BIOTECHNOLOQY IN ORQAN DONATION AND TRANSPLANTATION

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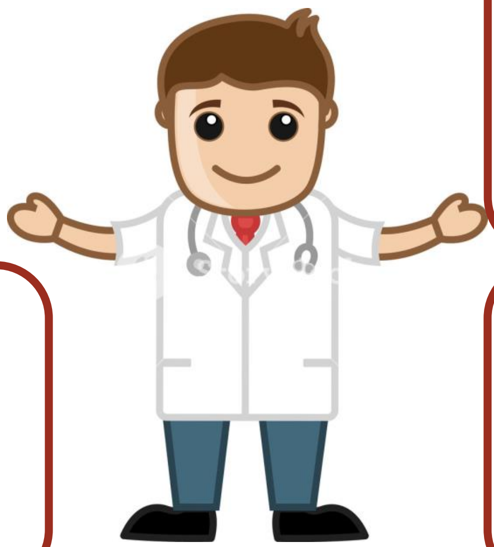
Main Ways of Restoring Organ Function to Patients in the Event of Serious Injury

Traditional pharmacological or surgical effects can correct disorders in the activity of a particular organ, but in not all cases…

Stimulation of the processes of regeneration in the body

Use of donor organs



Replenishment of organs functions with the help of devices of non-biological origin

Cultivation of tissues and organs

The Potential of the Modern Technologies of Regenerative Medicine

Augmenting organ function

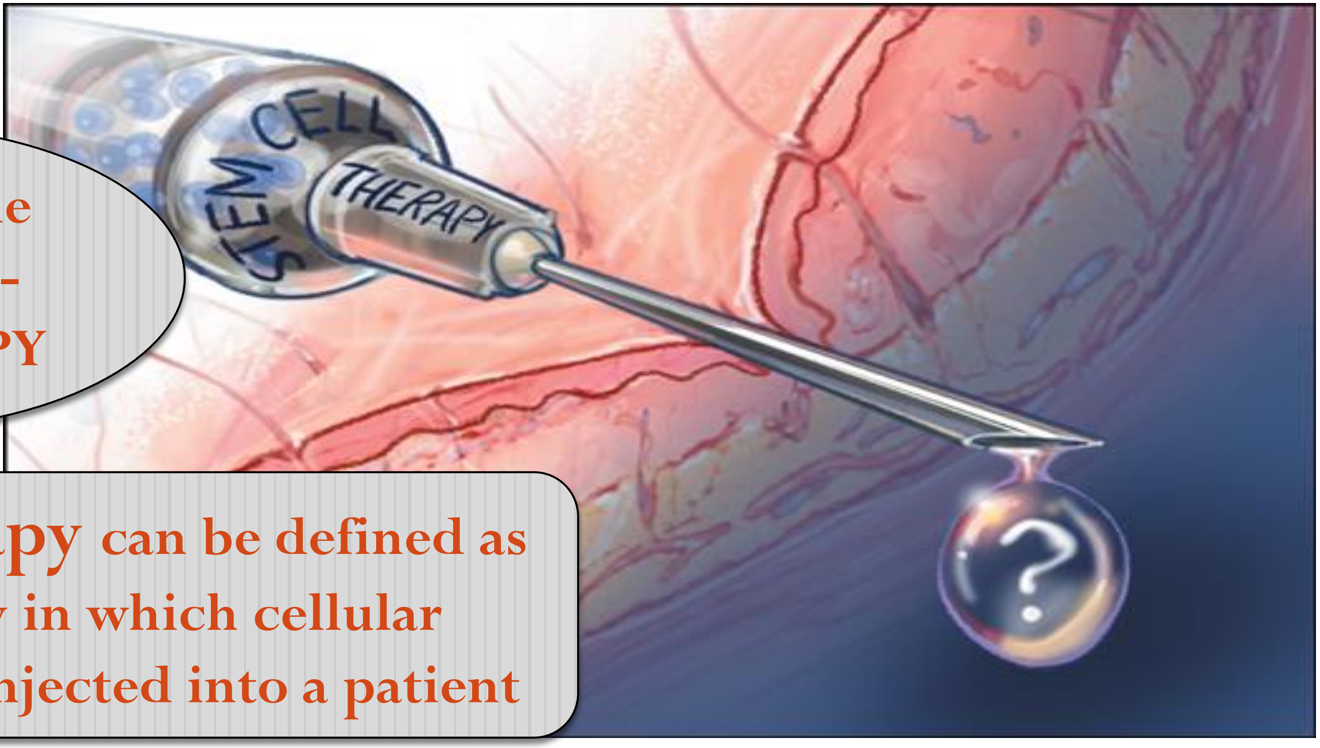
Allowing regeneration of deteriorated organs and tissue

Repairing damaged organ

- Researchers are exploring possible regenerative medicine applications in organ transplantation in order to benefit better results.
- **The present lecture** discusses the strategies that are being implemented to regenerate or bio-engineer human organs for clinical purposes.
- It also highlights the limitations of the regenerative medicine. The full potential of this field needs to be further explored.

STIMULATION OF THE REGENERATION PROCESSES IN ORGANISM

- Pharmacological effects. Drug Therapy.
- Procedure for the stem cells introduction into the body that have the ability to turn into full-fledged functional cells of the body.

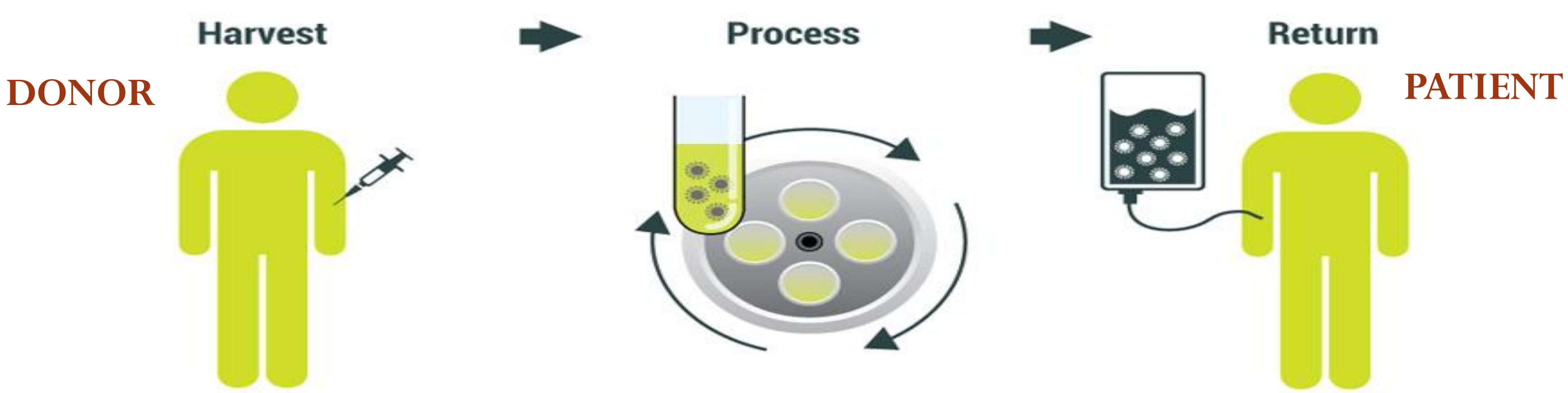


This technique has the name - CELL THERAPY

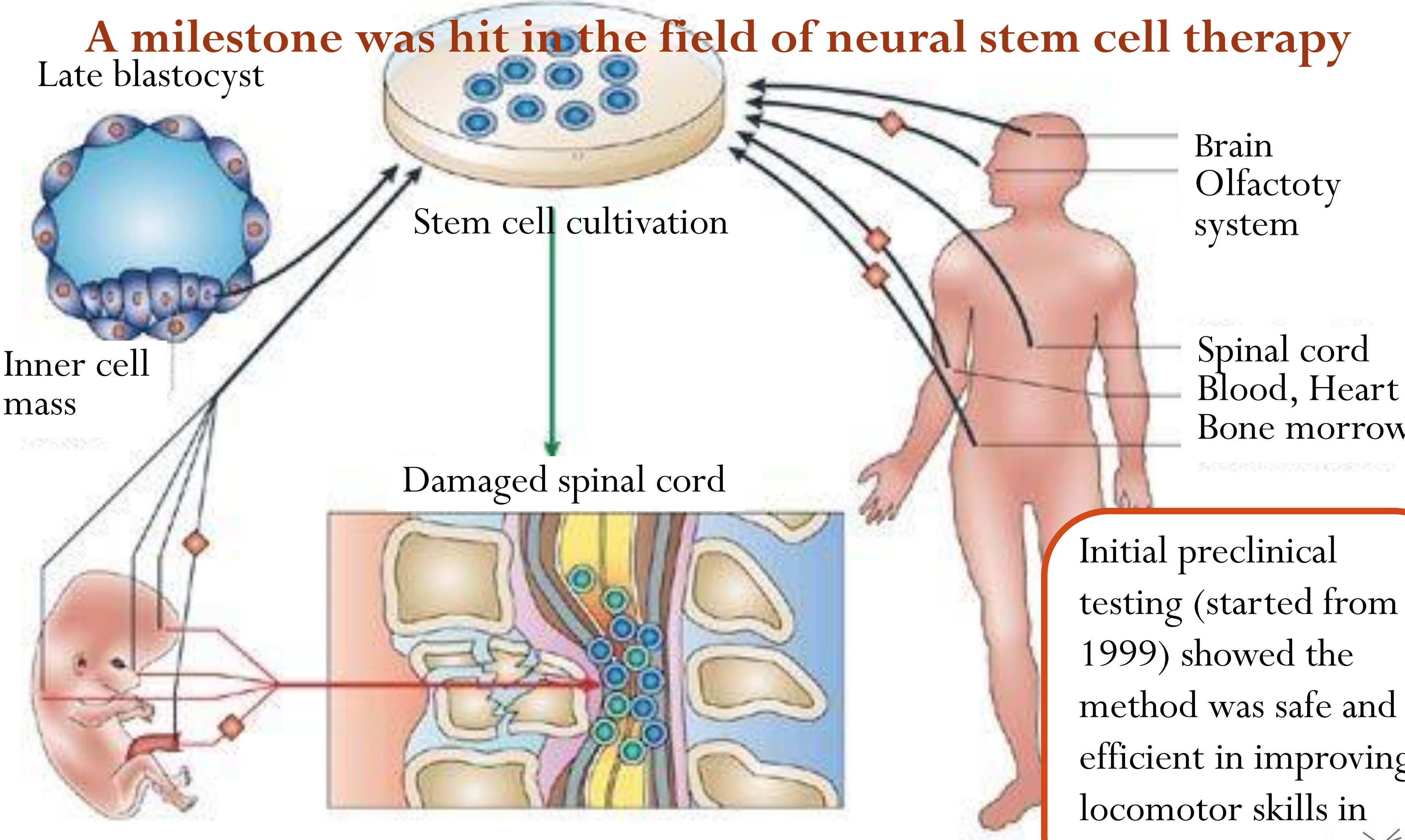
Cell Therapy can be defined as a therapy in which cellular material is injected into a patient

DIVISIONS OF CELL THERAPY

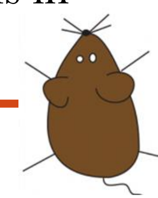
- **Transplantation of human cells from a donor to a patient.** It has strong prospects for future growth. Therapeutic applications include neural stem cell therapy, mesenchymal stem cells therapy and others such as hematopoietic stem cell transplantation.
- **The practice of injecting animal materials to cure disease.** This practice, lacks any medical evidence of effectiveness and can have very serious consequences.



A milestone was hit in the field of neural stem cell therapy



Initial preclinical testing (started from 1999) showed the method was safe and efficient in improving locomotor skills in animal models.



In 2009 - the US Food nd Drug Administration granted permission to the company Geron to initiate the world's first human clinical trial of an embryonic stem cell-based therapy for acute spinal cord injury.

Advantages and disadvantages of Stem Sell Therapy

- Cell therapy can be performed with much less risk to the patient.
- Cell therapy can also be applied to patients who are severely ill and would not be able to tolerate organ transplantation.
- This approach has already given a positive result in treatment heart attacks, strokes, neurodegenerative diseases, diabetes …
- It holds a promising future.

