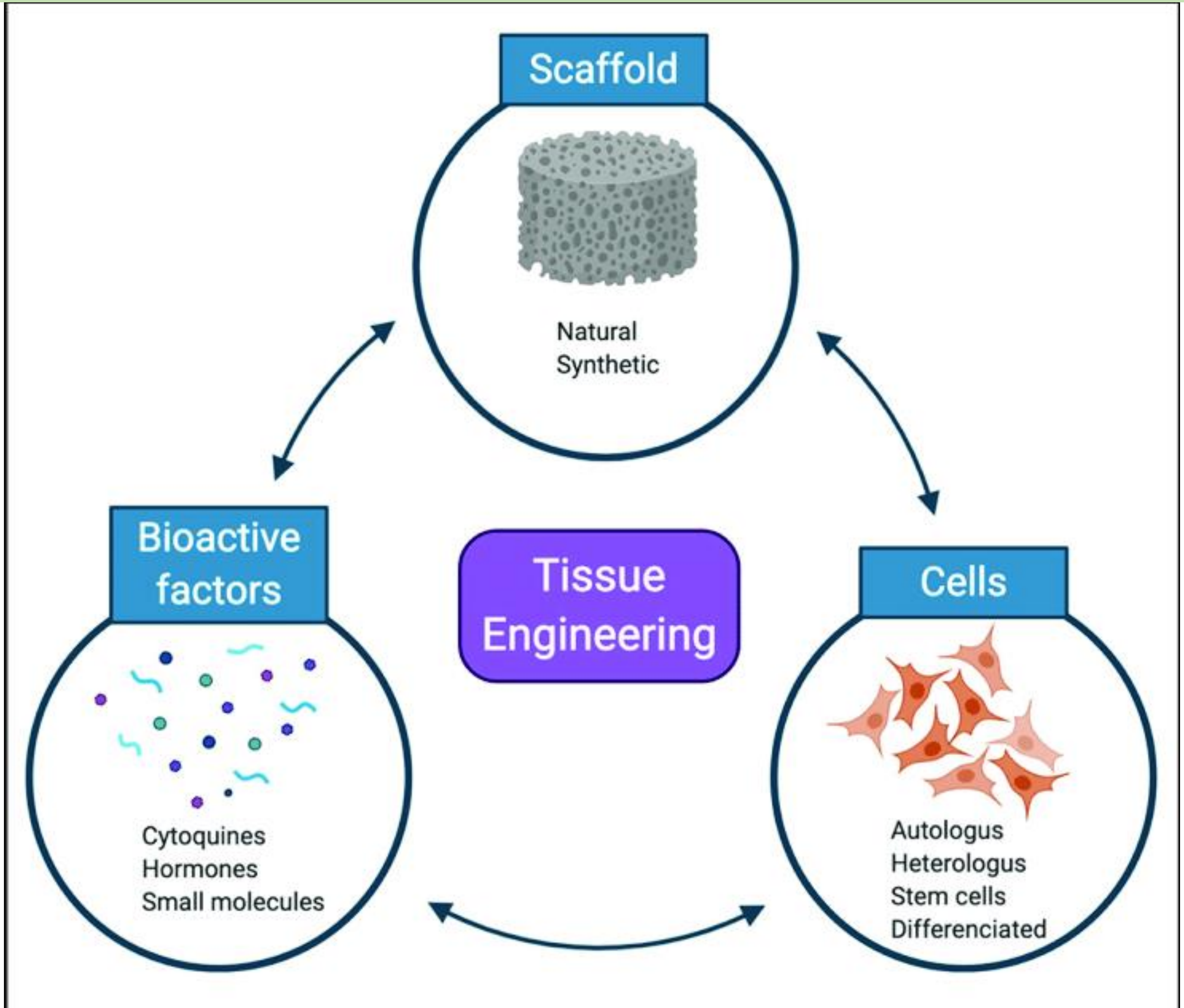
Tissue Engineering

- **Tissue engineering** is the use of a combination of cells, engineered materials, methods, and suitable biochemical and physicochemical factors to restore, maintain, improve, or replace different types of biological tissues.

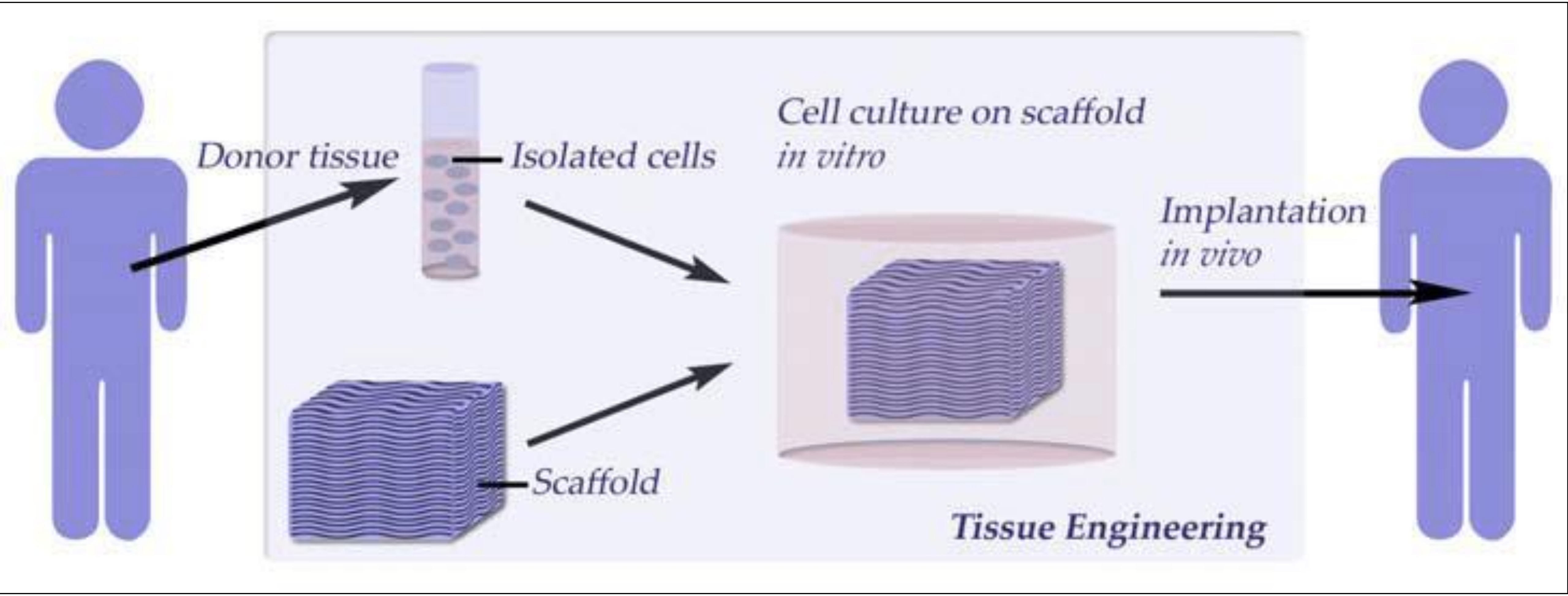
- The term has also been applied to efforts to perform specific biochemical functions using cells within an artificially-created support system (e.g. an artificial pancreas, or a bio artificial liver).

