The pulmonary circulation facing the flow

Insights into the pathophysiology of vascular remodeling

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cardiogen
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Centre de Référence des Maladies Cardiaques Héréditaires

European Reference Network
for rare or low prevalence complex diseases
Network
Respiratory Diseases
(ERN-LUNG)

European Reference Network
for rare or low prevalence complex diseases
Network
Heart Diseases
(ERN GUARD-HEART)

Disclosure

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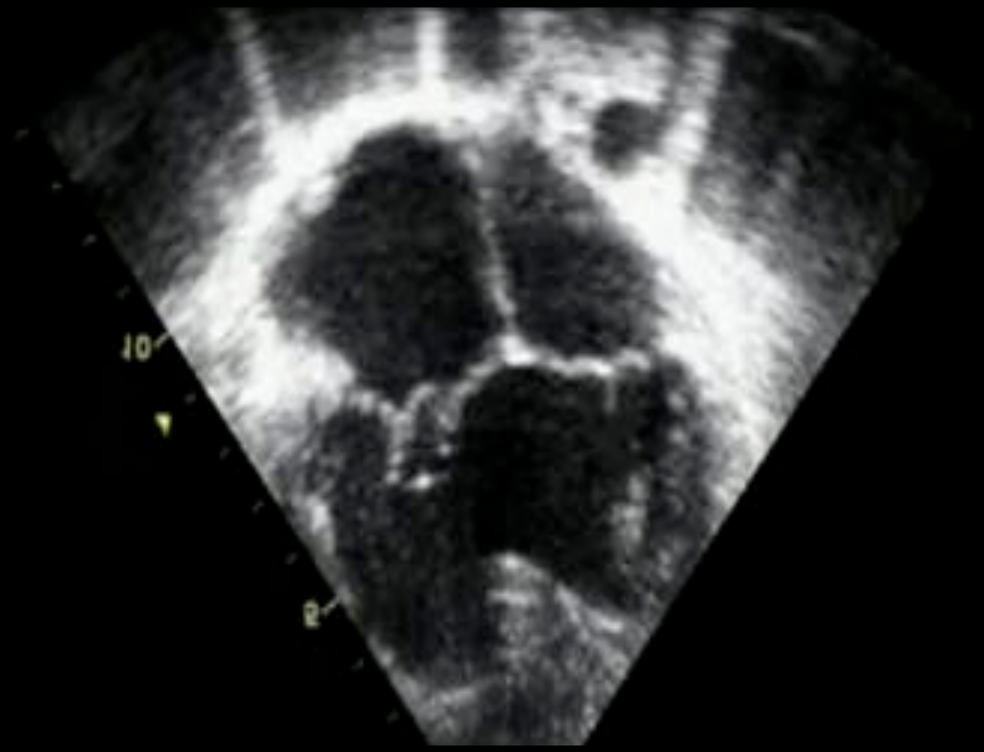
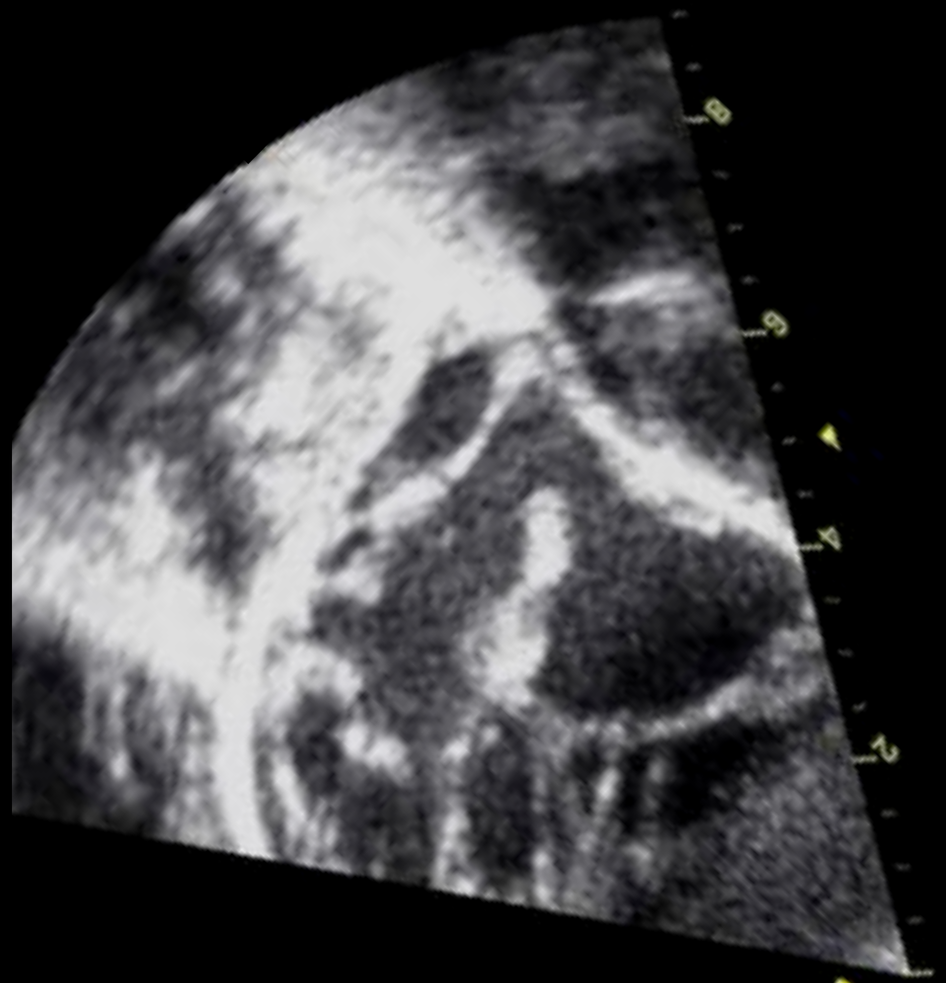
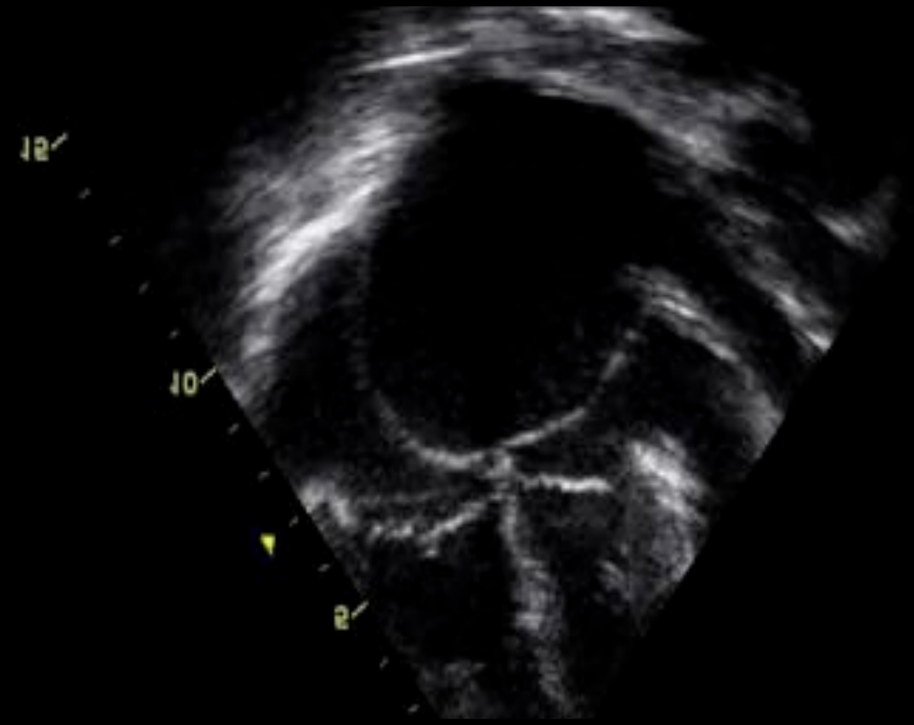




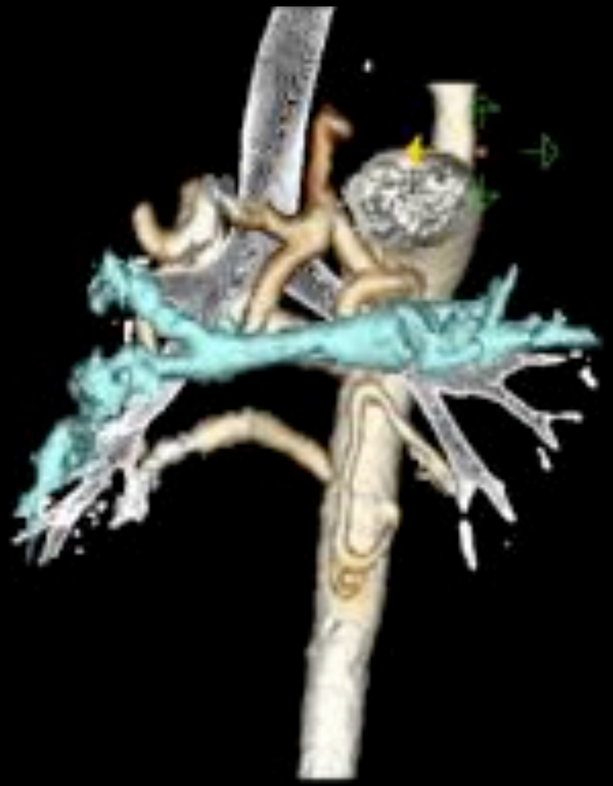
Flow-associated pulmonary hypertension (hyperkinetic)

Congenital systemic pulmonary shunt: *same physiology ?*

Cor triatriatum



Pulmonary atresia VSD

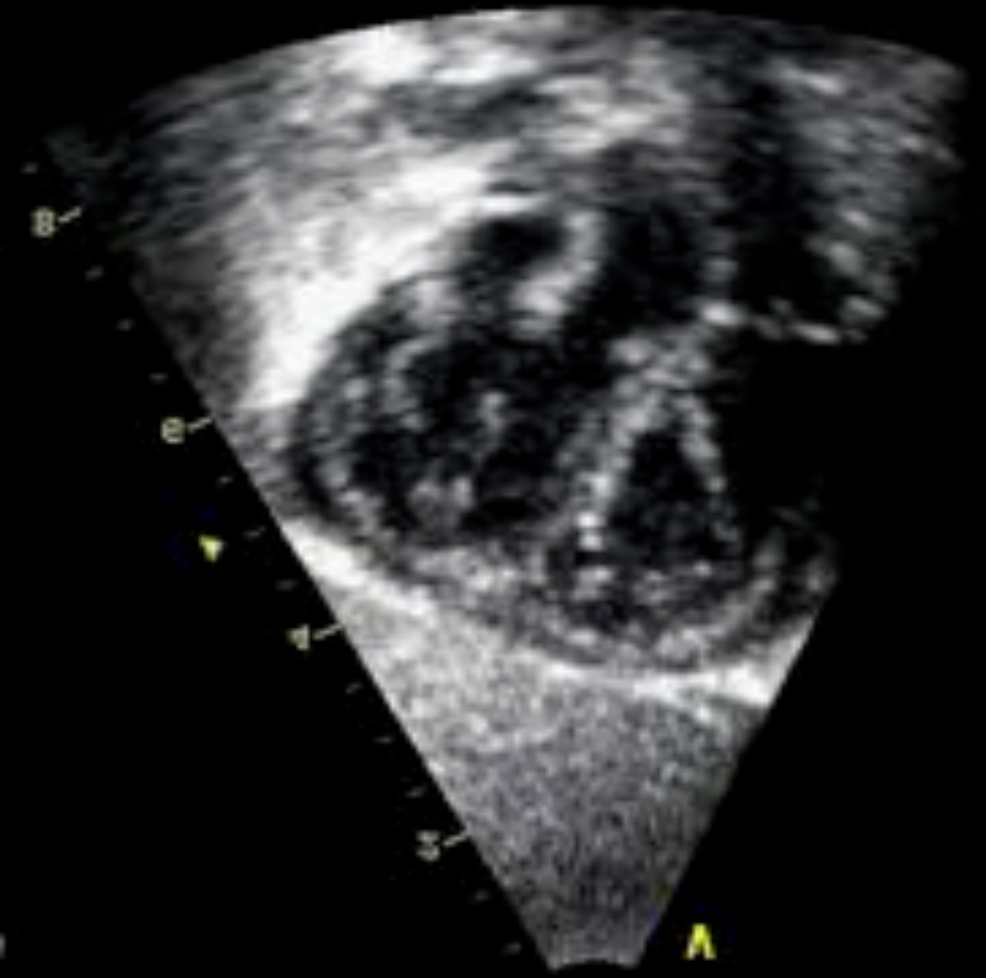


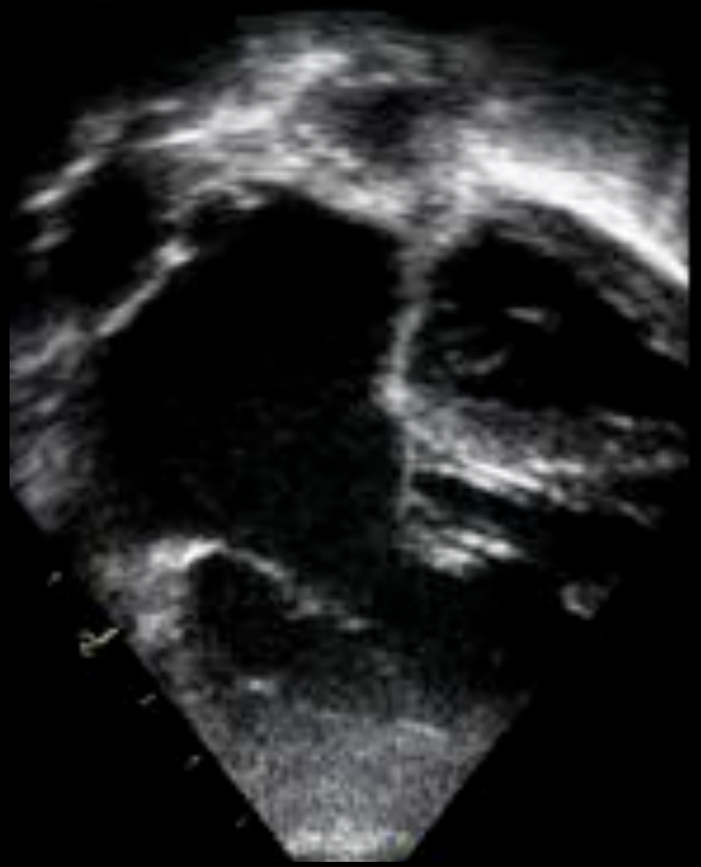
Pulmonary venous congestion

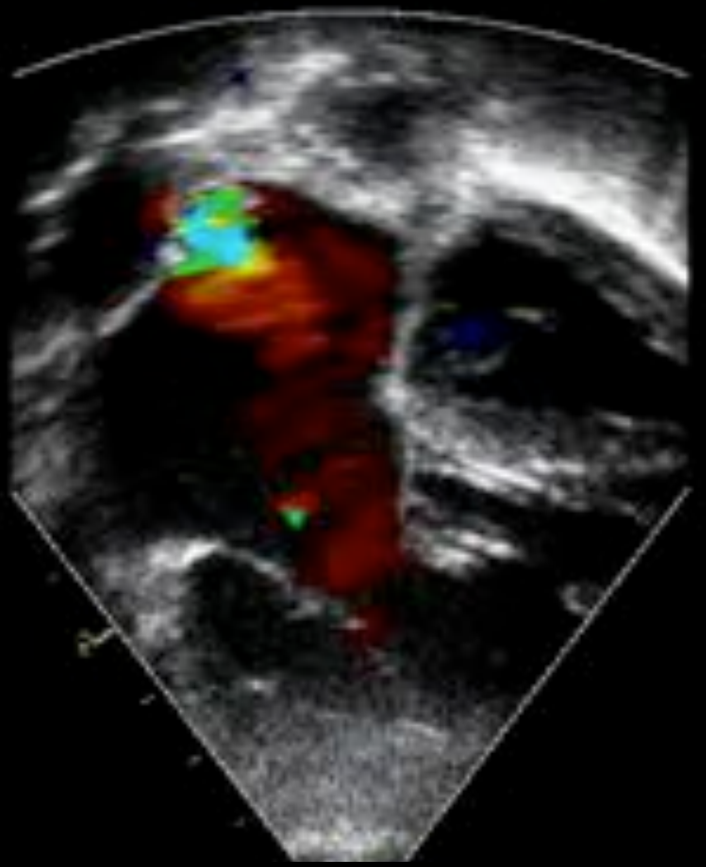
Segmental PH

Never shunt in TGA

TCPC







« Bizarre » physiopathologies with atypical/unknown vascular remodelling

Modified Classification of PH:Nice 2018

1.Pulmonary Arterial Hypertension
- **1.1 Idiopathic PAH**
- **1.2 Heritable PAH**
- **1.2.1. BMPR2**
- **1.2.2.ALK-1,endoglin,SMAD9, CAV1, KCNK3**
- **1.2.3 Unknown**
- **1.3 Drugs and toxins induced**
- **1.4 Associated with:**
- **1.4.1 Connective tissue disease**
- **1.4.2 HIV infection**
- **1.4.3 Portal hypertension**
- **1.4.4 Congenital Heart diseases**
- **1.4.5 Schistosomiasis**

1’ Pulmonary Veno Occlusive Disease and/or Pulmonary Capillary Hemangiomatosis

1.'' PPHN

2. Pulmonary Hypertension Due to Left Heart Disease
- **2.1 Left Ventricular Systolic Dysfunction**
- **2.2 Left Ventricular Diastolic Dysfunction**
- **2.3 Valvular disease**
- **2.4 Congenital / acquired left heart inflow/outflow tract obstruction**

3.Pulmonary Hypertension Due to Lung Diseases and/or Hypoxia
- **3.1 Chronic obstructive pulmonary disease**
- **3.2 Interstitial lung disease**
- **3.3 Other pulmonary diseases with mixed restrictive and obstructive pattern**
- **3.4 Sleep-disordered breathing**
- **3.5 Alveolar hypoventilation disorders**
- **3.6 Chronic exposure to high altitude**
- **3.7 Developmental lung diseases**
- **3.7.1 Congenital diaphragmatic hernia**
- **3.7.2 Bronchopulmonary dysplasia**

4.Chronic Thromboembolic Pulmonary Hypertension

5.Pulmonary Hypertension with Unclear Multifactorial Mechanisms
- **5.1 .Hematologic disorders:chronic hemolytic anemias myeloproliferative disorders splenectomy,**
- **5.2 Systemic disorders, Sarcoidosis, pulmonary Langerhans cell histiocytosis, Lymphangioleiomyomatosis, neurofibromatosis, vasculatis**
- **5.3 Metabolic disorders: Glycogen storage disease, Gaucher disease, thyroid disorders**
- **5.4 Others: Segmental PAH, tumoral obstruction, fibrosing mediastinitis, chronic renal failure**

M3C

2013/18 Nice
Clinical Classification of PAH Associated with CHD

A. Eisenmenger Syndrome

Includes all large intra- and extra-cardiac defects which begin as systemic-to-pulmonary shunts and progress with time to severe elevation of PVR and to reversal (pulmonary-to-systemic) or bidirectional shunting; cyanosis, secondary erythrocytosis, and multiple organ involvement are usually present.