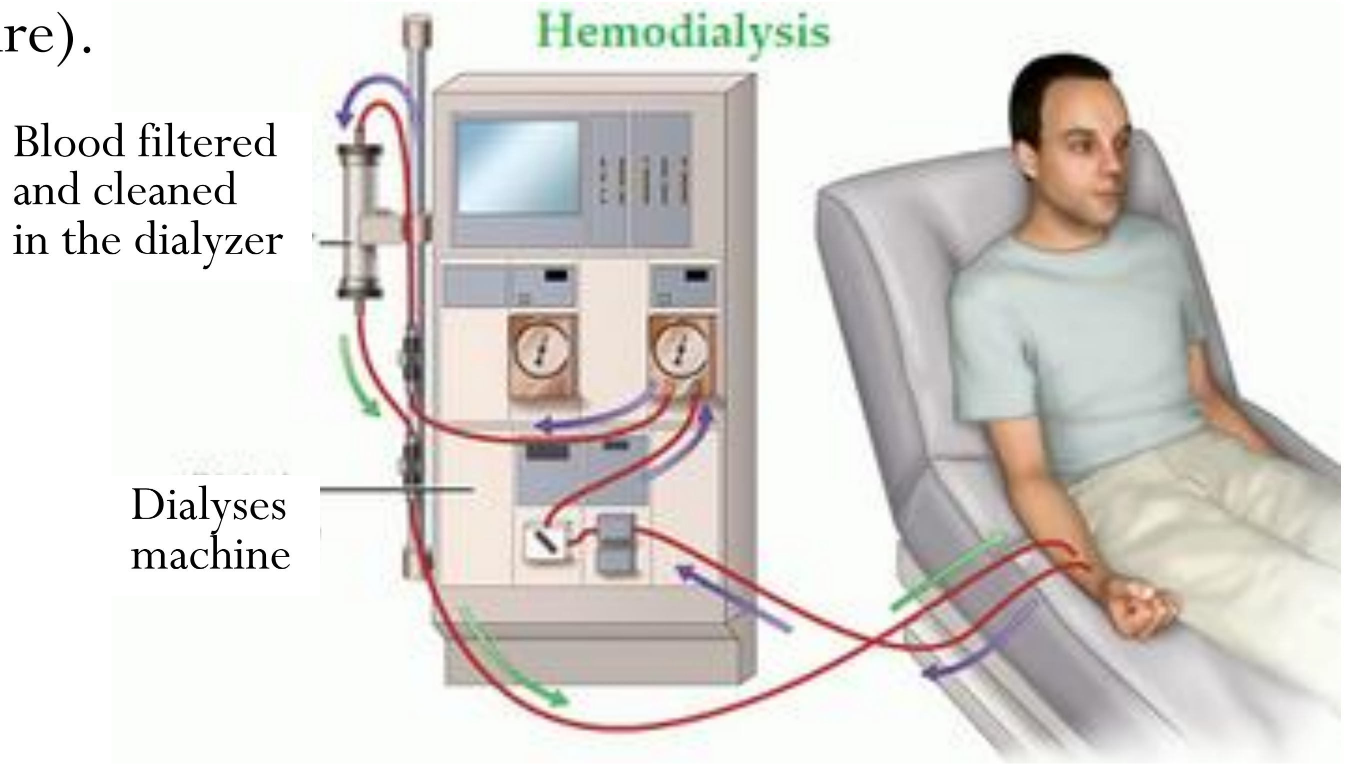
BUT

- Cell therapy is challenged by:
  - shortage of donor cells,
  - poor cell survival,
  - low transplant efficiency,
  - may lack true regeneration.
- Cell therapy is applicable only to eliminate relatively small damage to organs.

Embryonic-Stem-Cell in vitro

Replenishment of the Organs Functions with the Appliances of the Nonbiological Apparatuses

- Large-sized devices, to which patients are connected for a certain time (for example, apparatus for hemodialysis in renal failure).



Ingalator

Replenishment of the Organs Functions with the Appliances of the Nonbiological Apparatuses

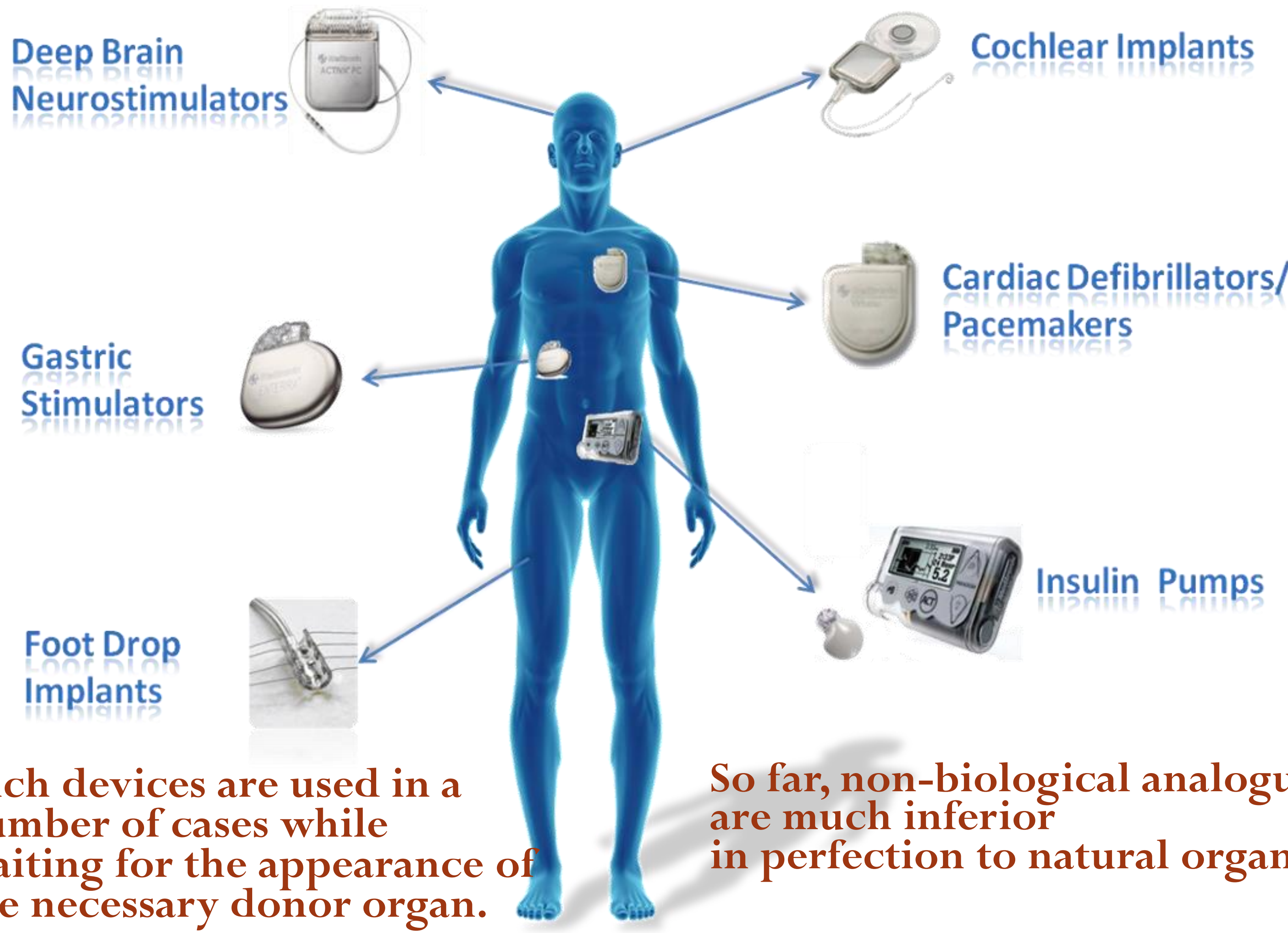
- Models of wearable devices, or devices implanted inside the body and help to remote health monitoring.
- There are options to do this, leaving the patient's own organ, however, sometimes it is removed, and the device takes over its functions, as in the case of the AbioCor artificial heart.

Healthcare wearable bracelet

Biosensor

AbioCor artificial heart

WIRELESS IMPLANTABLE MEDICAL DEVICES

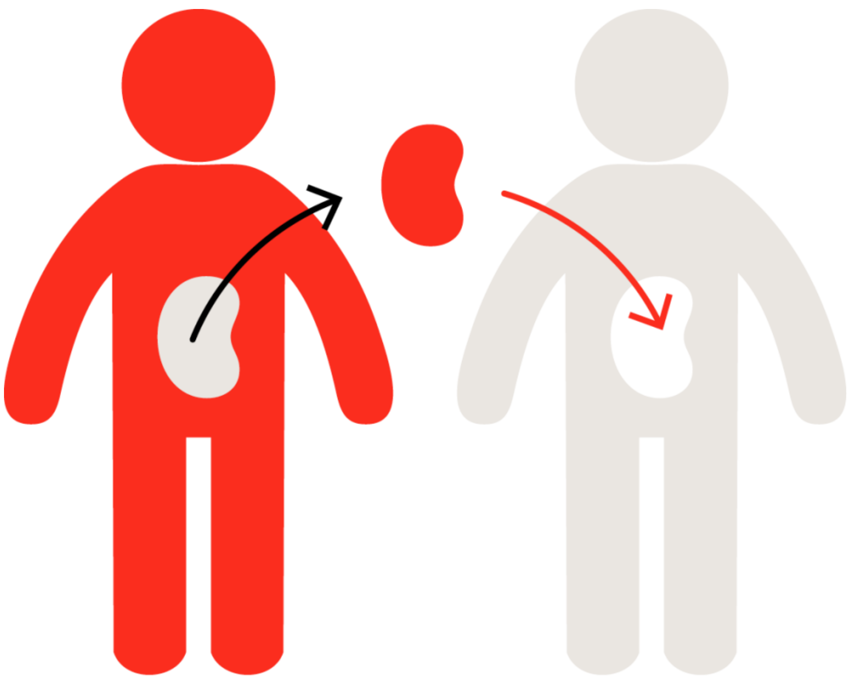


Such devices are used in a number of cases while waiting for the appearance of the necessary donor organ.

So far, non-biological analogues are much inferior in perfection to natural organs.

USE OF DONORS ORGANS

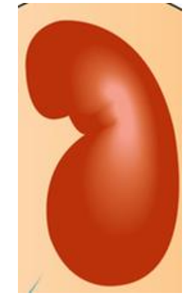
- Donor organs, transplanted from one person to another, are already widely and sometimes successfully used in clinical practice.



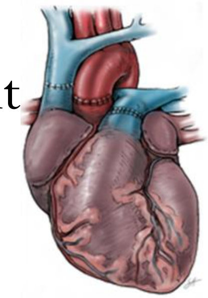
However, this direction faces a number of problems:

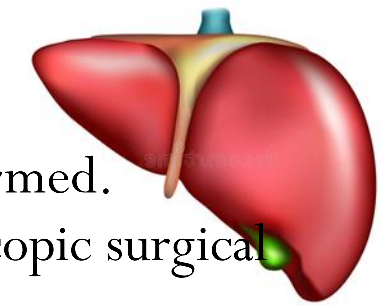
- a serious shortage of donor organs,
- the possible reaction of the rejection of a foreign body by the immune system,
- the threat of infection.

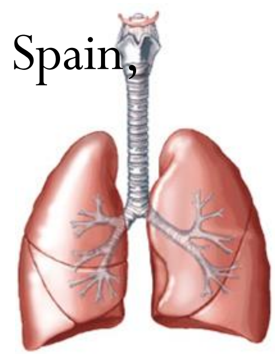
Timeline of Historical Events and Significant Milestones

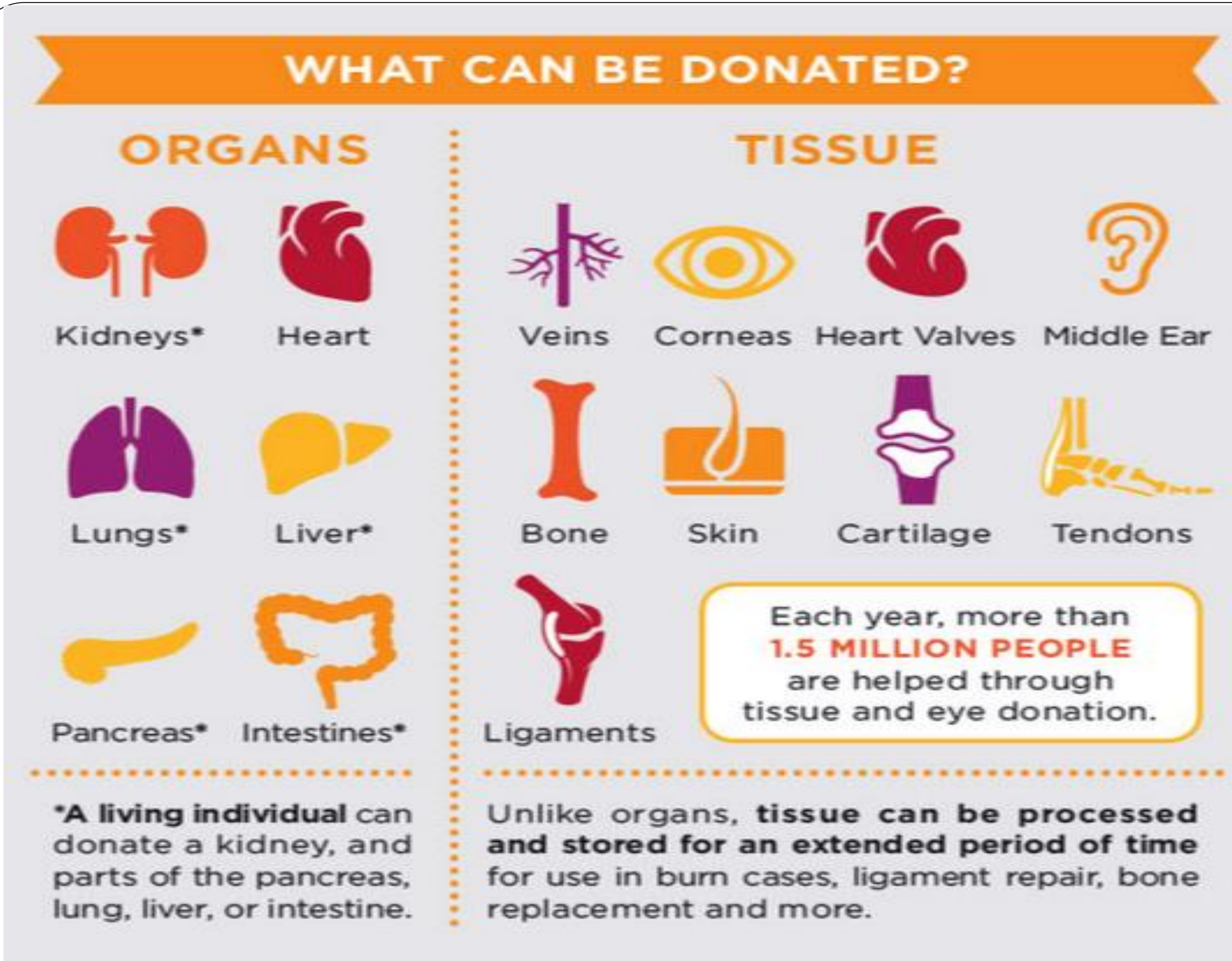


- **1869–1960** – First transplant of separate organs, such a heart, lung, cornea, skin, liver, kidneys, bone morrow, performed.
- **1983–1984** – First successful lung and heart/liver combined transplant performed.
- **1983** – First successful single lung transplant with significant recipient survival (more than 6 years).
- **1987** – First successful intestine transplant performed.
- **1988** – First split-liver transplant surgery performed.
- **1990** – First successful living donor lung transplant was performed.
- **1995** – First removing of living donor kidney through laparoscopic surgical methods.
- **1998–2001** – First successful hand transplant performed in France, Spain, USA.
- **2005** – First successful partial face transplant performed in France.
- **2010** – First successful full face transplant conducted in Spain.