- A commonly applied definition of tissue engineering, as stated by Langer and Vacanti is "**An interdisciplinary field that applies the principles of engineering and life sciences toward the development of biological substitutes that restore, maintain, or improve [biological tissue] function or a whole organ**".

Tissue Engineering



Tissue Engineering



Regenerative Medicine

- Regenerative medicine deals with the **"process of replacing, engineering or regenerating human or animal cells, tissues or organs to restore or establish normal function"** (Mason & Dunnill, 2008). This field holds the promise of engineering damaged tissues and organs by stimulating the body's own repair mechanisms to functionally heal previously irreparable tissues or organs.
- Regenerative medicine also includes the possibility of growing tissues and organs in the laboratory and implanting them when the body cannot heal itself. When the cell source for a regenerated organ is derived from the patient's own tissue or cells, the challenge of organ transplant rejection via immunological mismatch is circumvented. This approach could alleviate the problem of the shortage of organs available for donation.
- Some of the biomedical approaches within the field of regenerative medicine may involve the use of stem cells. Examples include the injection of stem cells or progenitor cells obtained through directed differentiation (cell therapies); the induction of regeneration by biologically active molecules administered alone or as a secretion by infused cells (immunomodulation therapy); and transplantation of in vitro grown organs and tissues (tissue engineering).
- The term regenerative medicine is increasingly conflated with research on stem cell therapies. Some academic programs and departments retain the original broader definition while others use it to describe work on stem cell research.

Tissue Engineering and Regenerative Medicine

- Tissue engineering and regenerative medicine aim to restore or remodel damaged or diseased tissues by developing in vitro biological substitutes that can repair, replace, maintain, or enhance organ function.
- Tissue engineering is based on three essential components that are cells of variable sources, guiding tissue formation scaffolds, and microenvironment and its inductive factors.
- The structure, mechanical properties, and chemical composition of the scaffold can influence cell behavior, including proliferation, differentiation, extracellular matrix synthesis, and secretion.

- Bioreactors and bioprinting are also applied to improve the interaction between cells and scaffolds in order to achieve cell adhesion, proliferation, migration, extracellular matrix secretion, and differentiation, resulting in newly formed constructs or tissues.
- Translation from the laboratory to applied clinics must consider the integration of implanted constructs achieving functional restoration and regeneration of the target tissue.

- Tissue engineering and regenerative medicine hold important ongoing innovative developments with the potential to revolutionize healthcare, proponing treatments, beyond just repairing lost tissue function, but regenerating the original characteristics of tissues and organs.

Tissue Engineering and Regenerative Medicine

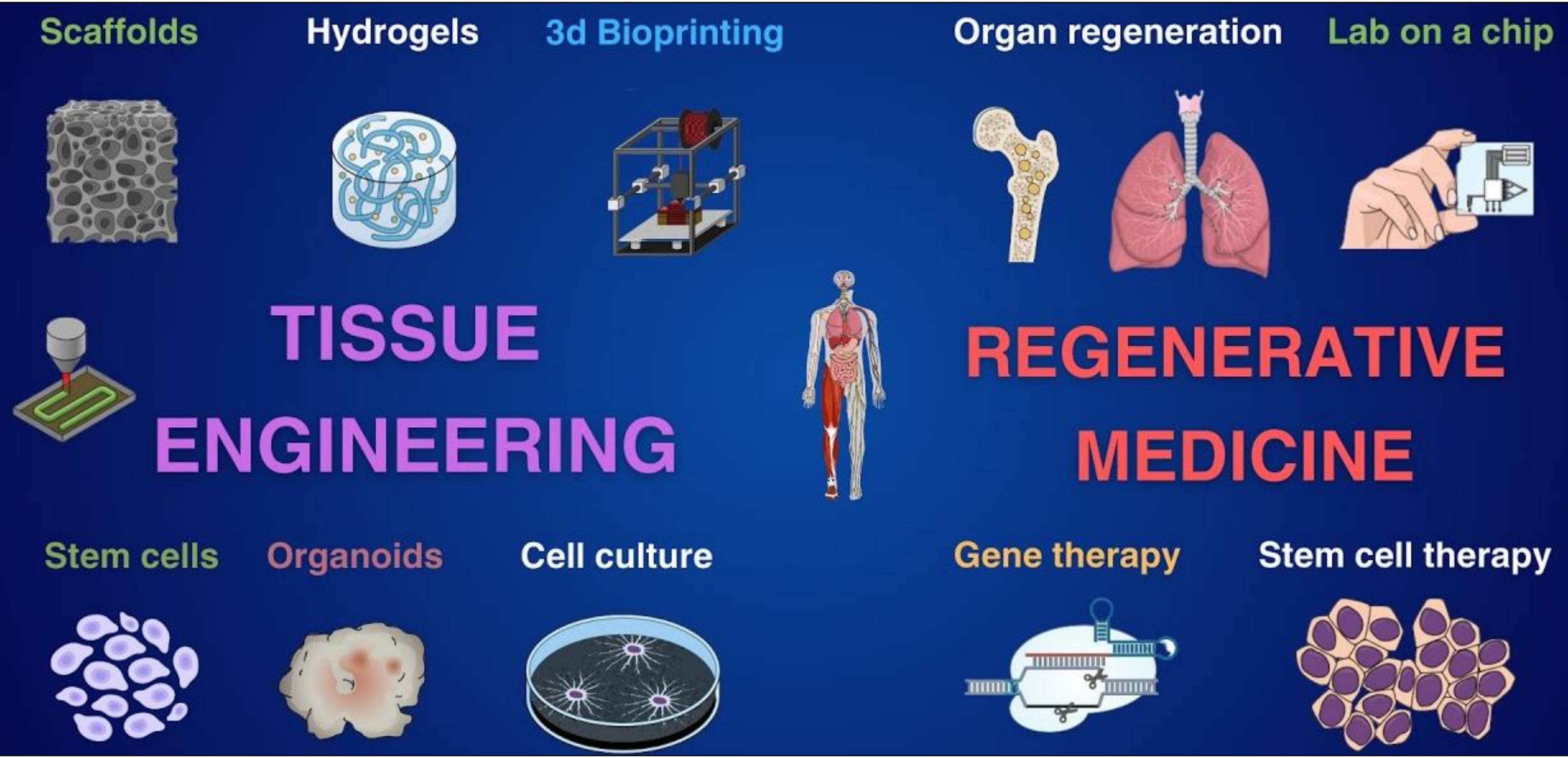
- Tissue and organ repair still represents a clinical challenge. Tissue engineering (TE) and regenerative medicine (RM) (TERM) is an emerging field focused on the development of alternative therapies for tissue/organ repair. This highly multidisciplinary field, in which bioengineering and medicine merge, is based on integrative approaches using scaffolds, cell populations from different sources, growth factors, nanomedicine, gene therapy, and other techniques to overcome the limitations that currently exist in the clinics. Indeed, its overall objective is to induce the formation of new functional tissues, rather than just implanting spare parts.

