Northeastern University

Department of Bioengineering

BIOMEDICAL ENGINEERING SOCIETY

"BOOSTING BIOETHICS & BIOPRINTING" WEBINAR SERIES

3D Bioprinting for Pediatric Congenital Heart Defect Repair: Current Technologies and Ethical Challenges

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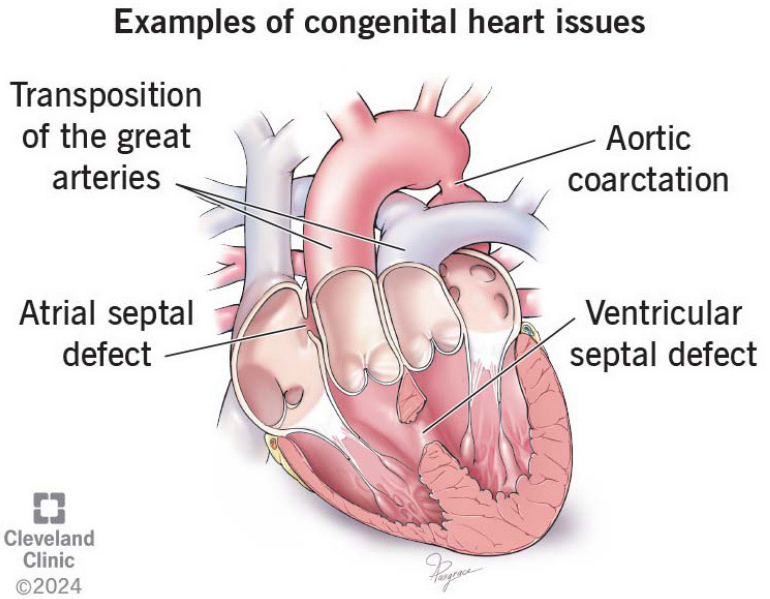
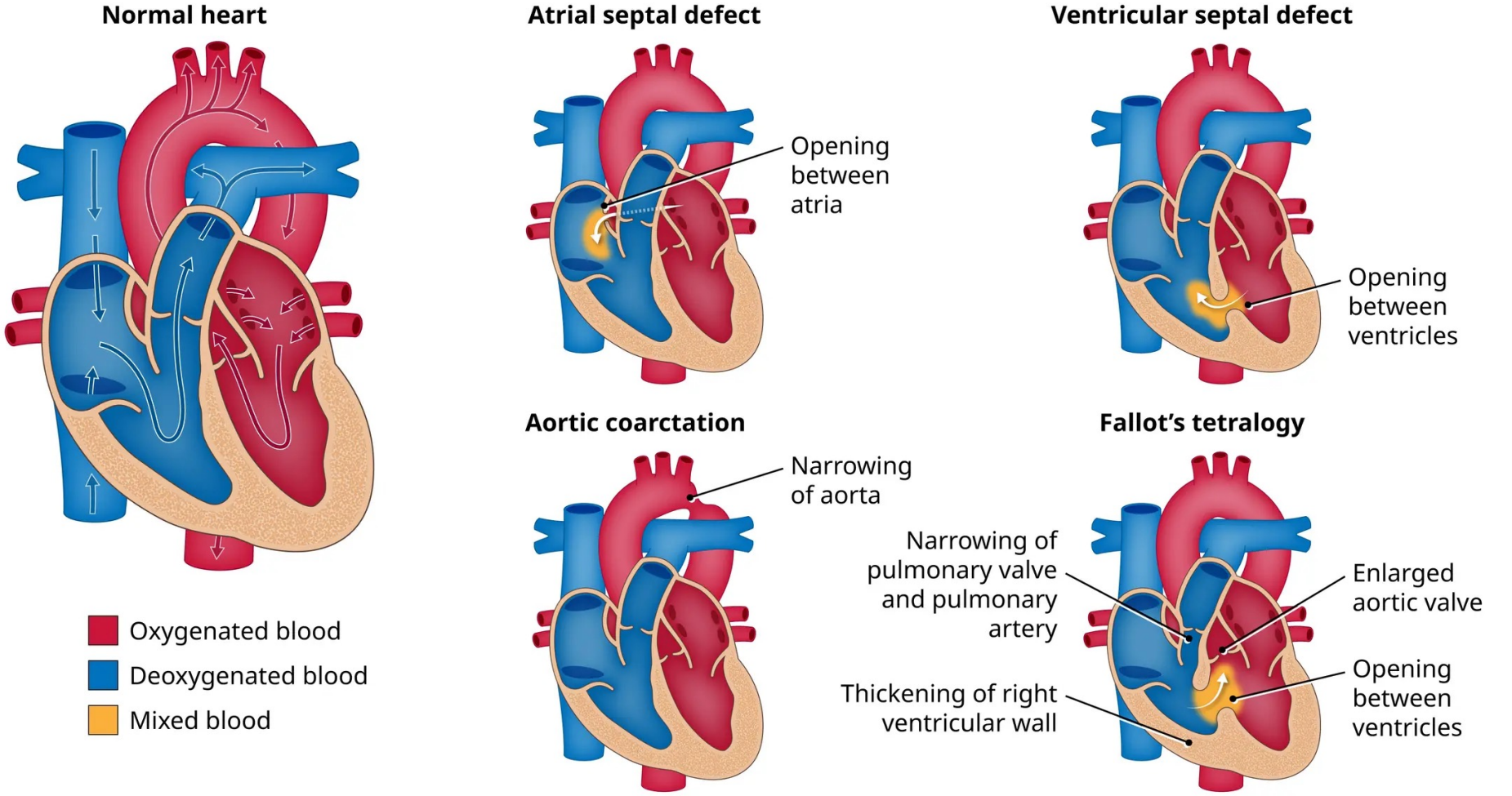
Pediatric Congenital Heart Defects (CHD)

- CHD are present in **1%** of newborns.
- Complications: defects in childhood development, cardiopulmonary anomalies, lead to other system/organ failure.
- Inpatient costs for a single-year cohort through age 10: **~ $1 billion**
- One of the **leading causes of death** in newborns.
- **25% of CHD** are considered critical – require surgery.