B. Left to Right Shunts
- *Operable*
- *Inoperable*

C. PAH with co-incidental CHD

D. Post-operative PAH

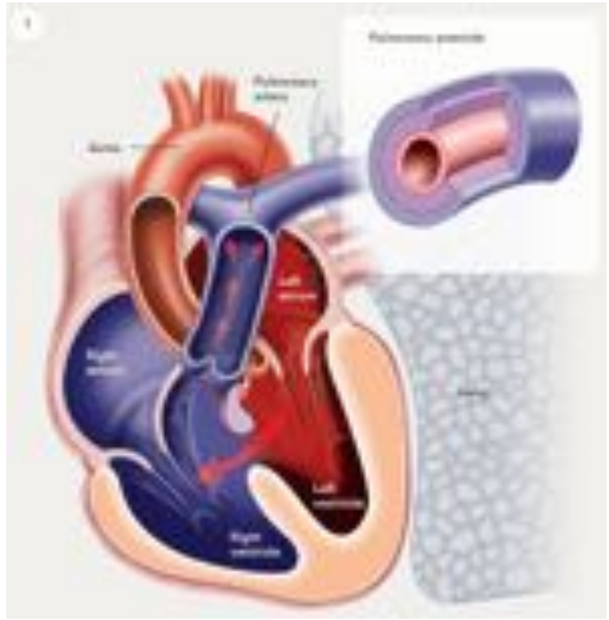
E. Never shunt/non classifiable

Definition of PAH based on mean PAP ≥ 25mmHg;
PVR provides essential information for CHD patients

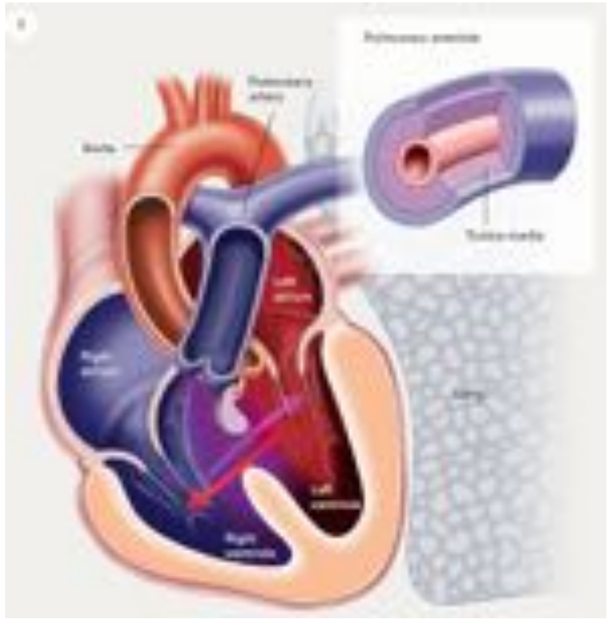
Ivy D et al. J Am Coll Cardiol 2013;62:D117–26

Left-to-right shunt: natural history/*physiology*

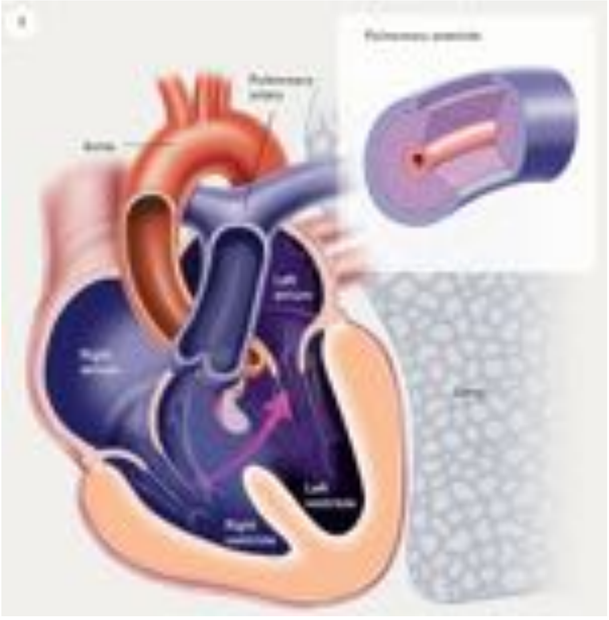
Systemic-to-pulmonary shunting can ultimately remodel the pulmonary vasculature to a characteristic irreversible phenotype similar to other forms of PAH.



ASD, VSD or complex defect, ↑ Qp and/or PAP, with left-to-right shunting

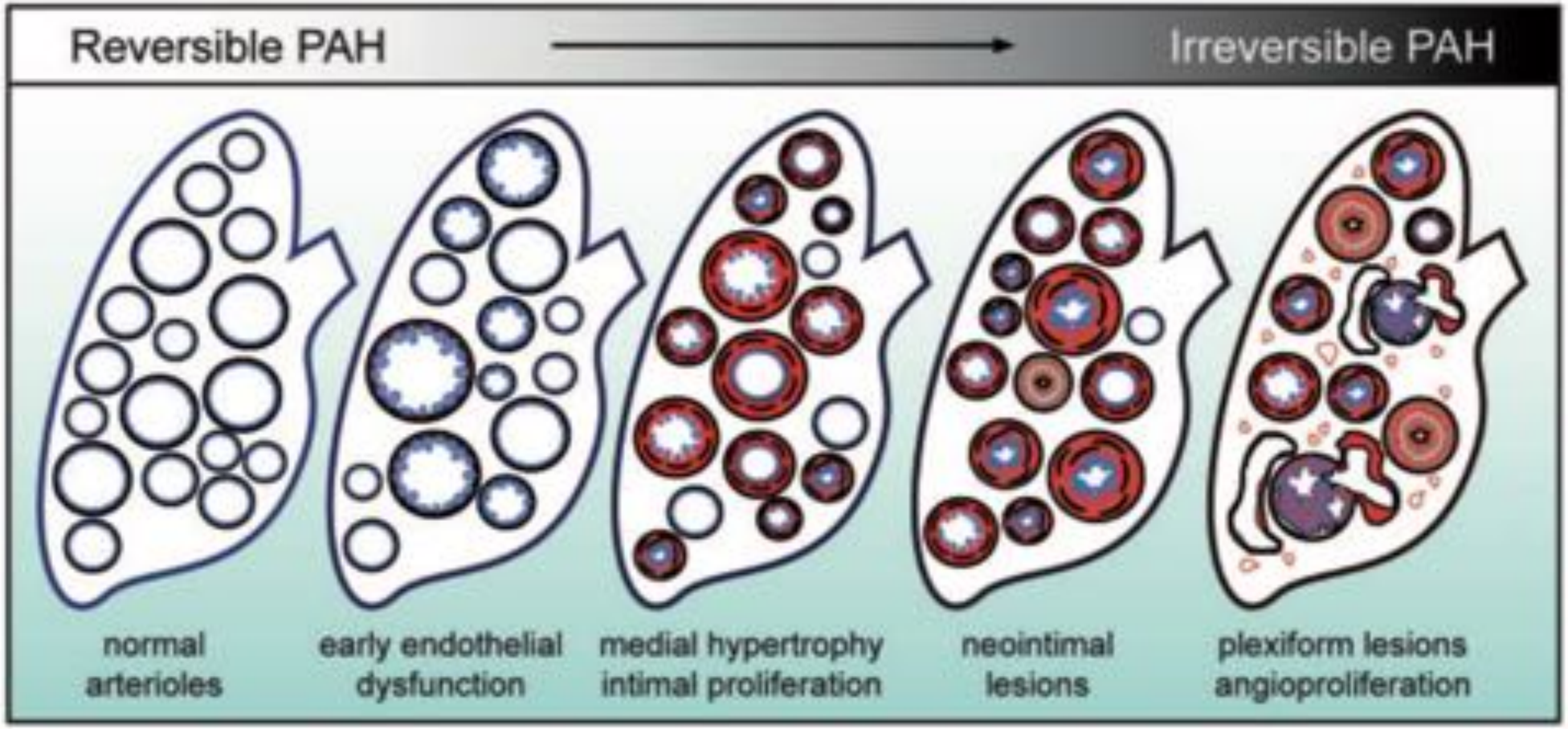


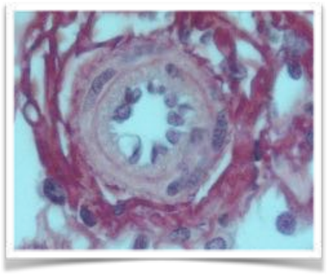
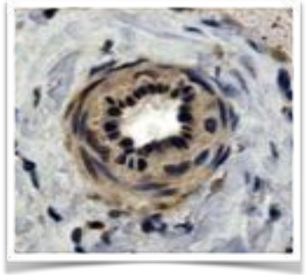
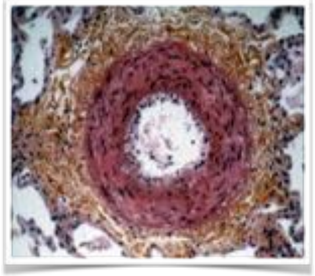
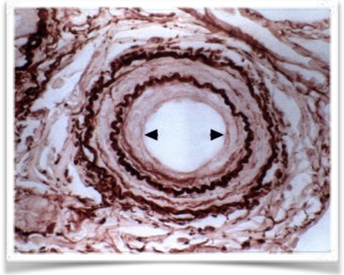
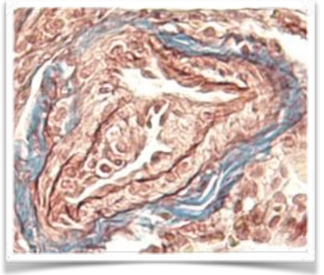
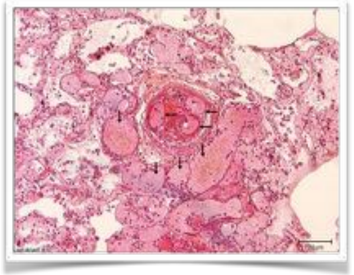
Over time, PVR ↑ resulting in bi-directional flow



Resistance ↑ further with reversal of shunt: right-to-left → Eisenmenger syndrome – patient becomes ↑ cyanotic

Left-to-right shunt: natural history/*pathology*



Shear stress and circumferential stretch

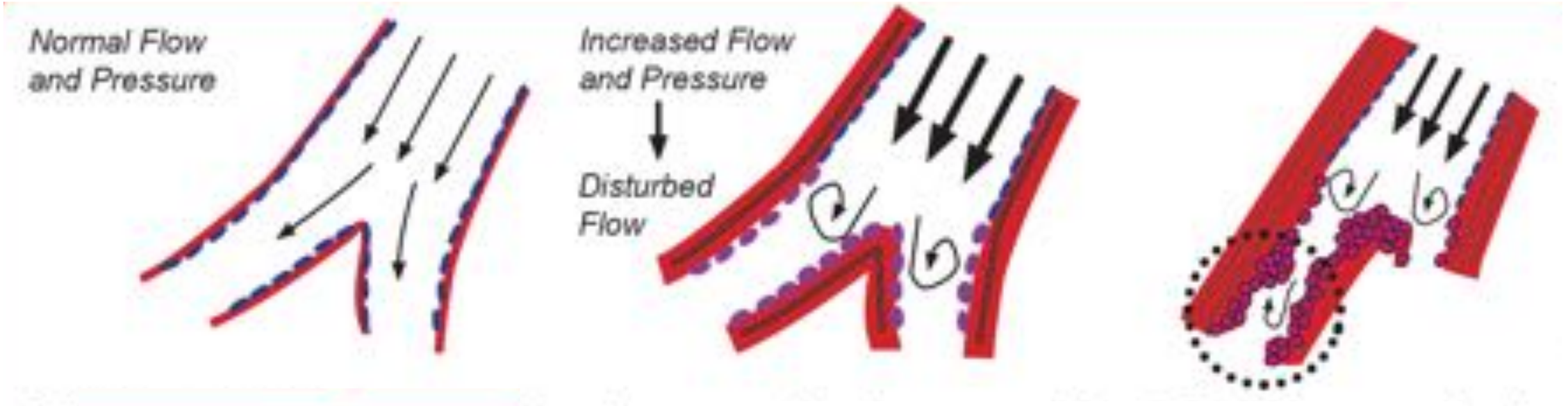
Shear stress

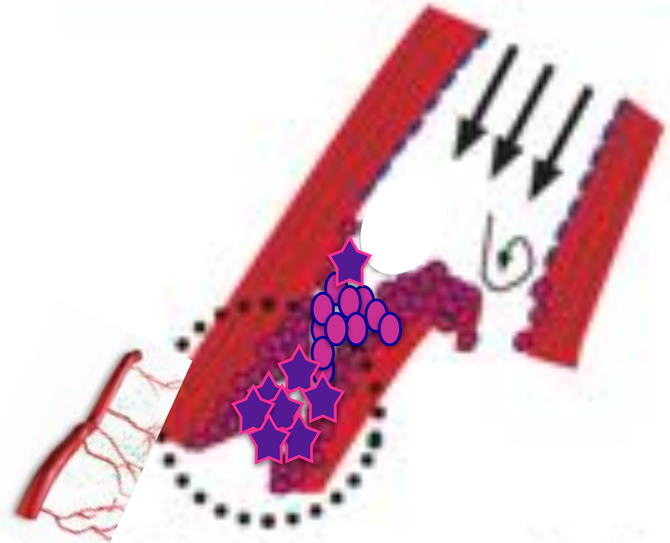
Circumferential stretch

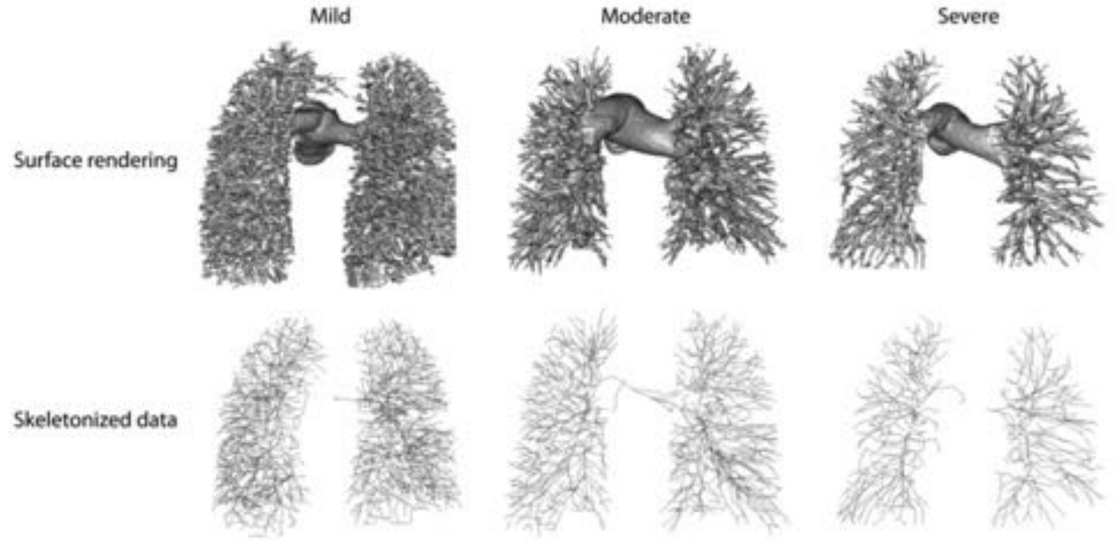
These hemodynamic forces are translated into biochemical signals