- Since its start, Tissue Engineering (TE) has been relying on three pillars (Salgado, Coutinho, & Reis, 2004a): scaffolds, cells, and growth factors.

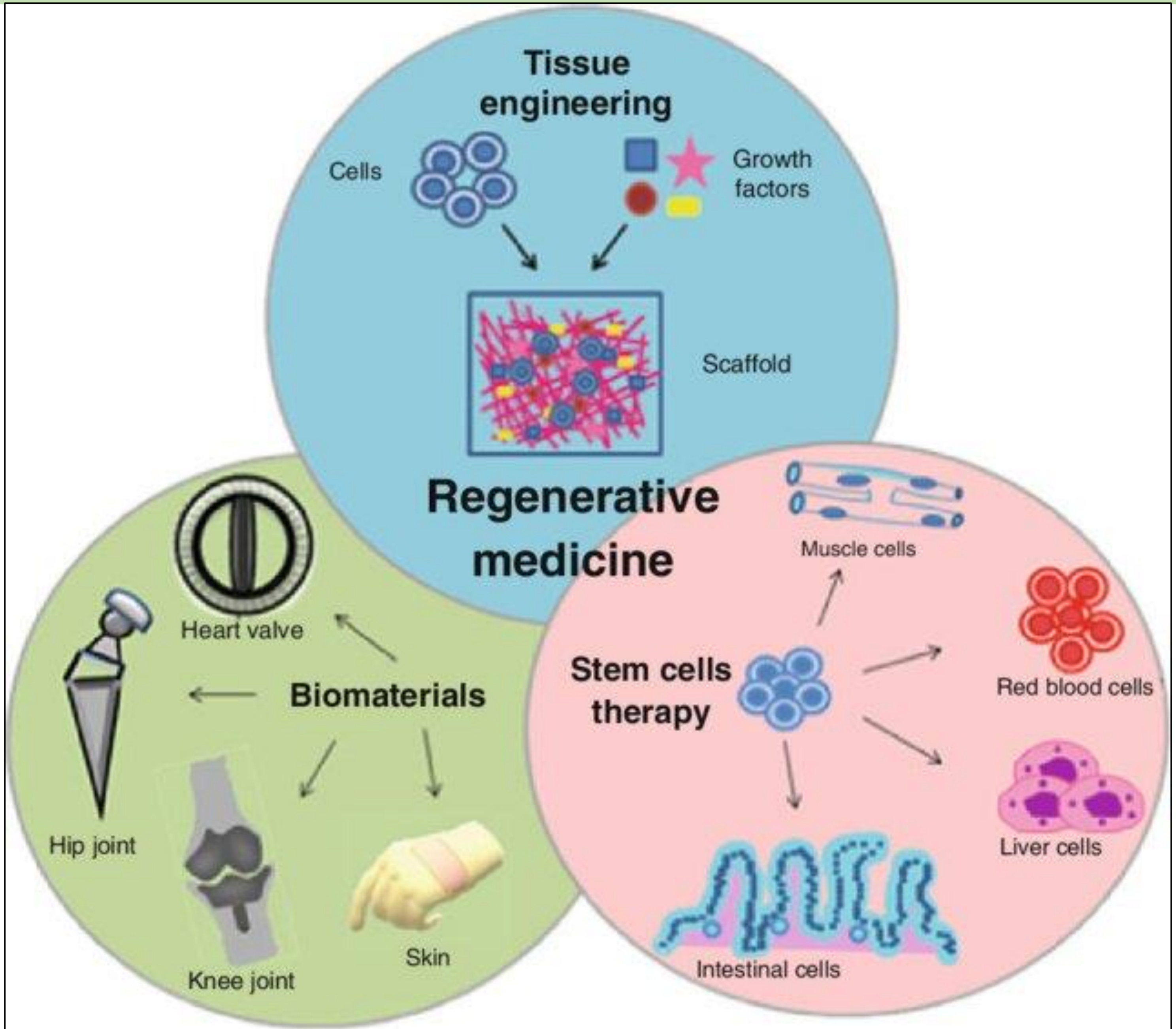
- On the other hand, regenerative medicine (RM) uses other strategies to induce organ regeneration including cell-based therapies, immunomodulation, gene therapy, nanomedicine, and TE itself.

- In fact, because of their similar objectives, these two fields have been merging in recent years, originating the broad field of tissue engineering and regenerative medicine (TERM).

