日本集中治療医学会第2回関東甲信越支部学術集会

2018年6月30日（土）、宇都宮

会長：布宮　伸教授（自治医科大学）
テーマ：臨床→集中治療∞

特別講演

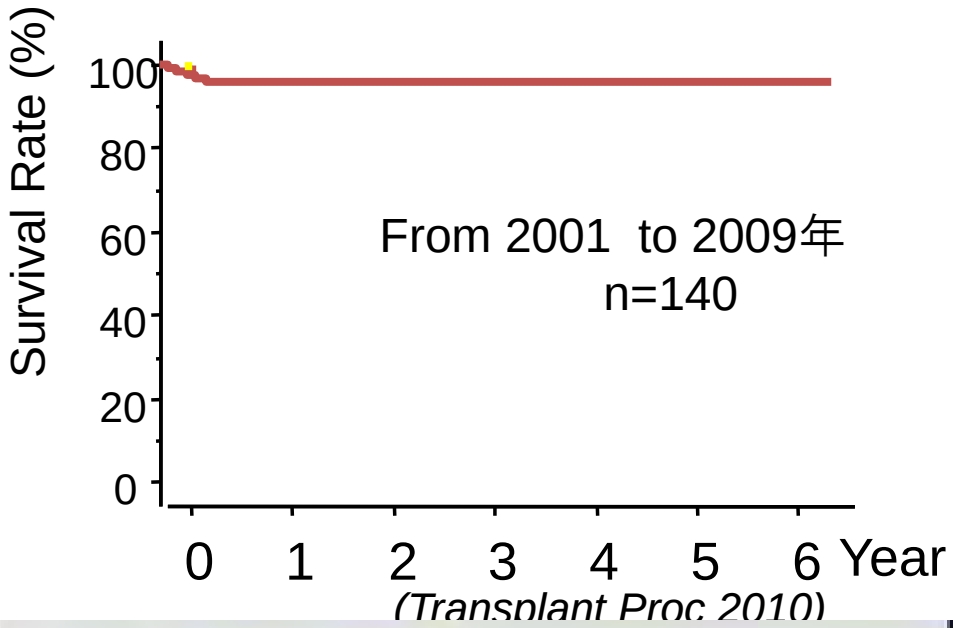
移植可能な臓器を造る
ー臓器灌流培養法の開発

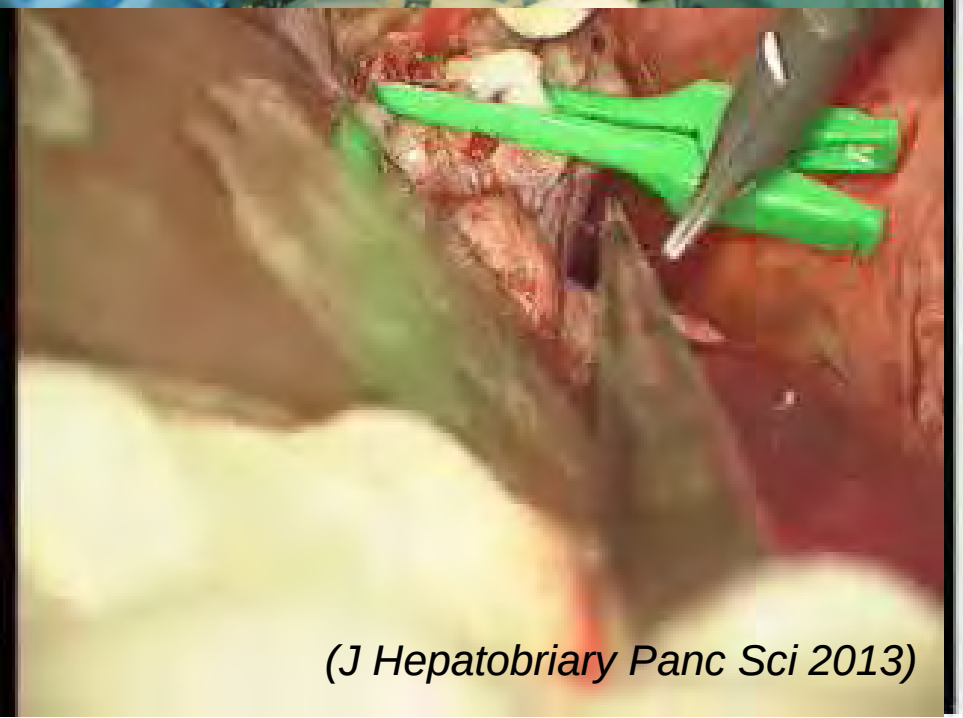
小林英司

慶応大学医学部　臓器再生医学講座

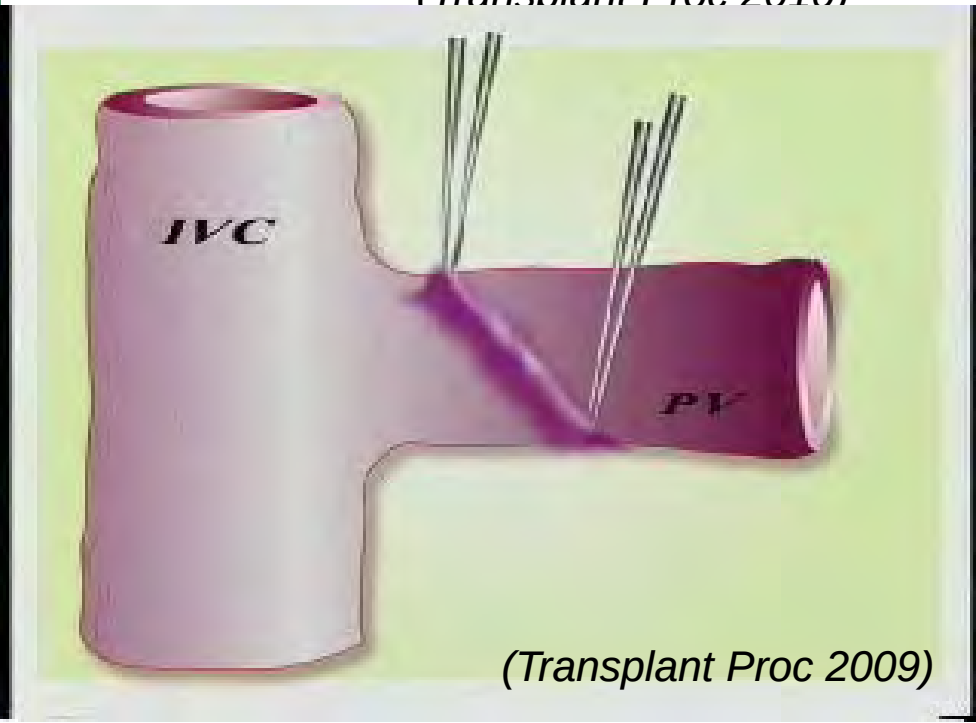
COI; E.K. is a medical adviser for ***Screen Ltd***.

Clinical Microsurgery in JMU





(J Hepatobriary Panc Sci 2013)



(Transplant Proc 2009)

イスタンブール宣言

(Istanbul, 30th April – 3rd May 2008)

1. Organ trafficking（臓器取引）、Transplant tourism（移植ツーリズム）、Transplant commercialism（移植商業主義）等の内容を明確して、人道的、社会的、国際的に問題があ

2. 死体（脳死、心停止）ドナー を増やすよう呼びかけること

3. 生体ドナーは、ドナー保護を 的な保障等の制度を国家的

152 professionals from 78 countries

（Ｌａｎｃｅｔ　２００８年7月6日）

TTS The Transplantation Society

ISN International Society of Nephrology 1960


The National Diet
（6 Sep ,2009）
参考人
自治医科大学先端医療技術開発センター
先端治療開発部門客員教授
小林 英司

NHKスペシャル　2009

人体“製造”

―再生医療の衝撃―

「移植可能な臓器作り」

戦略分類	小動物による プルーフ・オフ・コンセプト実験	大型動物でのPOC
（1） 丸ごと使う （動物工場）	膵（*Kobayashi T, et al. Cell 2010*） 膵（*Yamaguchi T, et al. Nature 2017*） 腎（*Usui J, et al. Am J Path 2012*） 肝（*Hata T, et al. Ann Surg 2013*）*	膵（*Matsunari H, et al. PNAS 2013*） 肝（*Fisher JE, et al. Liver Transplant 2013*） 肝（*Hsu H, et al Transplant Proc 2017*）* 豚ヒトキメラ（*Wu J, et al Cell 2017*）
（2） 発生原器を使う （胎仔原器グラフト）	腎（*Matsumoto K, et al. Stem Cells 2012*）* 腎（*Mae S, et al Nature Communi 2013*） 腎（*Taguchi A, et al . Cell Stem Cell 2014*） 肝（*Takebe T, et al. Nature 2013*））	膵（*Hammerman M, et al. Organogenesis 2012*） 腎（*Yokote S, et al. PNAS 2015*）*
（3）Vitroでの再構築 （脱細胞充填） （組織再構築） （Ex vivo再構築）	心（*Ott HC, et al. Nature Med 2008*） 腎（*Ross EA, et al. JASN 2009*） 肝（*Uygun BE, et al. Nature Med 2010*） 肺（*Ott HC, et al. Nature Med 2010*） 心（*Sekine H, et al. Nature Com 2013*）*	腎（*Orland G, et al. Ann Surg 2012*） 肝（*Yagi H, et al. Cell Transplant 2012*） 心（*Kitahara H, et al. Interact Cardio Thrac Surg 2016*） 脾臓、骨髄を含め多種臓器 （*Kobayashi E, et al. unpublished*）*

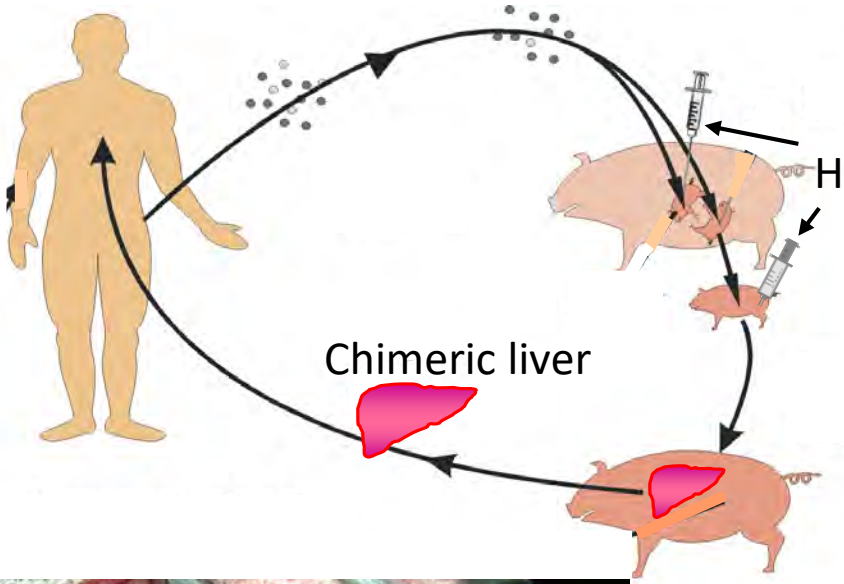
＊著者ら

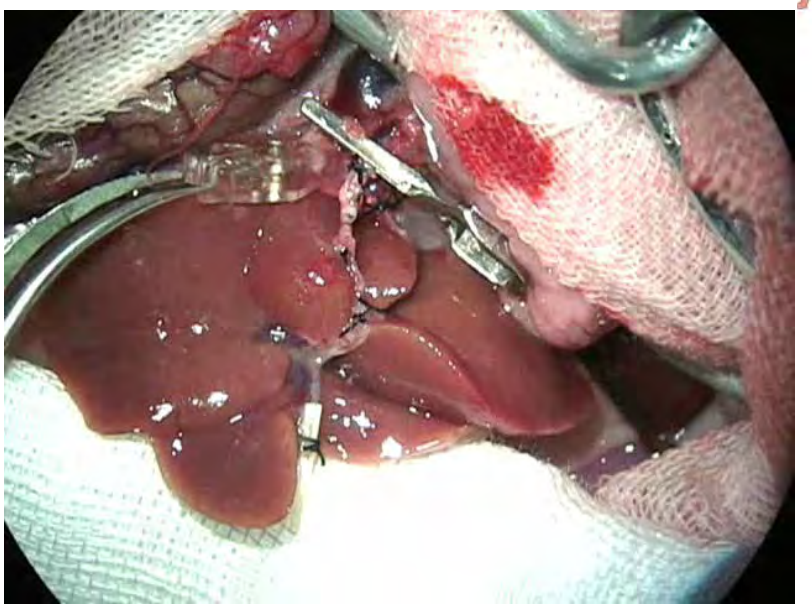
（2013年内閣府生命倫理委員会資料　小林英司 より改編）

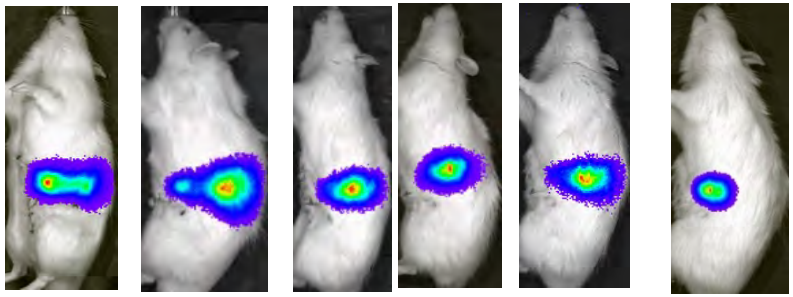
戦略（1）

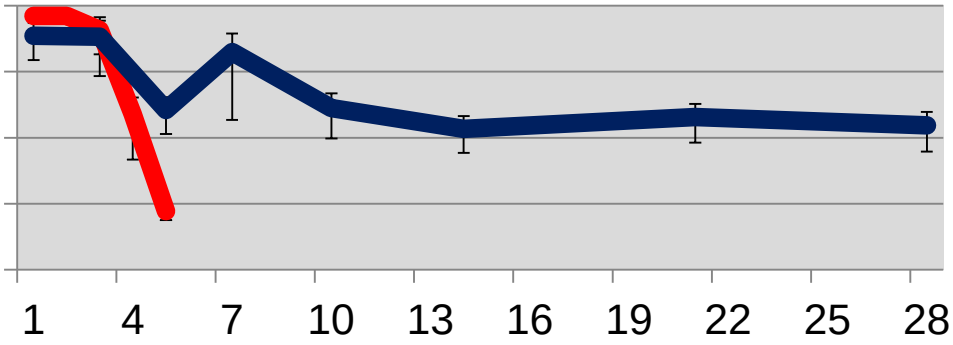
Creating the human liver in Pigs
動物の体内でヒトの臓器を作る