Trusty P. et al, *J. Thorac. Cardiovasc. Surg.*, 2020

Congenital Heart Disease



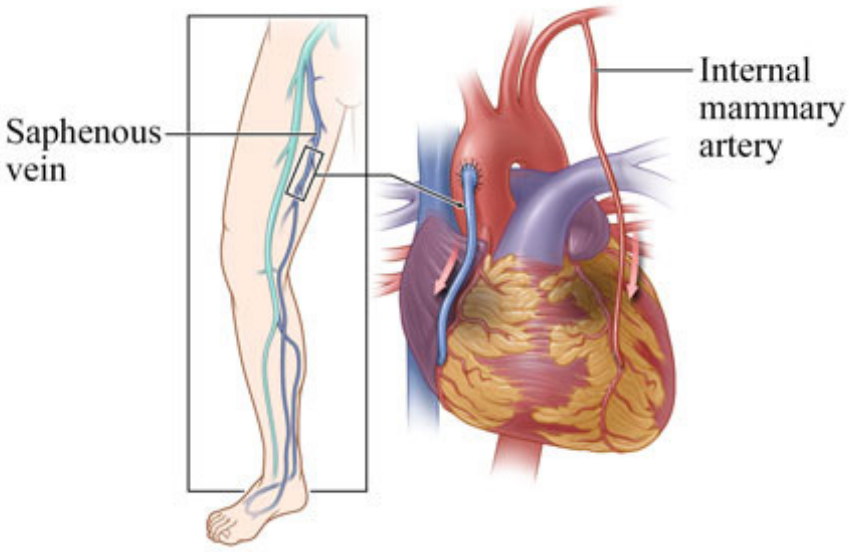
Current surgical approach to fix CHD

- Synthetic grafts: polyethylene terephthalate (Dacron) or polytetrafluoroethylene (PTFE or Gore-Tex).
  - High risk of thrombosis, need lifetime anticoagulation therapy.
  - Material-related failures, including stenosis, calcification, or infection.
  - Mismatched geometry and mechanical property.
  - **Cannot grow** with pediatric patients, leading to *repeated surgery*

PTFE graft, GORE Medical

Current surgical approach to fix CHD

- Tissue grafts: more biocompatible and can grow with the children
  - Allografts
    - availability
    - immune rejection
    - disease transmission
    - chronic inflammation.
  - Autologous Vein
    - not available for pediatric patients (need to save for later).
  - Can not match the complex geometry of CHD



bypass graft using saphenous vein

Desired graft properties for CHD repair

- Good biocompatibility
- Low immune response
- Without the need of lifetime anticoagulation drug
- Can grow and remodel with the children (need to have cells?)
- Can match the exact size and geometry of the complex CHD for better hemodynamic profile (3D bioprinting ?)

Tissue-engineered Vascular Grafts in Children With CHD: Intermediate Term Follow-up

- The graft is made of a woven fabric of poly-l-lactide acid or polyglycolic acid and a 50:50 poly (l-lactic-co-ε-caprolactone) copolymer.
- The graft is seeded with autologous bone marrow mononuclear cells, and implanted into patient.
- In 2001, the ethics committee at Tokyo Women's Medical University approved the implantation of TEVGs in human subjects.