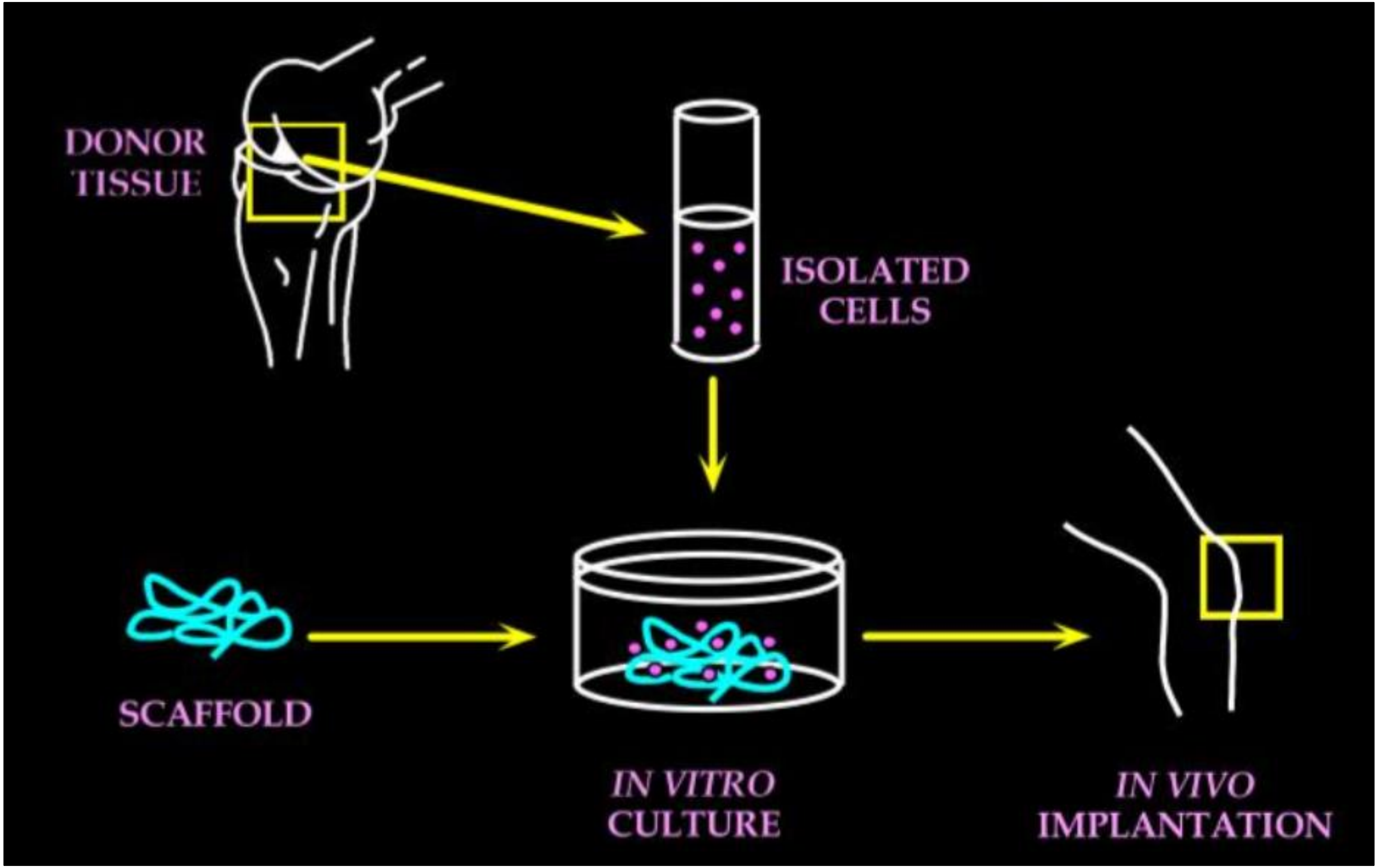
Tissue Engineering and Regenerative Medicine



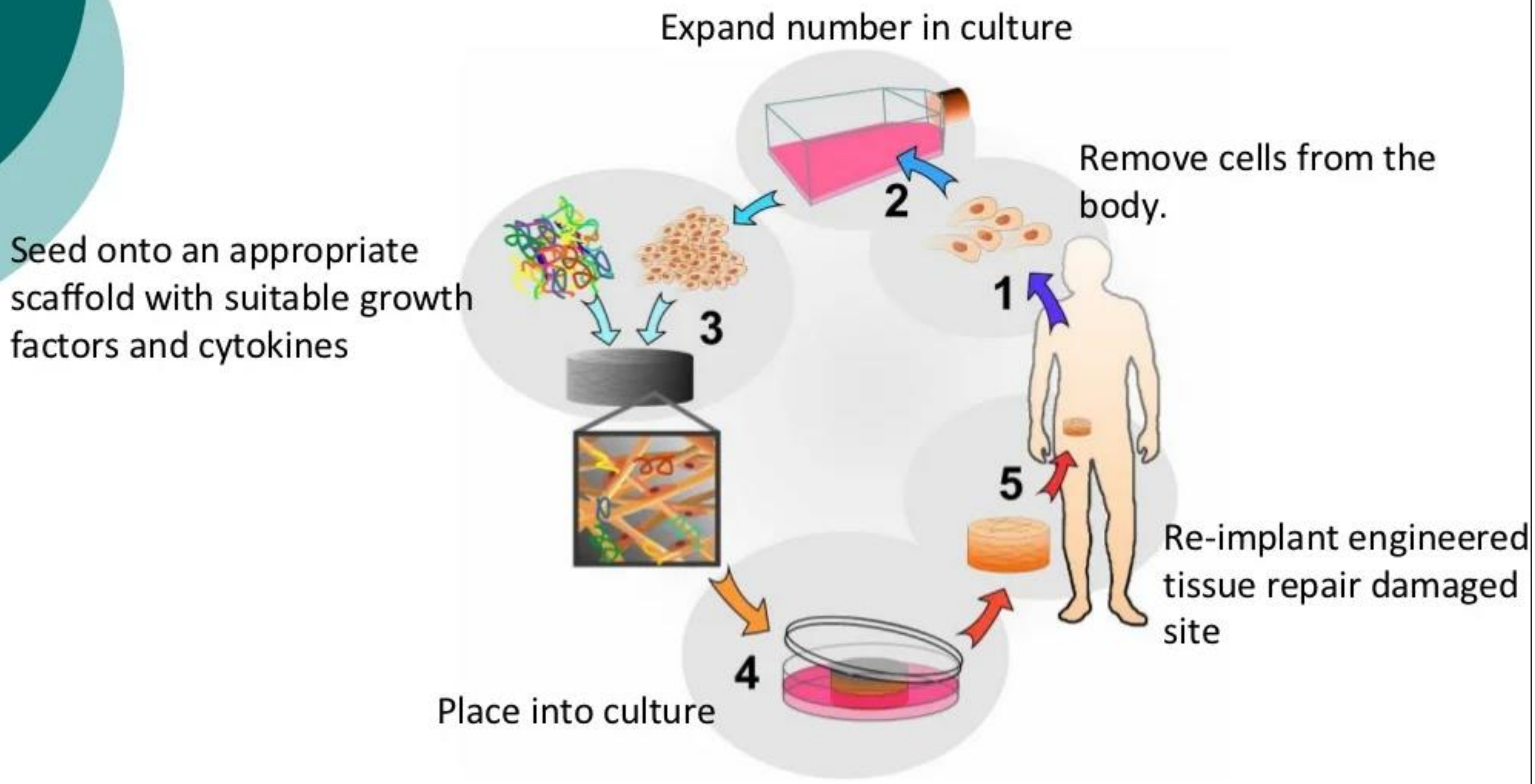
Tissue Engineering and Regenerative Medicine