京都大学　畑先生、上本教授



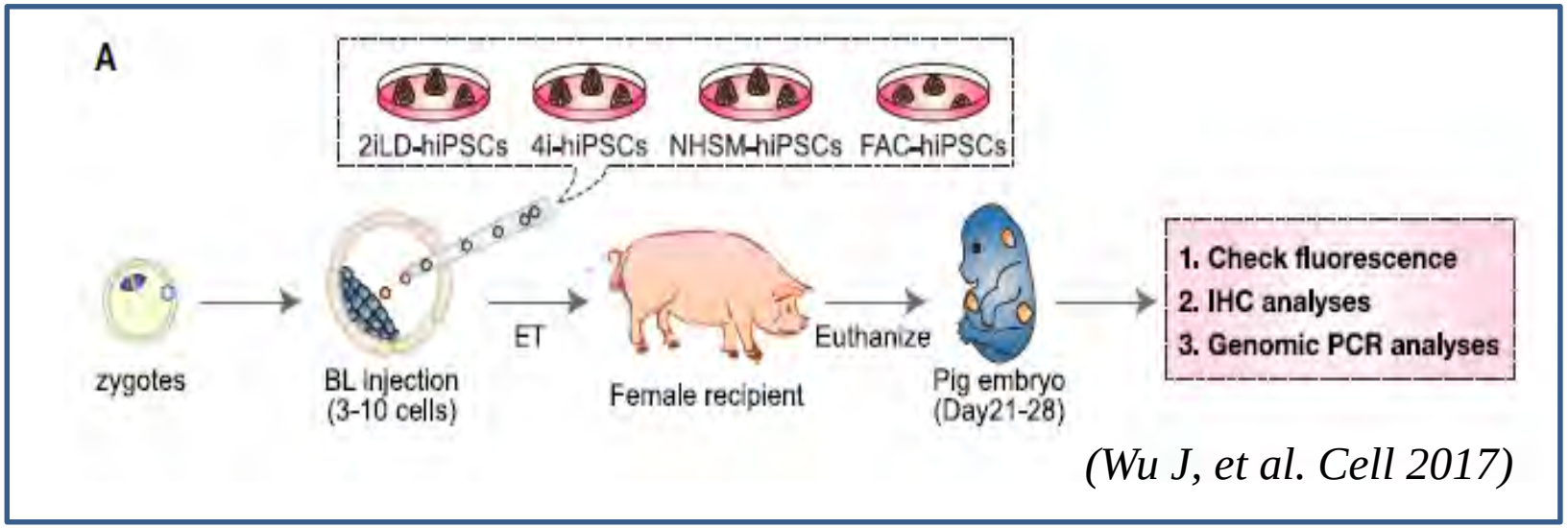




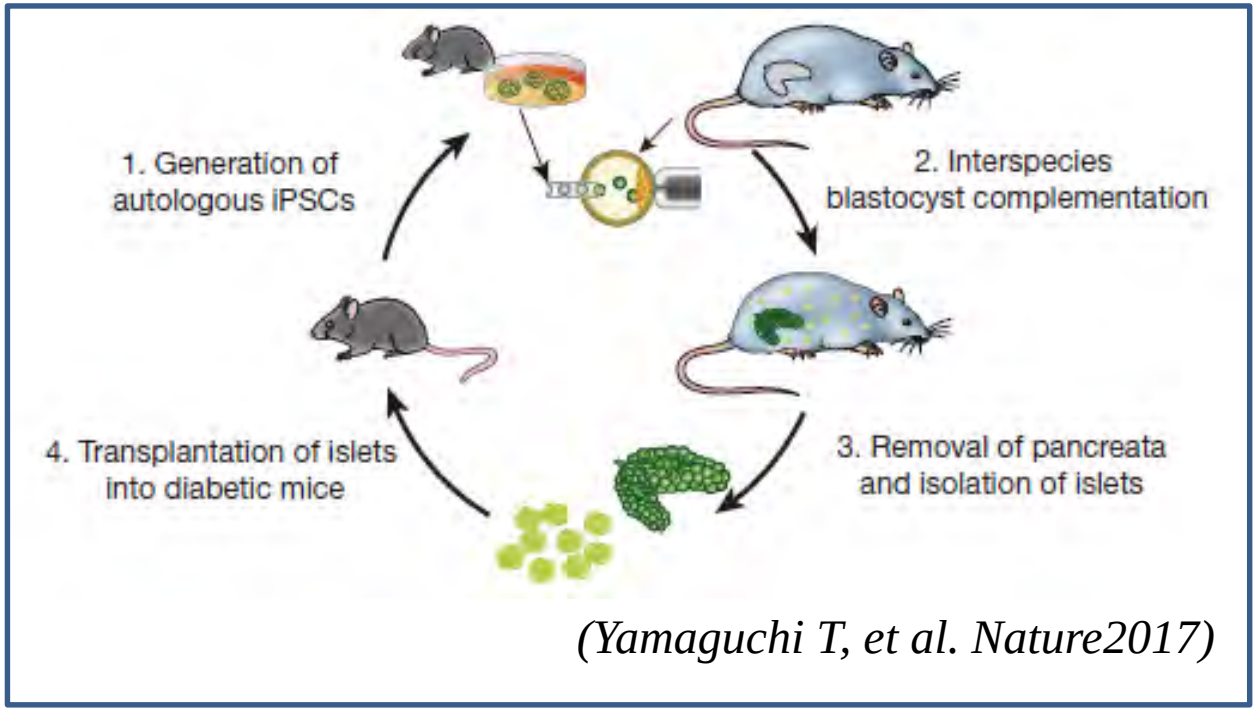


(Hata T, et al. Ann Surg 2013)

白熱する「動物の体内でヒトの臓器を作る」 第3次ブタブーム

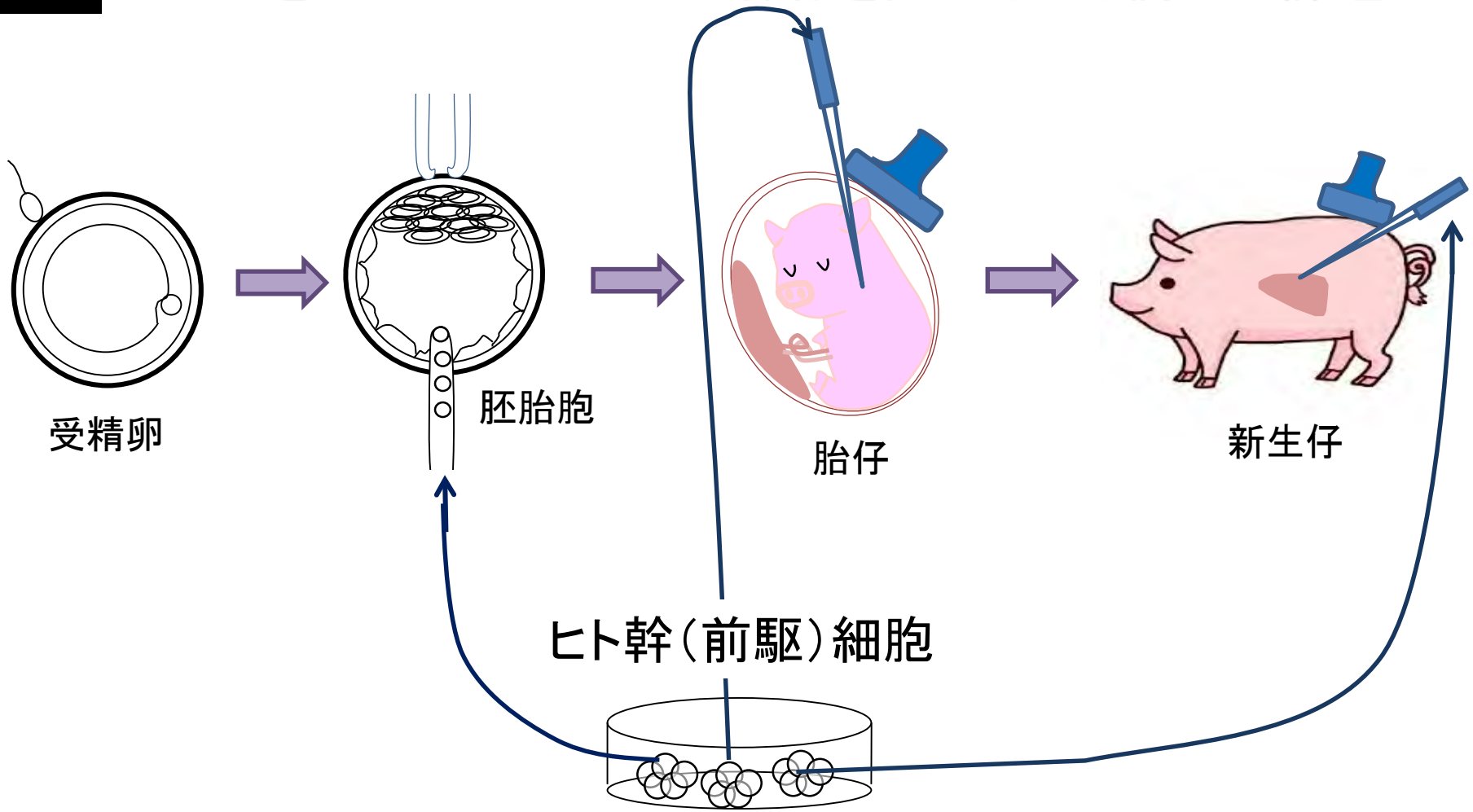


(Wu J, et al. Cell 2017)



(Yamaguchi T, et al. Nature2017)

課題　ブタをbioreactorとしてヒト臓器を発生させる倫理的課題



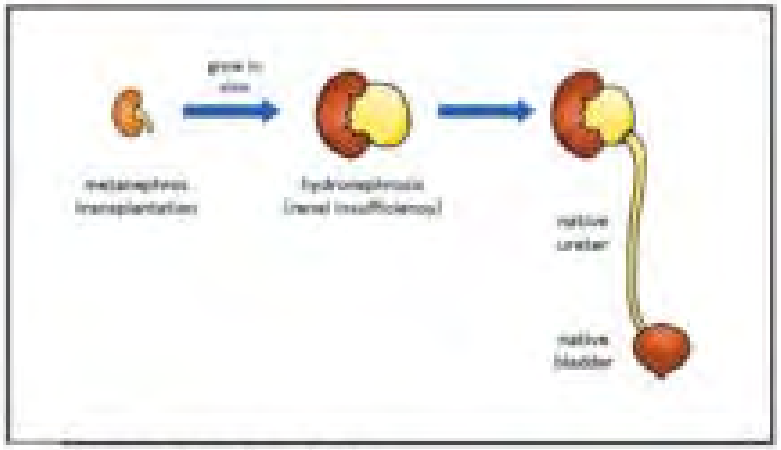
キメラ率	全身	（+）	（±）	（±）
	局所	（++）	（++）	（+）

（小林英司、第4回動物性集合胚の取り扱いに関する作業部会、2014年3月20日、文部科学省）

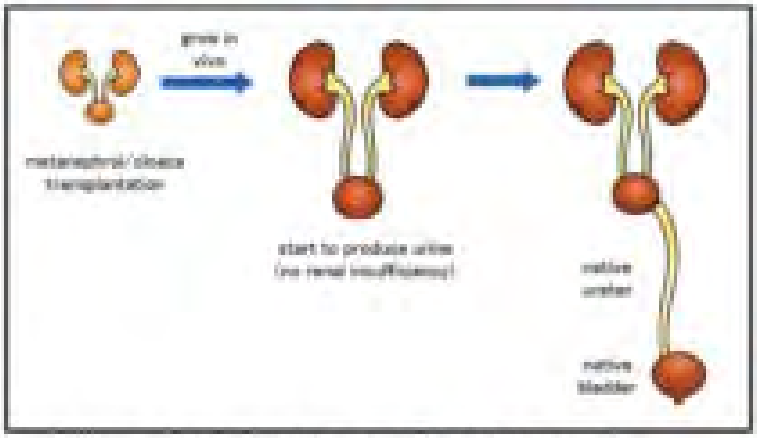
戦略（2）

臓器の芽を患者体内で育てる

慈恵医大　横尾教授、 明治大学　長嶋教授



Conventional method



Step-wise peristaltic ureter (SWPU) system

(Yokote T, et al. PNAS 2015 より改変)