WHAT CAN BE DONATED?
ORGANS
Kidneys*
Heart
Lungs*
Liver*
Pancreas*
Intestines*
*A living individual can donate a kidney, and parts of the pancreas, lung, liver, or intestine.
TISSUE
Veins
Corneas
Heart Valves
Middle Ear
Bone
Skin
Cartilage
Tendons
Ligaments
Each year, more than 1.5 MILLION PEOPLE are helped through tissue and eye donation.
Unlike organs, tissue can be processed and stored for an extended period of time for use in burn cases, ligament repair, bone replacement and more.

Types of grafts

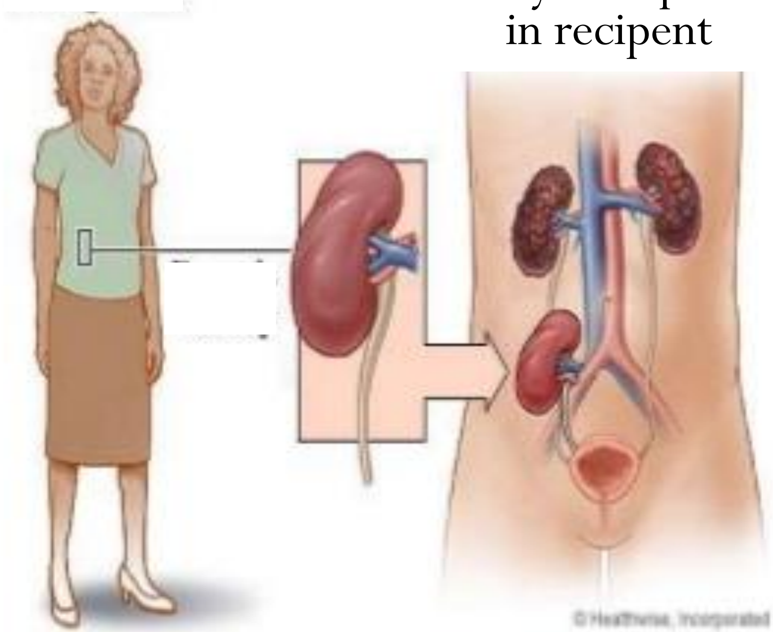
- Autologous graft (autograft) : within an individual, auto transplantation
- Syngeneic graft (syngraft, isograft) : identical twins, isotransplantation
- Allogeneic graft (allograft, homograft) : non-identical, allotransplantation
- Xenogeneic graft (heterologous graft, heterograft) : between species, xenotransplantation

Graft is a cell, a piece of living tissue or organ, that are transplanted surgically.

Who Can Donate?

LIVING DONOR

Kidney transplanted
in recipent



DESEASED DONOR

Organs for life

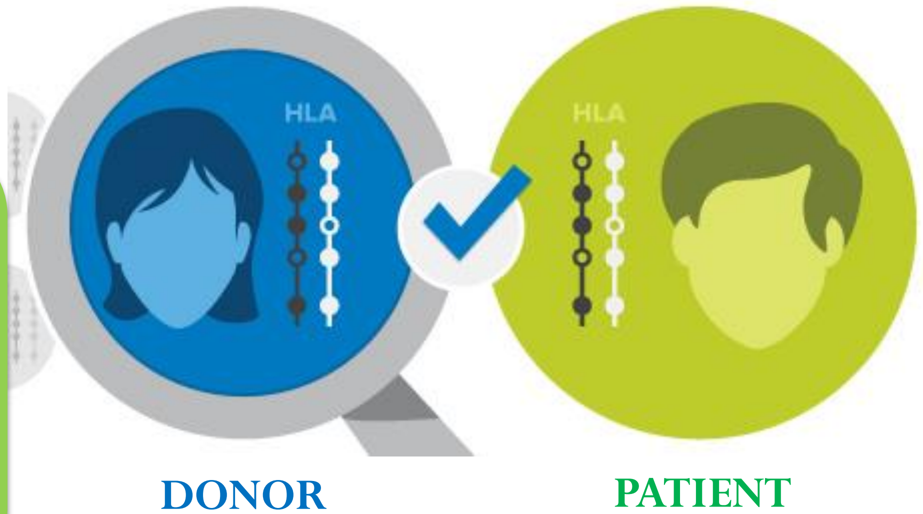
- All people should consider themselves potential organ and tissue donors – regardless of age, health, race, or ethnicity. No one is too old or too young to be a donor.
- Some major religions have released official statements or policies about donation.

More about Matching Donors with Recipients

The factors, in how organs are matched, usually include:

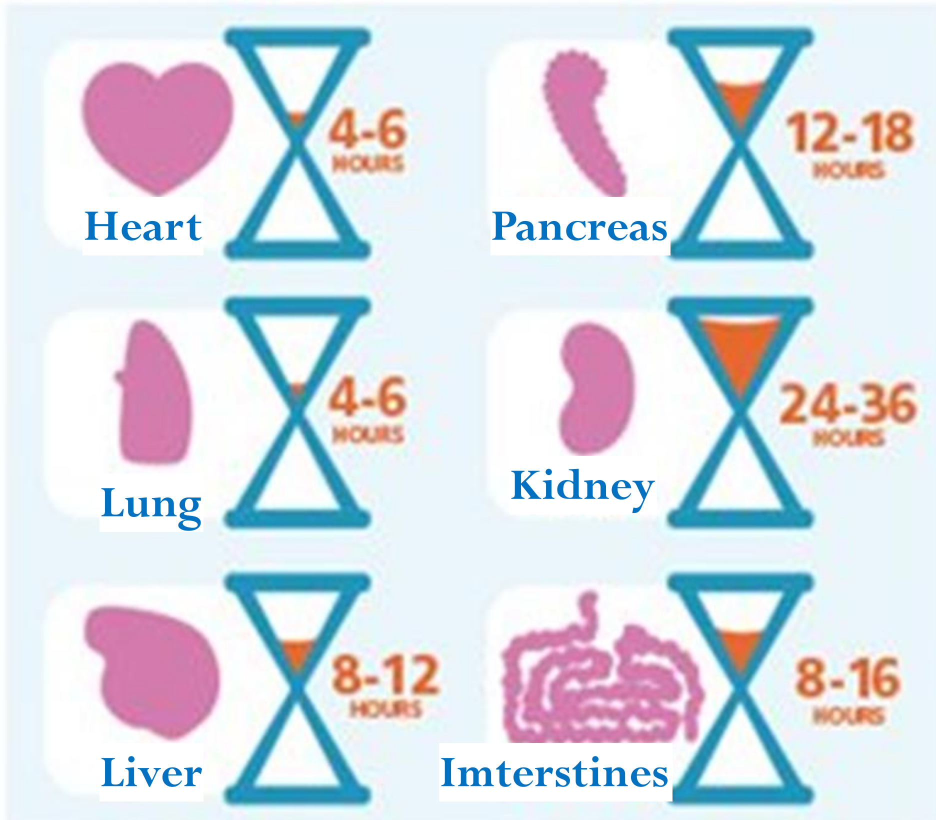
- Blood type,
- Body size,
- Severity of patient's medical condition,
- Distance between the donor's hospital and the patient's hospital,
- The patient's waiting time.

Transplantation can take place if the patient is available:
they can be contacted,
they has no current infection or other temporary complicating reason.



Recovering and Transporting Organs

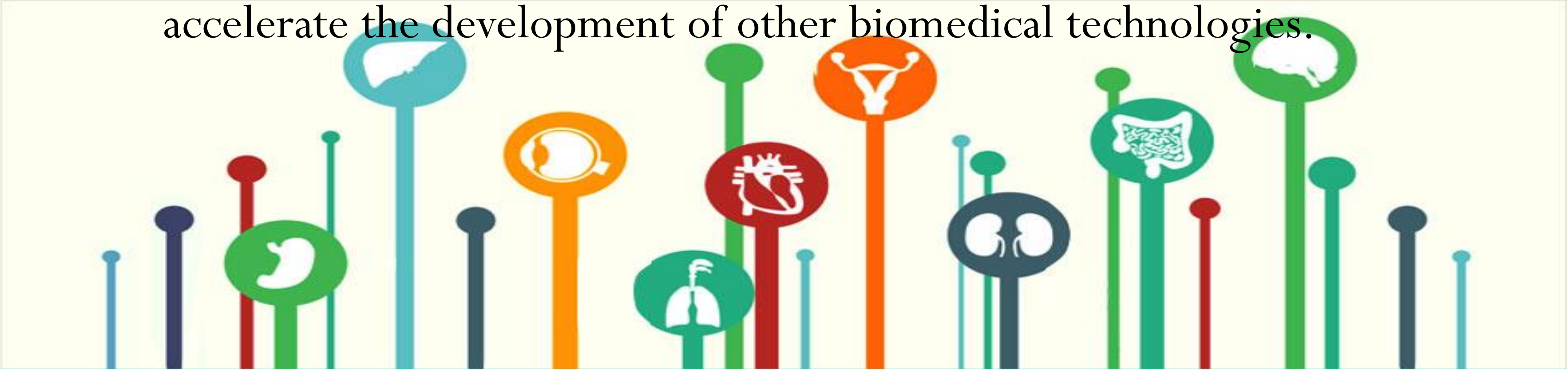
- Some organs can survive outside the body longer than others.
- While the search for matching recipients is under way, the deceased donor's organs are maintained on artificial support.
- Machines keep blood containing oxygen flowing to the organs. The condition of each organ is carefully monitored by the hospital medical staff.



The medical team trying to save the patient’s life and the transplant team are never the same team

Risks of exceeding preservation time

- Even a functional, transplantable organ may be turned down by the transplant centers through exseeding preservation limits.
- Each year, this phenomenon contributes to thousands of abdominal, thoracic, heart, lung and other organs being discarded. Most likely, many of these organs could have been successfully transplanted under the right circumstances. The resulting impact on waiting-list patients is profound.
- A successful large-scale organ preservation research effort would create a breadth of new capabilities that could make more organs available, improve transplant outcomes and mitigate risks, decrease costs, accelerate the development of other biomedical technologies.

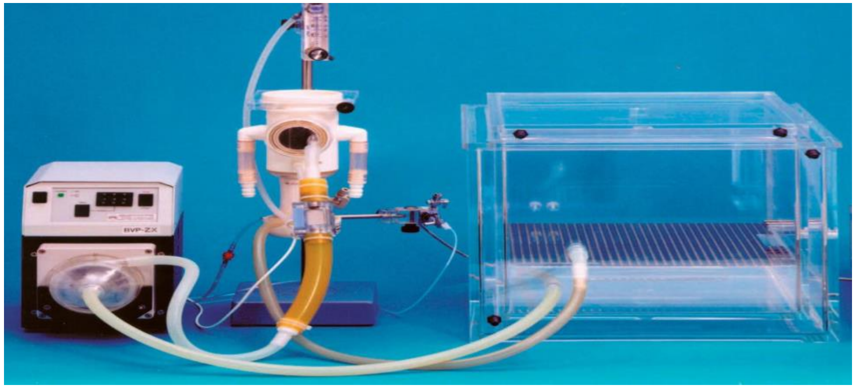


Promising Strategies that Use Perfusion Circuits

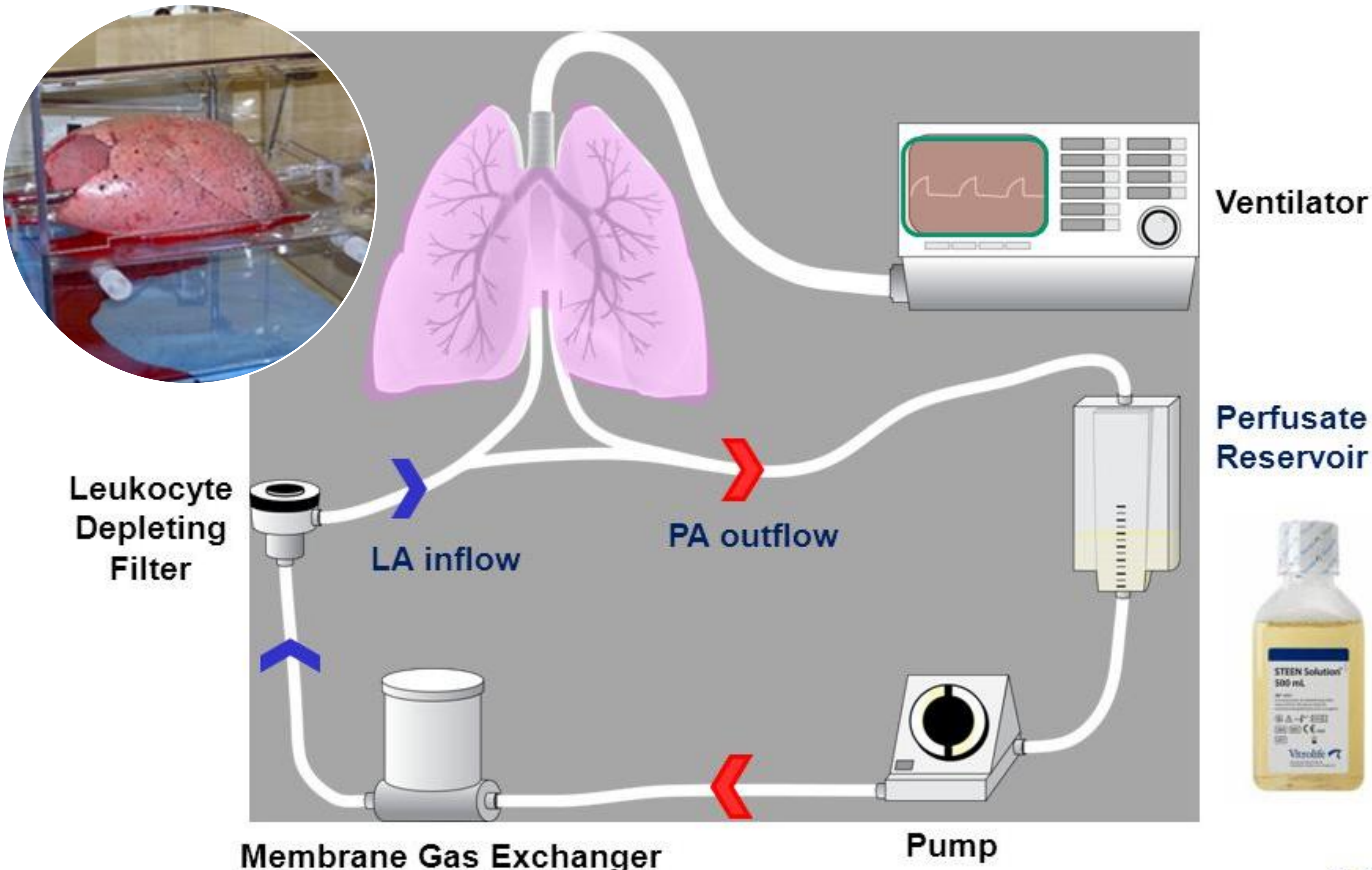
Perfusion platforms can:

- minimize effects after cellular stress and tissue injury during donor death and organ procurement,
- decrease the risks of inflammation and organ rejection following transplantation,
- enable therapeutic intervention before transplantation.