Process of tissue engineering



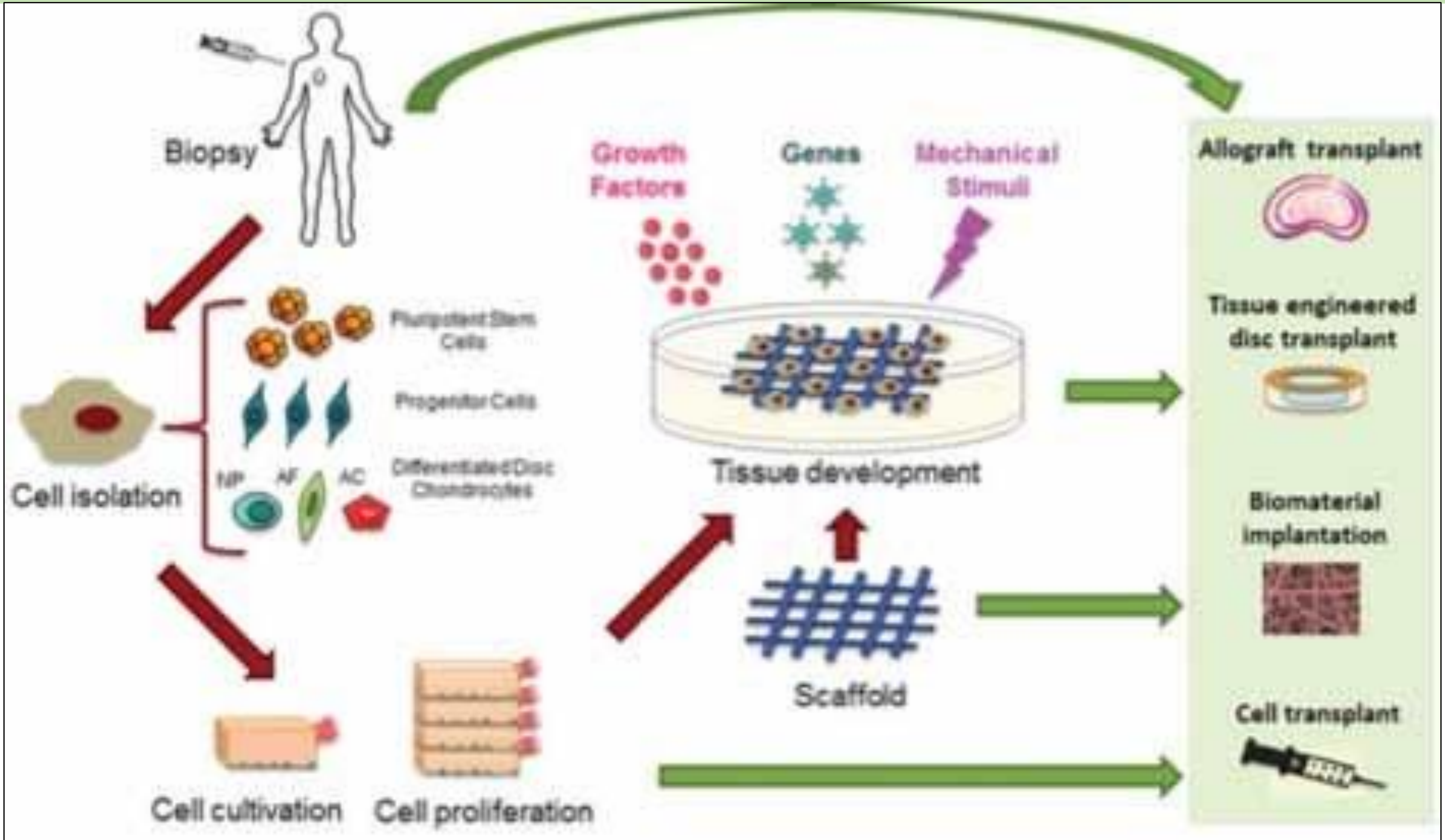
Tissue Engineering and Regenerative Medicine



Process of tissue engineering

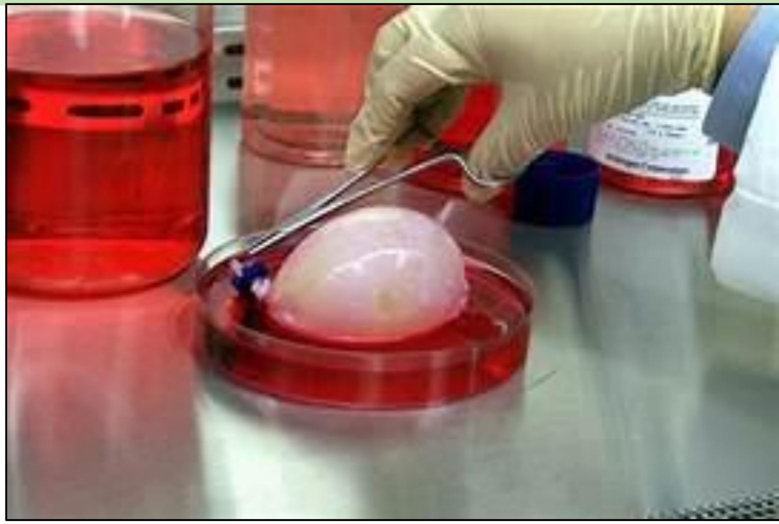
- (1) Start building material (e.g., extracellular matrix, biodegradable polymer).
- (2) Shape it as needed.
- (3) Seed it with living cells.
- (4) Bathe it with growth factors.
- (5) Cells multiply & fill up the scaffold & grow into three-dimensional tissue.
- (6) Implanted in the body.
- (7) Cells recreate their intended tissue functions.
- (8) Blood vessels attach themselves to the new tissue.
- (9) The scaffold dissolves.
- (10) The newly grown tissue eventually blends in with its surroundings.

Tissue Engineering and Regenerative Medicine



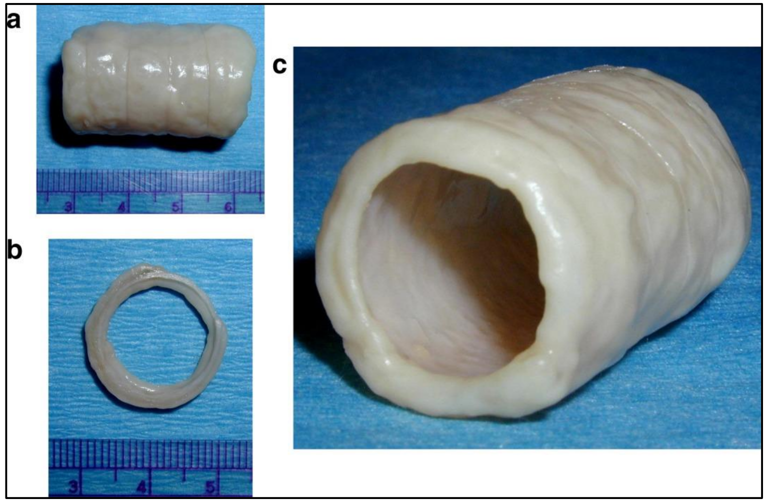
Examples of Tissue Engineering

- Bioartificial windpipe
- Bioartificial liver device
- Artificial pancreas
- Cartilage (heart) in a jar
- Tissue engineered airway
- Tissue engineered vessels
- Artificial bone marrow
- Artificial bone
- Oral mucosa
- Artificial skin
- Foreskin