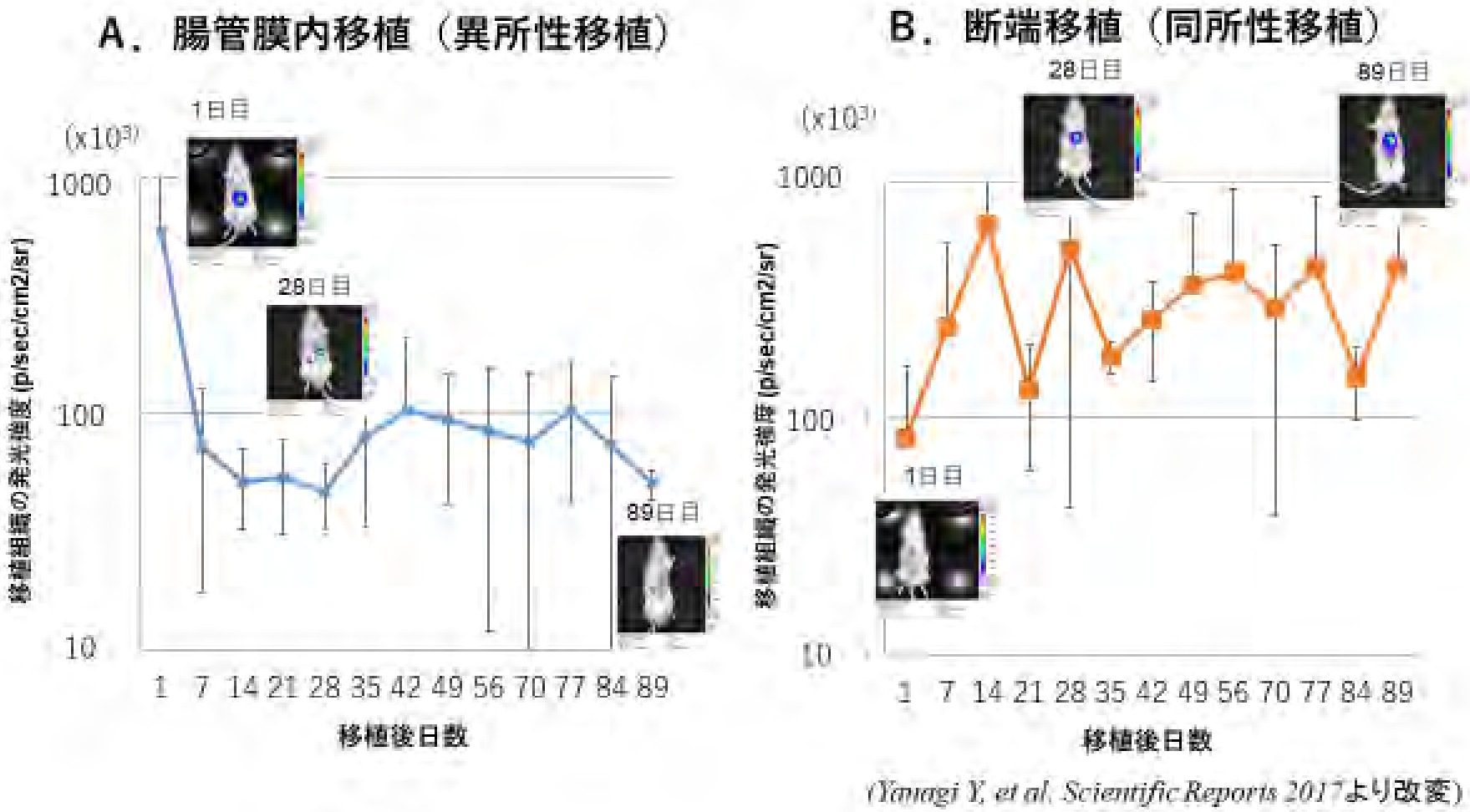
戦略（2）

‘3Dプリンターで作った肝臓の芽’を患者に移植する

九州大学　柳先生、田口教授
佐賀大学　中山教授

(Yanagi Y, et al. Scientific Report 2017)

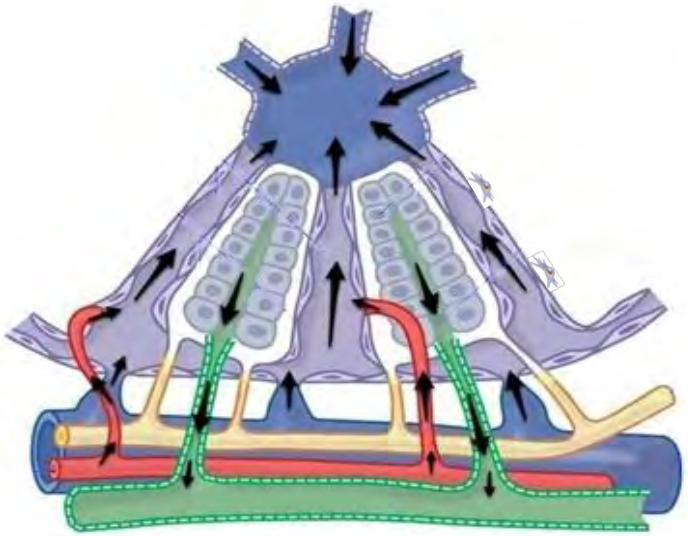
肝芽は、異所性では育たない



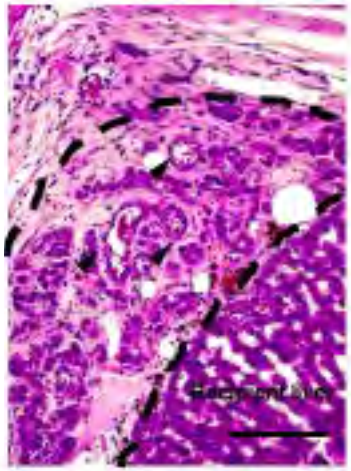
(Yanagi Y, et al. Scientific Reports 2017より改変)