Hemodynamic forces → reaction in vessels → messengers → cellular response

Increased flow & pressure are the essential triggers for the development of PH in CHD



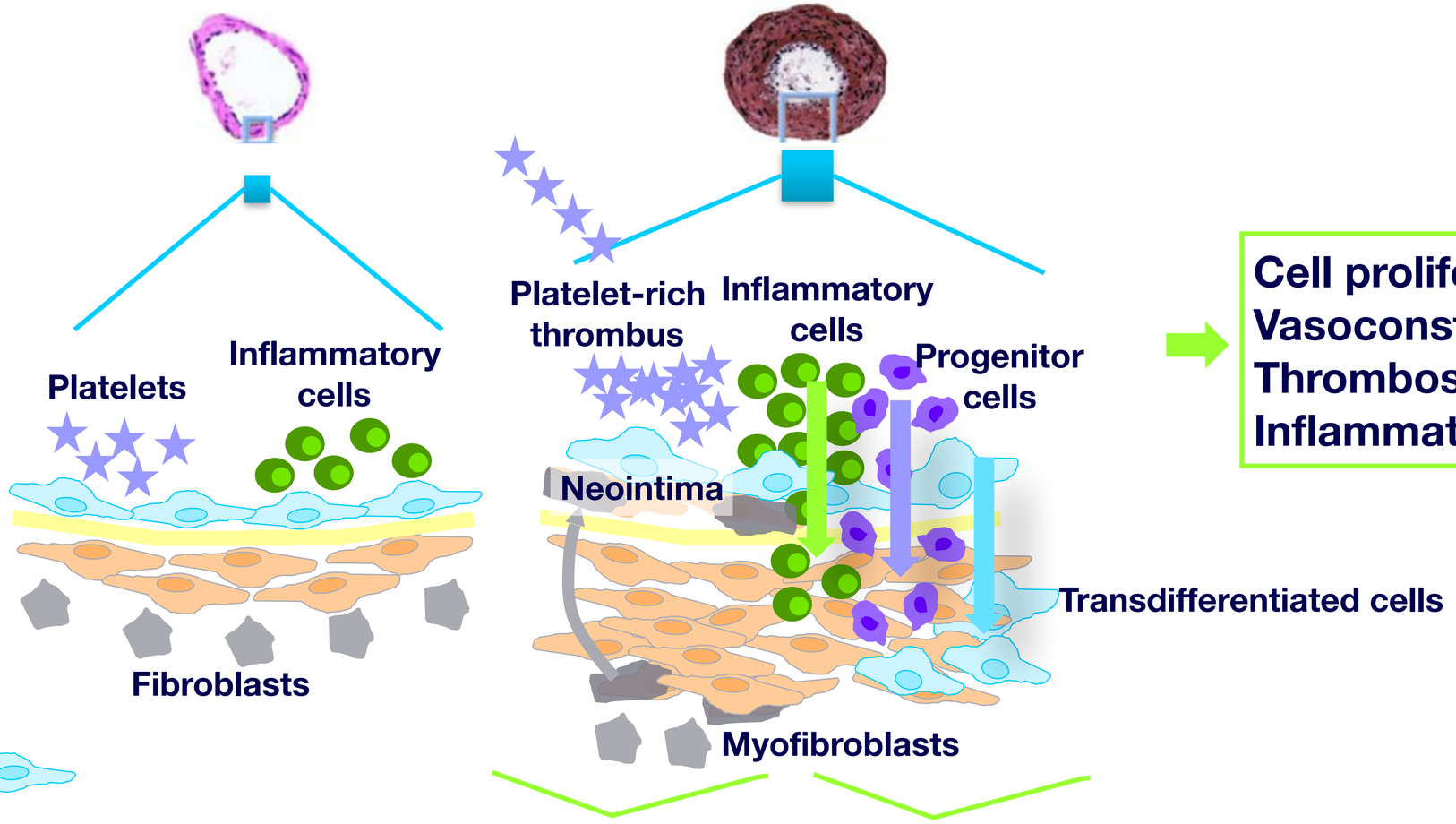




Fractal dimensions in PH

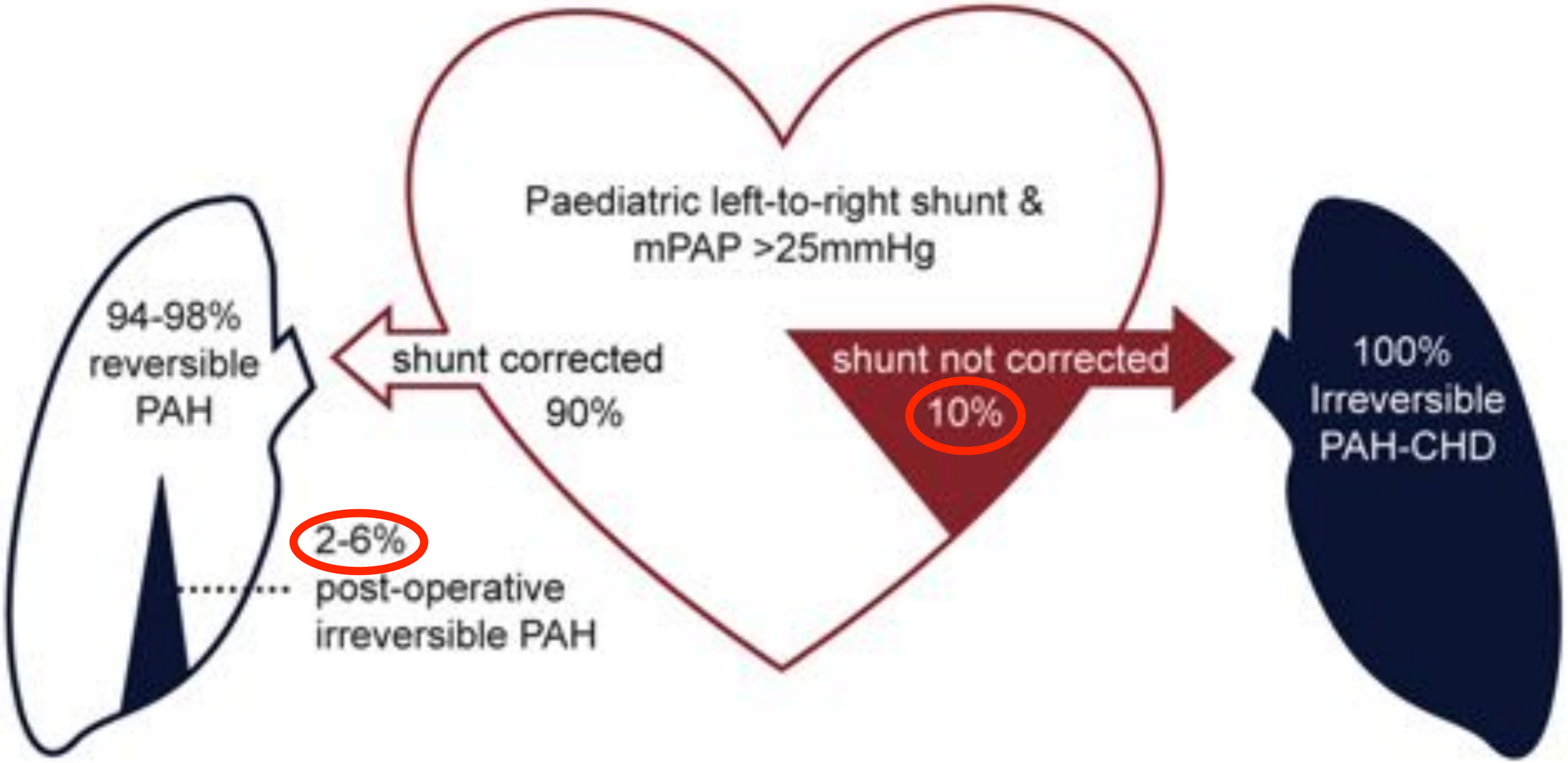
Dickinson, AJP 2013;
van der Feen, Eur Heart J 2017
Moledina S et al. Heart 2011

Reaction in vessels

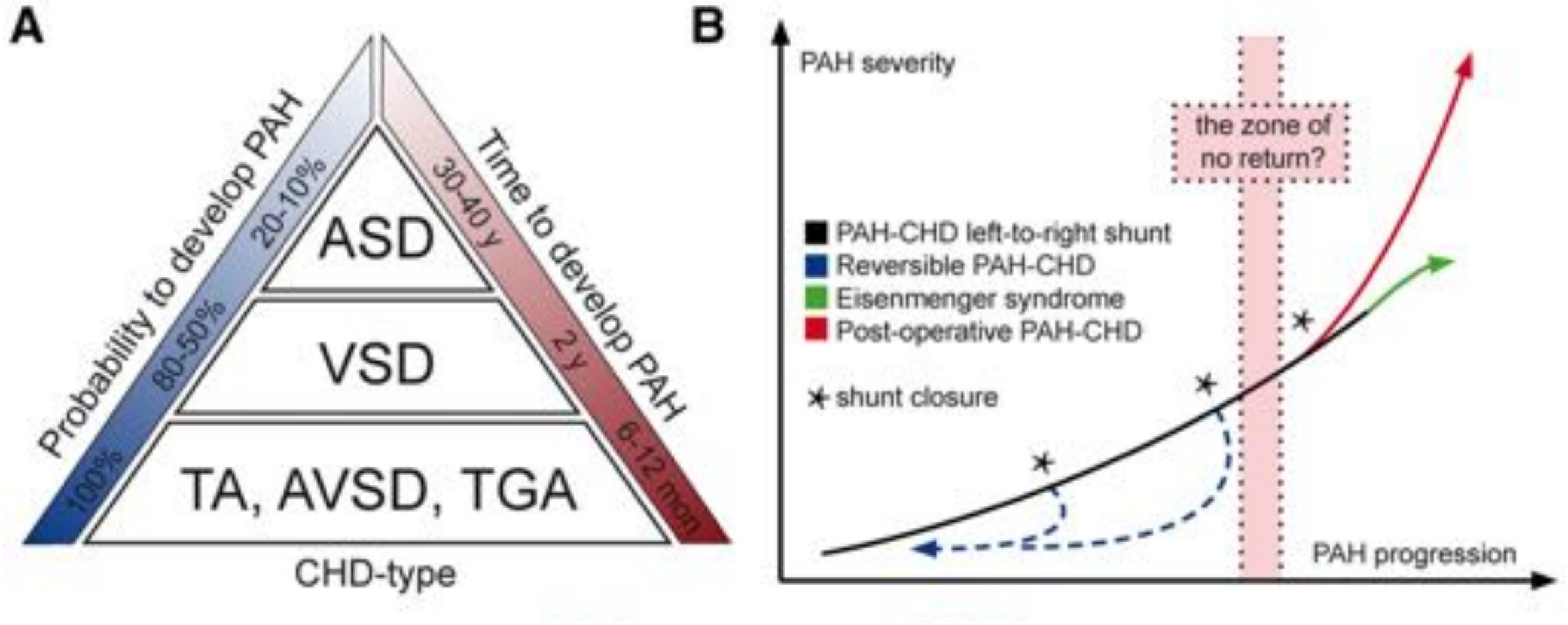


Cell proliferation
Vasoconstriction
Thrombosis
Inflammation

Chan SY and Loscalzo J. J Mol Cell Cardiol 2008; 44:14-30.

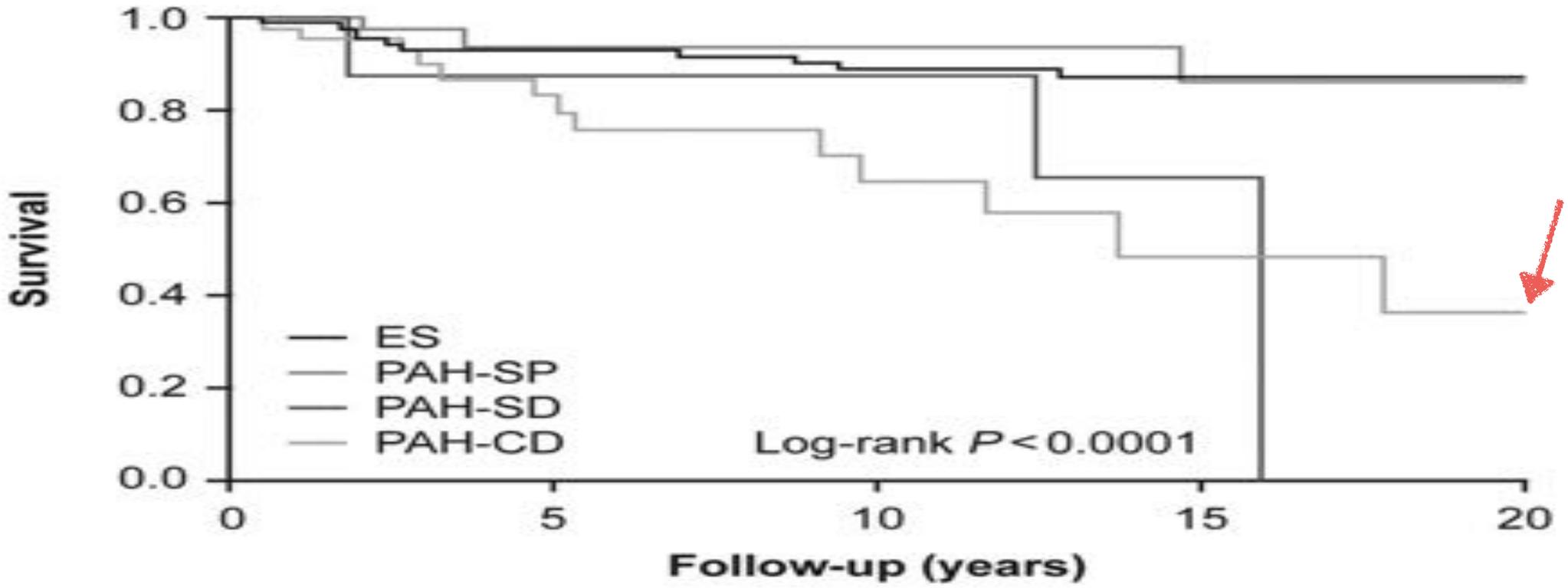


From: Pulmonary arterial hypertension in congenital heart disease: translational opportunities to study the reversibility of pulmonary vascular disease
Eur Heart J. 2017;38(26):2034-2041.



From: Pulmonary arterial hypertension in congenital heart disease: translational opportunities to study the reversibility of pulmonary vascular disease
Eur Heart J. 2017;38(26):2034-2041.

Kaplan–Meier survival curve of 192 patients with pulmonary arterial hypertension associated with congenital heart disease categorized according to the four clinical subgroups



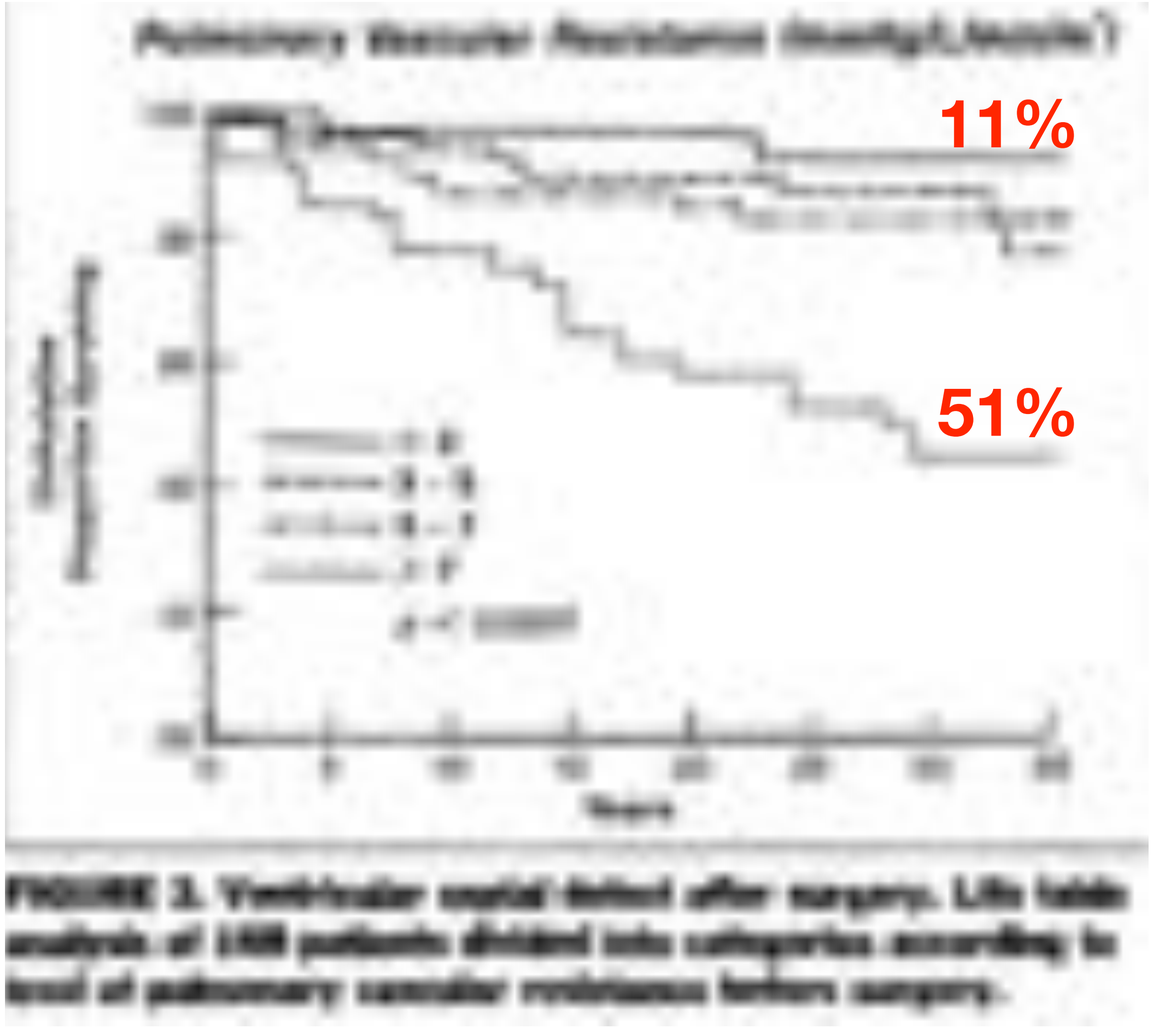
Patients at risk

ES	90	71	59	52	48
PAH-SP	48	22	18	11	10
PAH-SD	10	4	4	2	0
PAH-CD	44	22	12	4	3

Manes A et al. Eur Heart J 2014;35:716-724

Recommendations for correction of CHD with prevalent systemic-to-pulmonary shunts

Recommendations			Class[a]	Level[b]	Ref.[c]
PVRi (WU · m²)	PVR (WU)	Correctable[d]			
<4	<2.3	Yes	IIa	C	317
>8	>4.6	No	IIa	C	317
4–8	2.3–4.6	Individual patient evaluation in tertiary centres	IIa	C	317



Moller JH, Patton C, Varco RL, Lillehei CW. Late results (30to35years) after operative closure of isolated ventricular septal defect from 1954 to 1960. Am J Cardiol 1991;68:1491–1497.