The possibility of rehabilitation of organs using of *ex vivo* organ perfusion promises to make larger amount of donor organs available that would otherwise be unsuitable for transplantation.



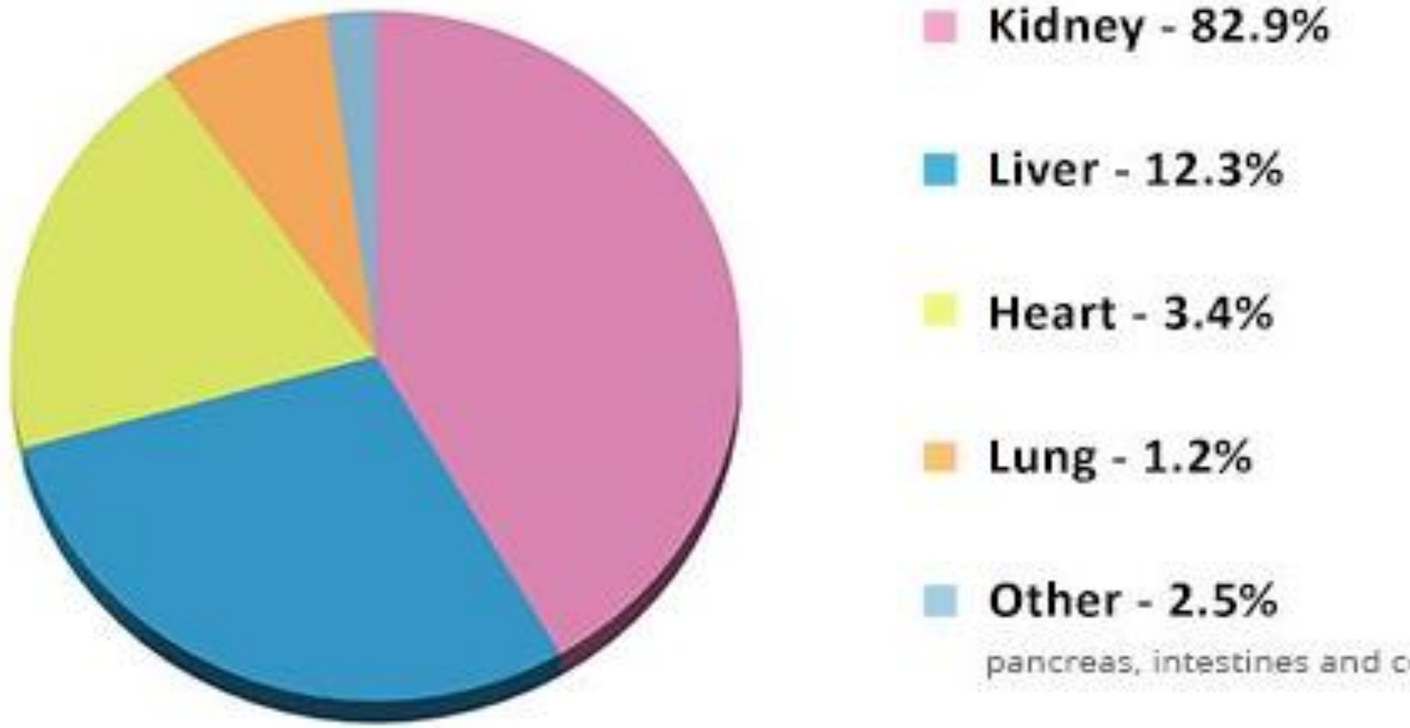
Ex-Vivo Lung Perfusion



TRANSPLANTING THE ORGANS

- The transplant operation takes place after the transport team arrives at the hospital with the new organ.
- The transplant recipient is typically waiting at the hospital and may already be in the operating room awaiting the arrival of the lifesaving organ.
- Surgical teams work around the clock as needed to transplant the new organs into the waiting recipients.

Organs People Are Waiting For (7/2017)



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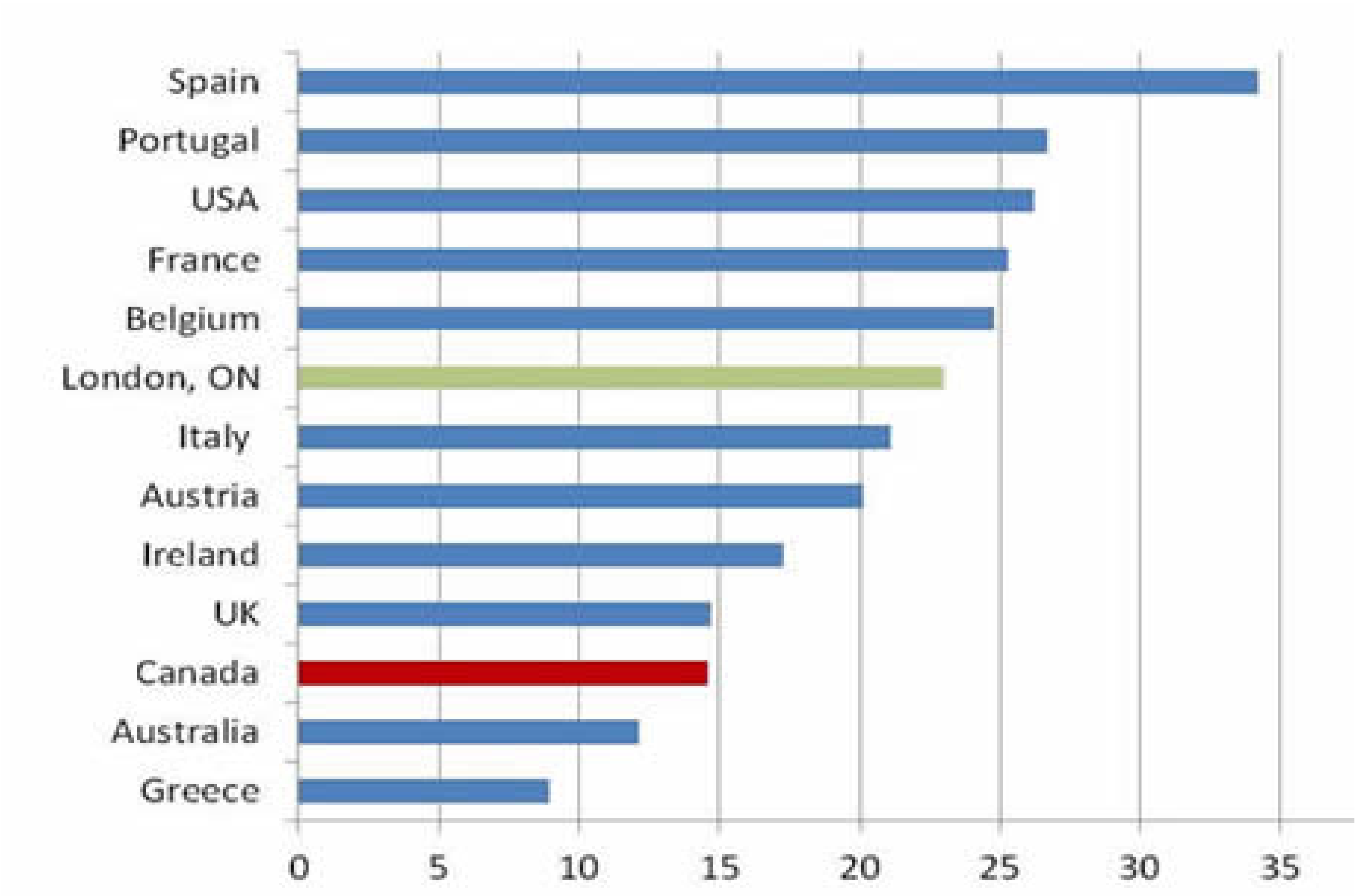
People die each day awaiting a transplant.

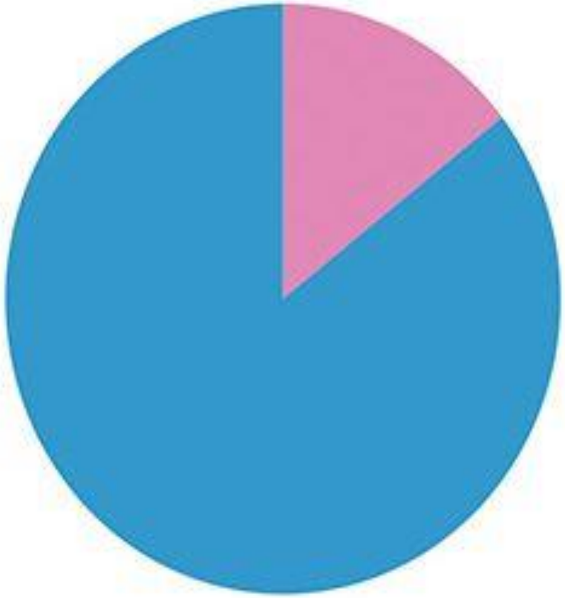
Every 10 MINUTES someone is added to the transplant waiting list.

Organ donation rate in 2016 (number of donors per million population)



Organs Donated by Deceased and Living Donors (2016)

number of donors per million population



Living – 5,976

Deceased – 35,360

- More than 4 out of 5 donations came from deceased donors, and 1 out of 5 donations from living donors.
- 33% of deceased donors was over the age of 50.
- 60% of deceased donors were male, 40% female.

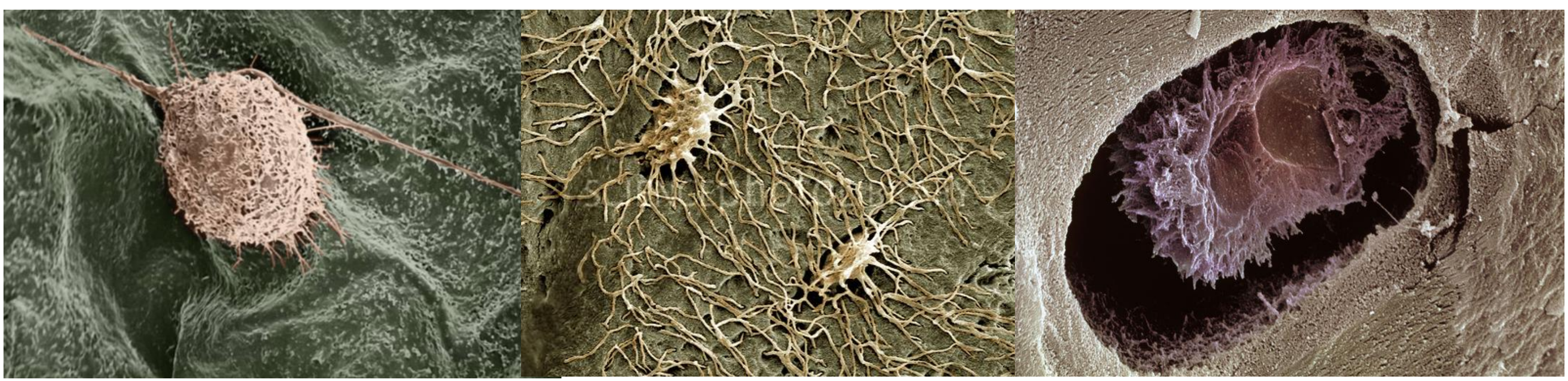
Some Bioethical Issues

- Obtaining consent and payment for an organ to be transplanted.
- Transplantation tourism and the socio-economic context. Limited supply of organs is causing serious crime of organ trafficking. Compensated donation (donors get money or other compensation in exchange for their organs) is common in Pakistan, India, China, and some other countries and helps in driving medical tourism.
- The gap between organ need and organ availability continues to widen despite very substantial public education efforts on organ donation.
- This background needs to be dealt with immediate attention and demands for finding an alternative source of organ transplantation.

Basic strategies of recovery or bio-engineering of human organs

Bio-engineering and regeneration technologies hold the promise to address two most urgent needs in organ transplantation:

- the identification of a new, potentially inexhaustible source of organs
- the immunosuppression-free transplantation of tissues and organs.