なぜ肝芽が異所では育たないか？

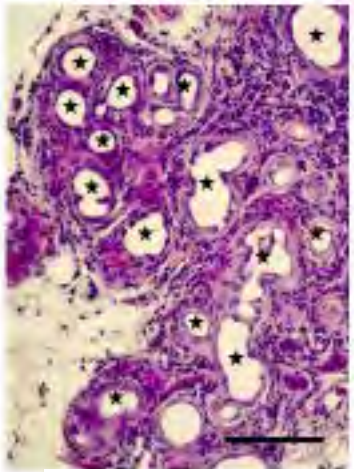
肝臓維持に必要な解剖学



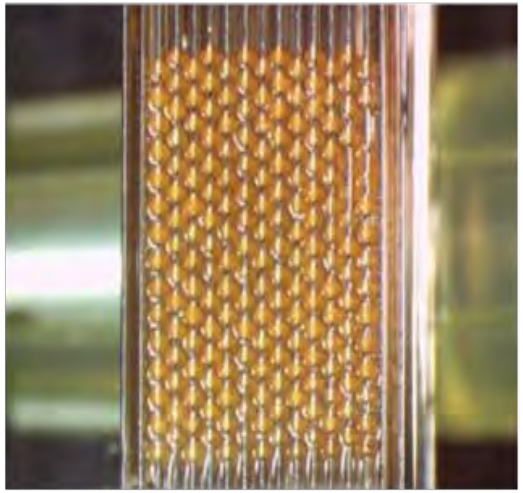
断端移植（同所性移植）

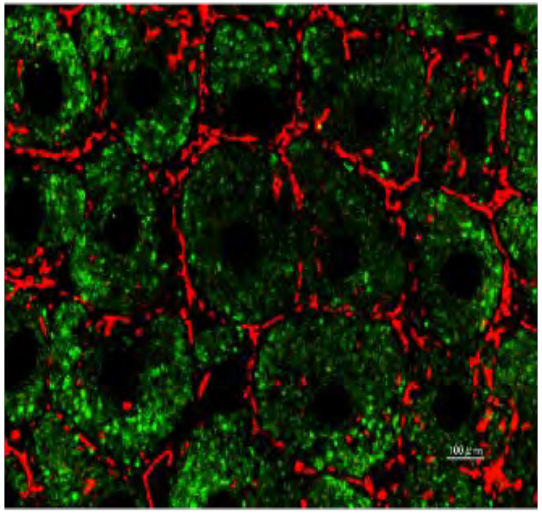


腸管膜内移植（異所性移植）



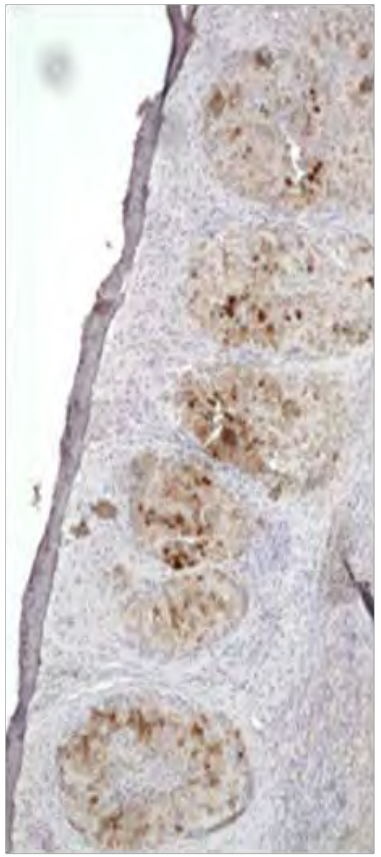
バイオ３Dプリンターで作製したヒト肝臓組織





緑ー肝細胞
赤ー血管内皮細胞

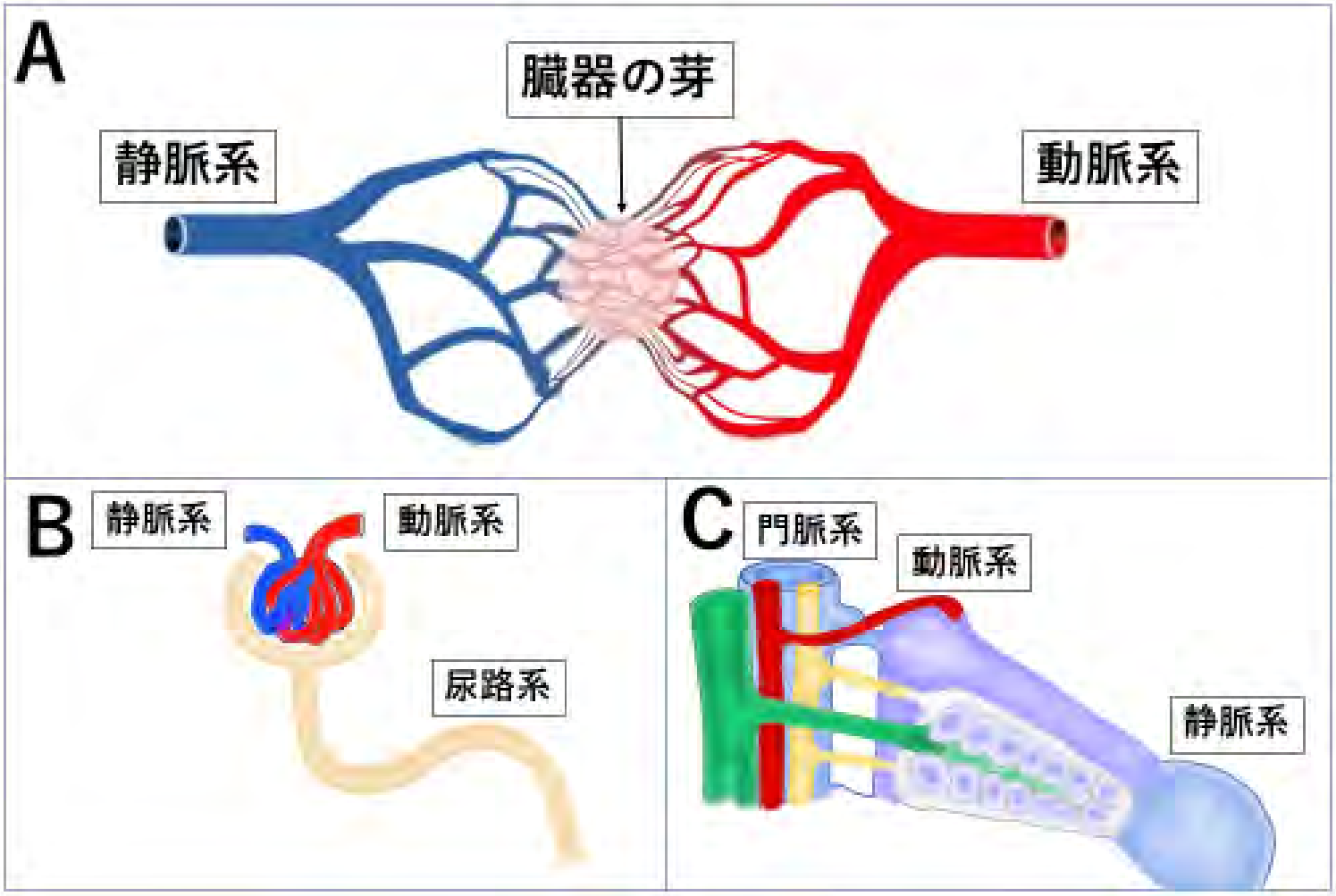
ラットの肝臓断端に移植したヒト肝臓組織が
ヒトアルブミンを産生



茶－ヒトアルブミンを産生する肝細胞

課題

外科技術を応用した「臓器の芽」の移植法（腎、肝）

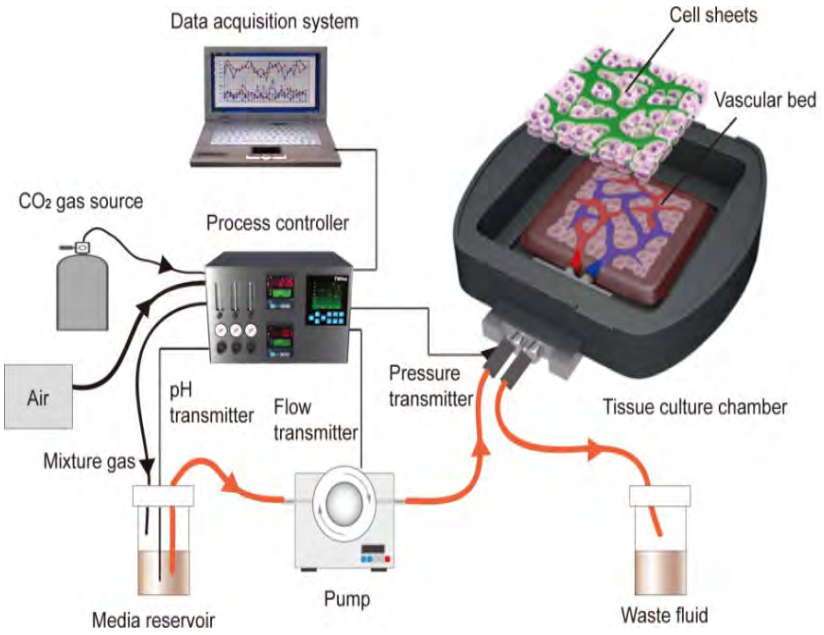


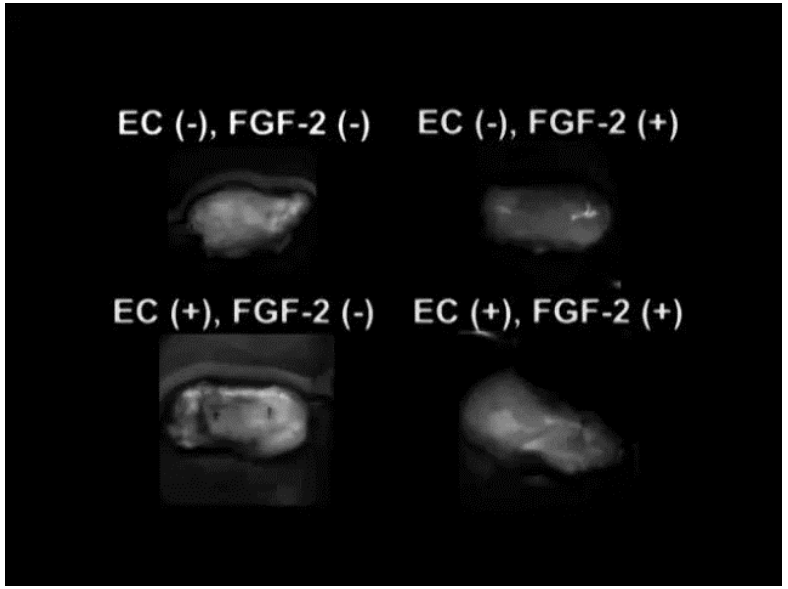
（小林英司、日本外科学会誌（総説）２０１８）

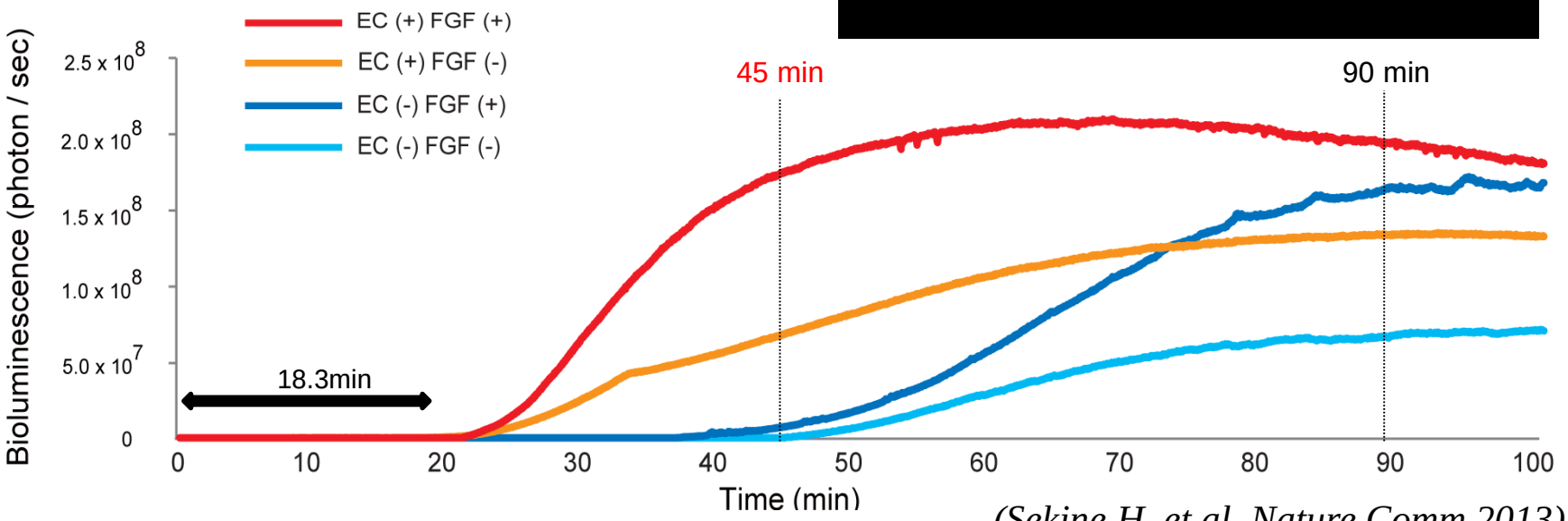
戦略 (3)

Ex vivoの還流で組織を積み上げる

東京女子医大　関根先生、清水教授



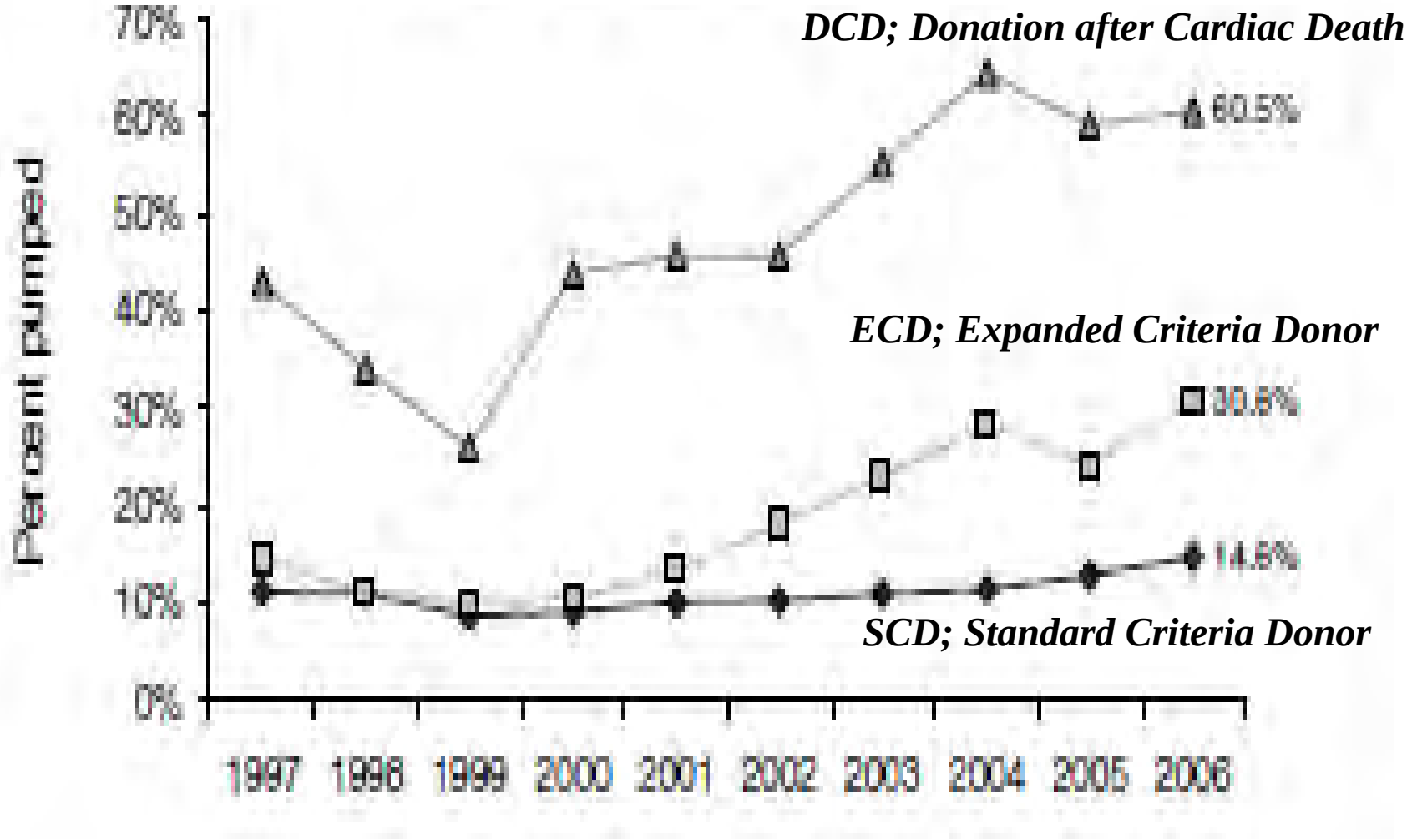




(Sekine H, et al. Nature Comm 2013)

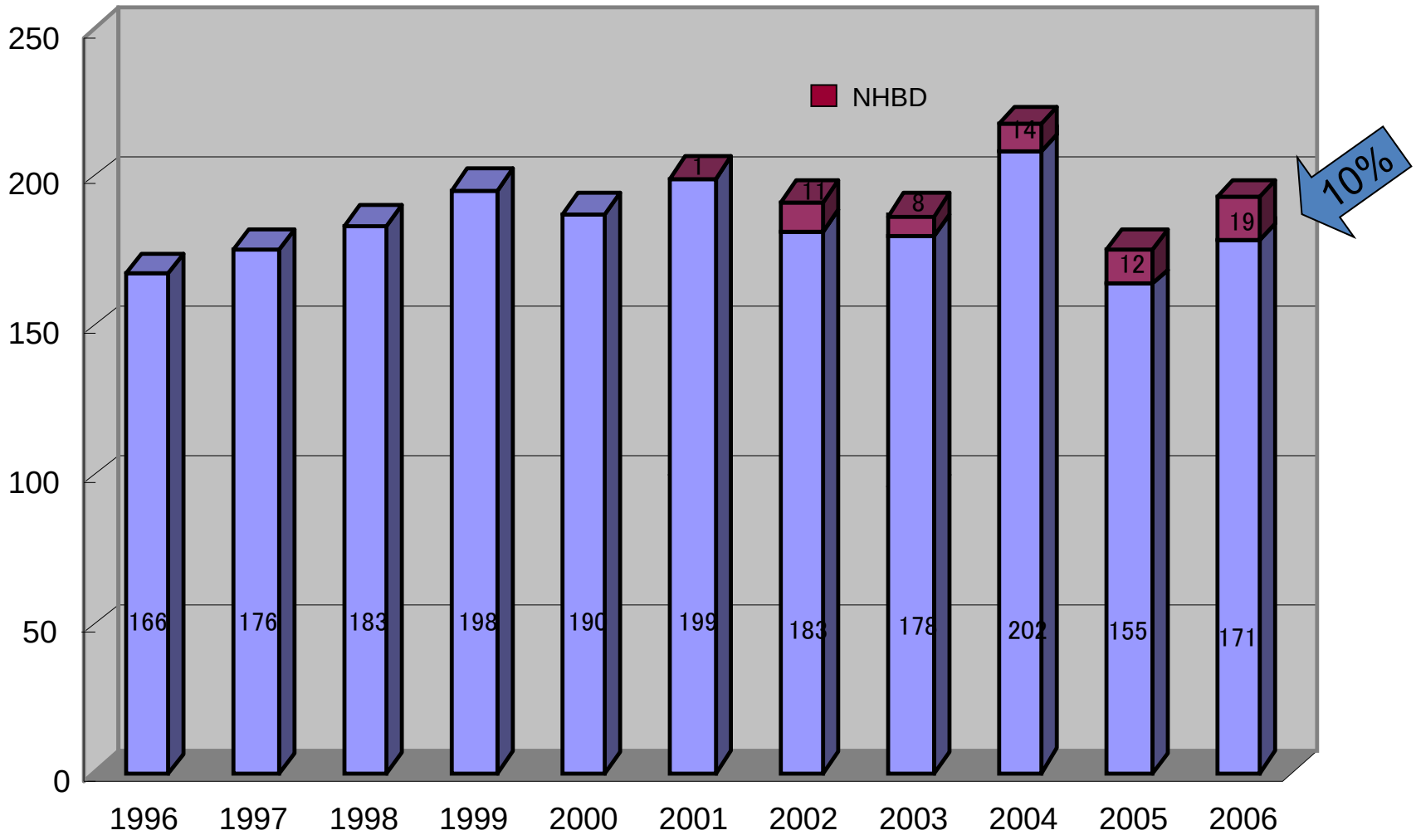
臓器を培養する？！

Organ Donation and Utilization in US (1997-2006)