Attempt to define group 2 (ex group B): Operable vs. Inoperable

Table 3. Recommended preoperative evaluation of pediatric patients with congenital systemic-to-pulmonary shunts, with the findings that may indicate a favorable or unfavorable response to correction of cardiac lesions

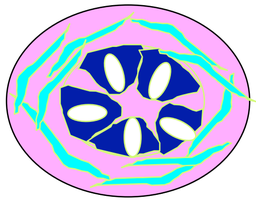
	Findings	
Source, features/parameters	Favorable	Unfavorable
Clinical history		
Age, years	<1	>2
Congestive heart failure/pulmonary congestion	Present	Absent
Tendency to respiratory disorders (inflammatory/infectious)	Yes	No
Failure to thrive	Yes	No
Use of anticongestive medication	Yes	No
Associated syndromes	No	Yes (Down syndrome)
Associated airway/lung disease	No	Yes
Physical examination		
Dyspnea	Present/overt	Mild/absent
Dynamic precordium	Present	Absent
Precordial murmur	Present	Absent
Second heart sound (pulmonic area)	Mildly increased split present	Loud split absent
Peripheral oxygen saturation, %	>93	<90
Associated airway obstruction/lung disease	No	Yes
Chest X-ray		
Size of the heart	Enlarged	"Hypertrophic"
Pulmonary vascular markings	Proeminent	Decreased distal markings
Congestion	Present	Absent
Parenchymal lung disease	Absent	Present
Transthoracic echocardiography		
Direction of flow across the communication	Left-to-right or bidirectional, but predominantly left-to-right	Bidirectional, predominantly right-to-left
Size of left cardiac chambers (posttricuspid shunts)	Enlarged	Not enlarged
Pulmonary-to-systemic blood flow ratio (Qp : Qs)	>3.0 : 1	>2.0 : 1
Right ventricular dysfunction	Absent	Present
Type of defects[a]	Simple lesions[b]	Complex anomalies[c]
Cardiac catheterization		
Pulmonary vascular resistance index, Wood units m^2	<6.0 (preferably, <4.0)	>8.0
Pulmonary-to-systemic vascular resistance ratio (PVR : SVR)	<0.3	>0.5

Pulm Circ 2014;4(2):330-341

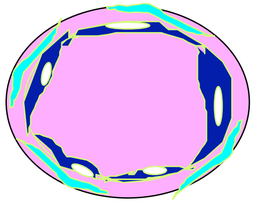
Normalisation of Flow (Haemodynamic Unloading) reverses PAH-CHD,

but not after a certain point of no return.

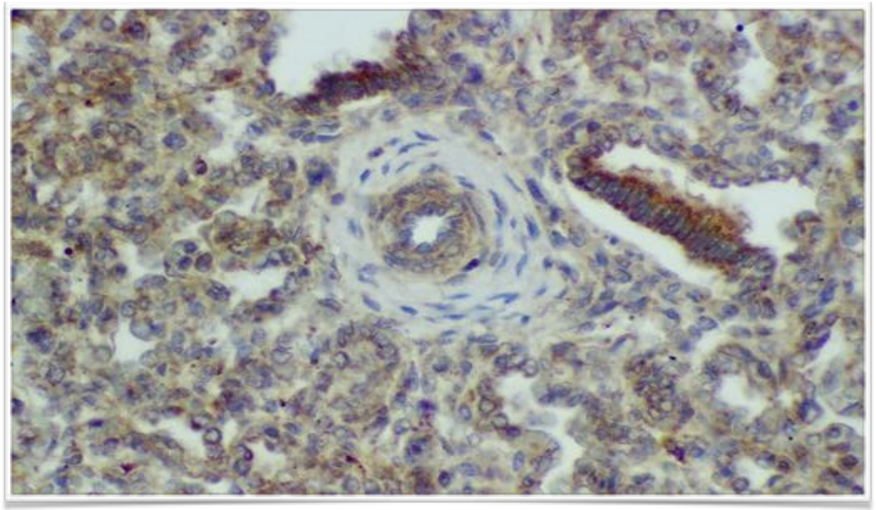
Which lesions are reversible in PAH-CHD ?



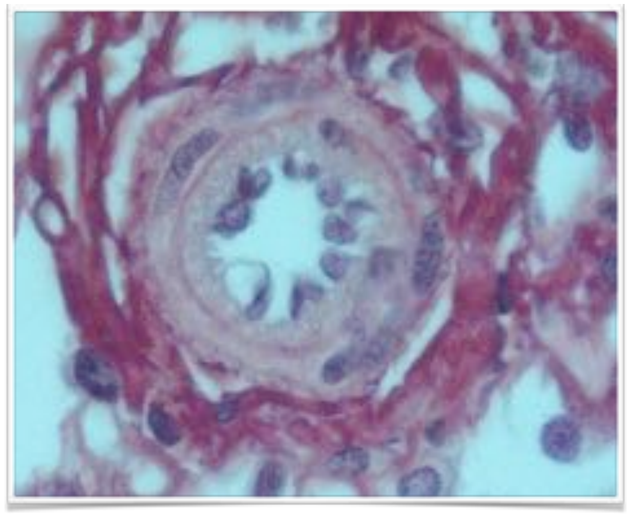
Foetus



Birth



Foetal arteriole

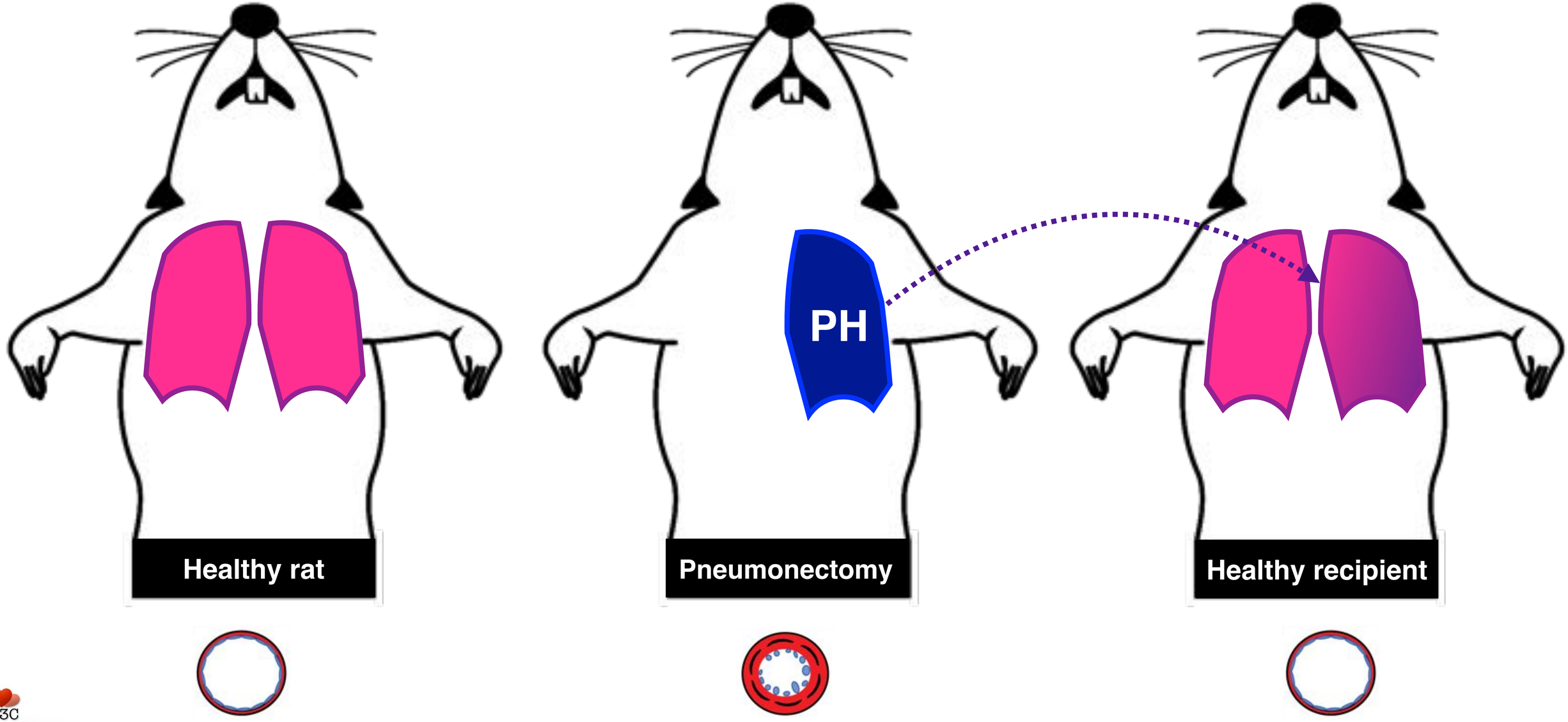


Mature arteriole

8 weeks

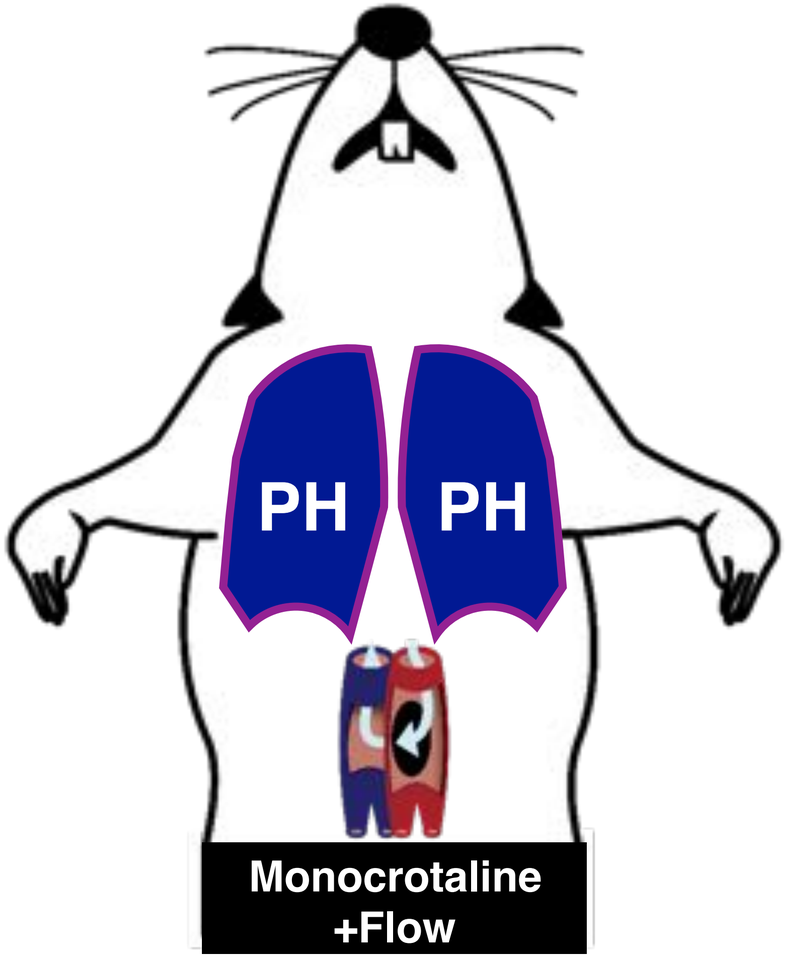
Wagenvoort, and Heath & Edwards

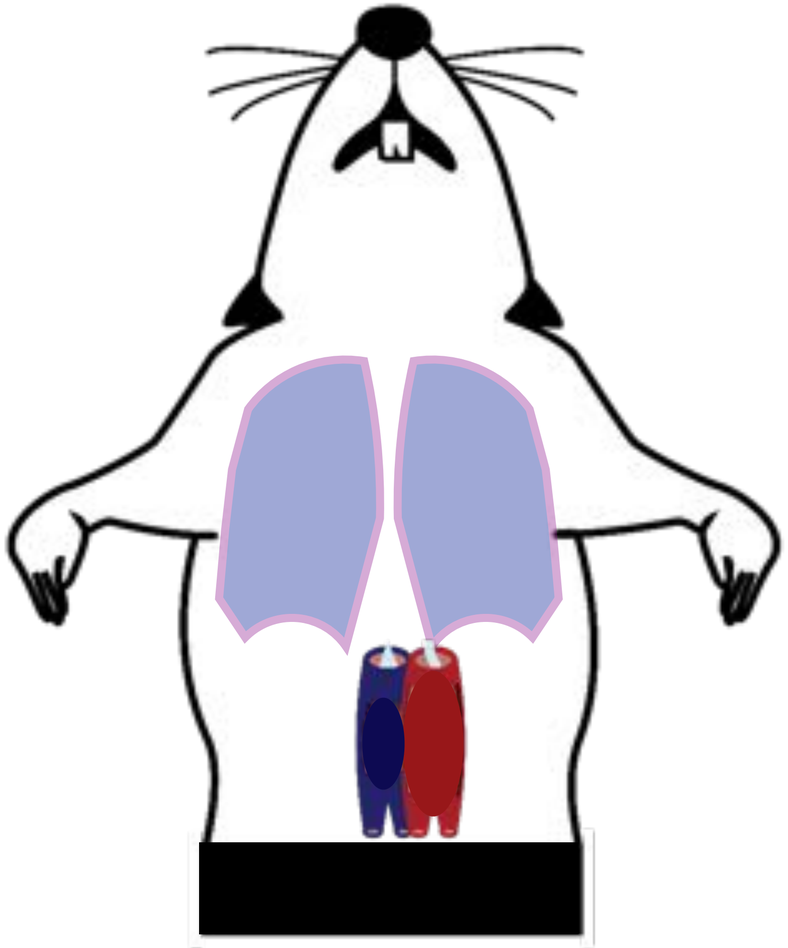
Reversibility in animal models



O'Blenes SB et al. J Thorac Cardiovasc Surg 2001;121:279–289.

Reversibility in animal models



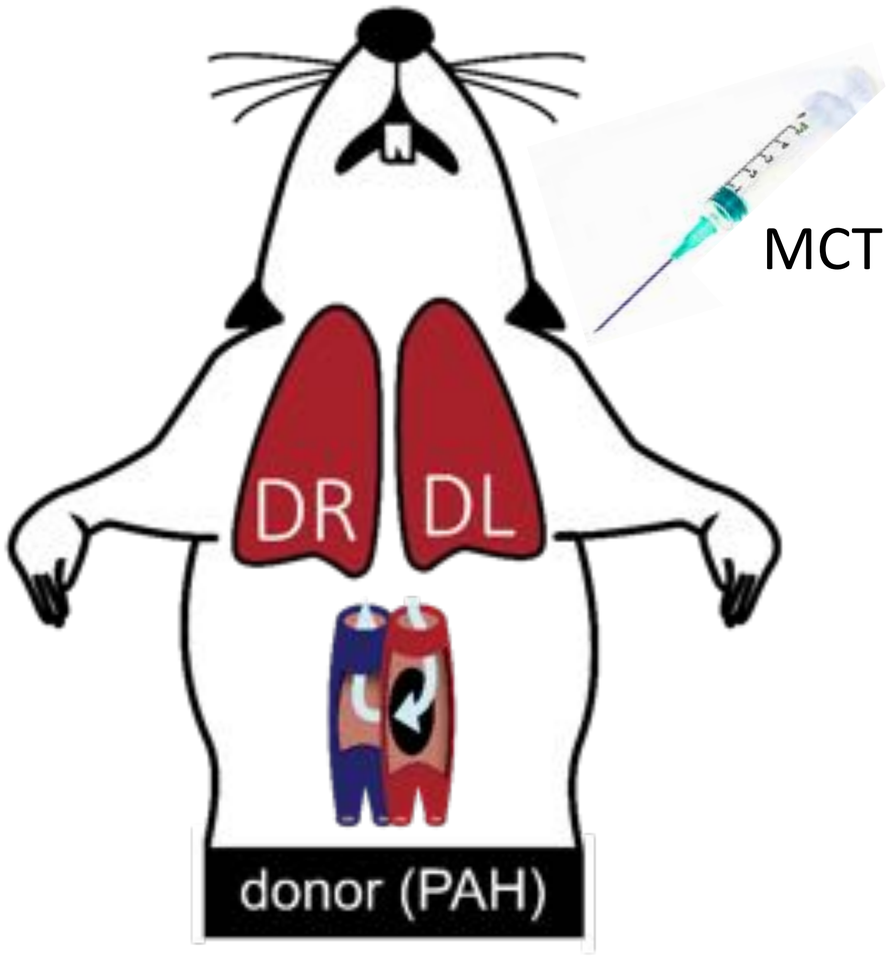


?