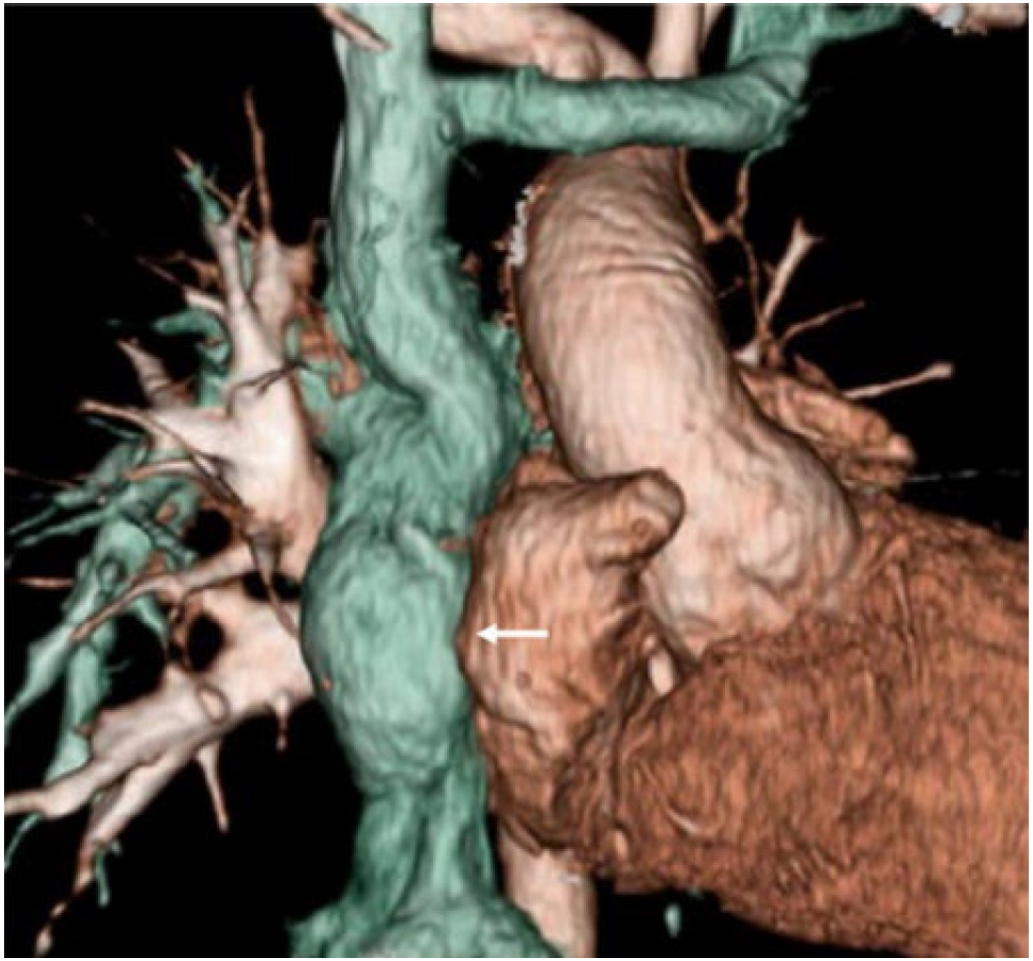
Sugiura *et al. Semin Thorac Cardiovasc Surg*. 2018 Feb 7;30(2):175–179.

Tissue-engineered Vascular Grafts in Children With CHD: Intermediate Term Follow-up

- The graft is implanted in 25 patients with CHD.

- Inclusion criteria for patient screening:
  - elective surgery.
  - age younger than 30 years.
  - full understanding of the procedure by the patient or family.
  - minimal extracardiac disease burden.
  - informed consent was obtained from each patient, or from the parent or guardian

- Patients were followed up with postoperatively in a multidisciplinary clinic.



3D CT image of the tissue-engineered vascular graft.

Patient	Main Diagnosis	Age	Graft Type	Graft Size
1	Asplenia, AVSD(A), small RV	2	PLA	16
2	Asplenia, SRV, TAPVC(Ib+III)	1	PLA	20
3	Concordant criss-cross heart, DORV, PAA, MS	8	PLA	18
4	TA(Ib)	22	PLA	24
5	PPA, ASD(II), sinusoidal communication	13	PLA	22
6	SRV, DORV, AVVA	4	PLA	20
7	Total sinus defect, ASD, TR(IV)	14	PLA	24
8	Asplenia, SLV, CAVVR	17	PLA	24
9	TA(Ib)	22	PLA	22
10	Polysplenia, SRV	4	PLA	12
11	HLHS, MA, IAA(A)	2	PLA	16
12	Asplenia, SRV, PAA, nonconfluent PA	2	PGA	16
13	SLV, lt AVVA	2	PGA	16
14	DORV, small LV, VSD, PS, ASD(II)	2	PGA	18
15	Polysplenia, cAVSD, DORV, PS	2	PGA	12
16	Asplenia, SRV, CA, TAPVC(Ib)	2	PGA	16
17	PPA, RA thrombosis, AFL, af	24	PGA	18
18	SRV, DIRV, PA, ASD(II)	1	PGA	16
19	Asplenia, SRV, PS, CA	11	PGA	18
20	Polysplenia, cAVSD, PS, CAVVR	2	PGA	14
21	DORV, VSD, small RV, PLSVC, TAPVC(IIb)	3	PGA	16
22	PPA, ASD(II), PS, Sinusoidal communication	5	PGA	18
23	SLV, DILV, PA, ASD, bilateral SVC	4	PGA	18
24	Asplenia, SRV	13	PGA	16
25	TA(IIc), SAS	2	PGA	18

AF	atrial fibrillation
AFL	atrial flutter
ASD	atrial septal defect
AVSD	atrioventricular septal defect
AVVA	atrioventricular valve atresia
CA	common atrium
cAVSD	complete atrioventricular septal defect
CAVVR	common atrioventricular valve regurgitation
CAVV	common atrioventricular valve
DILV	double-inlet left ventricle
DIRV	double-inlet right ventricle
DORV	double-outlet right ventricle
HLHS	hypoplastic left heart syndrome
IAA	interruption of aortic arch
LV	left ventricle
MA	mitral atresia
MS	mitral stenosis
PA	pulmonary artery
PAA	pulmonary artery atresia
PLA	polylactide acid
PLSVC	persistent left superior vena cava
PPA	pure pulmonary atresia
PS	pulmonary stenosis
RA	right atrium
RV	right ventricle
SAS	subaortic stenosis
SLV	single left ventricle
SRV	single right ventricle
SVC	superior vena cava
TA	tricuspid atresia
TAPVC	total anomalous pulmonary venous connection
TR	tricuspid regurgitation
VSD	ventricular septal defect