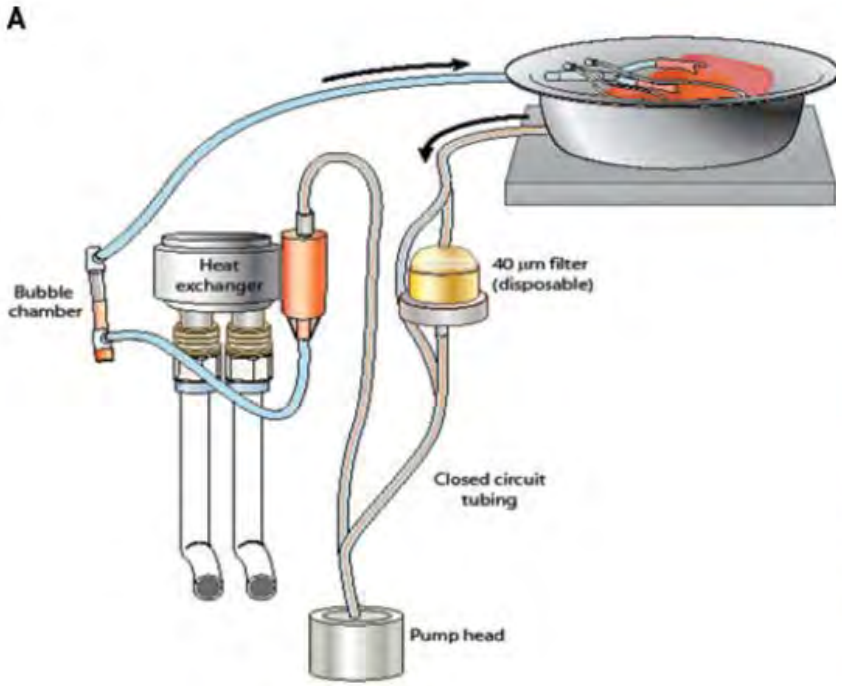
(R.S.Sung, et al. Am J Trasuplntation 8(2); 922, 2008)

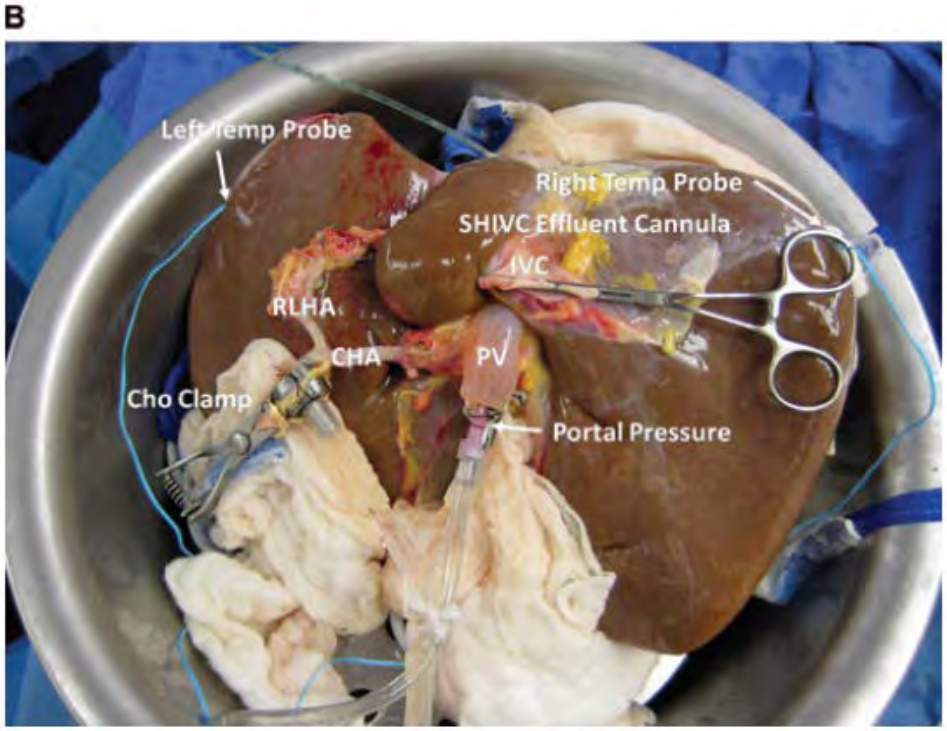
Impact of NHBD liver transplantation in King's college Hospital



(Muiesan P. 2008)

Hypothermic Machine Preservation in Human Liver Transplantation: The First Clinical Series





(Guarrera JV, et al. Am J Transplant 2009)

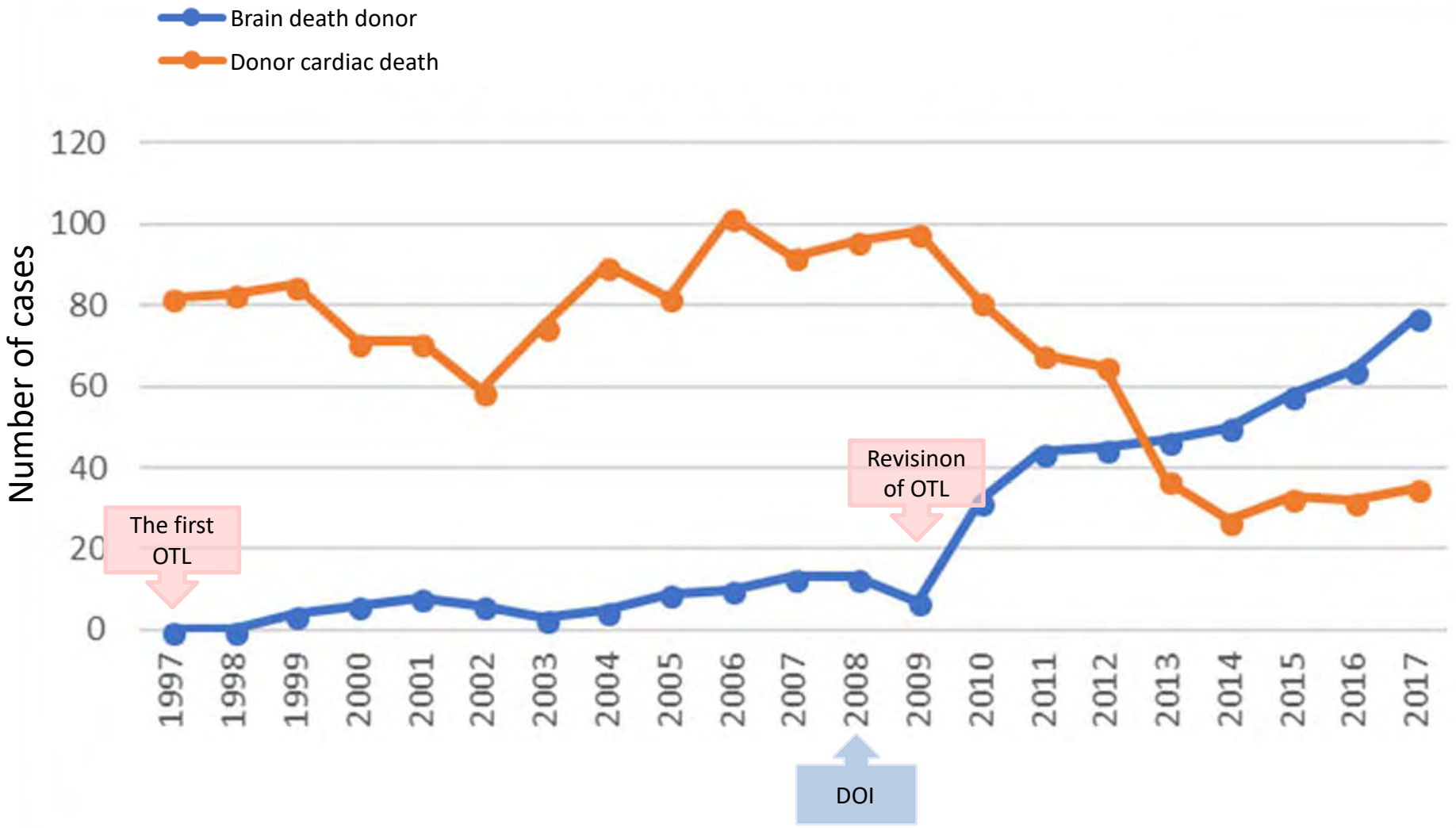
Table 3: Patient outcomes

	Machine perfusion (HMP)	Cold storage (CS)
Primary nonfunction	0	0
Early allograft dysfunction	1* (5%)[1]	5 (25%)
Vascular complications (total)	0	1
Hepatic artery stenosis		1
Biliary complications (total)	2	4
Early bile leak	1	1
Biliary stricture	1	3
Hospital length of stay (days)	10.9 ± 4.7[2]	15.3 ± 4.9
Actual graft and patient survival	18/20 (90%)	18/20 (90%)
Deaths with fuctional grafts	2 Cardiovascular death at 1 month Pneumonia and sepsis at 3 months	2 Recurrent cancer at 5 months Recurrent HCV and sepsis at 7 months

[1] $p = 0.08$, [2] $p = 0.006$.
*Technically met criteria but occurred in the setting of early acute cellular rejection.

(Guarrera JV, et al. Am J Transplant 2009)

Changing Patterns of Organ Donation in Japan ten years after the Declaration of Istanbul



(Kobayashi E. Transplantation in press)

An Oxygenated and Transportable Machine Perfusion System Fully Rescues Liver Grafts Exposed to Lethal Ischemic Damage in a Pig Model of DCD Liver Transplantation

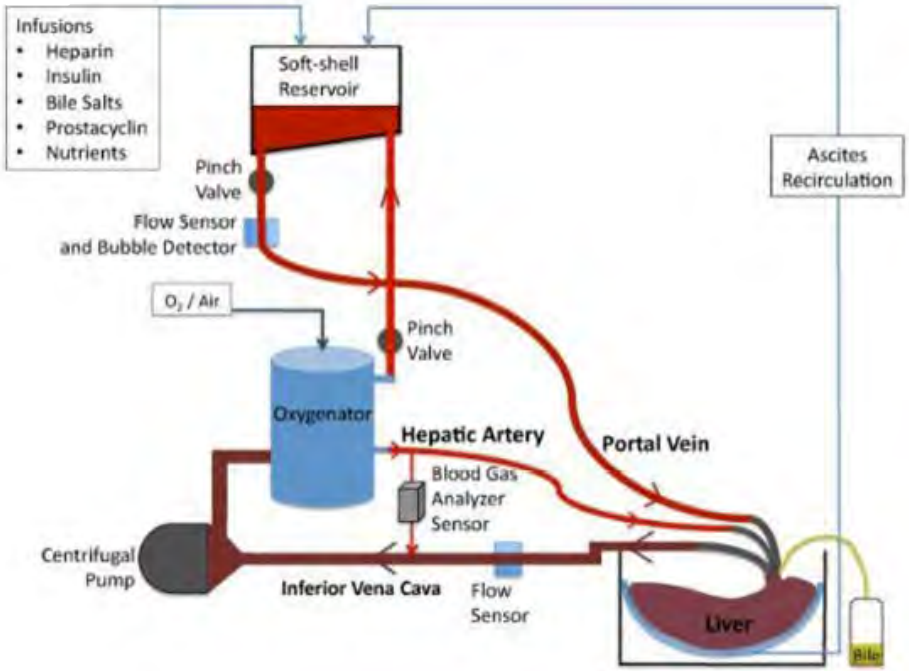
(Compagnon P, et Transplantation 2017)

A randomized trial of normothermic preservation in liver transplantation

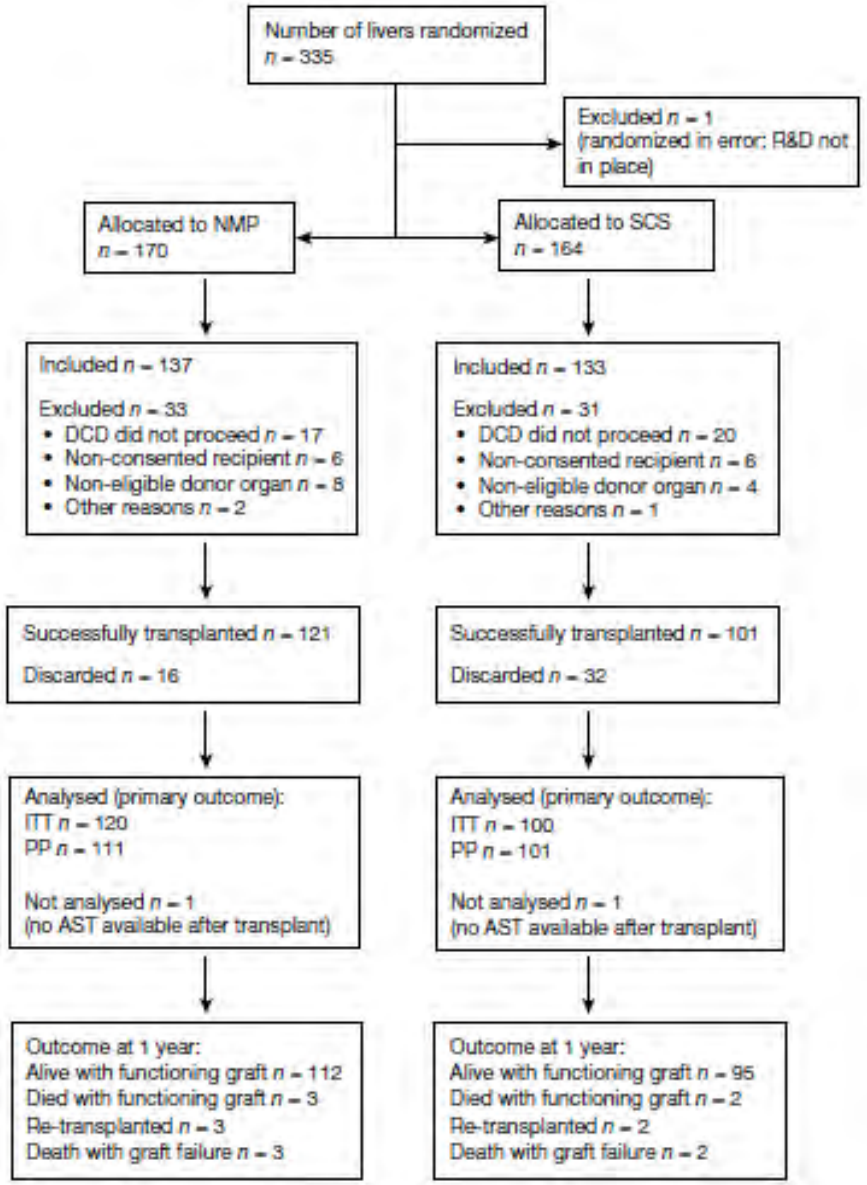
NMP device and circuit.
OrganOx metra (generation 1)