Tissue Engineered Bladder
https://bme240.eng.uci.edu/students/06s/cphuang/index.html

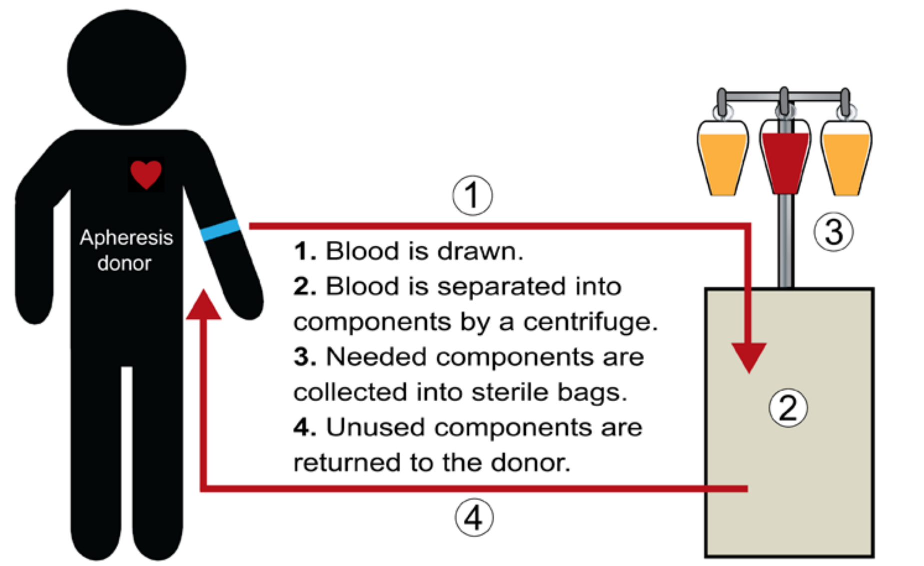
https://www.youtube.com/watch?v=W_nz49wcHK8

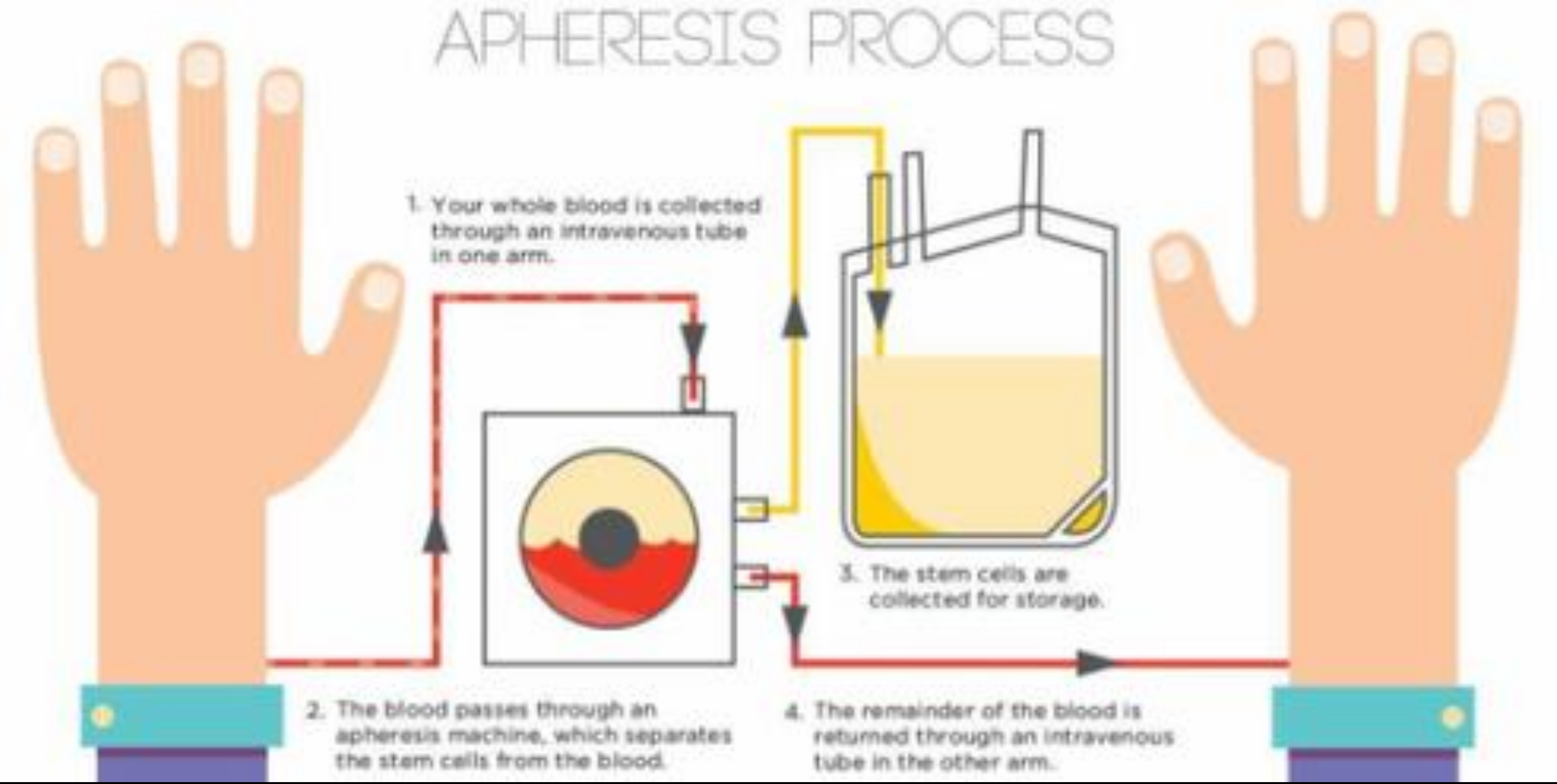


Tissue Engineered Trachea
(Kojima and Vacanti, 2013)

Process of tissue engineering

- **From fluid tissues** such as blood, cells are extracted by bulk methods, usually centrifugation or apheresis (process of separating blood components using a machine for various biomedical applications).
- **From solid tissues**, extraction is more difficult. Usually the tissue is minced, and then digested with the enzymes trypsin or collagenase to remove the extracellular matrix (ECM) that holds the cells. After that, the cells are free floating, and extracted using centrifugation or apheresis.





CELLS AS BUILDING BLOCKS

- Tissue engineering utilizes living cells as engineering materials.