Patient	F/U Years	Status	Stenosis Requiring PTA	Postoperative Year of the PTA	Thrombus
1	14.9	Alive	No		No
2	14.8	Alive	No		No
3	13.7	Dead	Yes (×1)	5.5	No
4	14.3	Alive	No		No
5	5.5	Dead	No		No
6	14.2	Alive	Yes (×1)	7.6	No
7	3.8	Dead	No		No
8	14.1	Alive	No		No
9	3.6	Dead	No		No
10	11.8	Dead	Yes (×3)	5.1, 6.1, 9.9	No
11	0.5	Dead	No		No
12	13.5	Alive	Yes (×4, stent)	1.5, 2.3, 4.3, 9.4	No
13	13.3	Alive	Yes (×1)	11.9	Yes
14	13.2	Alive	No		No
15	13.2	Alive	Yes (×1)	13.1	No
16	10.1	Dead	No		No
17	12.9	Alive	No		No
18	12.9	Alive	Yes (×2)	3.8, 4.8	No
19	12.8	Alive	No		No
20	12.8	Alive	No		No
21	12.5	Alive	No		No
22	12.5	Alive	No		No
23	12.4	Alive	No		No
24	11.9	Alive	No		No
25	1.8	Dead	No		No

PTA percutaneous transluminal angioplasty

8 patients died

7 patients had stenosis and required PTA

Cause of Death of 8 Patients

Patient	Cause of Death	Year After Implantation
3	PLE, DIC, MOF	13.7
5	Sudden death	5.5
7	SLE, Aspergillus pneumonia, MOF, DIC	3.8
9	Sudden death (arrhythmia)	3.6
10	Pancytopenia, DIC, MOF	11.8
11	CHF, LOS	0.4
16	Severe AVVR, LOS	10.1
25	Sudden death	1.8

AVVR aorto-ventricular valve regurgitation
CHF congestive heart failure
DIC disseminating intravascular coagulopathy
LOS low output syndrome
MOF multiple organ failure
PLE protein loosing enteropathy
SLE systemic lupus erythematosus

Patients died of other complications, not directly from graft failure (occlusion)

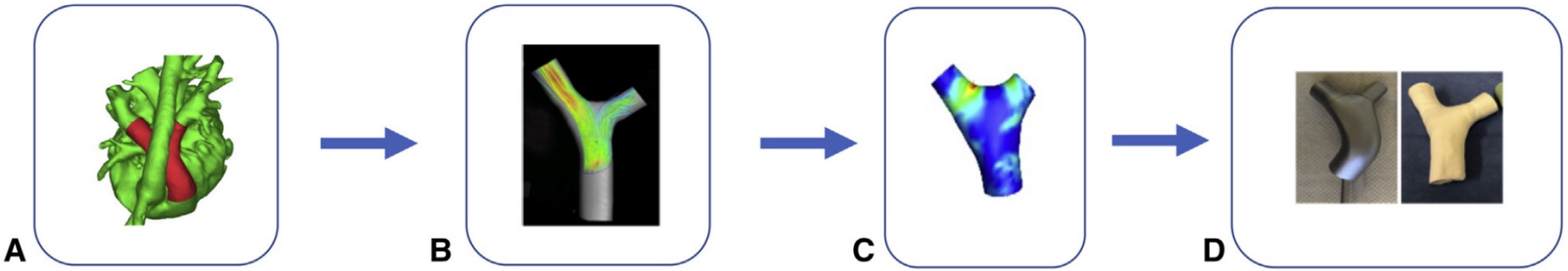
Tissue-engineered Vascular Grafts in Children With CHD: Summary

- 25 patients follow-up ranging from 1~15 years.
- There was no graft-related mortality during the follow-up period.
- No evidence of aneurysmal formation, graft rupture, graft infection, or calcification.
- 7 (28%) patients had asymptomatic graft stenosis and underwent successful balloon angioplasty.
- 8 (32%) patients died from complications (not directly from graft failure).
- Avoidance of anticoagulation therapy would improve patients' quality of life.
- Tissue-engineered vascular grafts have feasibility in pediatric cardiovascular surgery.