Different Cell Types in the Body

Cultivation of artificial organs

- Organs can be artificially grown both in the human body and outside the body.
- In some cases, it is possible to grow an organ from the cells of the person to whom it is going to be transplanted.
- A number of methods have been developed for the cultivation of biological organs, for example, using special devices operating on the principle of 3D printer.

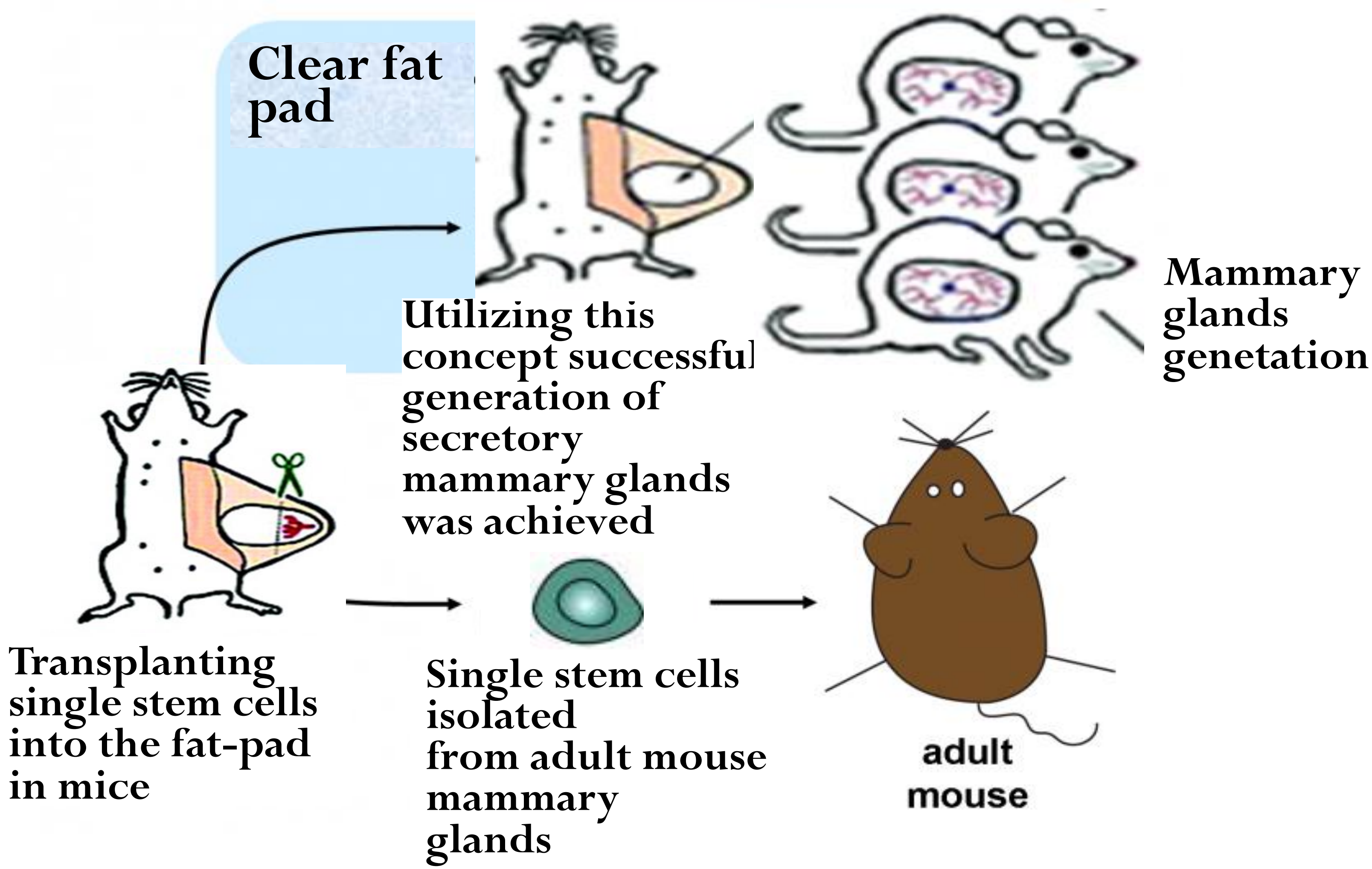
Ott HC et al., Perfusion-decellularized matrix: using nature's platform to engineer a bioartificial heart, Nature Medicine, 2008

AREAS OF APPLICATIONS

- Artificial Bone
- Artificial Skin
- Bionics
- Biomedical Engineering
- Cochlear Implant
- Ocular prosthetic
- Organ Anatomy
- Organ Transplant
- Plastic Surgery
- Prosthesis
- Tissue Scaffold
- Decellularization and others.

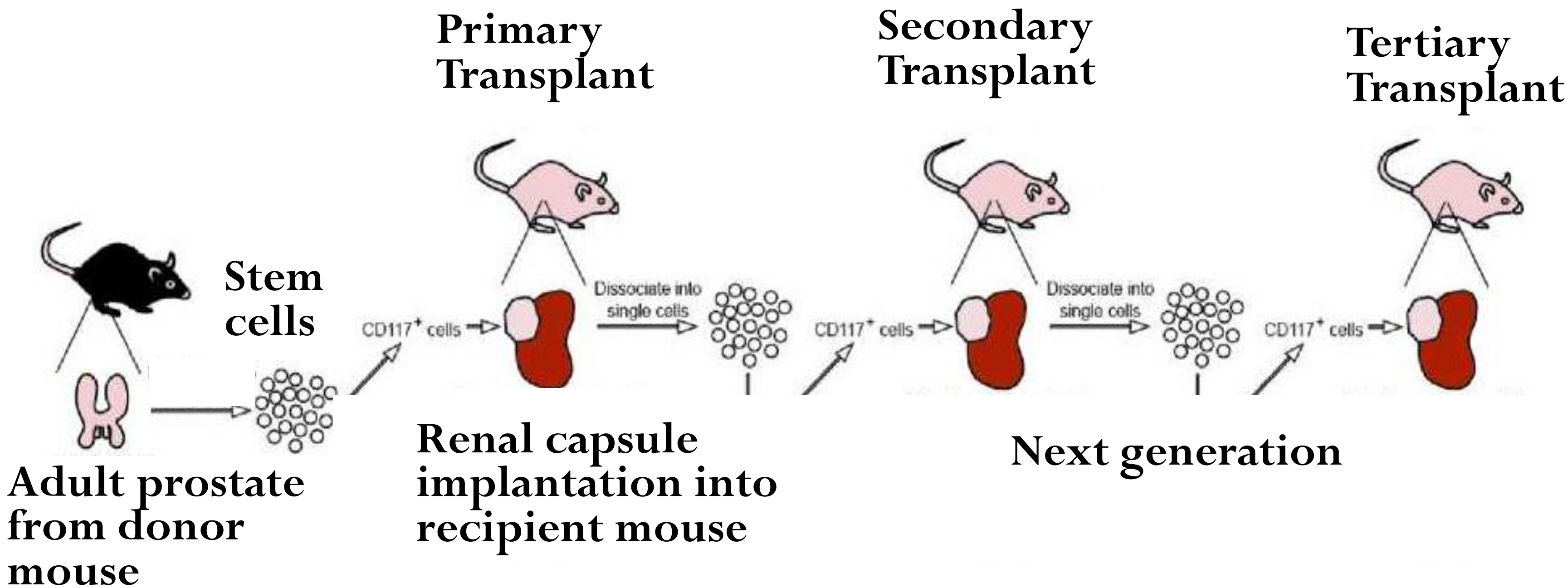
ARTIFICIAL ORGANS

Generation of a functional organ from a single adult tissue stem cell



Ref.: Generation of a functional mammary glands from a single stem cell.
Shackleton M, Vaillant F, Simpson KJ at all. **Nature. 2006** Jan 5; 439(7072):84-8.

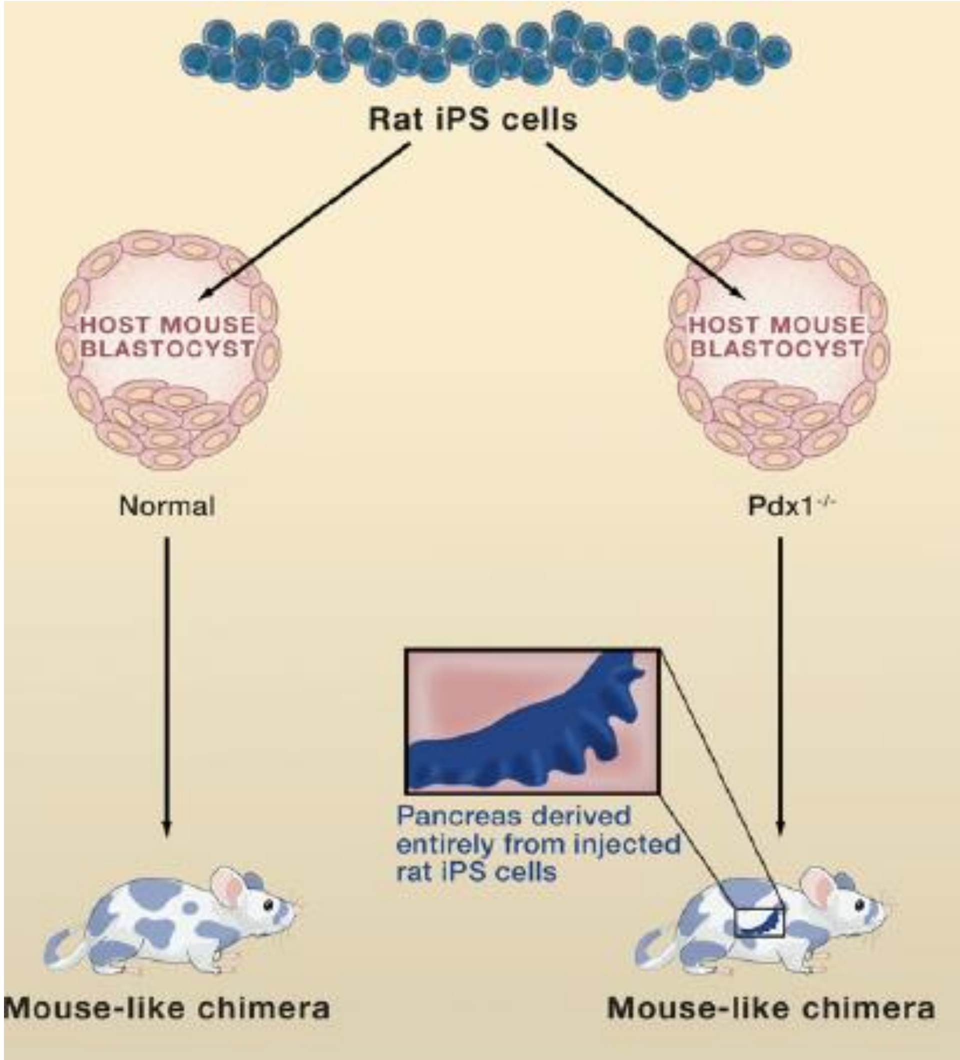
Generation a functional prostate from a single stem cell



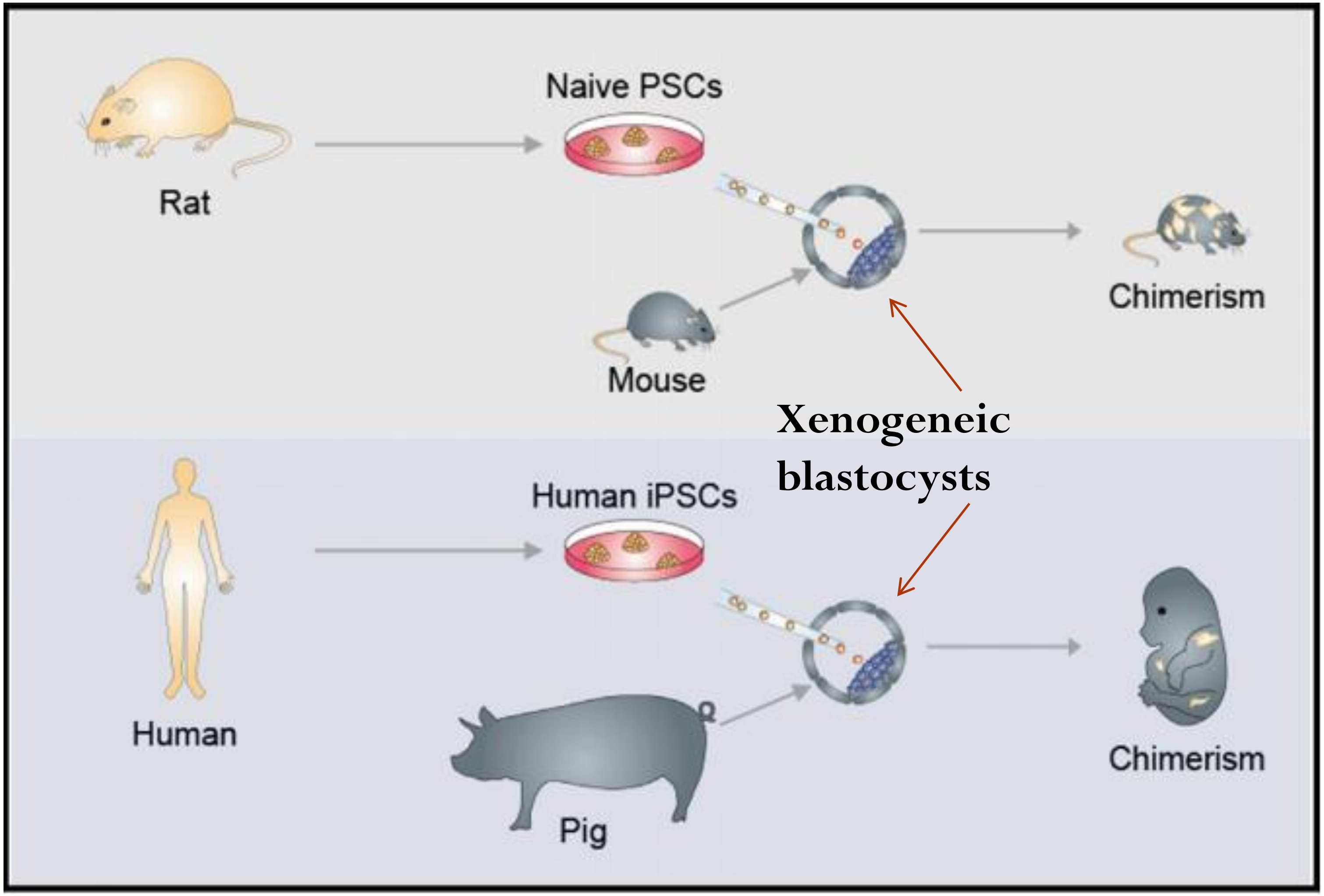
The approach bases on using a colony-formation *in vivo* assay and an *in vivo* renal capsule transplantation.