- Examples include using living fibroblasts in skin replacement or repair, cartilage repaired with living chondrocytes,

CELLS (TYPES) AS BUILDING BLOCKS

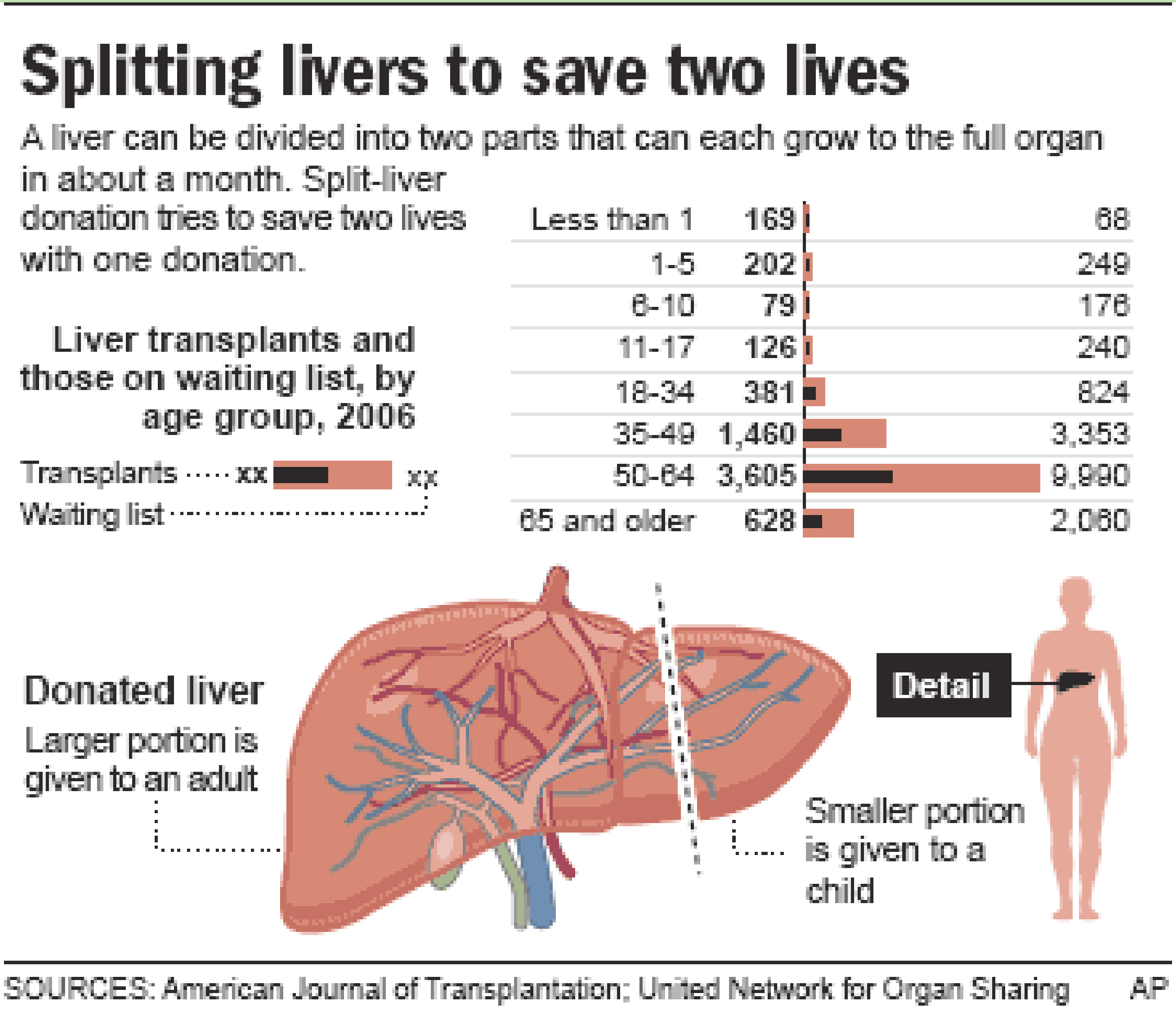
Cells are often categorized by their source:

- **Autologous cells** are obtained from the same individual to which they will be reimplanted. Autologous cells have the fewest problems with rejection and pathogen transmission, however in some cases might not be available.
- **Allogeneic cells** come from the body of a donor of the same species. While there are some ethical constraints to the use of human cells for in vitro studies, the employment of dermal fibroblasts from human foreskin has been demonstrated to be immunologically safe and thus a viable choice for tissue engineering of skin.
- **Xenogenic cells** are these isolated from individuals of another species. In particular animal cells have been used quite extensively in experiments aimed at the construction of cardiovascular implants.
- **Isogenic cells** are isolated from genetically identical organisms, such as twins, clones, or highly inbred research animal models.
  - Primary cells are from an organism.
  - Secondary cells are from a cell bank.
- **Stem cells** are undifferentiated cells with the ability to divide in culture and give rise to different forms of specialized cells. According to their source stem cells are divided multipotent, pluripotent and totipotent.

Organ transplantation

Autotransplant	**Transplanted within the same person’s body**
Allotransplant	**Transplants are performed between two subjects of the same species (mostly performed)**
Isotransplant	**Transplanted from a donor to a genetically identical recipient (such as an identical twin). Anatomically identical and do not trigger an immune response.**
Xenotransplant	**Transplanted (heterologous) from one species to another. Risky due to increased risk of non-compatibility, rejection, and disease carried in the tissue. Examples include porcine heart valves.**
Split transplants	**A deceased-donor organ, usually a liver, may be divided between two recipients, especially an adult and a child. This is not usually a preferred option because the transplantation of a whole organ is more successful.**