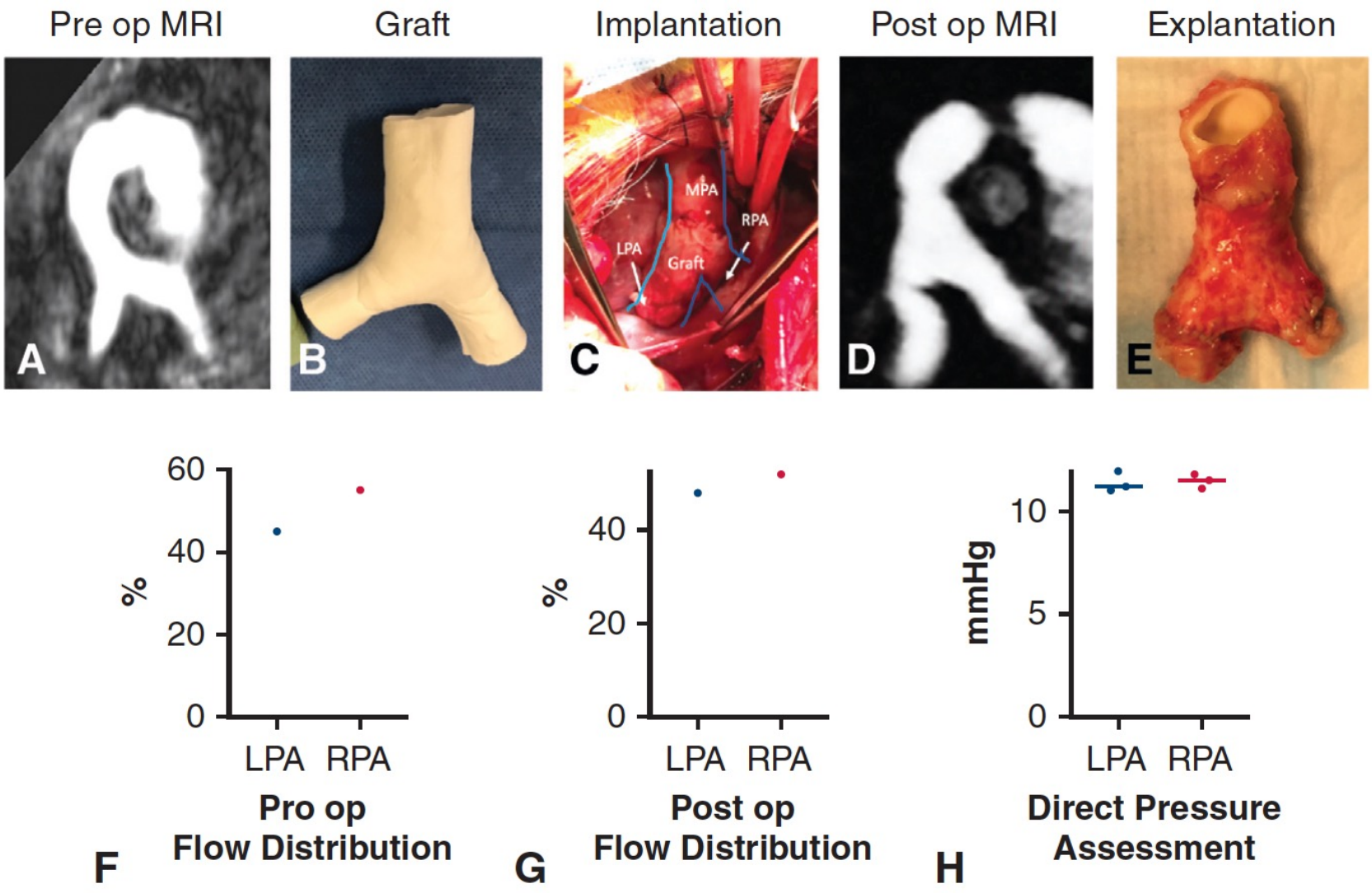
In vivo implantation of 3-D printed customized branched tissue engineered vascular graft in a porcine model

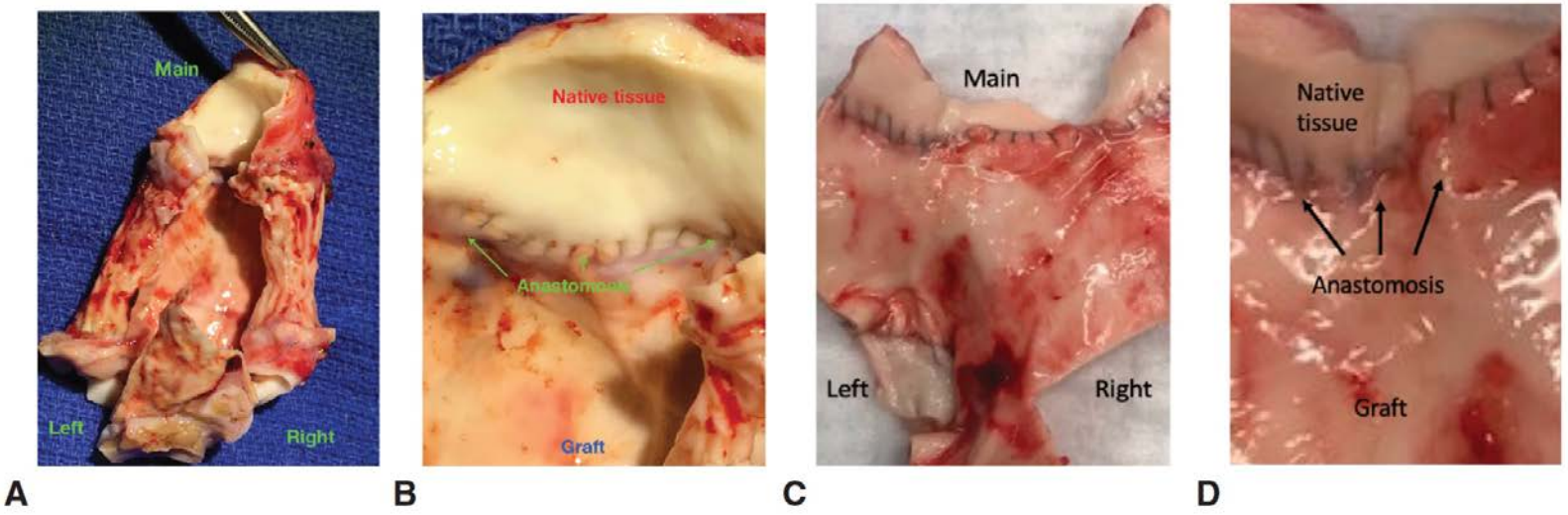
- 3-D printed graft made of PCL/PLCL was implanted in porcine model (2 pigs).
- Follow up for 1 month.