Image of liver during
normothermic machine perfusion

A randomized trial of normothermic preservation in liver transplantation



NMP device and circuit; OrganOx



Four types of Non-Heart Beating Donor (NHBD)

Categories I: dead on arrival

Categories II: unsuccessful resuscitation

} **uncontrolled**

Categories III: withdrawal of life-supporting therapy

Categories IV: cardiac arrest in a brain-dead donor

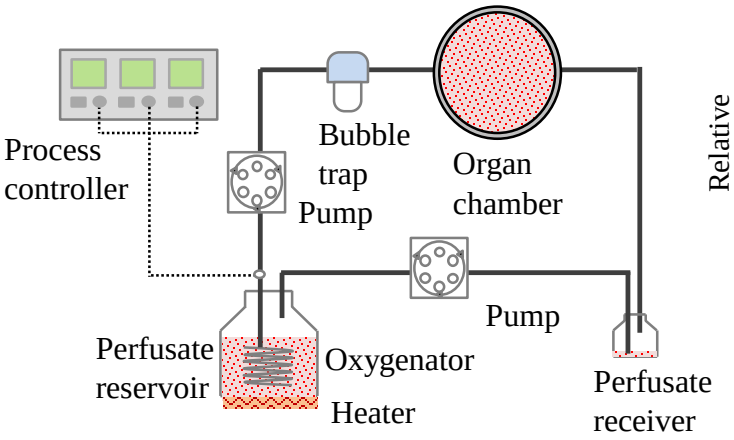
} **controlled**

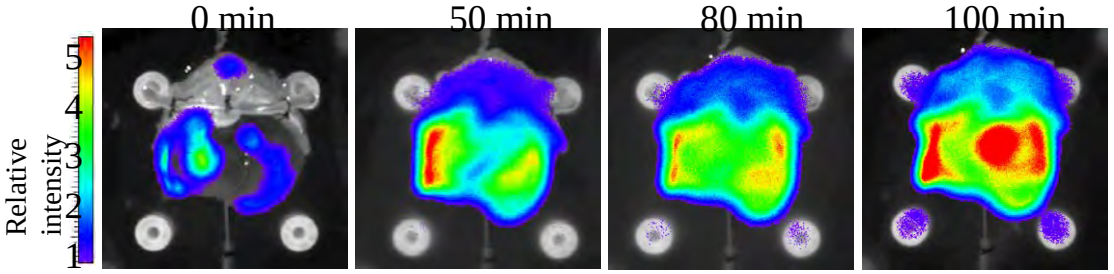
(Kootstra G, et al. Transplant Proc 1995)

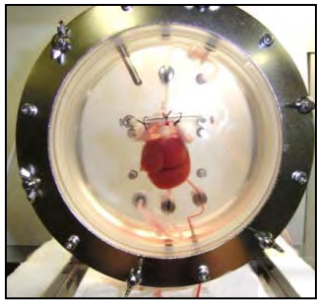
Don Quixote Project for Organ Resurrection

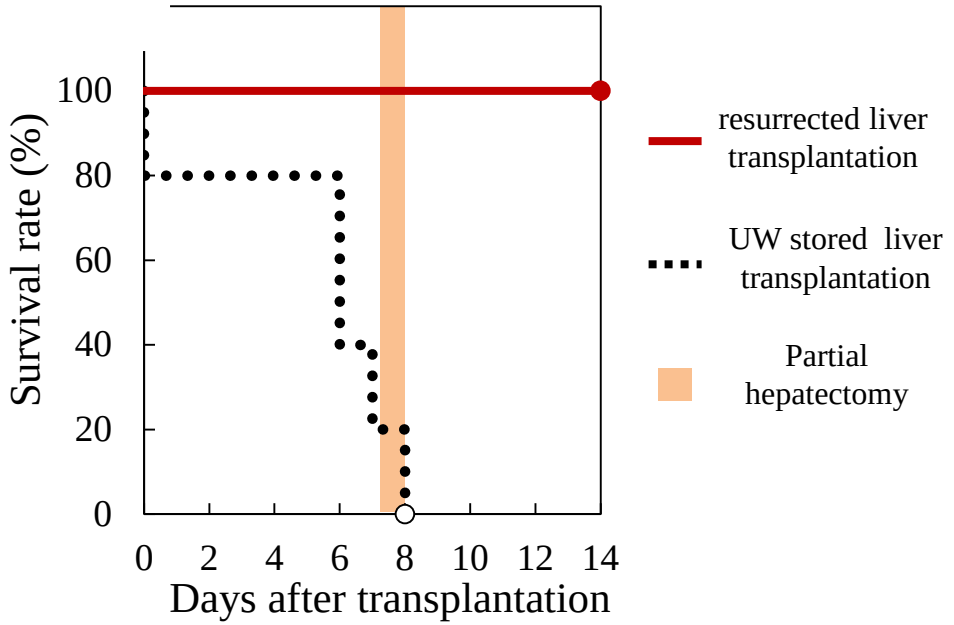
(Kobayashi E with Screen Ltd. from 2015)

Hypothermic temperature effects on organ survival and restoration





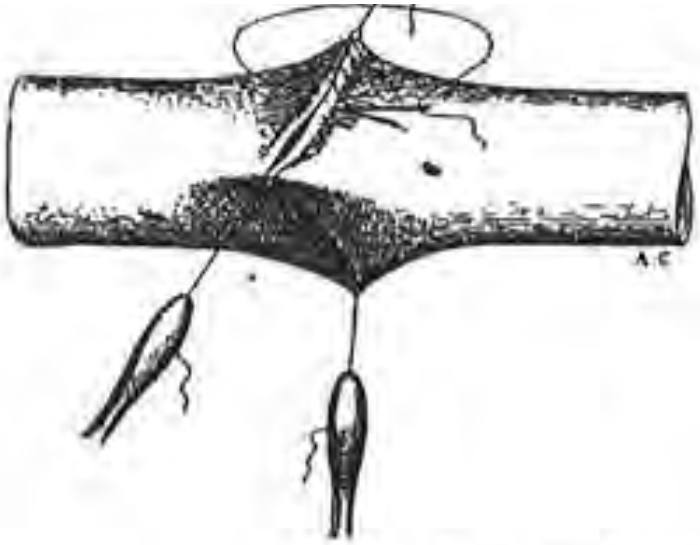




(Ishikawa J, et al. Scientific Reports 2015)

In 1912, **Dr. Alexis Carrel** was awarded **the Nobel Prize** in Physiology or Medicine for pioneering vascular suturing techniques

(1873-1944)



A New techniques in vascular sutures in 1902

The Culture of Whole Organs

Author(s): Alexis Carrel and Charles A. Lindbergh
Source: *Science*, New Series, Vol. 81, No. 2112 (Jun. 21, 1935), pp. 621-623
Published by: American Association for the Advancement of Science
Stable URL: http://www.jstor.org/stable/1660192
Accessed: 13-11-2017 02:50 UTC

Background

1812
Le Gallois Proposal for idea

1866
de Cyron Frog heart
Beating for 48 hours
Liver Urea production

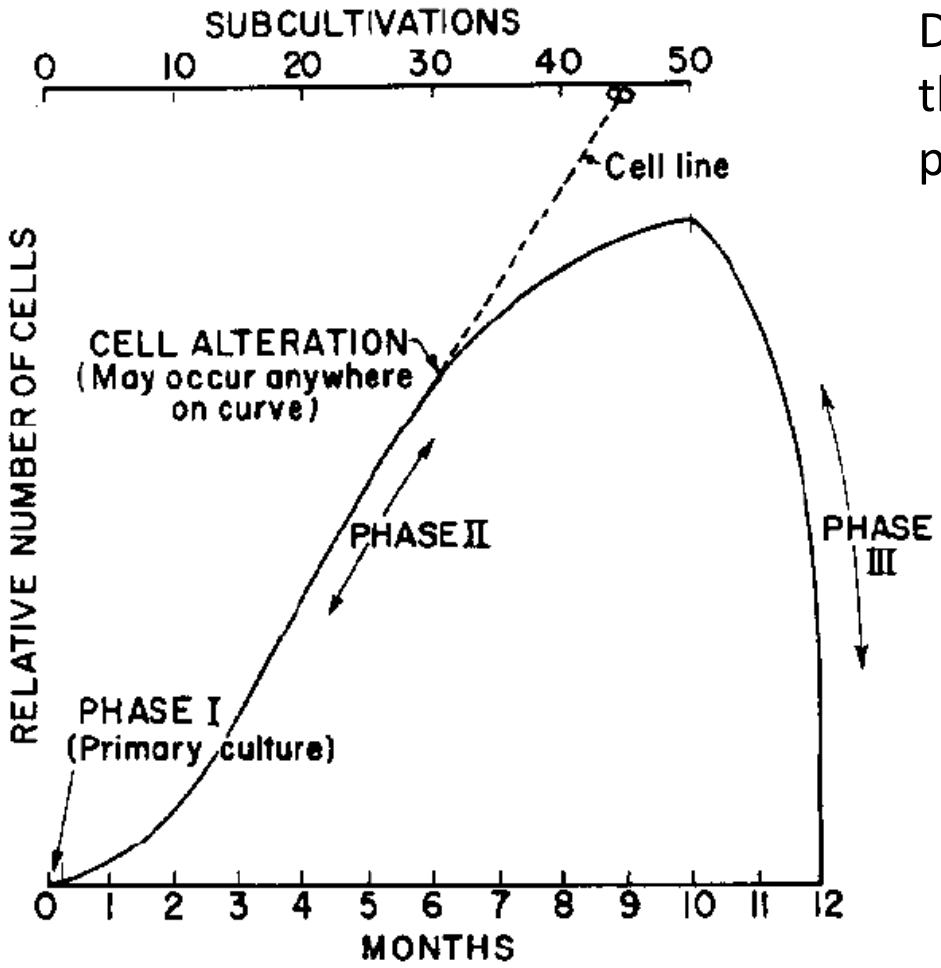
Brown-Sequard Brain circulation

A cat's thyroid gland, varying in weight from 85 to 110 mgs, demands about 230 cc of nutrient fluid.

The solutions contain protein split-products, hemin, cysteine, insulin, thyroxine, glutathione, vitamin A, ascorbic acid, blood serum, et. The apparatus is kept in an incubator at a temperature of 37-38C.

Thyroid glands were kept more than 20 days with pulsating arteries and active circulation.

THE SERIAL CULTIVATION OF HUMAN DIPLOID CELL STRAINS

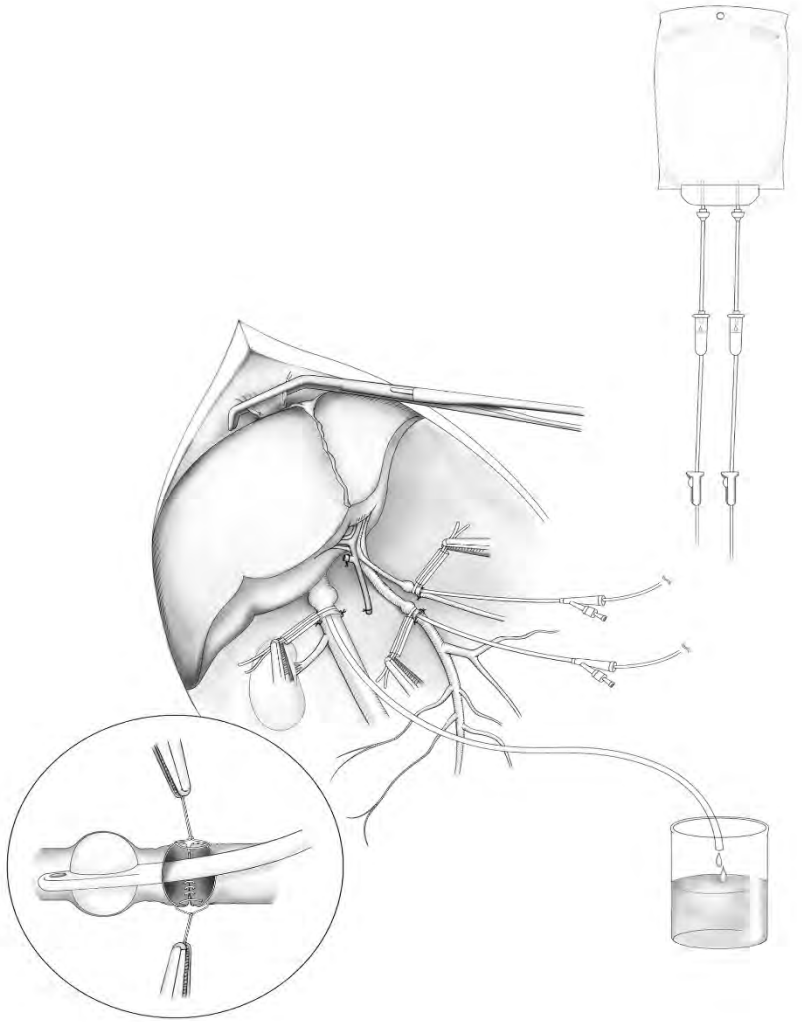


Dagrammatic representation of the history of cell strains and the phenomenon of cell alteration.