Ref.: Generation of a prostate from a single stem cell. Leong KG, Wang BE, Johnson L et all. **Nature. 2008** Dec 11; 456(7223):804-8.

Generation of organs using a blastocyst complementation system

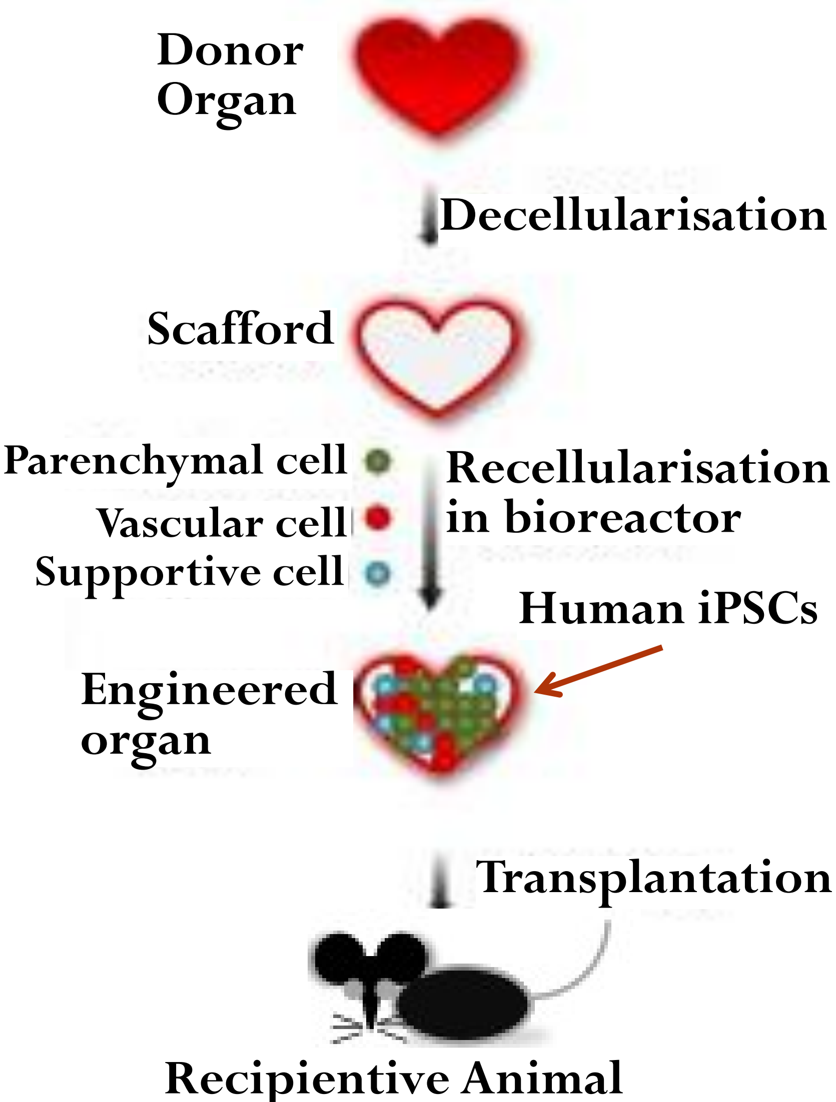


- This approach can be used to create chimeric animals that have organs belonging to another species.
- A missing organ can be generated from exogenous cells.
- The concept was successfully applied to generate pluripotent stem cell – derived rat pancreas and kidney.
- Ref.: **Generation of rat pancreas in mouse by interspecific blastocyst injection of pluripotent stem cells**. Kobayashi T, Yamaguchi T, Hamanaka et al. **Cell. 2010** Sep 3; 142(5):787-99.

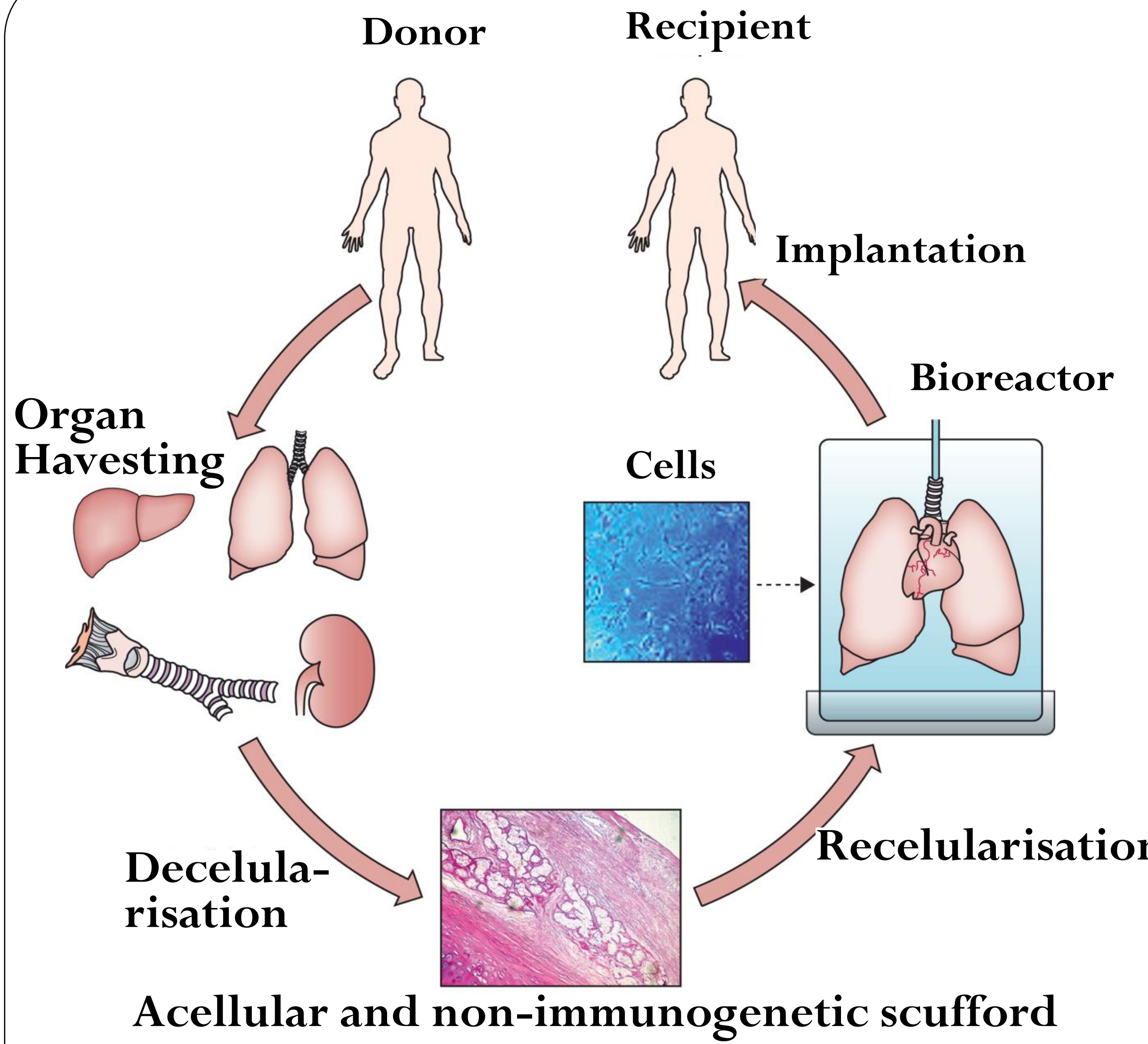


- This blastocyst complementation system may provide a novel approach for organ supply by generating specific human organs from various PSCs injected into xenogeneic sources.

Decellularization of matrix bioscaffolds and recellularization with stem cells



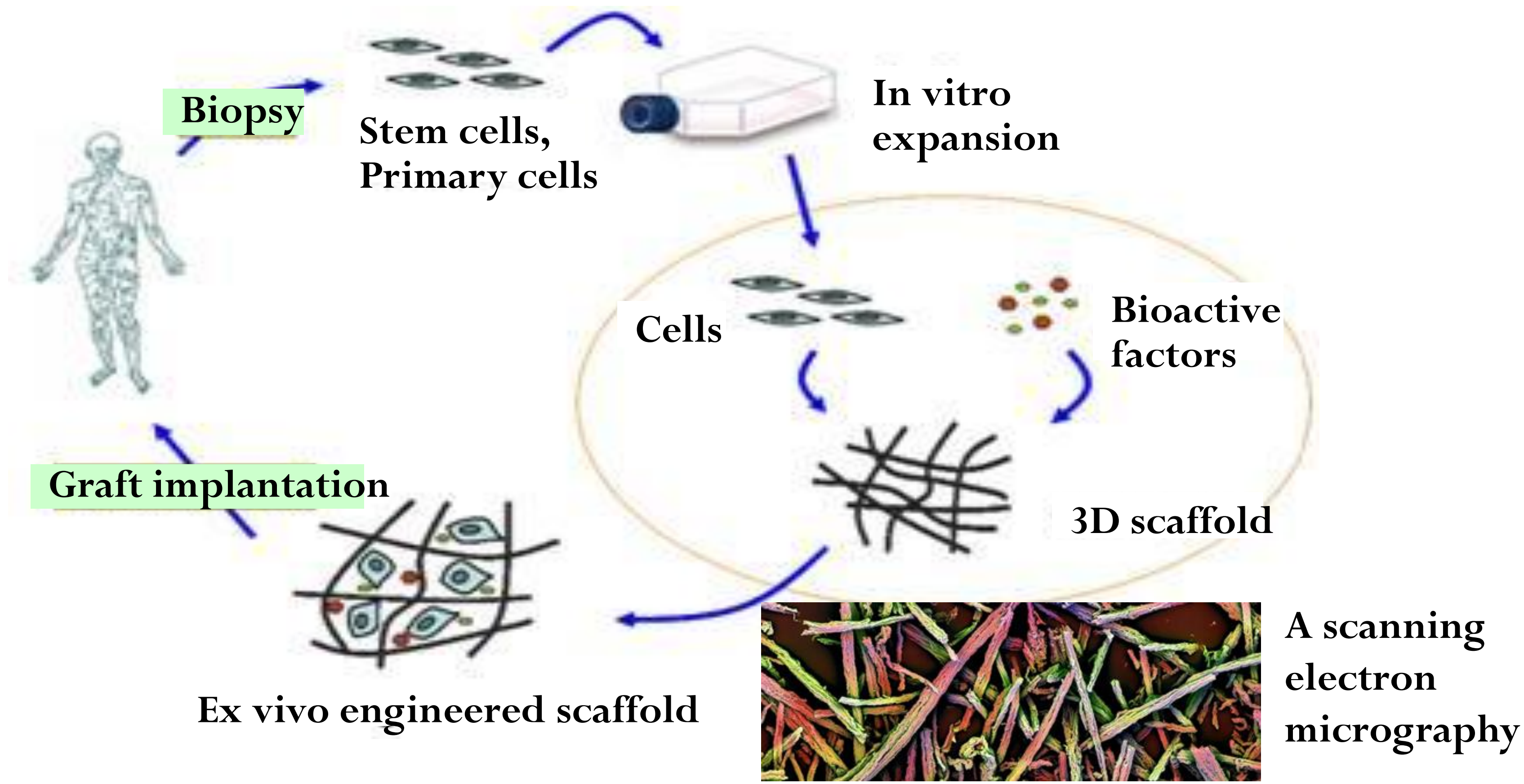
- Decellularized organs provide the ideal transplantable scaffold with all the necessary microstructure and extracellular cues for cell attachment, cell differentiation, cell vascularization, and cell function.
- **Ref.: Repopulation of decellularized mouse heart with human induced pluripotent stem cell-derived cardiovascular progenitor cells**.*Lu TY, Lin B, Kim J et all.* ***Nat Commun***. *2013.*
- Macchiarini *et al.* (***Lancet, 2008***) performed the first adult stem cell grown trachea transplant, obtained by decellularizing deceased donor trachea. This approach has been extended to treat patients with tracheal cancer.



Literature shows successful utilization of decellularized scaffolds for tissue-engineering of lung (*Petersen TH et all, Science. 2010*), liver (*Baptista PM et all, Hepatology. 2011*), urinary bladder (*Rosario DJ et all, Regen Med. 2008*), urethra (*El-Kassaby AW et all, Urol. 2003*), and blood vessel (*Amiel GE et all, Tissue Eng. 2006*) and other organs.