Hibino, et al. *The Journal of Thoracic and Cardiovascular Surgery,* Volume 159, Issue 5, May 2020

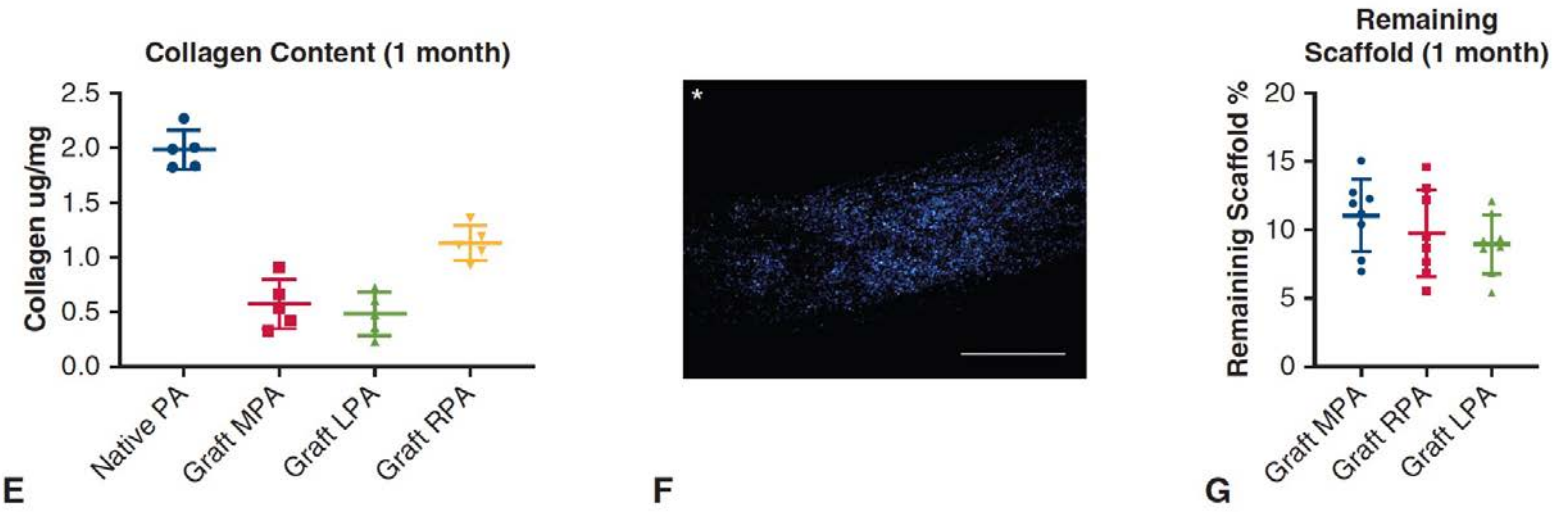


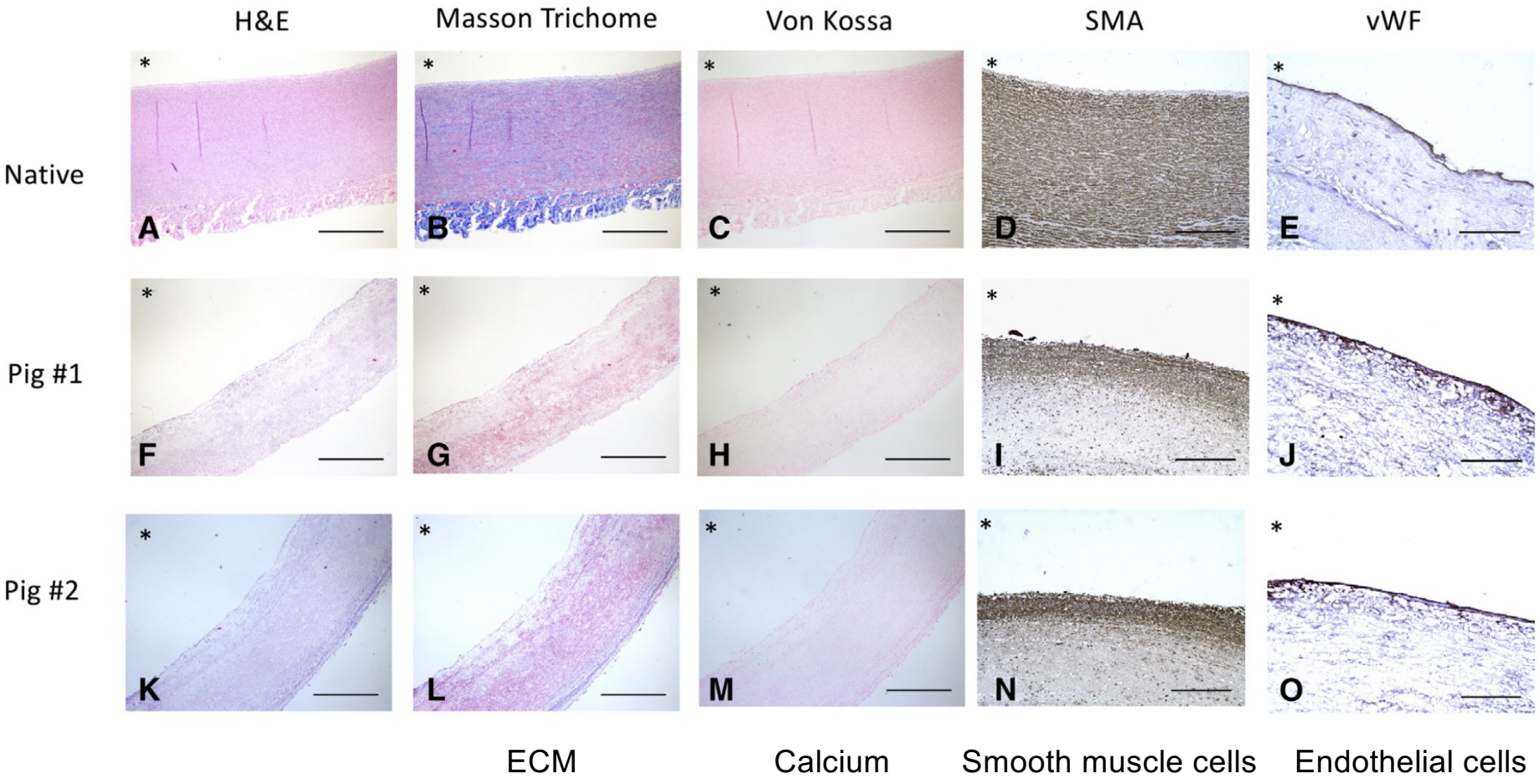
The biodegradable nanofiber material composed of a 1:1 ratio of polycaprolactone (PCL) and poly-L-lactide-co-ε-caprolactone (PLCL) were electrospun to coat the 3D-printed mandrel