Phase I, or the primary culture, terminates with the formation of the first confluent sheet. **Phase II** is characterized by luxuriant growth necessitating many subcultivations. Cells in this phase are termed "cell strains". An alteration may occur at any time giving rise to a "cell line" whose potential life is infinite. Conversely, cell strains characteristically enter **Phase III** and are lost after a finite period of time.

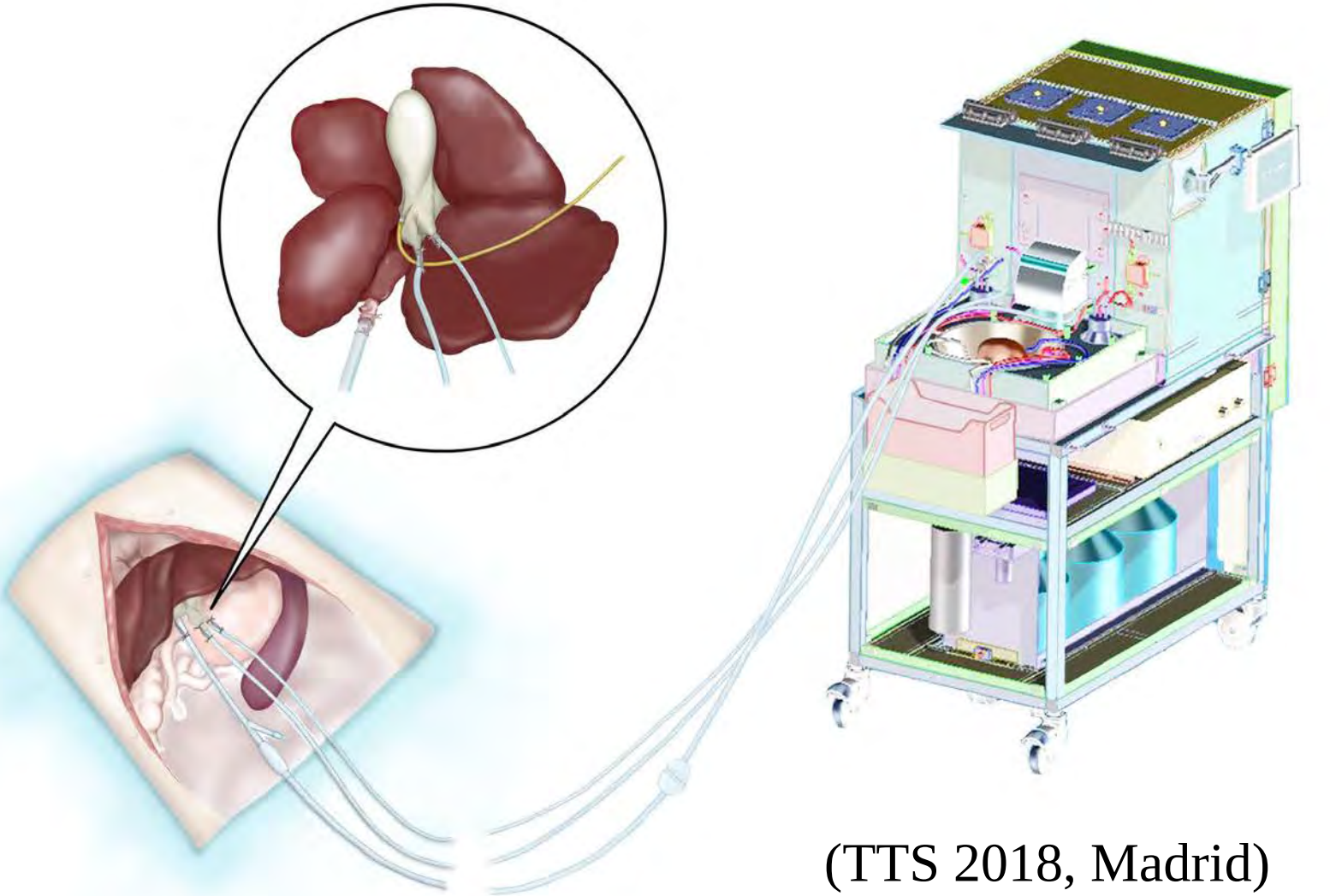
(L. Hayflick and PS Moorhead. Experimental Cell Research 1961)

"In-Site" Perfusion Technique for Rinse Solution in Liver Transplantation



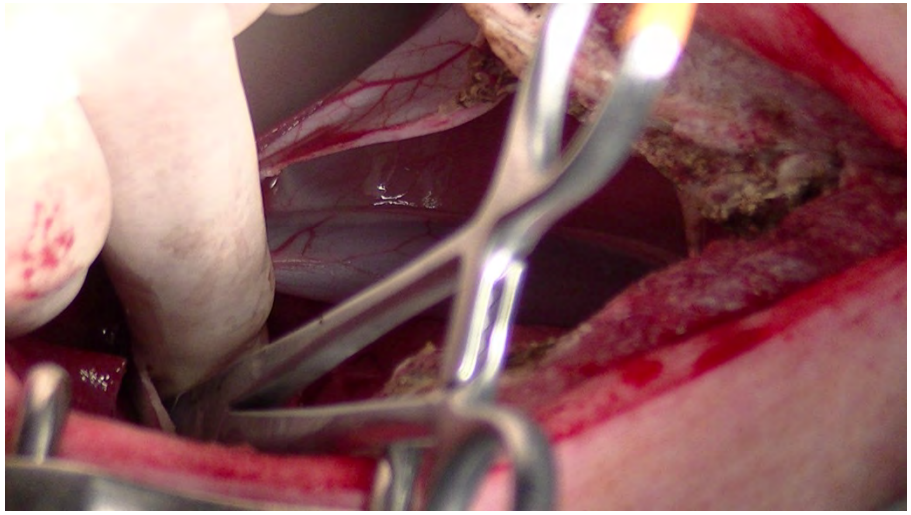
(Kobayashi E. Transplantation Direct2017)

A Novel Machine Perfusion System Continuously from Ex-vivo to “In-situ” on Marginal liver transplantation



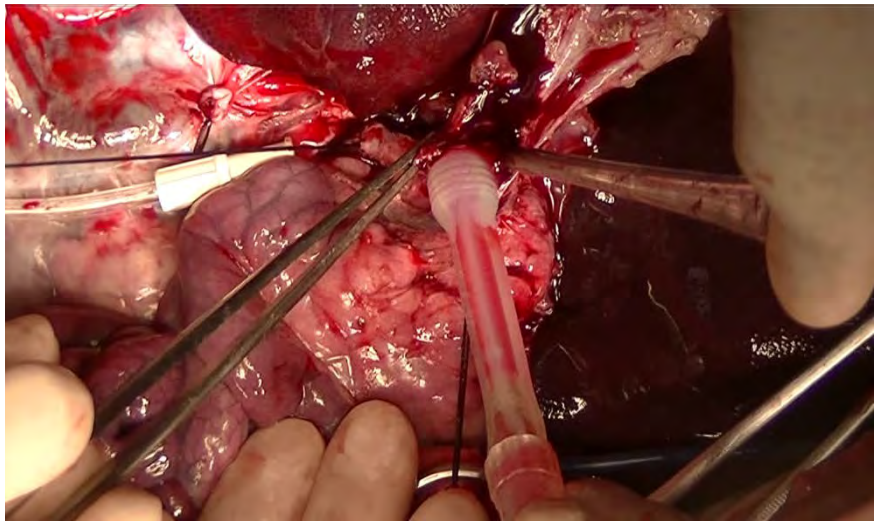
(TTS 2018, Madrid)

Donor Cardiac Death (DCD) Model in the Pig



A. Clamping of the thoracic Aorta

B. Systemic Injection of KCL solution

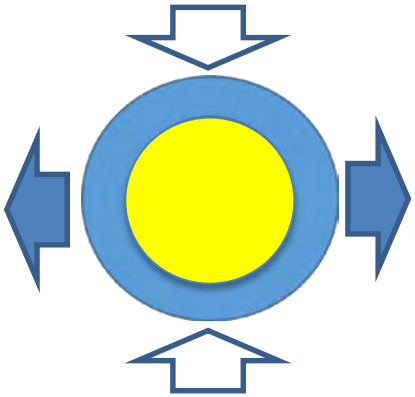
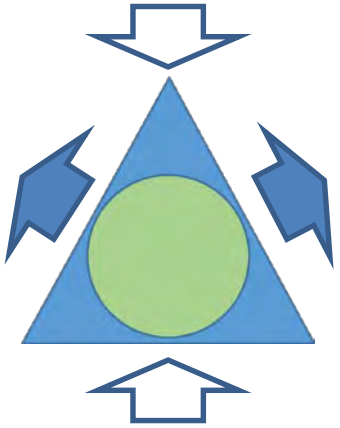
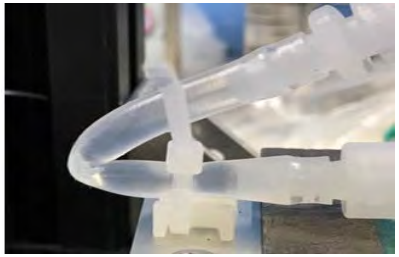


C. Donation of the marginal liver graft

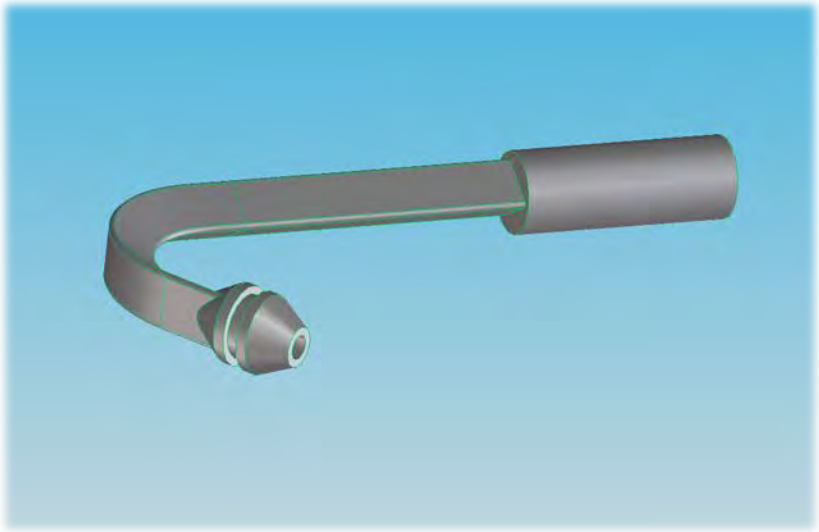
D. Massive colts from the graft liver

The kink less character

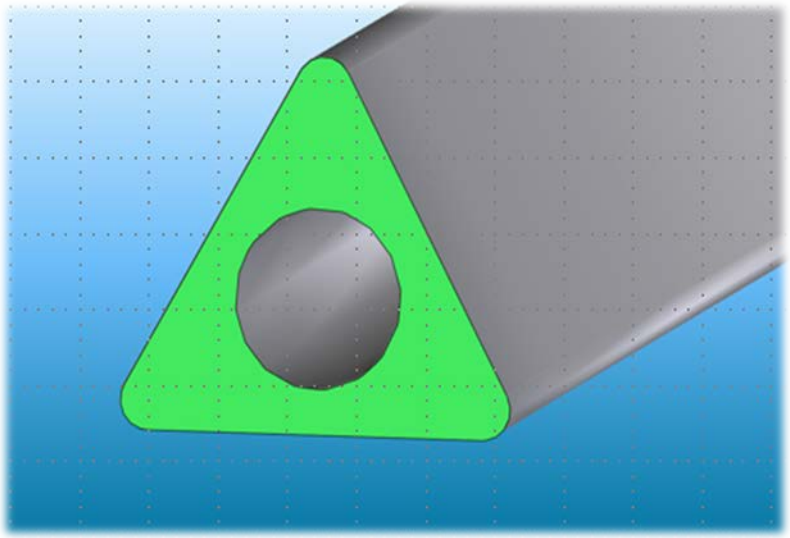
The rigidity mechanism

	The cylindrical tube	The triangular tube
Stress diffusion	Applied stress 	
Collapsed shapes image		
	 The flow was stopped at 150°	 Non-stop

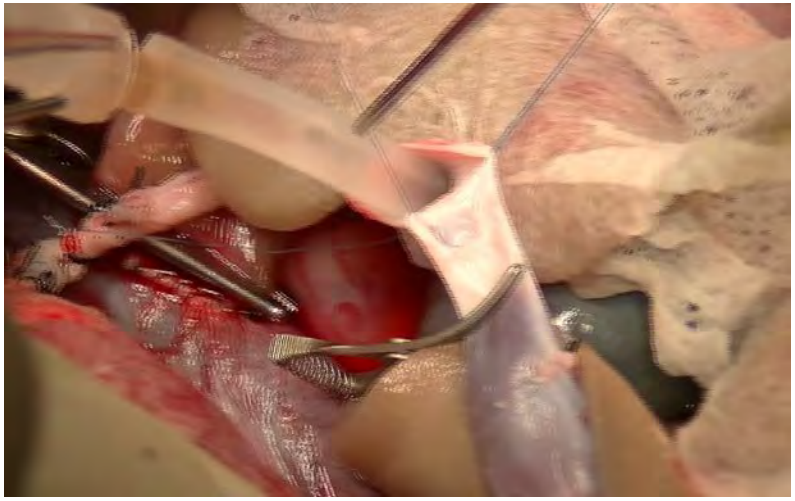
New technology for sewing vessels while perfusing organs