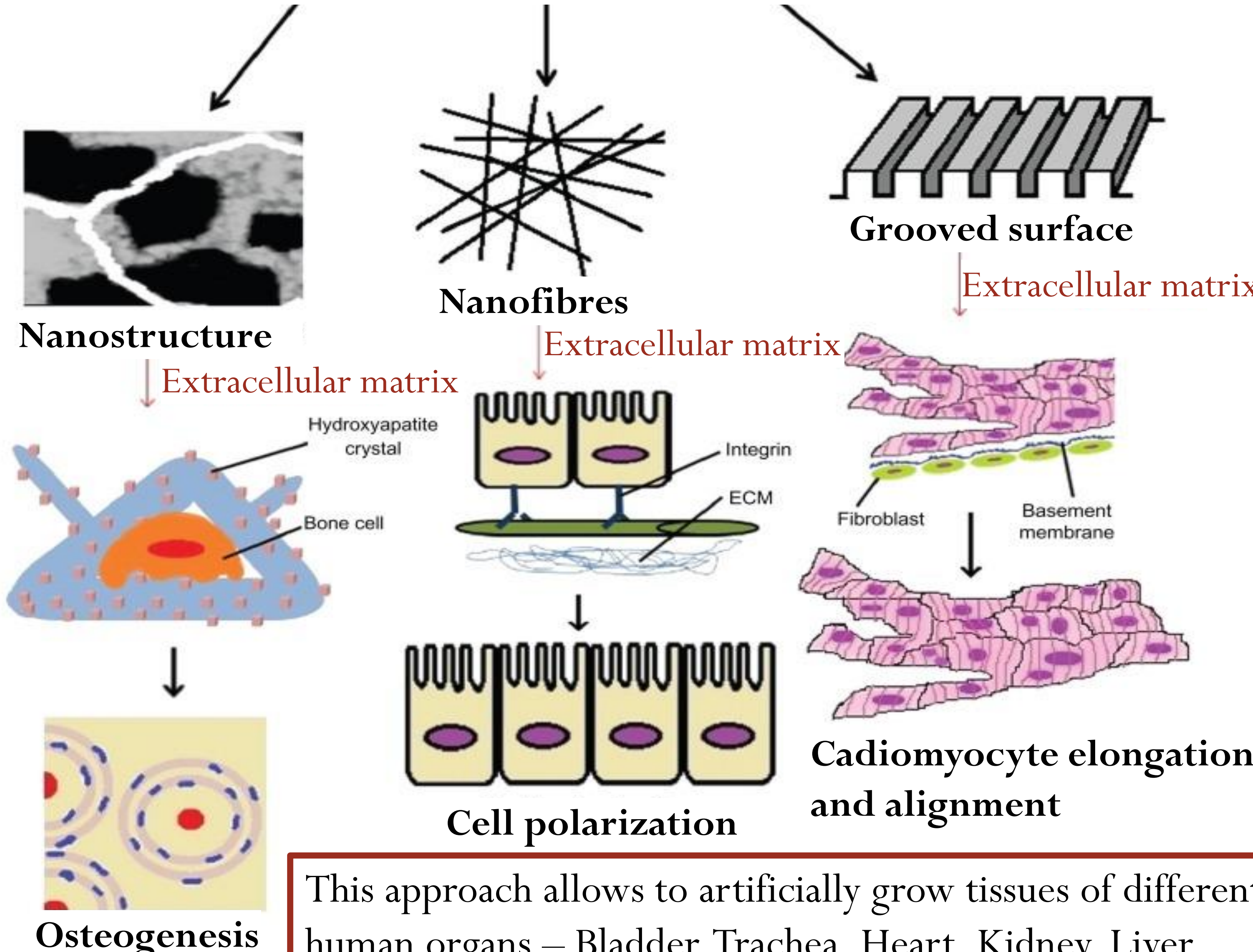
Extracellular matrix scaffolds are currently used for arterial grafts, heart valves, skin reconstruction, urinary tract reconstitution, and orthopedic applications. Some of the scaffolds are available commercially.

Basis of Tissue-engineering



Concurrent growth in the fields of material bio-engineering and cell biology has led to the possibility to grow tissue-engineered organs.
Bio-engineered organs have advantage of not prone to transplant rejection as they rely on a patient's own cells.

Different Scaffold Fabrication Methods Using Nanomaterials



This approach allows to artificially grow tissues of different human organs – Bladder, Trachea, Heart, Kidney, Liver, Ovaries, Thymus, Ear, Skin.

Organ printing

- Organ printing is a new emerging technology which involves three sequential steps:

Preprocessing in which digitized image reconstruction of a natural organ or tissue is obtained

Processing in which actual organ printing is done by layer-by-layer placement of cells or cell aggregates into a 3D environment

Post-processing involving perfusion of printed organ and accelerated organ maturation

Bio-printers

Bio-printed human ear

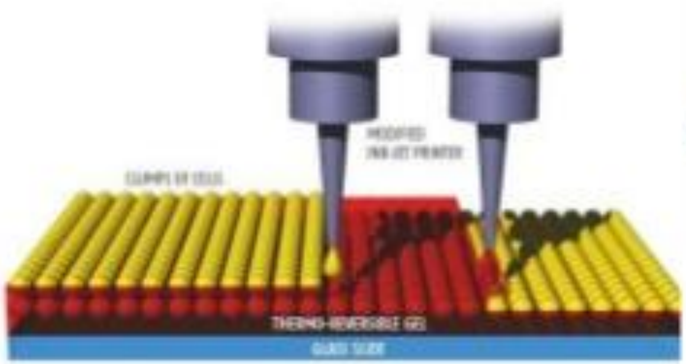
Organovo company was the first to commercialize 3D bio-printing technology (Doyle K. *Bioprinting: From patches to parts. Gen Eng Biotechnol. 2014*).

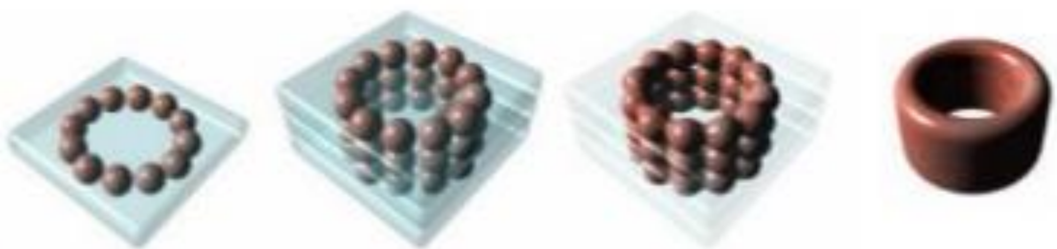
Bioprinting process flow

Computer model → Printing → Post-processing

3 important components: Bioink, Biopaper, Bioprinter.

With adding growth factors

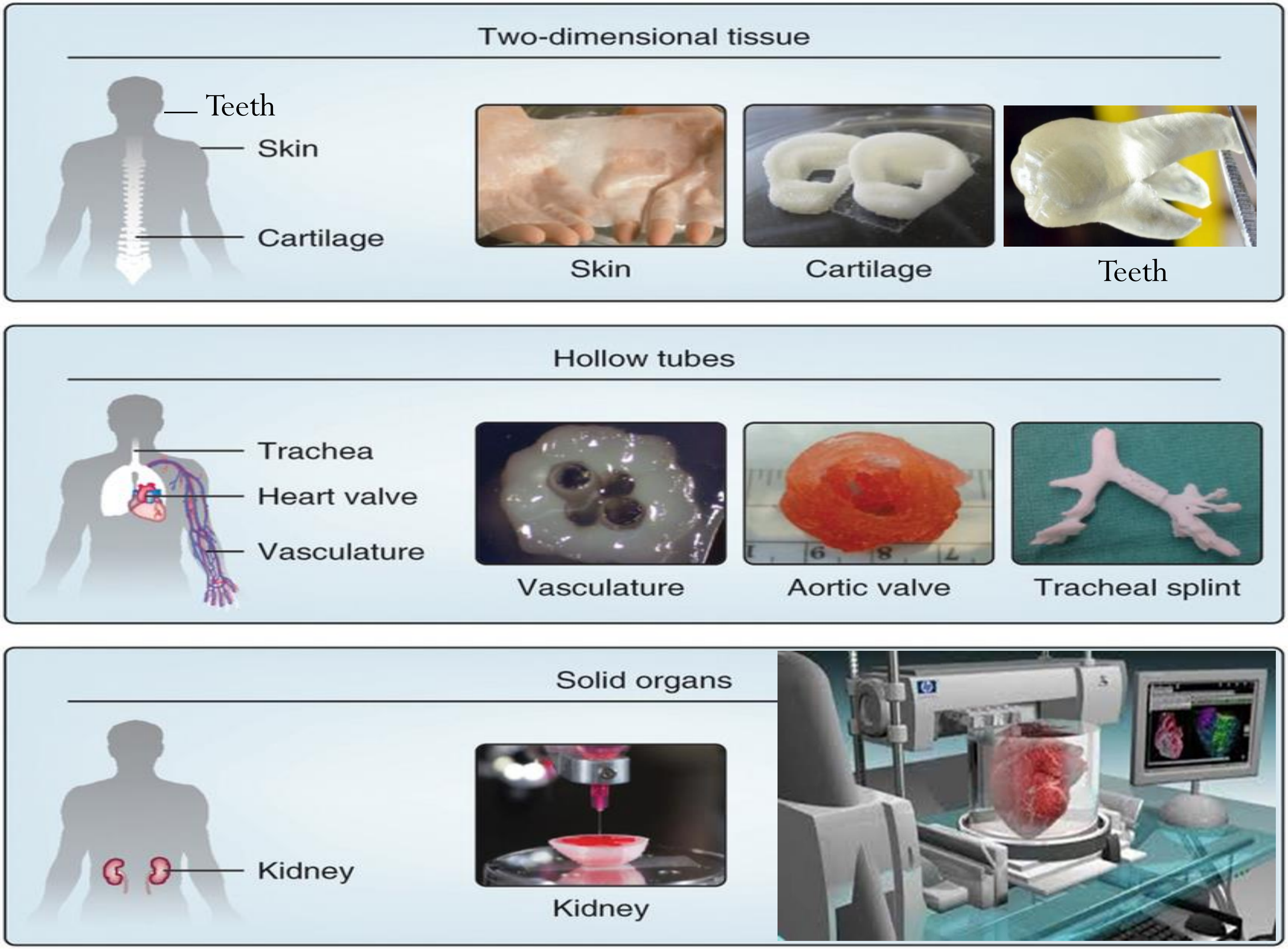




Bioink printed into layer of biopaper gel	Additional layers printed to build object	Bioink fuse together and biopaper dissolves	Final living tissue

Bioink (cells of sp. organ), Biopaper (collagens, nutrients)

Examples of human-scale bioprinted tissues



Limitations of Organ Bio-Engineering

- All the successful implanted bio-engineered organs are hollow organs, whereas the bio-engineering of modular organs such as cardiac, renal, hepatic, and pancreatic is still far from the realm of possibility.
- The reported clinical implantations of bio-engineered organs are with short follow-up and insufficient discussion of complications and limitations that may occur with time.
- The iPS cell technology has great potential, but it is important to evaluate the methodologies for iPS cell generation for their safety and efficacy.
- The present technology does not seem to produce adequate bioreactors to mimic *in vivo* conditions, i.e., temperature, nutrient, and oxygen concentration.
- Decellularizing process potentially may lead to loss of surface structure and composition resulting in reduced mechanical properties as compared with those of the normal native organs.

XENOTRANSPLANTATION

- **Xenotransplantation** is the transfer of living cells, tissues and/or organs from non-human animal species into humans (although technically, it could be the other way around or between any two species.

Why is xenotransplantation important?

- There is a worldwide shortage of supply of organs for clinical transplantation, and many people die waiting for organs to become available.
- The development of xenotransplantation can be seen as serving several purposes:
  - to be a complete substitute for human organs;
  - to supplement human organs, thus easing the current shortage available for transplantation;
  - to be a "bridge" organ before a "destination" organ can be found.
- Xenotransplantation is currently not a recognized medical practice in industrialized countries, but has the potential to encompass a broad spectrum of applications.

Potential risks

- **Xenosis** is the infection of humans by agents such as bacteria or viruses that are derived from animals. The infection may or may not result in symptoms of human disease. It is a question of human safety, the worst-case scenario could be a major new epidemic.
- **The risk of rejection** in xenotransplantation primarily involves the immune system attacking the transplanted tissue and not recognizing it.

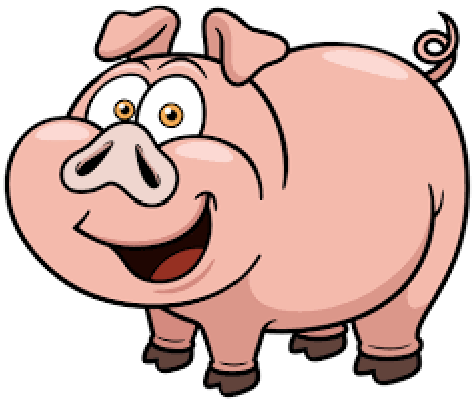


How to overcome the rejection phenomenon

To alter the patient's immune system to increase its tolerance to transplanted tissues, cells or organs so that no immune response is initiated.

To change the cells, tissues and organs of the donating animal, especially by deleting certain animal genes and replacing them with human genes.