Pre op MRI
Graft
Implantation
Post op MRI
Explantation
A
B
C
D
E
MPA
RPA
LPA
Graft
60
40
20
0
%
LPA RPA
Pro op
Flow Distribution
F
40
20
0
%
LPA RPA
Post op
Flow Distribution
G
10
5
0
mmHg
LPA RPA
Direct Pressure
Assessment
H


Main
Left
Right
A
Native tissue
Anastomosis
Graft
B
Main
Left
Right
C
Native tissue
Anastomosis
Graft
D

1 month


Collagen Content (1 month)
Collagen ug/mg
2.5
2.0
1.5
1.0
0.5
0.0
Native PA
Graft MPA
Graft LPA
Graft RPA
E
*
F
Remaining Scaffold (1 month)
Remaininig Scaffold %
20
15
10
5
0
Graft MPA
Graft RPA
Graft LPA
G


H&E
Masson Trichome
Von Kossa
SMA
vWF
Native
Pig #1
Pig #2
A
B
C
D
E
F
G
H
I
J
K
L
M
N
O
ECM
Calcium
Smooth muscle cells
Endothelial cells

In vivo implantation of 3-D printed customized branched tissue engineered vascular graft in a porcine model

- **Summary**:
  - 3D printed graft can match the geometry of native vessel to provide physiological hemodynamics.
  - Feasibility of 3D printed graft to remodel and grow in pig model.

- **Limitations of the study**:
  - Only tested in low pressure system (pulmonary artery)
  - Contain only biomaterials using 3D printing (no cells)
  - Need anti-coagulation throughout
  - Only 1 month, degradation of materials in the long term and structural integrity unknown.

Hibino, et al. *The Journal of Thoracic and Cardiovascular Surgery,* Volume 159, Issue 5, May 2020

Ethical Issues in 3D bioprinting for CHD

Moral validity of developing 3D bioprinting for CHD

- Why and for what purpose are we ready to print organs?
  - The need for organ transplantation.
  - The shortage of organ donation around the world.

- Ethical issue: difficult to distinguish between therapy and human improvement, technological immortality.

- The majority of governmental and international organizations now see this technology as morally justified if it has a therapeutic effect.
  - In the case of CHD, it is for treating children with the defect, instead of human improvement, so is morally justified.

Risk and benefits

- The benefits of custom-made 3D bioprinted tissue need to be balanced by the inherent known and unknown risk of transplants in humans.

- There should be special considerations for the use of 3D bioprinted tissues in children and adolescents.
  - Accommodate fast growth
    - If graft can not grow, then need repeated surgery, the risk and benefit is in question.
  - Accommodate to exercise and sport activity in children
    - Use of anticoagulation increase risk of bleeding.
  - Need to consider the long-term benefit and risk, beyond short-term benefit.
    - Can not use autologous vein (if use, could negatively impact child tissue development, also need to save for later).

Safety concerns

- Source of cells (autologous, allogenic, xenogenic)
- Cell type (embryonic, mesenchymal, iPSCs, etc)
  - Whether these cells are safe to use (not transmit disease, cause cancer or immune rejection).

- Biomaterials
  - Biocompatibility, toxicity, tumorigenicity.
  - Degradation of biomaterials, tissue integration
  - In pediatric patient, whether the construct grow with the children (reduce the need for multiple surgeries).

Issue with cell source

- Human embryonic stem cells (ESCs)
  - ethical issue, both legal and moral.
    - ✓bioethical problems of determining the moral status of an embryo
    - ✓legal pregnancy termination, and human participation in the experiments.
  - other factors may influence the ethical acceptability of using allogeneic cells.
    - ✓obtaining stem cells from donors who have been pressured, coerced, or have not given informed consent.
    - ✓barriers for commercialization (the application of 3D printing technology using ESCs in industrial and commercial purposes?)

Issue with cell source

- induced pluripotent stem cells (iPSCs)
  - Can be obtained from patients (self).
  - no ethical issues of ESCs or xenogeneic cells.
  - Issue with iPSCs
    - reprogramming and differentiation far from perfect.
    - risk of tumorigenicity.

Issue with cell source

- Allogenic cells
  - more availability than autologous cells
  - ethical issues:
    - donor confidentiality,
    - informed donor consent
    - donor cell ownership.

  - basic rules of medical ethics – Primum non nocere (First, do no harm).
    - the moral nature of the action
    - the intention of the agent
    - the means of action
    - the possible adverse effects
    - the proportionality between good and bad effects

Issue with cell source

- xenogeneic cells
  - more availability
  - ethical issues:
    - social and religious aspects of animal cell utilization.
    - problems with personal identity.
    - patients with religious beliefs may disagree with the use of cells of certain animal species.