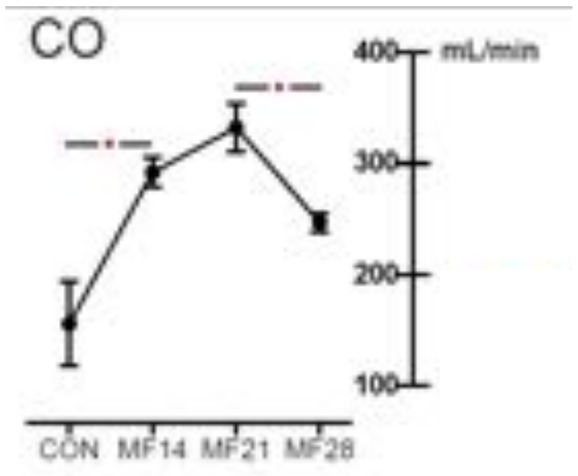
It is unknown if suppression of increased blood flow leads to reversibility of toxic induced or hypoxia induced PH

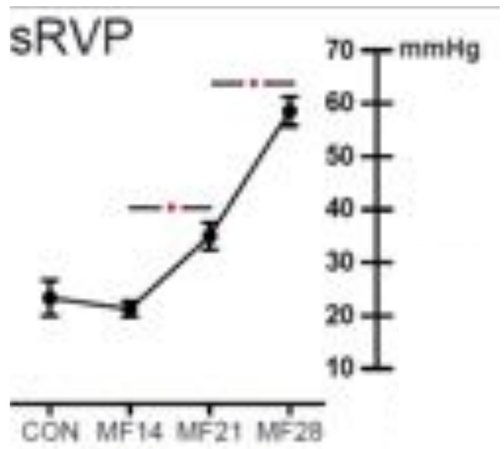
Dickinson MG, Bartelds B, Borgdorff MA, Berger RM. The role of disturbed blood flow in the development of pulmonary arterial hypertension: lessons from preclinical animal models. Am J Physiol Lung Cell Mol Physiol 2013;305:L1–L14.

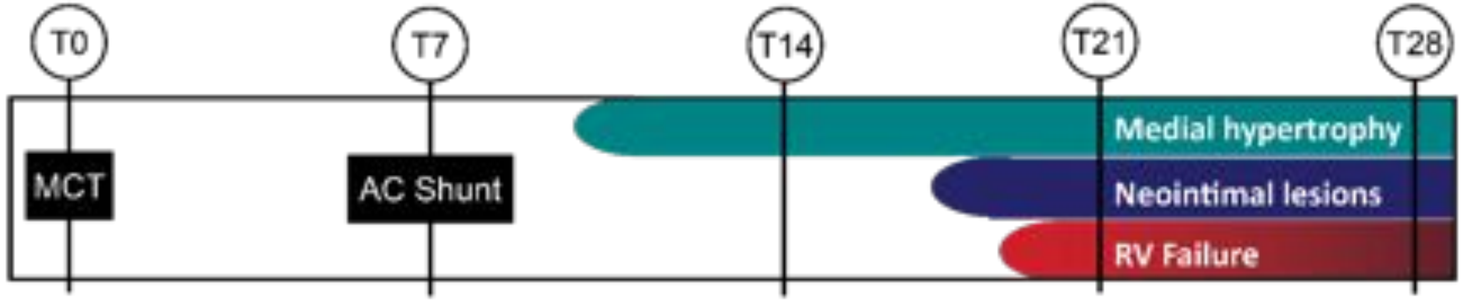
Progressive PAH

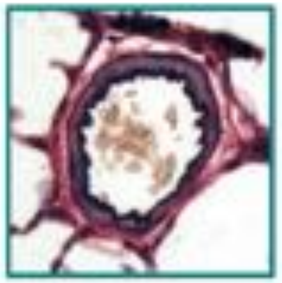


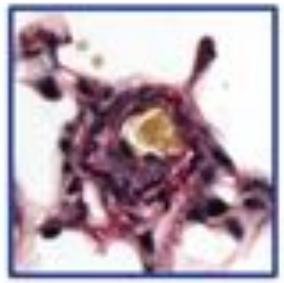
MCT







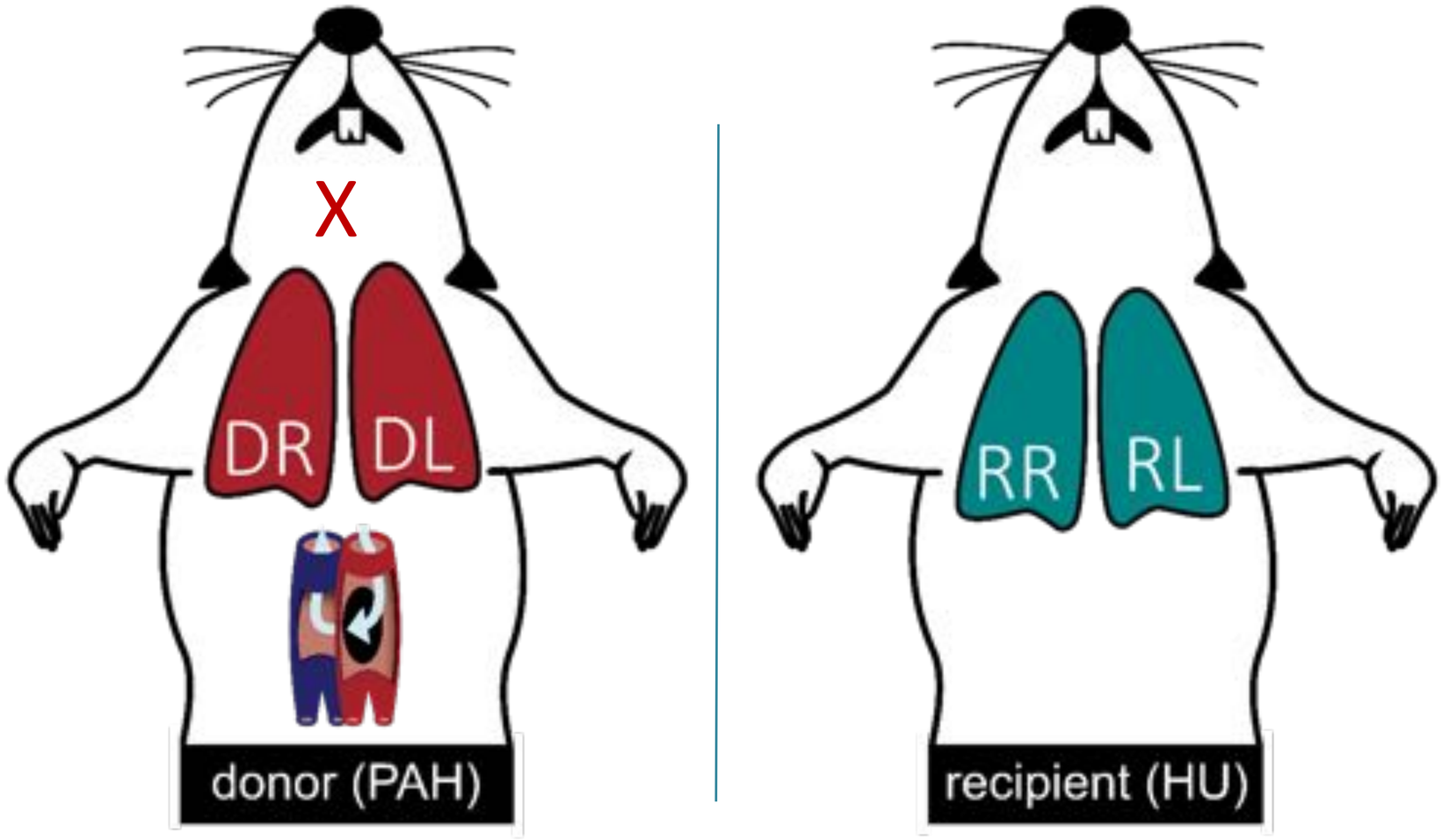




Diederik van der Feen

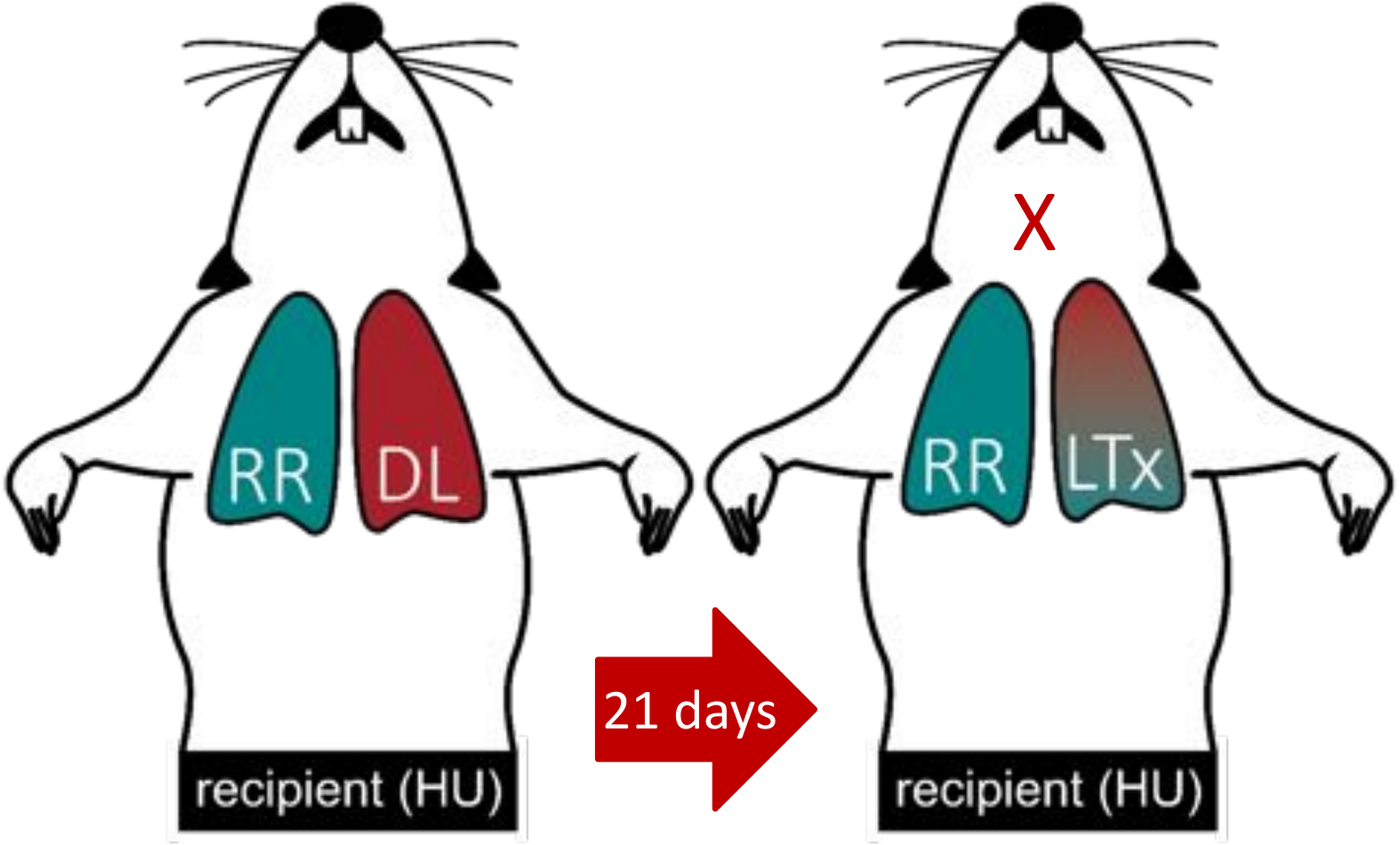
Haemodynamic Unloading by Lung Transplantation

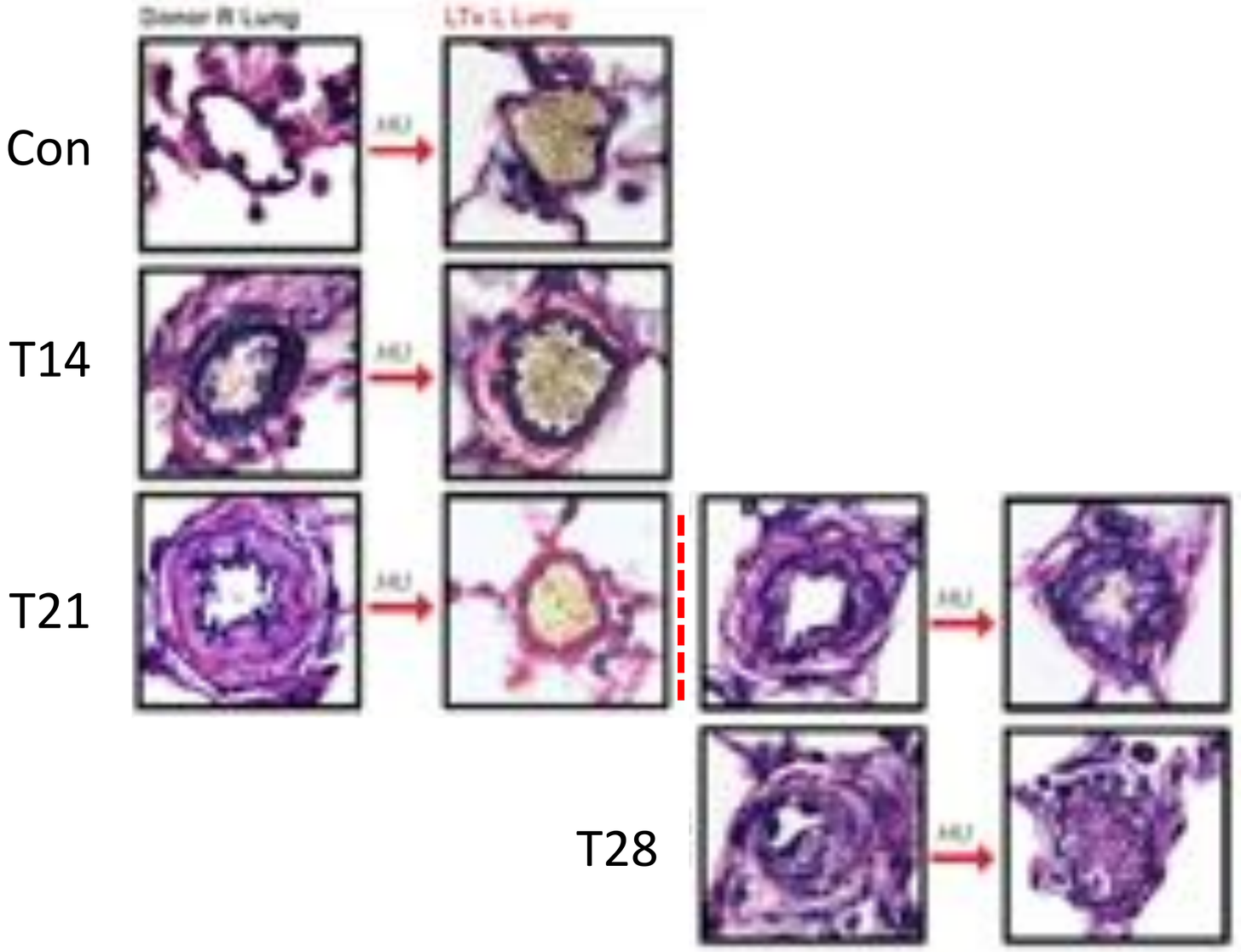
Sham
14 days
21 days
28 days

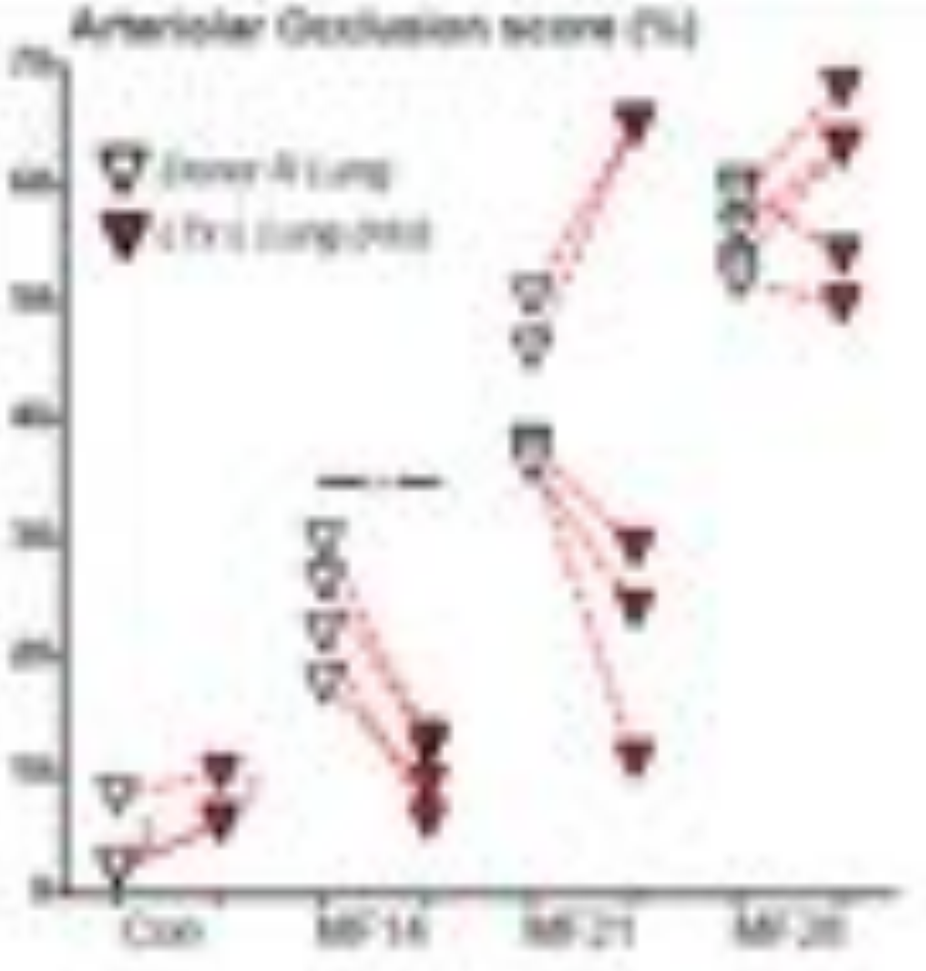


Diederik van der Feen

21 days of Haemodynamic Unloading







Diederik van der Feen

Mechanisms of disease progression in PAH-CHD

CHD an ideal ‘human model’ to study the mechanisms involved in early disease progression, disease reversal and transition to irreversibility :