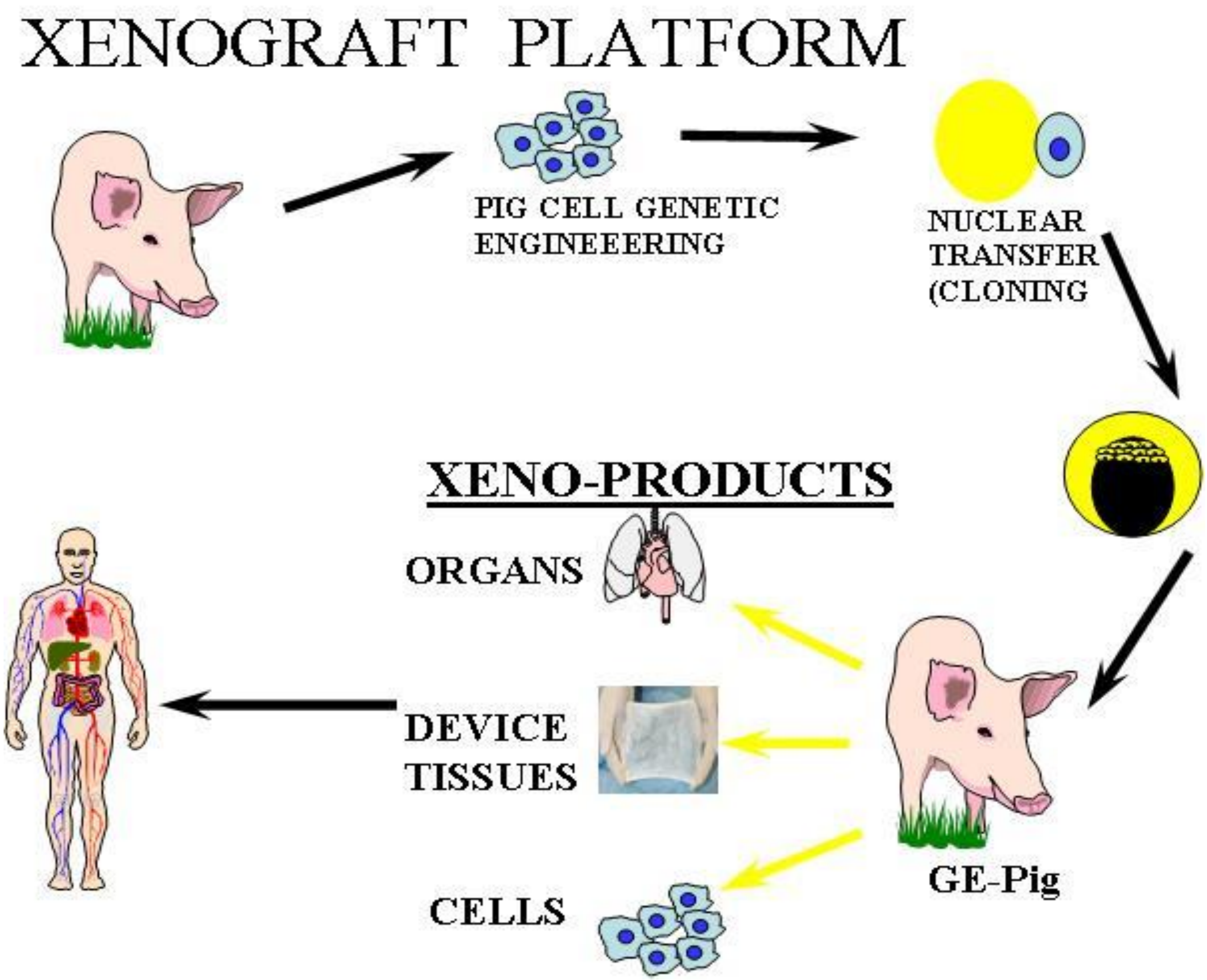
What animals may be suitable donors?



- Even though non-human primates, such as apes or baboons, are genetically closest to humans, the transfer of infective agents could be more likely between closely related species.

However, some new pig endogenous retroviruses, have been shown to infect human cell lines in vitro.

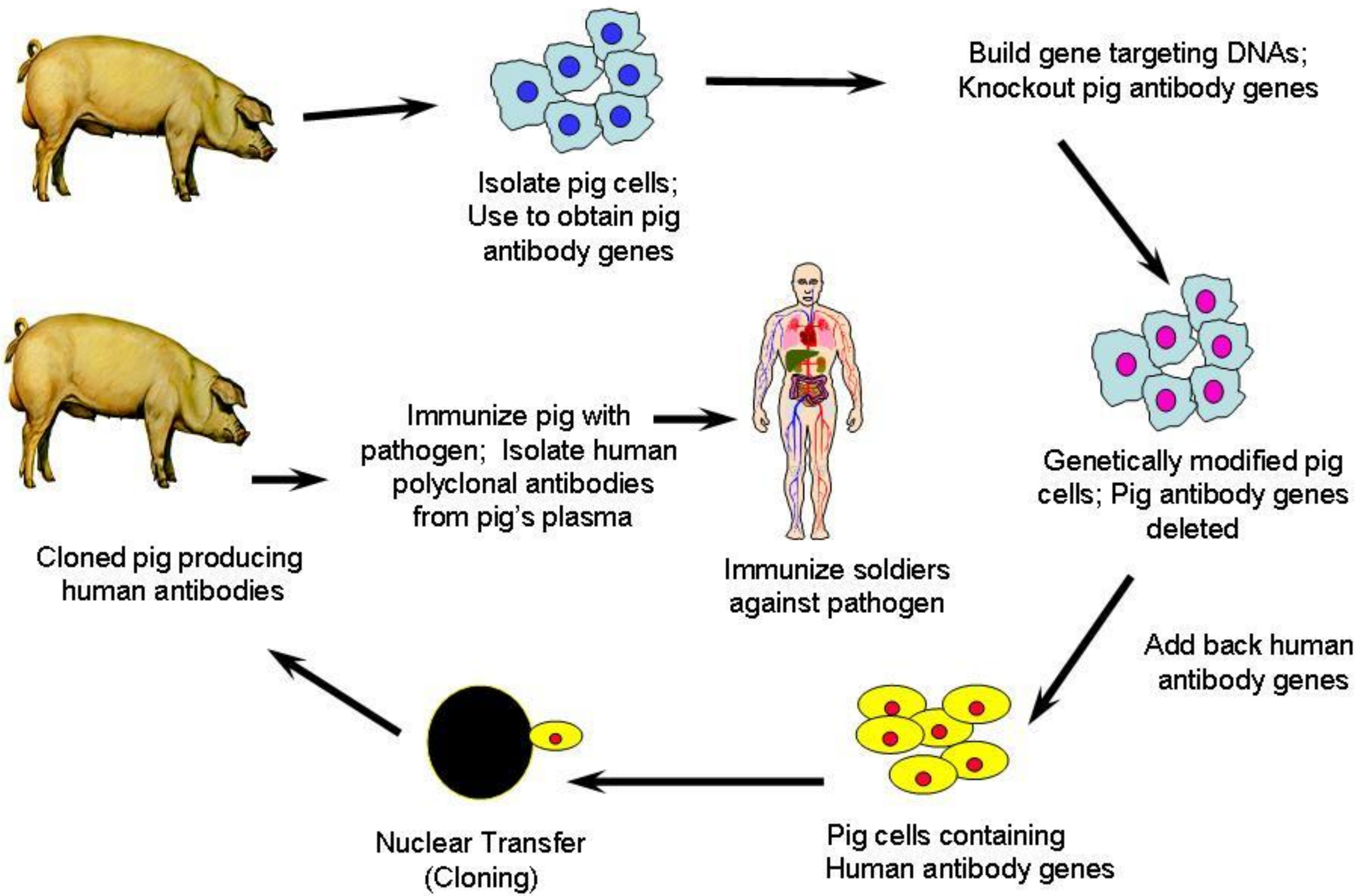
- Pig's organs are the most comparable in size to adult human recipients.
- They quickly grow to maturity.
- They can be raised solely for transplantation purposes in clean environments.
- They may pose less risk of infection to humans than non-human.



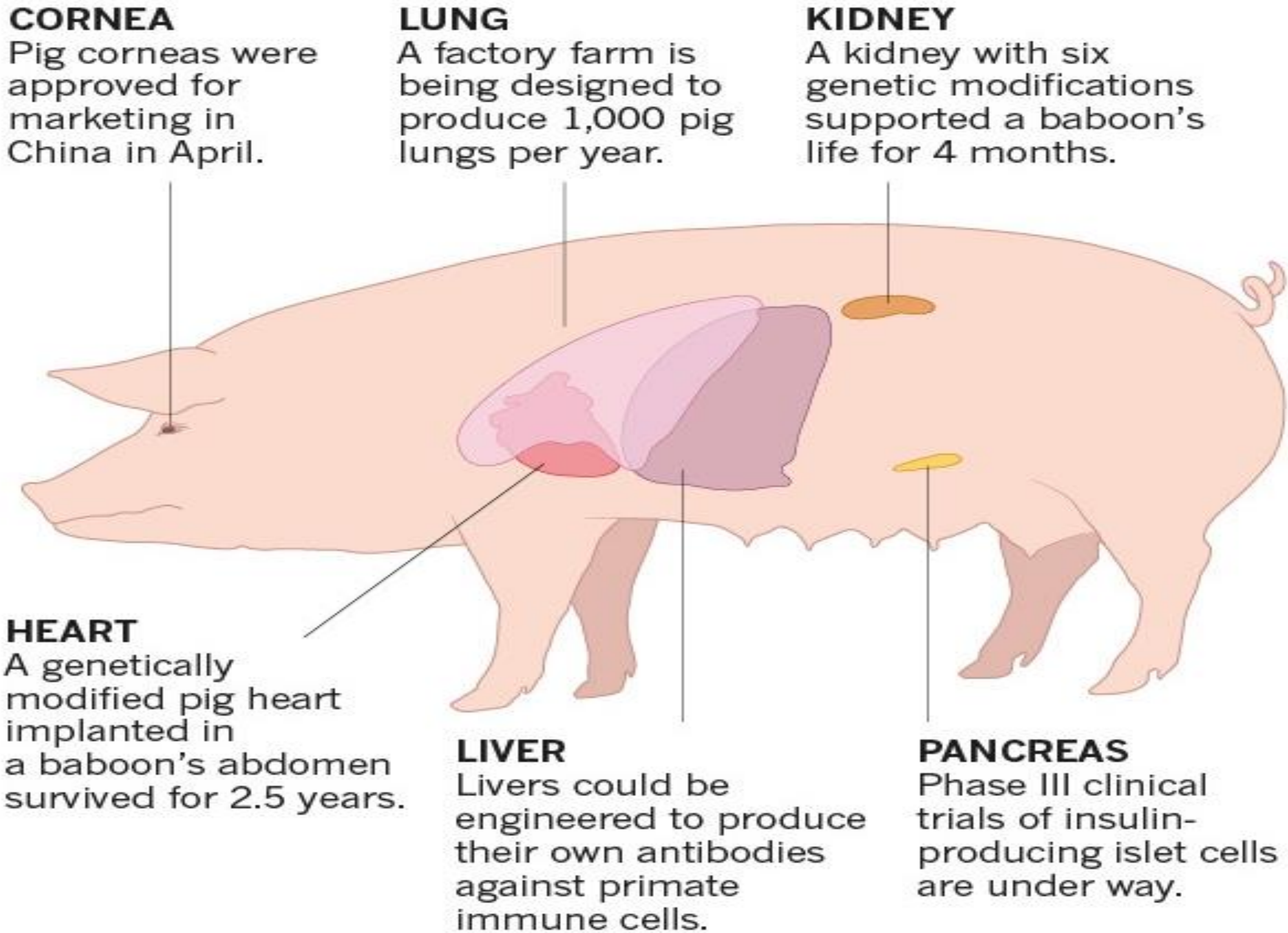
Medical science already uses animal parts for various therapeutic reasons, such as replacement heart valves from pigs.

However, these products have been chemically treated – they are not functional, living tissue. This makes them different from the living cells, tissues and organs used in xenotransplantation.

Generation of "Immunopig"



Some perspective directions of pig-to-human transplantation



Ref.: New life for pig-to-human transplants. Sara Readon, *Nature, 2015*.

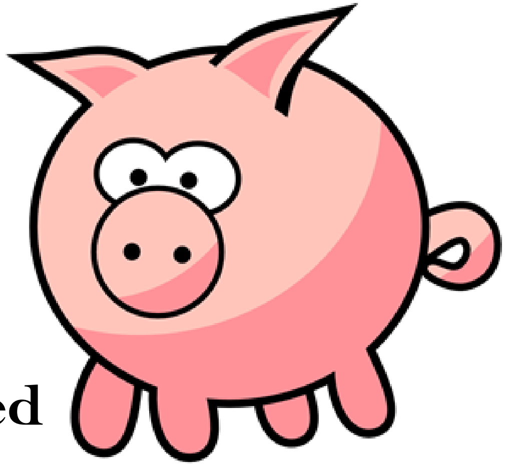
Regulatory status in the world

- The US Food and Drug Administration has updated its 1996 Public Health Service *Guidelines for Infectious Disease Issues in Xenotransplantation*. It has also approved clinical trials involving xenotransplantation.
- In Great Britain, clinical trial applications may be submitted to the United Kingdom Xenotransplantation Interim Regulatory Authority. Established in 1997, it has received no clinical trial applications to date. They are still in progress.
- The World Health Organization hosted a consultation with international experts and published guidelines in 1998 on preventing and managing infectious disease associated with xenotransplantation.
- In 1999, the Council of Europe's Parliamentary Assembly called for a moratorium until the technology is evaluated and guidelines, which balances ethical, medical, scientific, legal, social and public health issues, are established.
- According to the Organisation for Economic Co-operation and Development, limited clinical trials involving xenotransplantation are planned or ongoing in some countries, such as the United States, Belgium, Spain and Germany.

Some Etical Issues

In the human-based side:

- It is better to keep this issue in the experimental level and clinical trials in a limited range due to lack of scientific certainties including diseases manifestations, virus transference from animal to human.
- Its transference or spread in the society and also the degree of pain which is imposed on the patient and his/ her relatives.



From the animal-based side:

- Starting this treatment process might destroy animal welfare broadly.
- Its genetic structure can be modified.
- The animal will not have the chance to live its instincts necessary to live.
- This method at its present level is neither justified nor verified from the ethical point of view.
- **BUT** if its benefit and safety is proved, compromising the animal welfare to save a human life is justifiable.

CONCLUSION

- The march of regenerative technology has changed our concepts of organ transplantation.
- Although we are not theoretically far-off from beginning to understand human regeneration, it still remains a science of future due to ethical limitations.
- Regardless of whether regenerative organ therapy succeeds, we believe it may generate new knowledge and new visions about how organs may be replaced in the future.
- It is the shared hope that regenerative medicine may one day augment organ transplantation by developing a new source of organs or potentially rehabilitating those that are not transplantable.

Thank you for the attention