Split transplant



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Domino transplant	

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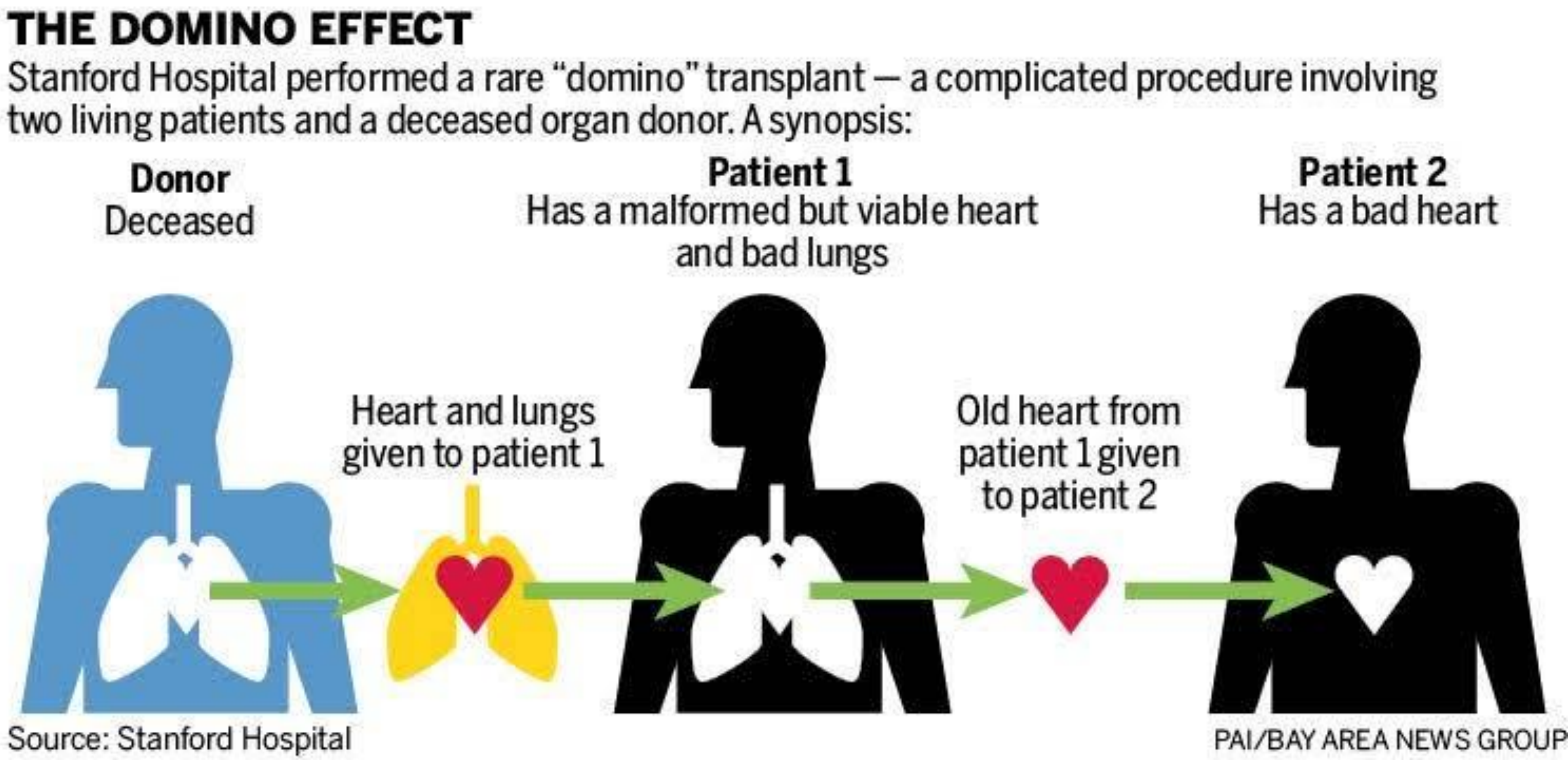

SCAFFOLDS

- Cells are often implanted or 'seeded' into an artificial structure capable of supporting **three-dimensional** tissue formation. These structures, typically called **scaffolds.**

- **Scaffolds usually serve at least one of the following purposes:**
  - Allow cell attachment and migration
  - Deliver and retain cells and biochemical factors
  - Enable diffusion of vital cell nutrients and expressed products
  - Exert certain mechanical and biological influences to modify the behavior of the cell phase

- To achieve the goal of tissue reconstruction, scaffolds must meet some specific requirements. A high porosity and an adequate pore size are necessary to facilitate cell seeding and diffusion throughout the whole structure of both cells and nutrients.
- Biodegradability is often an essential factor since scaffolds should preferably be absorbed by the surrounding tissues without the necessity of a surgical removal.

MATERIALS

- Many different materials (natural and synthetic, biodegradable and permanent) have been investigated. Examples of the materials are collagen and some polyesters.

- New biomaterials have been engineered to have ideal properties and functional customization: injectability, synthetic manufacture, biocompatibility, non-immunogenicity, transparency, nano-scale fibers, low concentration, resorption rates, etc.

- A commonly used synthetic material is PLA - polylactic acid. This is a polyester which degrades within the human body to form lactic acid, a naturally occurring chemical which is easily removed from the body.

MATERIALS

- Scaffolds may also be constructed from natural materials: in particular different derivatives of the extracellular matrix have been studied to evaluate their ability to support cell growth.

- Protein materials, such as collagen or fibrin, and polysaccharidic materials, like chitosan or glycosaminoglycans (GAGs), have all proved suitable in terms of cell compatibility, but some issues with potential immunogenicity still remains.

- Functionalized groups of scaffolds may be useful in the delivery of small molecules (drugs) to specific tissues.

Scaffolds – Important characteristics

Table 1. Outline of the properties to be considered when tissue engineering systems for regenerative medicine.

Properties	Design Considerations
Biocompatibility	The compatibility of a scaffold with the cells is of paramount importance. The scaffold should not illicit an immune response when inserted into the body.
Biodegradability	The ability of a scaffold to be biodegraded either through enzymatic or hydrolytic action is advantageous.
Electrical conductivity	Scaffolds which are conductive are able to influence the behavior of cells as a response to the electrical signals present in cell signaling.
Morphology	The morphology of the scaffold is vitality important as it impacts how the cells interact with the scaffold. The porosity of the scaffold ensures that cell infiltration can occur as well as the transfer of nutrients through the system.
Mechanical characteristics	This refers to characteristics such as the stiffness, elasticity and relaxation modulus of the scaffold. These influence cell behavior as well as the ability of the scaffold to mimic the natural microenvironment.
Ease of manufacturing	The cost of manufacturing, ease of the processes and the storage requirements are all factors which must be considered if the scaffold is to be produced on a large scale.

https://www.mdpi.com/1420-3049/26/9/2518

Applications

- Tissue engineering covers a broad range of applications, in practice the term has come to represent applications that repair or replace structural tissues (i.e., bone, cartilage, blood vessels, bladder, etc). These are tissues that function by virtue of their mechanical properties.
- A closely related (and older) field is cell transplantation. This field is concerned with the transplantation of cells that perform a specific biochemical function (e.g., an artificial pancreas, or an artificial liver).
- Tissue engineering solves problems by using living cells as engineering materials.

- These could be artificial skin that includes living fibroblasts, cartilage repaired with living chondrocytes, or other types of cells used in other ways.
- Tissue engineered heart valves offer a promising alternative for the replacement of diseased heart valves avoiding the limitations faced with currently available bioprosthetic and mechanical heart valves.
- Tissue-engineered skin is a significant advance in the field of wound healing and was developed due to limitations associated with the use of autografts.

Pros

- **Permanent cure for a disease or disorder**
- **No chance of rejection**
- **No need to wait for donor**
- **Organ donation will not be required**

Cons

- Ethical issues
- Expensive
- Difficulty in constructing suitable scaffolds
- Intensive research is required in understanding each tissue and organ to engineer them
- Cells have to remain alive inside the body and continue to function, which is difficult for researchers to establish for complex organs

Texts and References

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Thank you for your attention