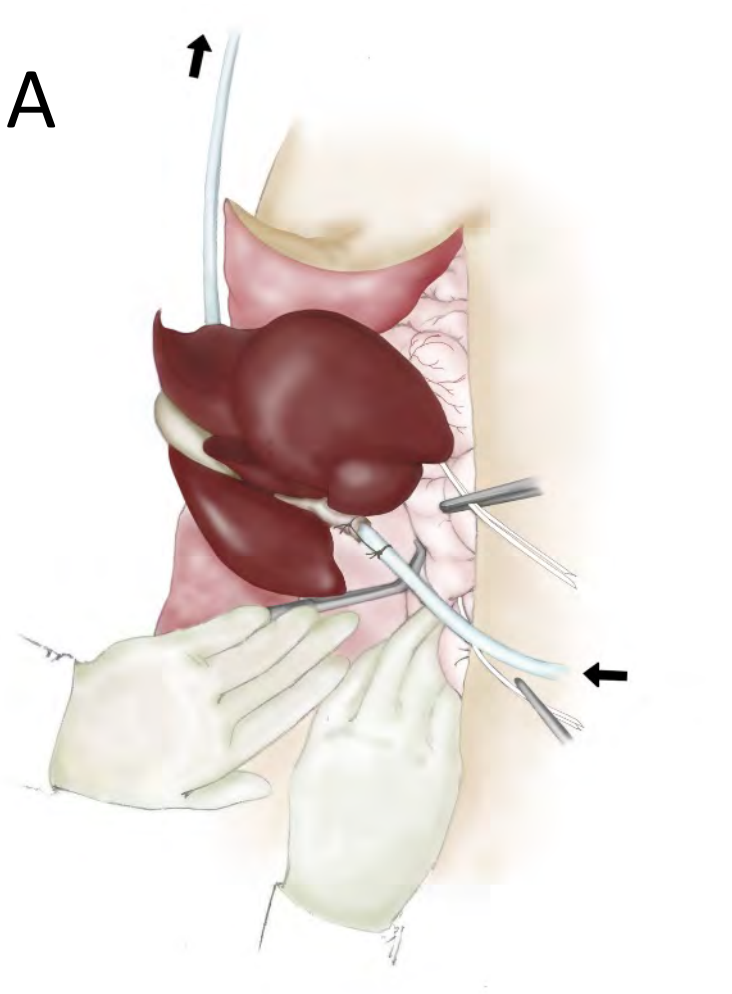
Triangular tube

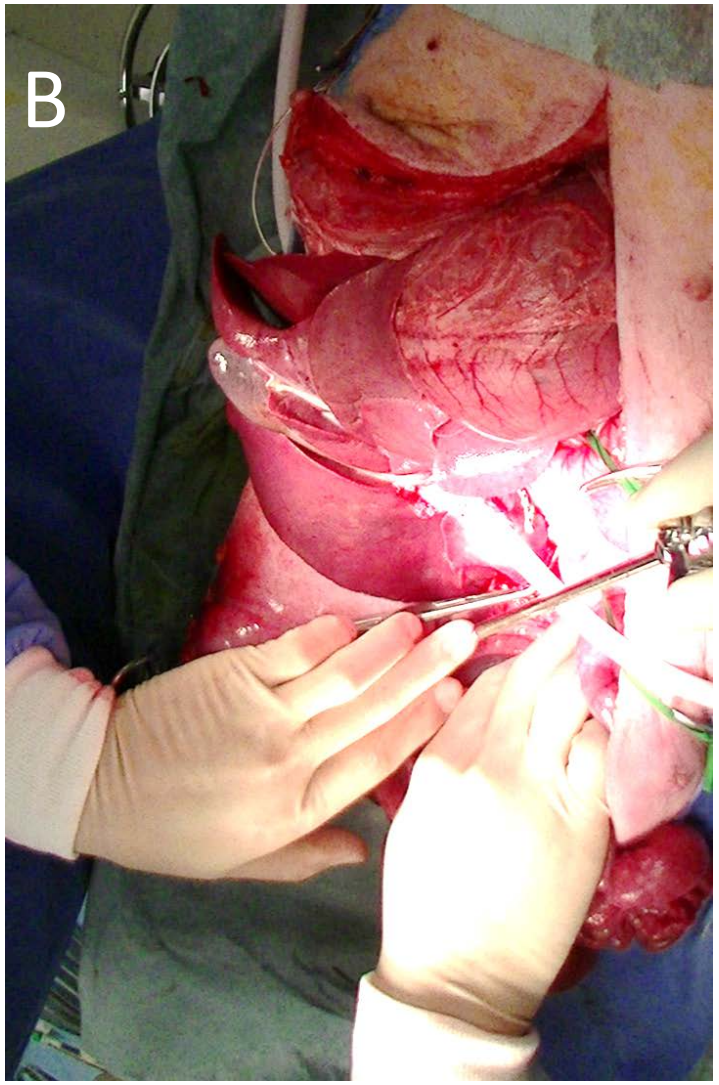


A cross section

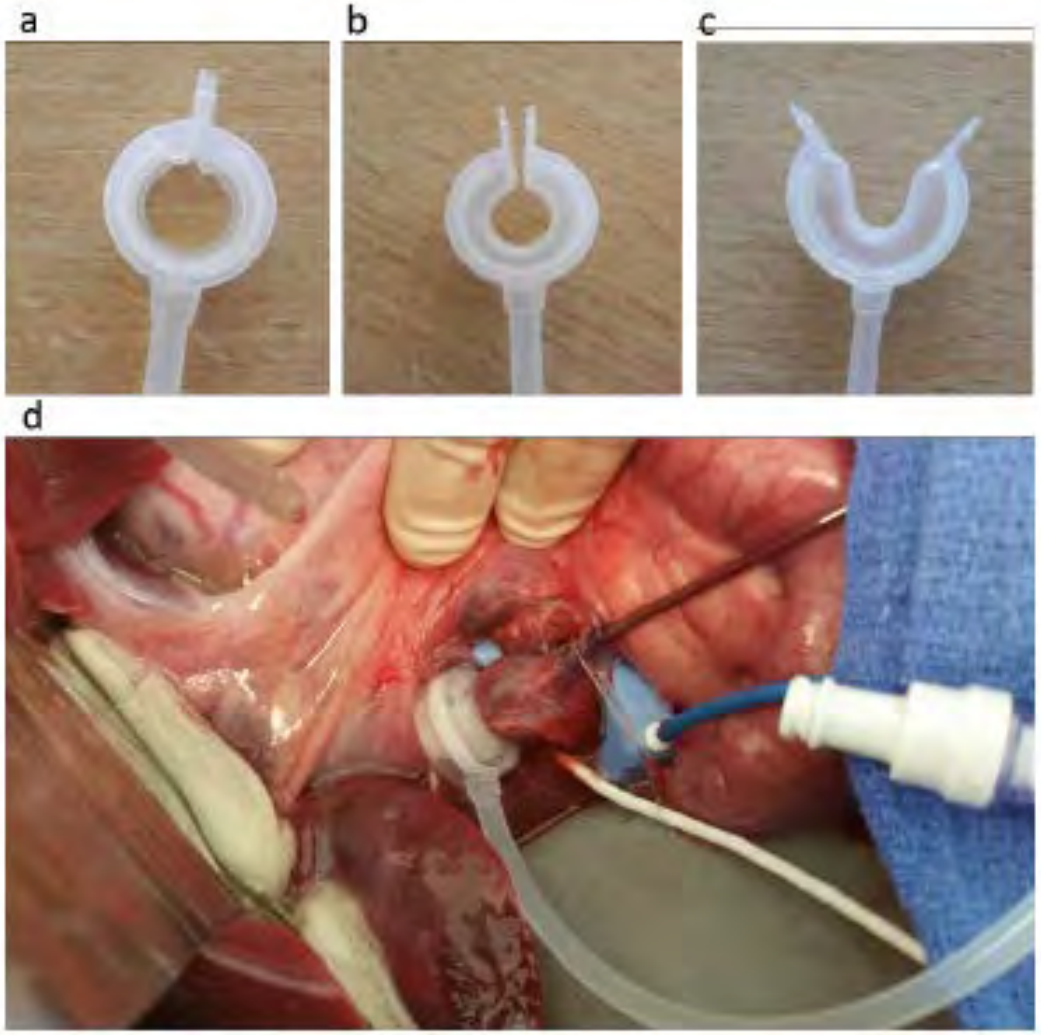


Ectopic Liver Transplantation for the damaged liver



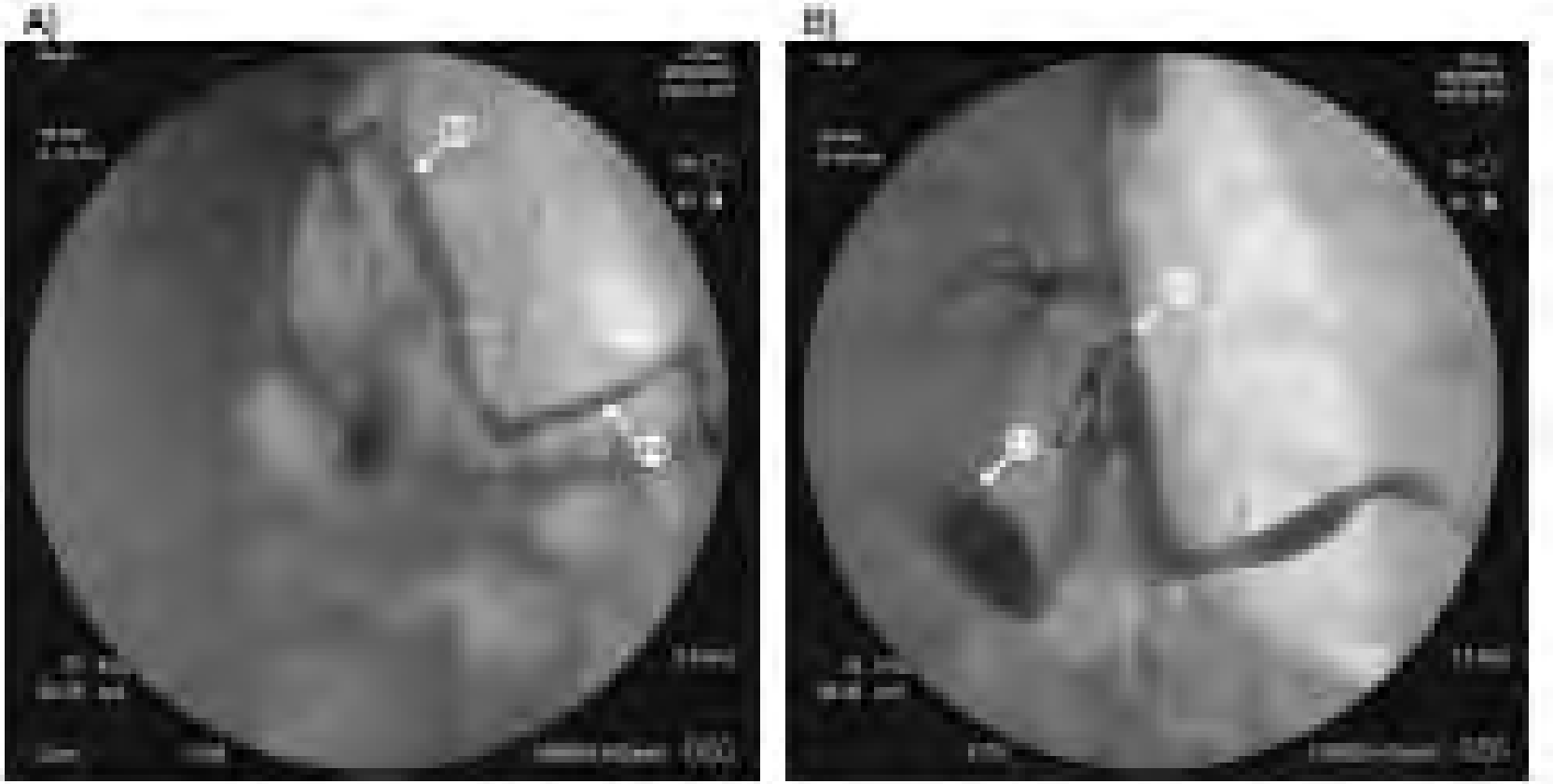


Modulating Portal Hemodynamics With Vascular Ring Allows Efficient Regeneration After Partial Hepatectomy in a Porcine Model



(Petru O, et al. Ann Surg 2018)

Angiography of portal vein after modeling portal hemodynamics in ectopic liver transplantation



(Yoshimoto S, et al, Transplantation Direct submitted)

Acknowledgement /共同研究者

慈恵医科大学
高血圧内科　横尾隆

明治大学　農学部
発生工学研究室　松成ひとみ、長嶋比呂志

北里大学
獣医学部　岩井聡美

慶應義塾大学　医学部
循環器内科　遠山周吾、藤田淳、福田恵一
心血管外科　川口新治、平野暁教、志水秀行
消化器外科　八木洋、北川雄光
消化器内科　杉本真也、佐藤俊朗

自治医科大学
先端医療技術開発センター　花園　豊

東京医科歯科大学
再生医学　関矢一郎

日本獣医生命大学
獣医看護　袴田陽二

京都大学　医学部
胆管膵・移植外科　八木真太郎、上本伸二

九州大学　医学部
小児外科　柳佑典、田口智章

佐賀大学　医学部
臓器再生医工学講座　中山功一

東京女子医科大学
先端生命医科学研究所　関根秀一、清水達也

国立成育医療センター
先端医療開発室　許懐哲、絵野沢伸

理化学研究所
多細胞システム形成研究センター
石川潤、辻孝