1-The trigger is known : flow

2-The onset and magnitude of the trigger can be modulated

3-The trigger can be removed and timely removal potentiates disease reversal

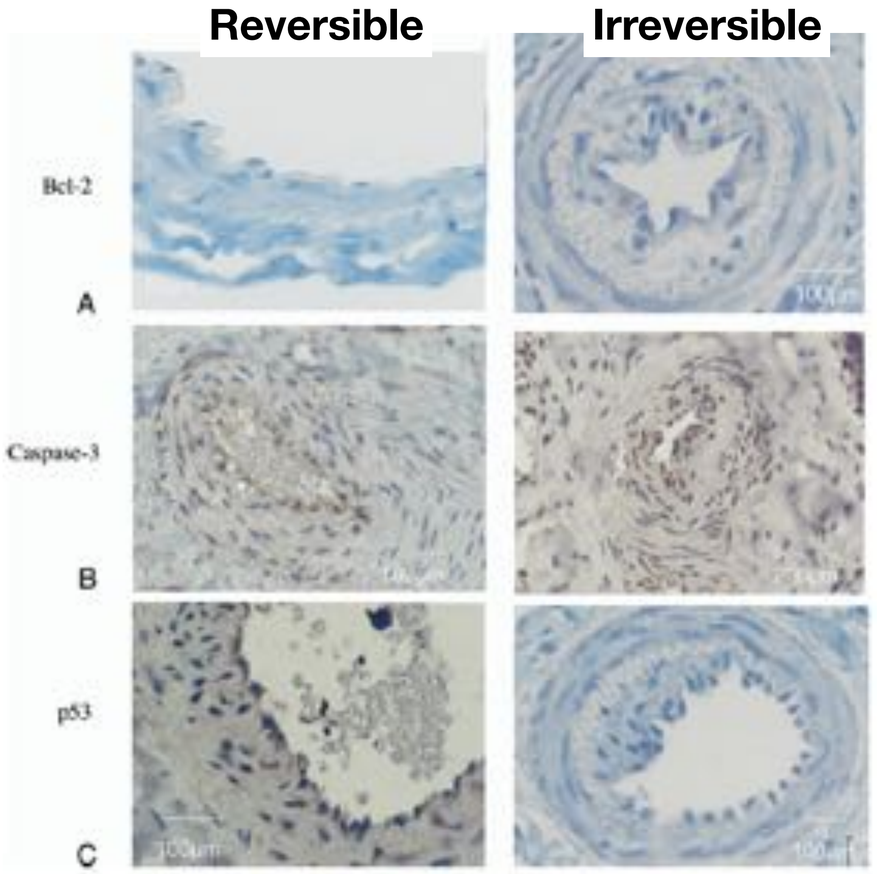
4-Persistence of the trigger leads to progressive PVD similar to other forms of PAH

5-The subgroup of patients that do not reverse despite trigger removal allows to identify conditions and mechanisms associated with irreversible disease

What could make PAH irreversible ?

1-Apoptosis and apoptosis resistance

Vascular immunostaining for markers of apoptosis in reversible and irreversible APAH-CHD

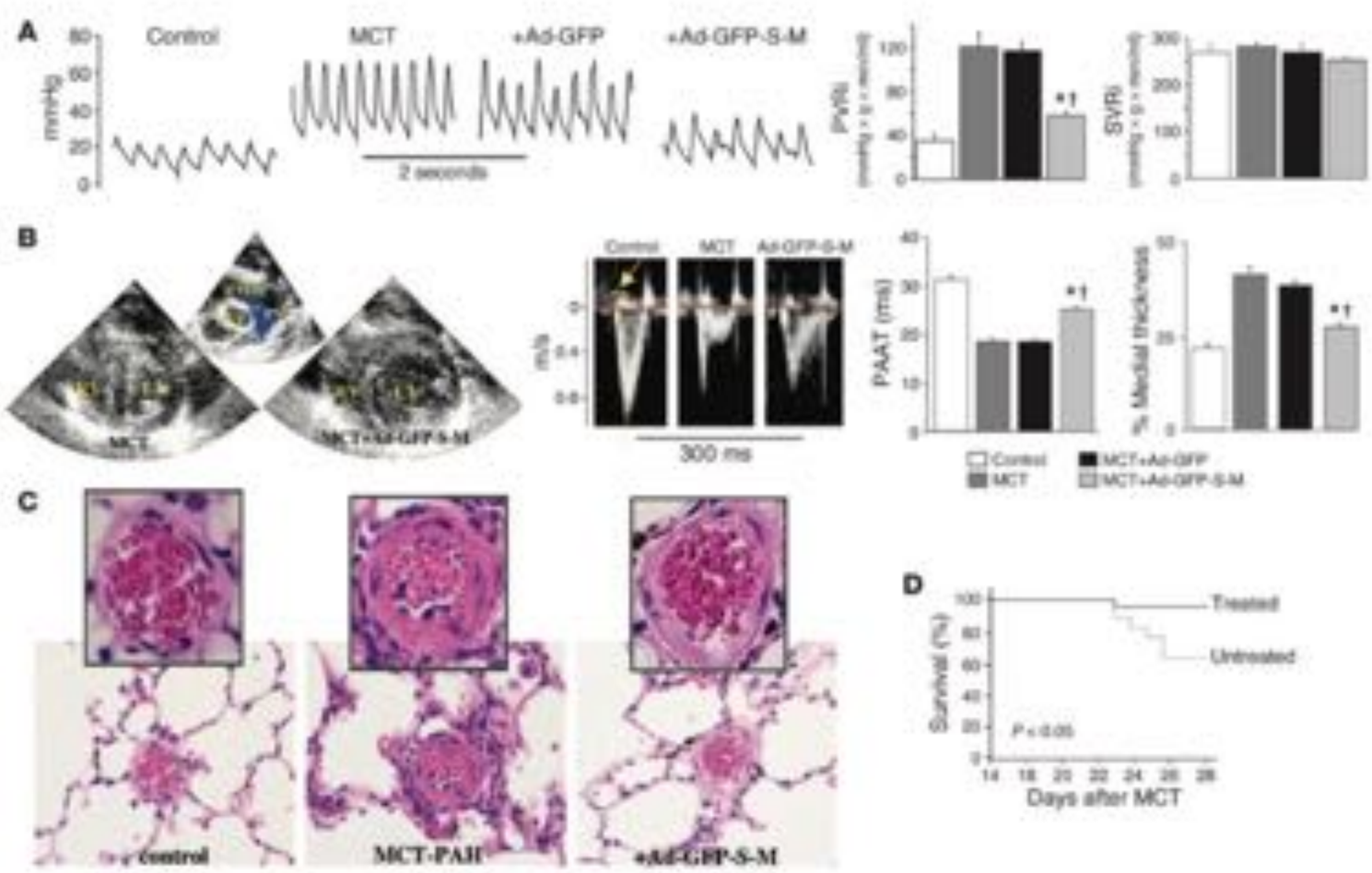


The antiapoptotic protein Bcl-2 is not expressed in reversible pulmonary hypertension (PHT), but by endothelial cells of severely damaged pulmonary arteries in irreversible PHT in all cases (A).
Endothelial cells of both groups expressed markers of apoptosis caspase-3 (B) and p53 (C).
The arrow indicates immunostaining in the endothelial layer.

Lévy M et al. JACC 2007;49:803-10

What could make PAH irreversible ?

1-Apoptosis and apoptosis resistance



Survivin, a marker for apoptosis-resistance, is expressed abundantly in patients with end-stage PAH and nearly absent in CHD without PAH

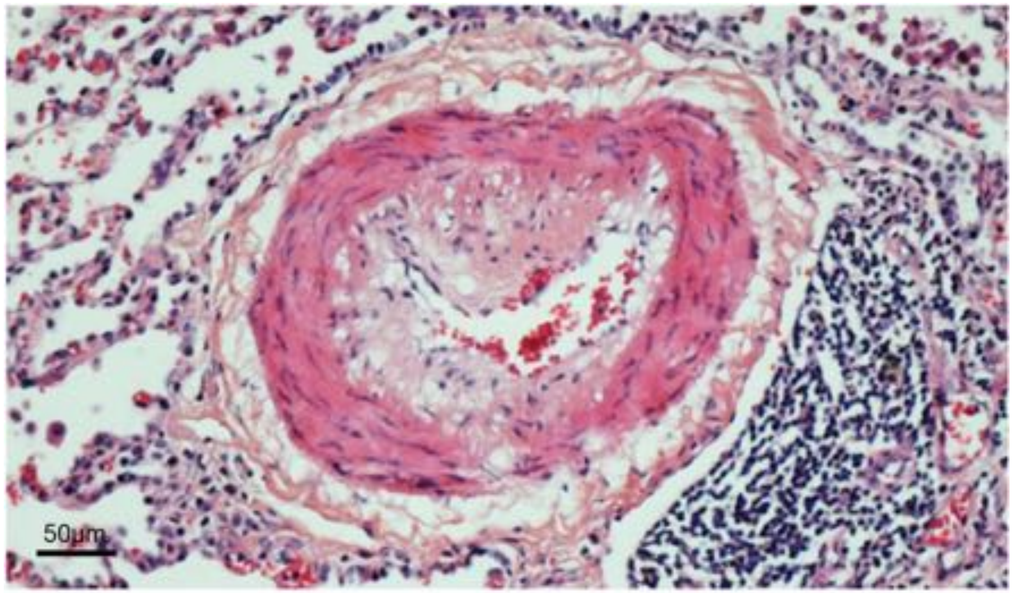
Gene therapy of rat MCT-PAH with Ad-GFP-Survivin-M improves hemodynamics, reduces remodeling of the resistance PAs, and prolongs survival

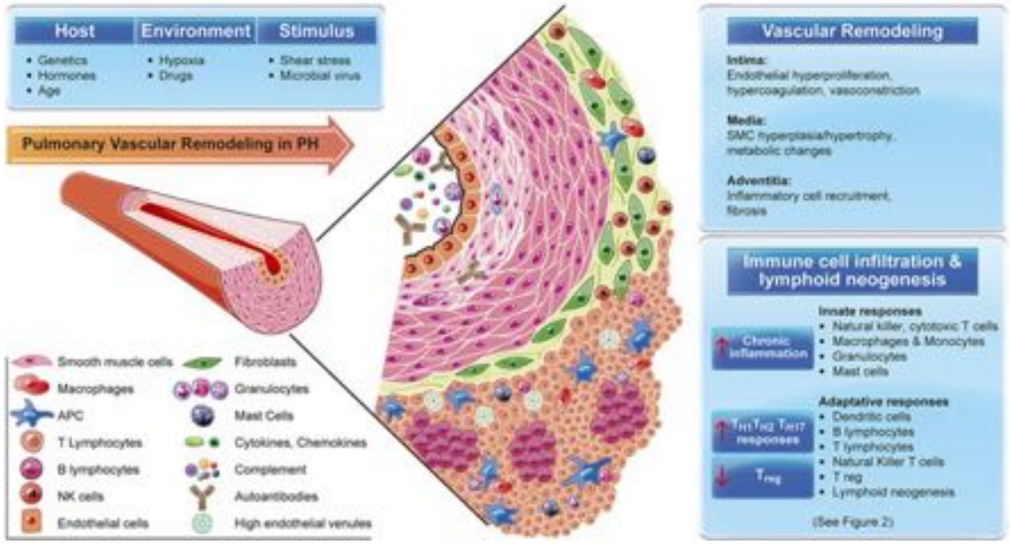
M3C

Mc Murtry MS et al. J Clin Invest 2005; 115:1479-91

What could make PAH irreversible ?

2-Inflammation





1. Inflammatory cytokines and macrophages are associated with disease progression in iPAH and PAH-CHD

2. Plexiform lesions can be observed in HIV-PAH, schistosomiasis PAH, scleroderma and lupus, some of these conditions being associated with reversible PAH

3. Animal models of the above conditions have proven reversibility of neointimal lesions with specific therapy

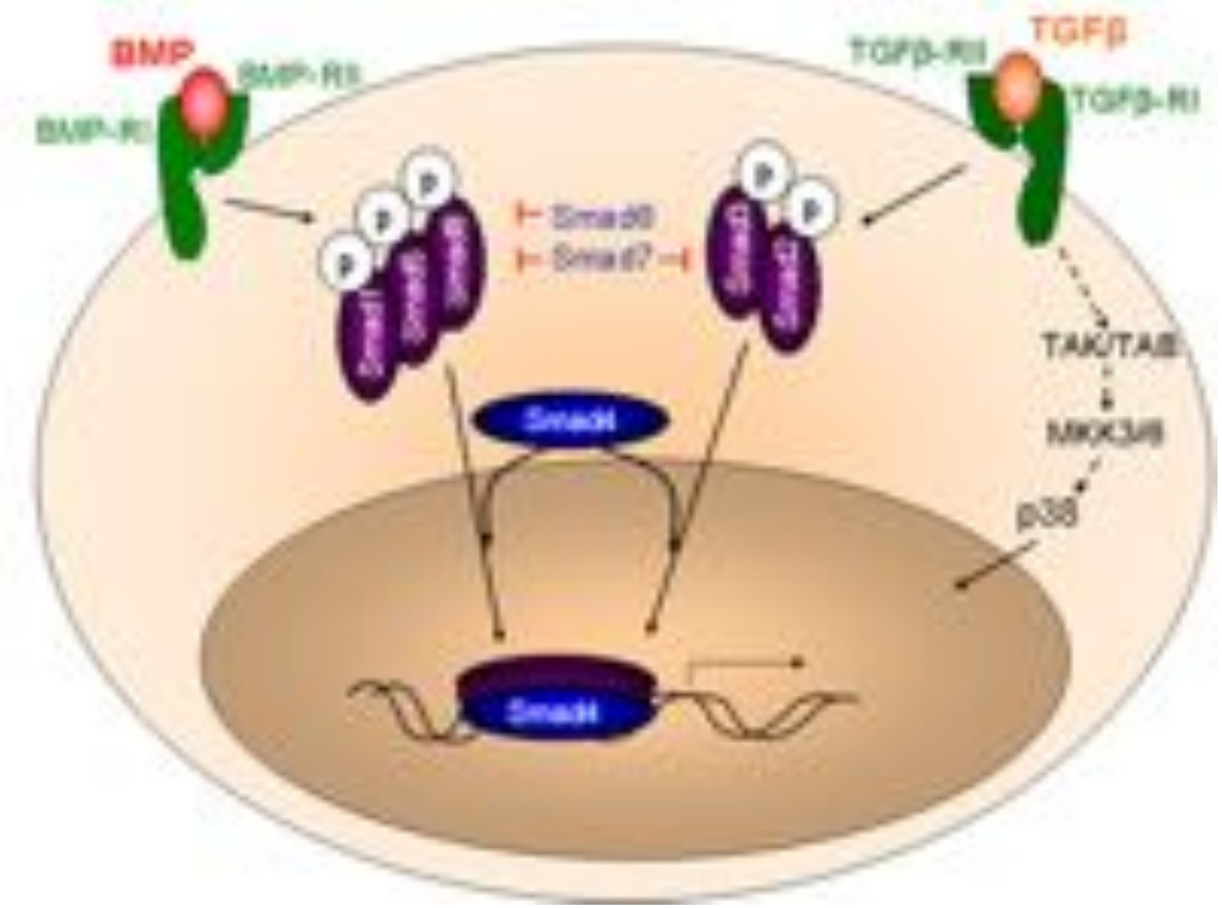
4. Anti-inflammatory drugs can be a valuable adjunct in the therapy of PAH-CHD

Rabinovitch M et al. Circulation

M3C

What could make PAH irreversible ?