Healthcare disparity

- Considering the cost of bioprinting, availability of expertise, trained workforce, machinery, and infrastructure.

- Who gets access and who is left behind becomes an issue.
  - As of 2018, there were an estimated 28 million uninsured Americans. This creates a much worse quality of life in low-income communities because of the struggle to afford basic healthcare.
  - Bioprinting will only serve to widen the gap in quality of healthcare for different socioeconomic classes if the right protections are not put in place, because of its high cost as of now.
  - Without mandated equal access for all, bioprinting will only benefit extremely wealthy folks for the foreseeable future.

Ownership

- Bioprinted is customed to patient-specific data, does patient own the bioprinted product ?

- What about the donor who contribute the cells ?
  - Besides ownership, Issue of donor privacy could arise

Ownership rights

- Bioprinting is currently a gray area in the legal arena, with little to no precedent of ownership of bioprinted objects.
  - In terms of the bioprinting machines, there is a question of whether or not they should be a patentable, profitable entity or categorized as a non-patentable medical treatment.
    - Patentable printers could easily lead to more innovation.
    - but they would limit treatment access to the most wealthy individuals in society.
- Ownership of the bioprinted organs themselves is undecided.
  - patients must have full control over their bodies, which would include a bioprinted organ they receive.

Societal and culture consideration

- Considerations of:
  - religious beliefs
  - use of animal cells
  - views on naturalness
  - public perceptions on the type of tissues/organs that should be or not be bioprinted.

Informed consent

- Media and scientific reports over enthusiastic, may over exaggerate the benefit of 3D bioprinting.
- Unknow risks, unrealistic expectations of benefits could impact the voluntariness of consent. misconceptions or unrealistic expectations must be avoided.
- It is crucial that researchers and media outlets do not promote hype around current advances.
- Patients need to be informed of the risk of the bioprinted tissues.
- Consent for human participation in medical trials including knowledge of the nature, duration, and purpose of the experiment, its methods, and associated risks

Confidentiality

- The principle of confidentiality implies that the circumstances of treatment and the patient's characteristics are kept confidential with the respect to the patient's life.
- Confidentiality helps to build trust relationships that are essential for effective and timely medical care.

Additional ethic issues of 3D bioprinting

- 3D bioprinting technology leads to the “digitalization” of objects of the material world (the boundaries between the physical world and the digital space erase).
  - who and to which extent shall be responsible for the translation of the anatomical image into digital: designers, biologists, or programmers?
  - Who will have the legal rights for the model?
  - Will it be possible to use the model without a patient’s consent?
  - Is it possible to apply the models commercially?
  - Issues of confidentiality and privacy arise regarding human digitization.
  - Privacy of the digital model.

Legal Issues in 3D bioprinting for CHD

Legal issues of 3D bioprinting into clinical practice

- Legal regulation of 3D bioprinting is complex.
- The issues become even more exacerbated as numerous participants are involved in the production chain of bioprinting.
  - Expertise from 3D model designers, medical professionals, engineers, biologists, lawyers, ethical committee, and insurance companies need to establish multi-stakeholder collaboration to form an acceptable path for the bioprinting technology development and introduction into clinical practice.

Clinical trials

- In the case of 3D bioprinting of human tissues and organs, specific issues arise regarding the design of human clinical trials.
  - Each 3D bioprinted treatment is unique and adapted to a specific individual, and therefore, results of each case cannot be fully extrapolated into future treatments.
  - Thus, standard approaches for clinical trials such as double-blind randomized control studies cannot be applied to 3D bioprinting technology.
  - The efficiency and safety of the custom-made organs cannot be tested on other individuals; therefore, in this respect; each patient becomes the first examinee. Consequently, the question arises of the ratio of risks to benefits and criteria of inclusion.

  - Nevertheless, while the 3D bioprinting are personalized, criteria and protocols for the procedures can be standardized based on the first clinical trials.
  - The study has to be ethically acceptable and safe for the patients.

Risks regulation and responsibility for product quality

- The regulatory framework for advanced therapy medicinal products (ATMPs) may serve as a guideline to different stages of 3D bioprinting production.
  - Who is primarily responsible for the quality of bioprinted products
    - the 3D bioprinting providers or medical organizations;
  - Who should be responsible for quality control;
  - Who should be liable in case of bioprinted organ quality claims from the recipient.

- Currently, there is no suitable framework, or special regulatory guidelines governing 3D bioprinting of tissues and organs and their further transplantation.

- Ethical evaluation and legal regulation of 3D bioprinting need to be developed.

Summary of ethical and legel issues moving forward

- 3D bioprinting for pediatric CHD is still at early stage, far from clinical application.

- Before that, several regulations should be adopted.
  - Need to develop informed consents for donation, material manipulation, storage, and its further use, including for commercial and research purposes.
  - Develop requirements for safety, quality, and efficiency of technological procedures and the end products obtained by 3D bioprinting taking into account the human rights and dignity.
  - Establish committees for creation and regulation of national guidelines on technical, legal, and ethical issues related to the development and application of 3D bioprinting technologies.
  - All patients including minors and incapable people need to be legally protected.
  - Establish regulations of turnover and limits of commercialization for 3D bioprinting technologies of human organs and tissues, as well as possible sanctions for illegal trafficking of artificial organs.

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