2-Tgf-b/BMPR signalling imbalance



Proliferation, anti-apoptotic, inflammation

1. **BMP-9**, an endogenous stimulator of BMP-signalling, has shown to reverse medial hypertrophy in BMPR2-deficient mice and MCT rats, but also neointimal lesions in SuHx rats.

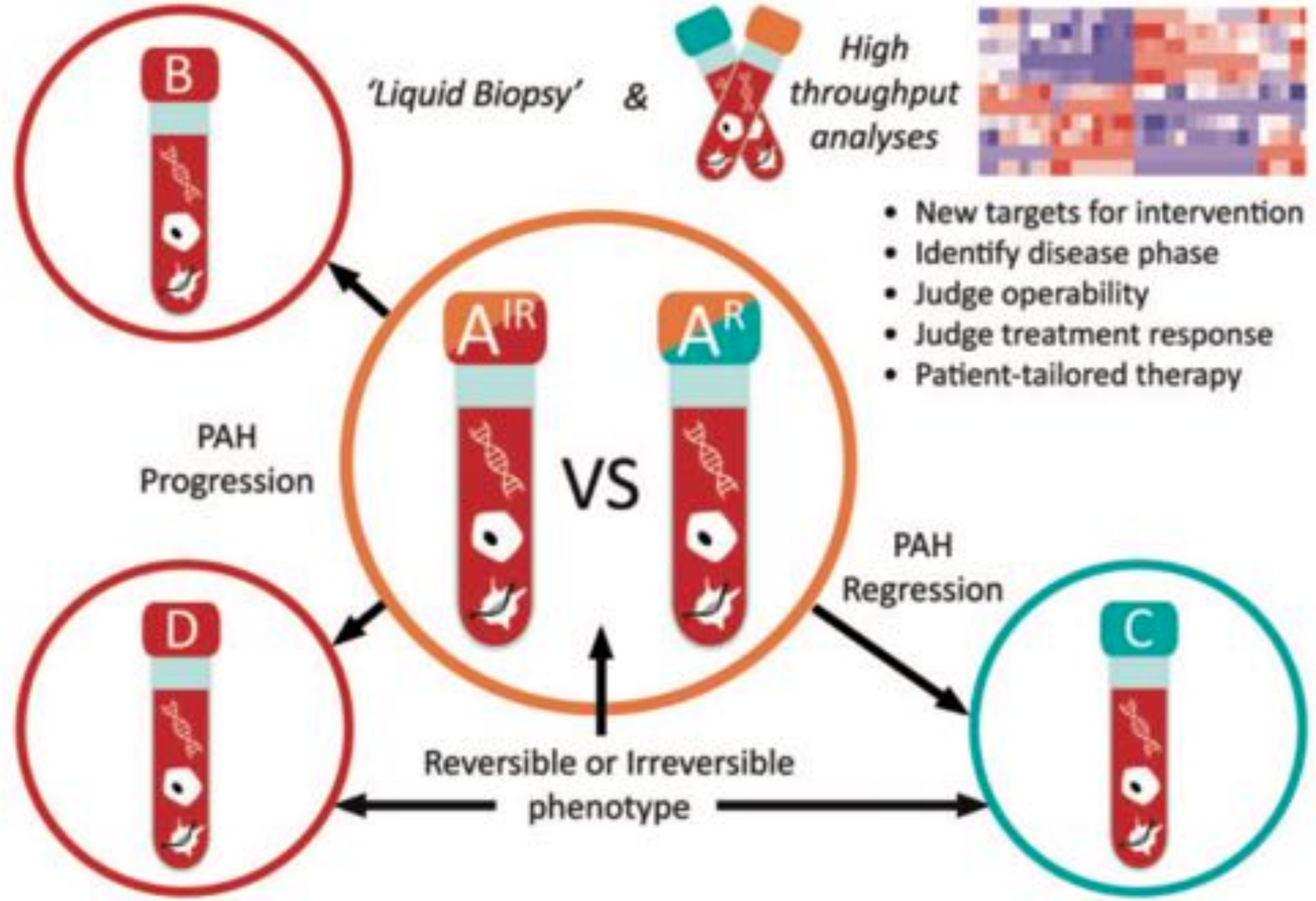
2. **FK506** (Tacrolimus) showed to (1) restore disturbed BMPR2-signalling and endothelial function in PAECs from IPAH patients, (2) prevent PAH progression in BMPR2-deficient mice, and (3) reverse established neointimal lesions in SuHx rats. Low-dose Tacrolimus was associated with relieved symptoms of right heart failure and improved WHO-FC3 IPAH patients.

3. **Elafin**, an endogenous serine protease inhibitor that enhances BMPR2 signalling, has been shown to reverse neointimal lesions in SuHx rats and reduce neointimal thickness of pulmonary arteries in cultured sections from lung explants of patients with PAH.

Spiekerkoetter EE. J Clin Invest 2013; 123:3600-13.
Long Lu L. Nat Med 2015;21:777-85.
Spiekerkoetter EE et al. Am J Respir Crit Care Med 2015;192:254-7.
Nickel NP et al. Am J Respir Crit Care Med 2015;191:1273-86.

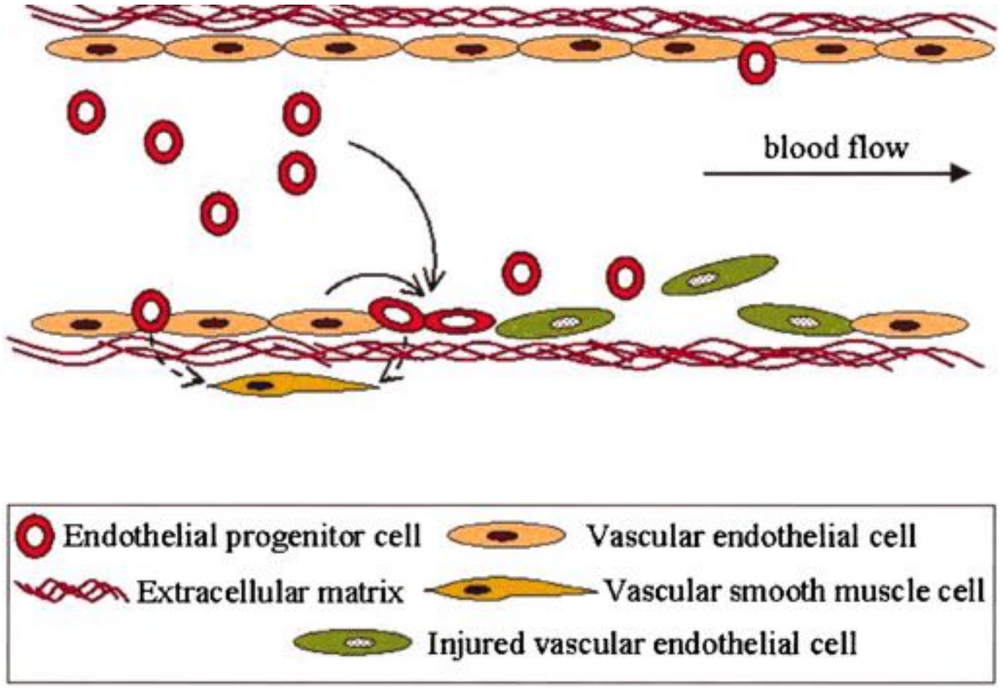
Comparison of human reversible to irreversible PAH-CHD

The liquid biopsy concept

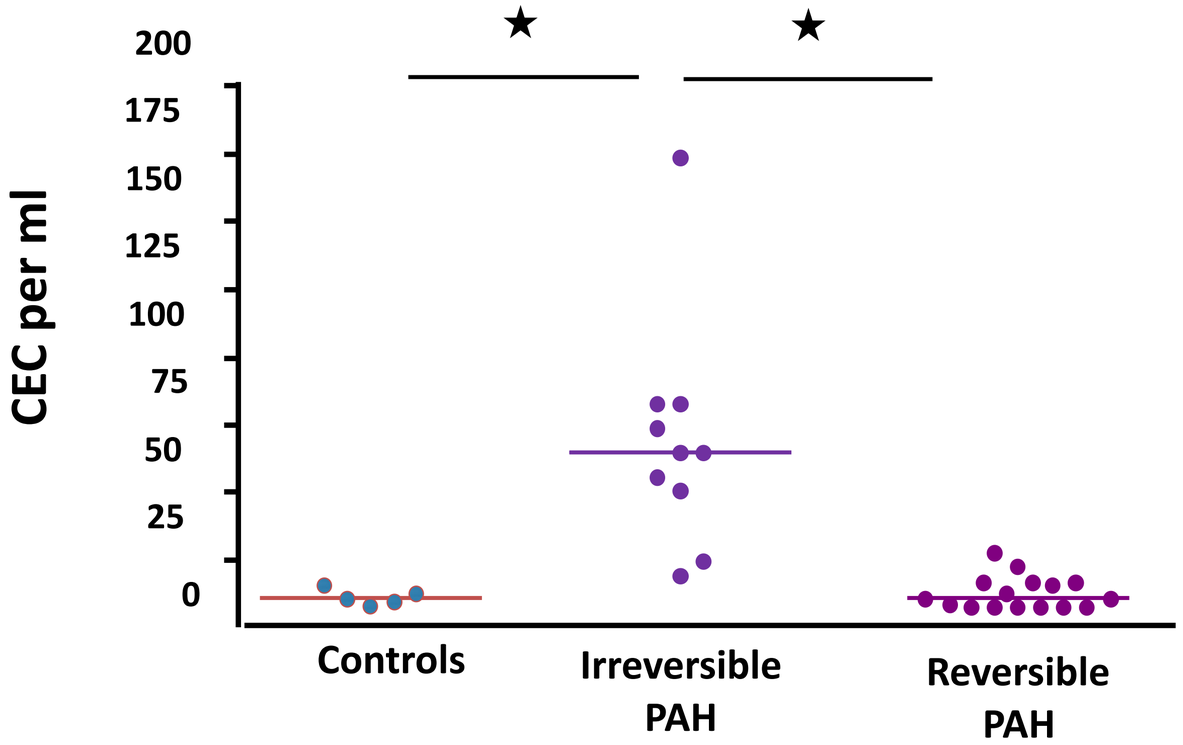


From: Pulmonary arterial hypertension in congenital heart disease: translational opportunities to study the reversibility of pulmonary vascular disease
Eur Heart J. 2017;38(26):2034-2041.

Circulating endothelial cells: A biomarker of irreversible PH secondary to CHD



CEC counts in peripheral venous blood of CHD patients with PH

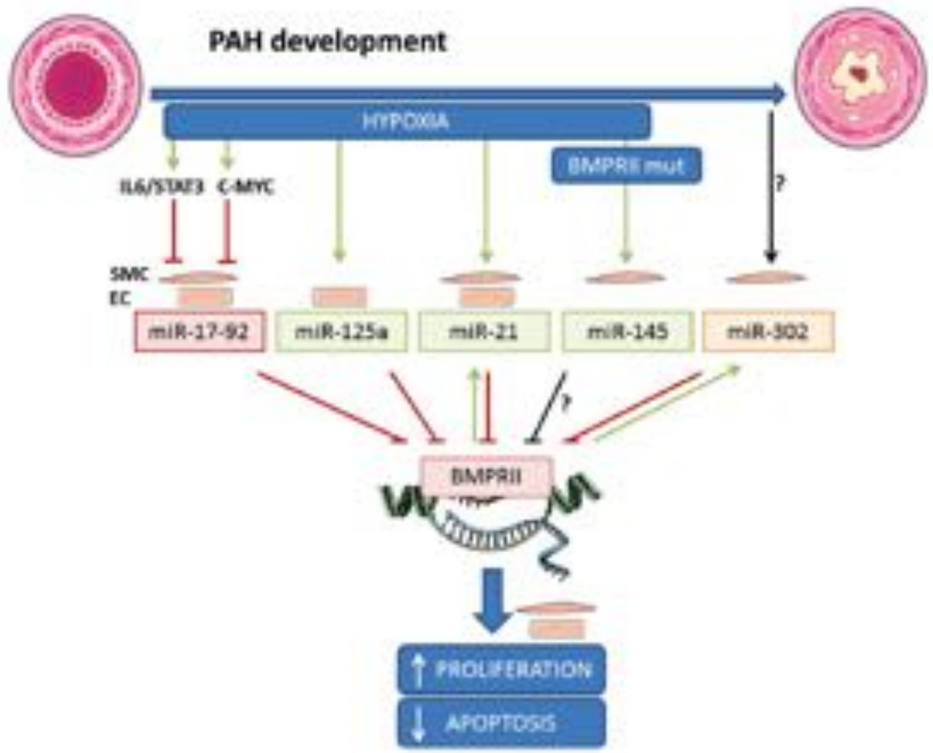


Smadja DM, et al. Circulation 2009; 119:374-81.

miRNAs : new players in the game

miRNAs are small non-coding RNAs that negatively, posttranscriptionally regulate the expression of target genes by interfering with both the stability of the target transcript as well as its translation

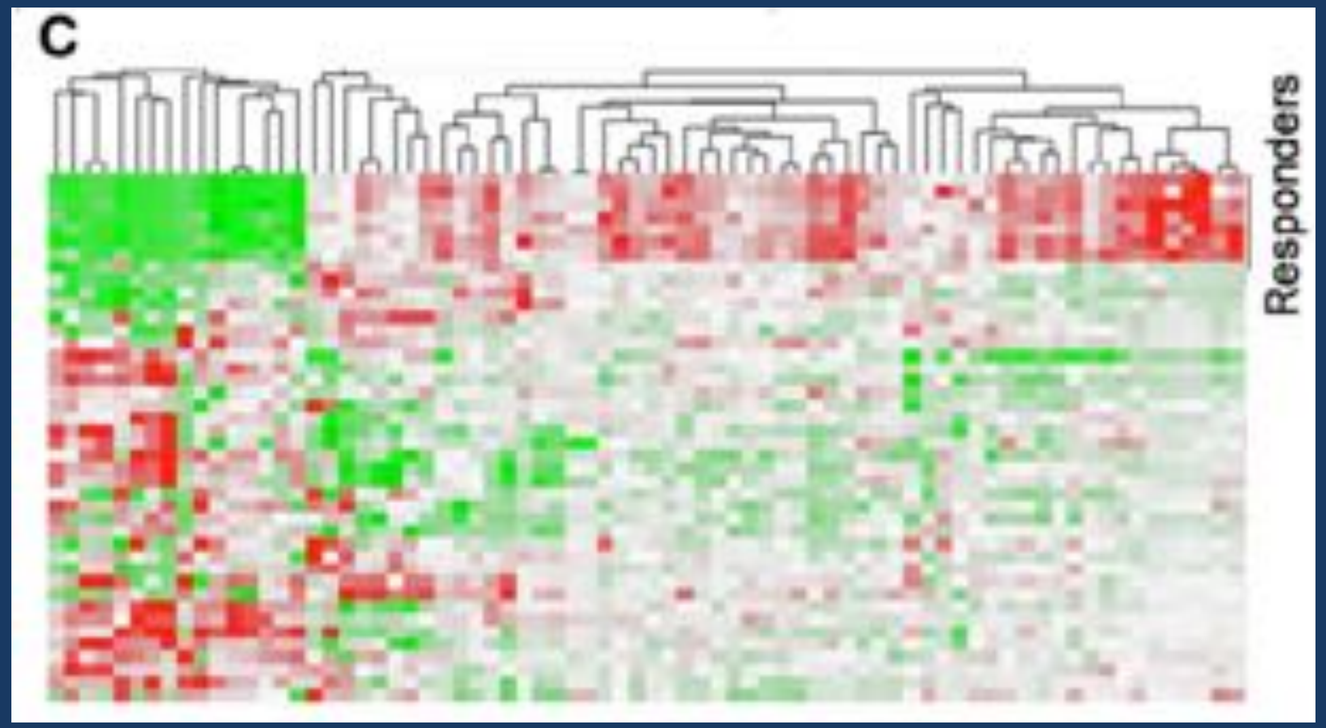
Fig. 4 miRNAs targeting BMPRII in the vascular wall. The illustration shows hypoxia and BMPRII mutations as regulators of miRNAs expression in endothelial or smooth muscle cells. These miRNAs negatively regulate BMPRII expression resulting in increased cell proliferation and impaired apoptosis. *Green arrows* indicate activation, *red arrows* represent inhibition, and black arrows correspond to unknown regulation. *EC* endothelial cells, *IL* interleukin, *miR* micro RNA, *mut* mutant, *SMC* smooth muscle cell, *STAT* signal transducer and activator of transcription



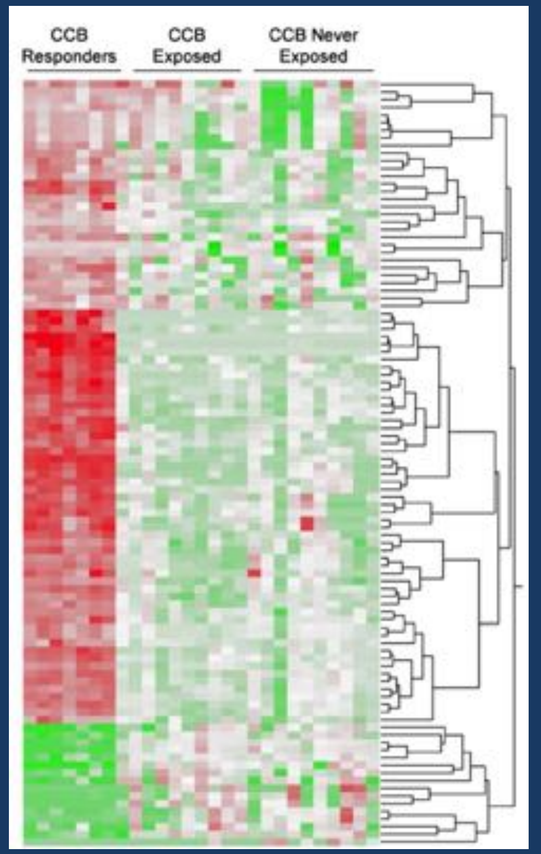
miR-145, miR-21 and the miR17/92 cluster, have been associated with the disrupted BMPRII pathway in PAH and can explain the incomplete penetrance of BMPR2 mutations

Heterogeneity in response to treatment of PAH and genetics

Peripheral Blood Gene Expression Levels Between Vasodilator-Nonresponsive and Responsive Pulmonary Arterial Hypertension Patients



Heat map of differentially expressed genes in AVTr vs non AVTr



Heat map of differentially expressed genes in AVTr vs non AVTr
Non AVTr exposed or not to CCB

Hemnes AR et al. Circulation. 2015;131:401-409.

How to identify new genes in pediatric PAH and PAH-CHD ?

Exome sequencing and pathway analysis

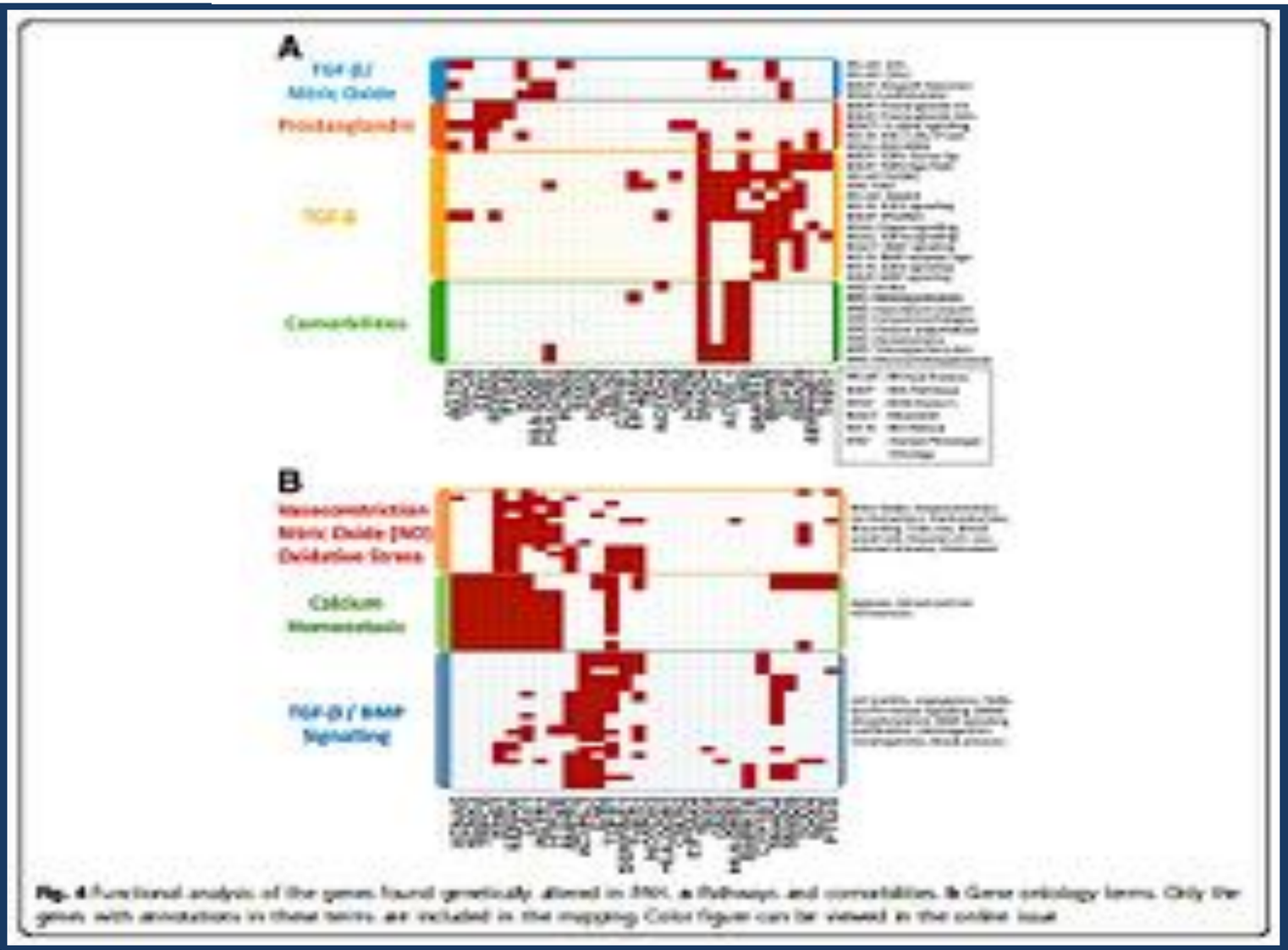


Fig. 4 Functional analysis of the genes found genetically altered in PAH. a Pathways and comorbidities. b Gene ontology terms. Only the genes with annotations in these terms are included in the mapping. Color figure can be viewed in the online issue

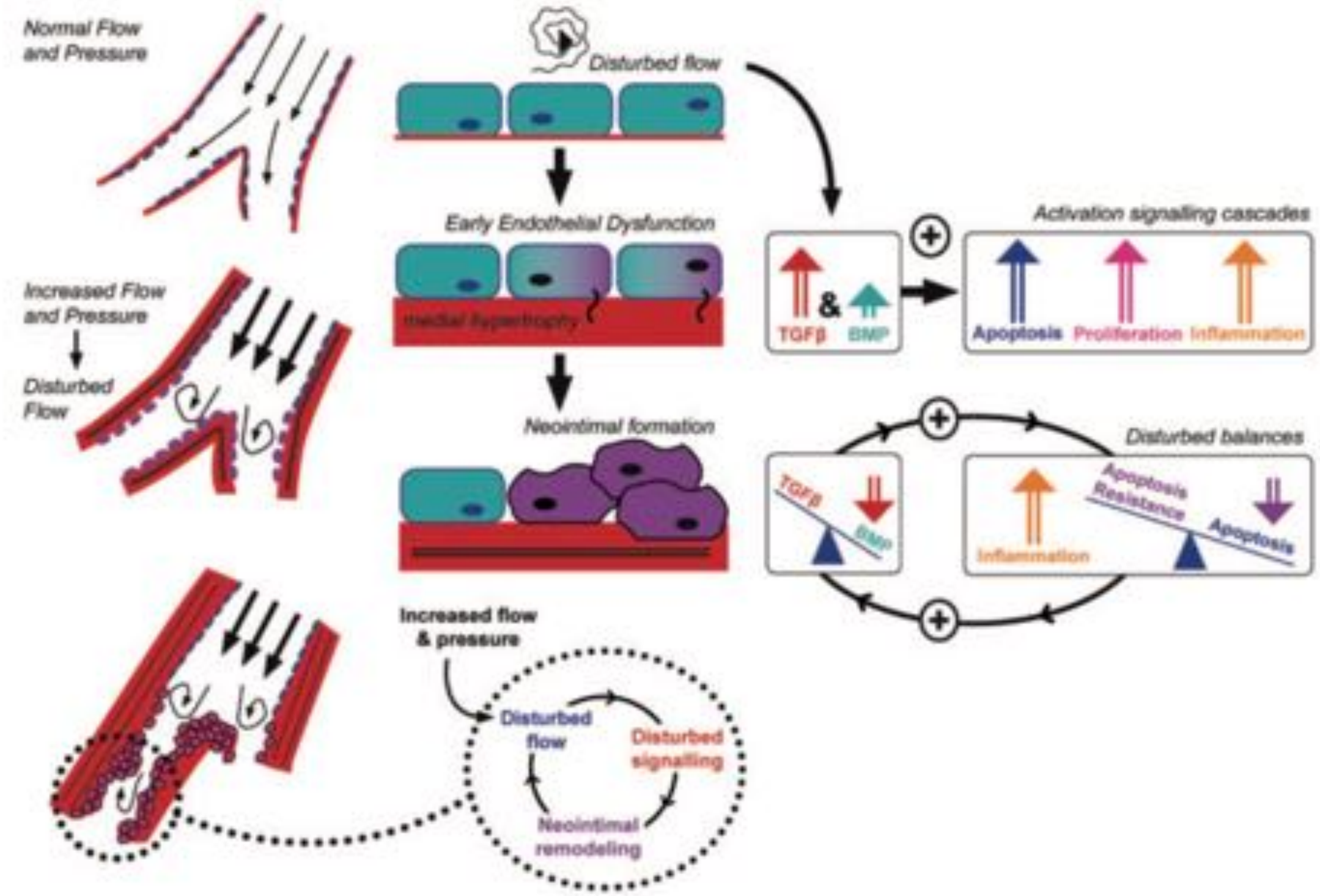
1- Opportunity for identifying key signaling pathways, both as potential drug targets and as biomarkers for patient selection

2- Mutations in genes with strong biological relevance to PH can contribute to PH susceptibility in at risk conditions.

3- PH might not be related to a global change in genomic content, but rather associated with altered dose of specific genes in crucial pathways (variants).

Garcia-Rivals et al. BMC Medical Genetics 2017

Summary **Flow PH in CHD**

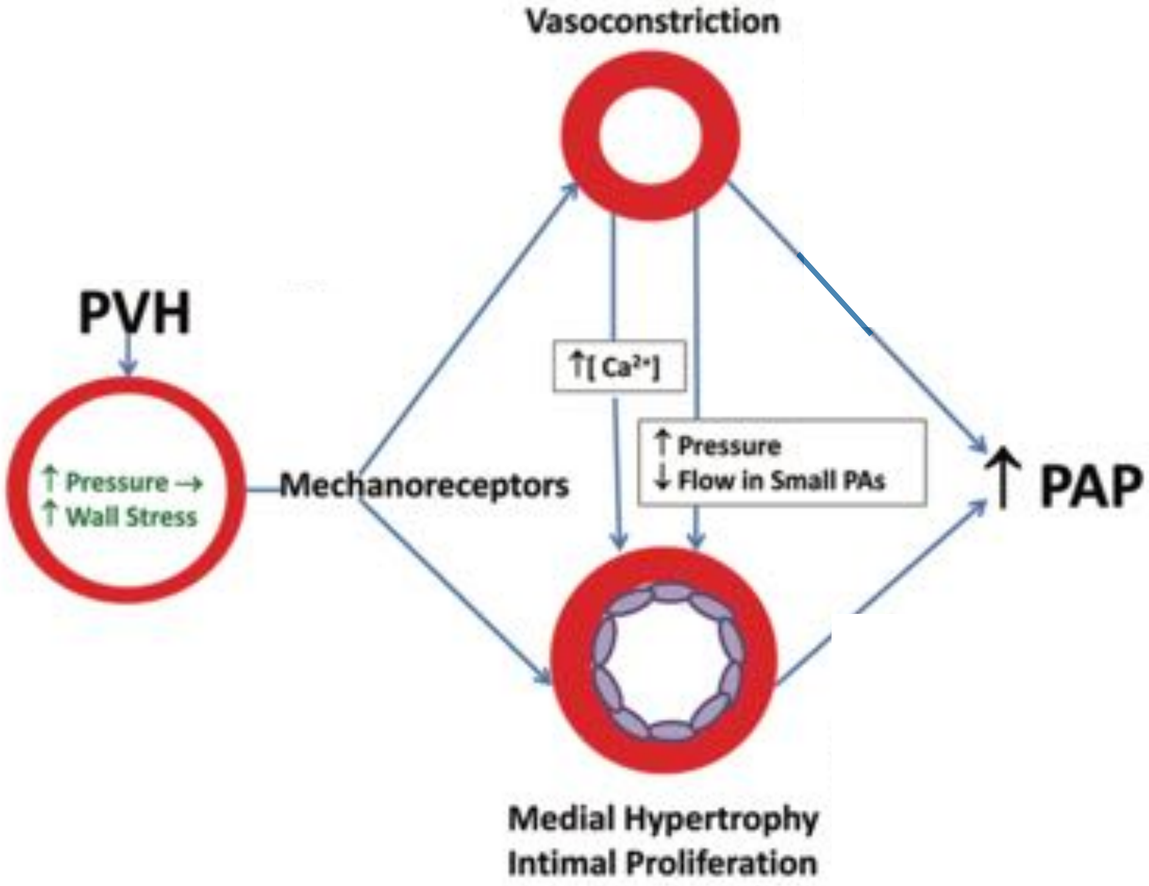


Pulmonary hypertension caused by pulmonary venous hypertension

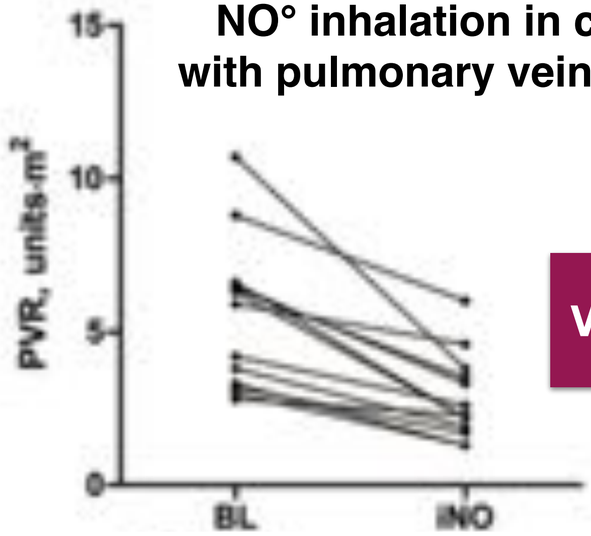
1. The effect of pulmonary venous hypertension (PVH) on the pulmonary circulation is extraordinarily variable, ranging from no impact on pulmonary vascular resistance (PVR) to a marked increase

2. PVH-PH is one of the very few “models” of increased PVR in which removal of the stimulus is often possible, which is followed by substantial reduction of PVR.

How pulmonary venous hypertension (PVH) causes increased pulmonary vascular resistance (PVR)

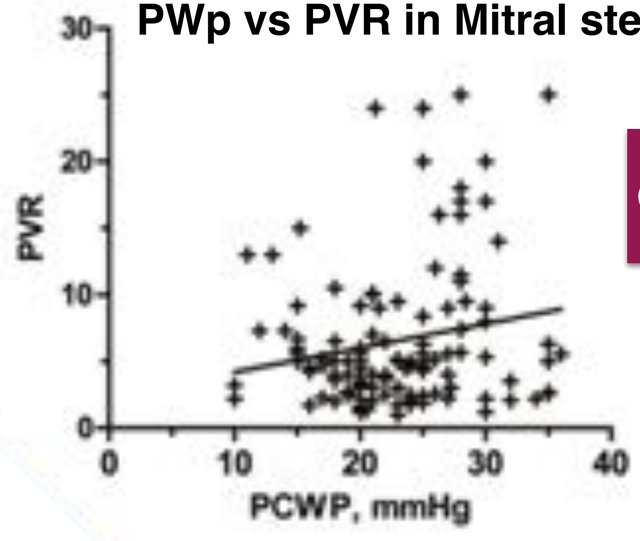


NO° inhalation in children with pulmonary vein stenosis



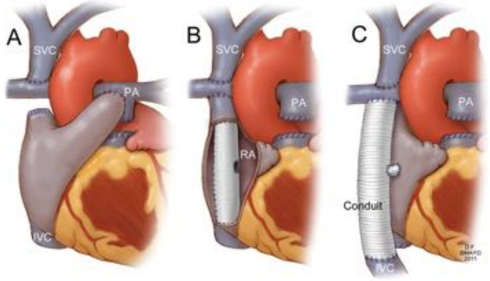
Vasoconstriction

PWp vs PVR in Mitral stenosis



Out-of-proportion

Wood P, Besterman E, Towers MK, McIlroy MB. The effect of acetylcholine on pulmonary vascular resistance and left atrial pressure in mitral stenosis. Br Heart J 1957;19: 279–286.
Atz AM, Adatia I, Jonas RA, et al. Inhaled nitric oxide in children with pulmonary hypertension and congenital mitral stenosis. Am J Cardiol 1996;77:316–319.



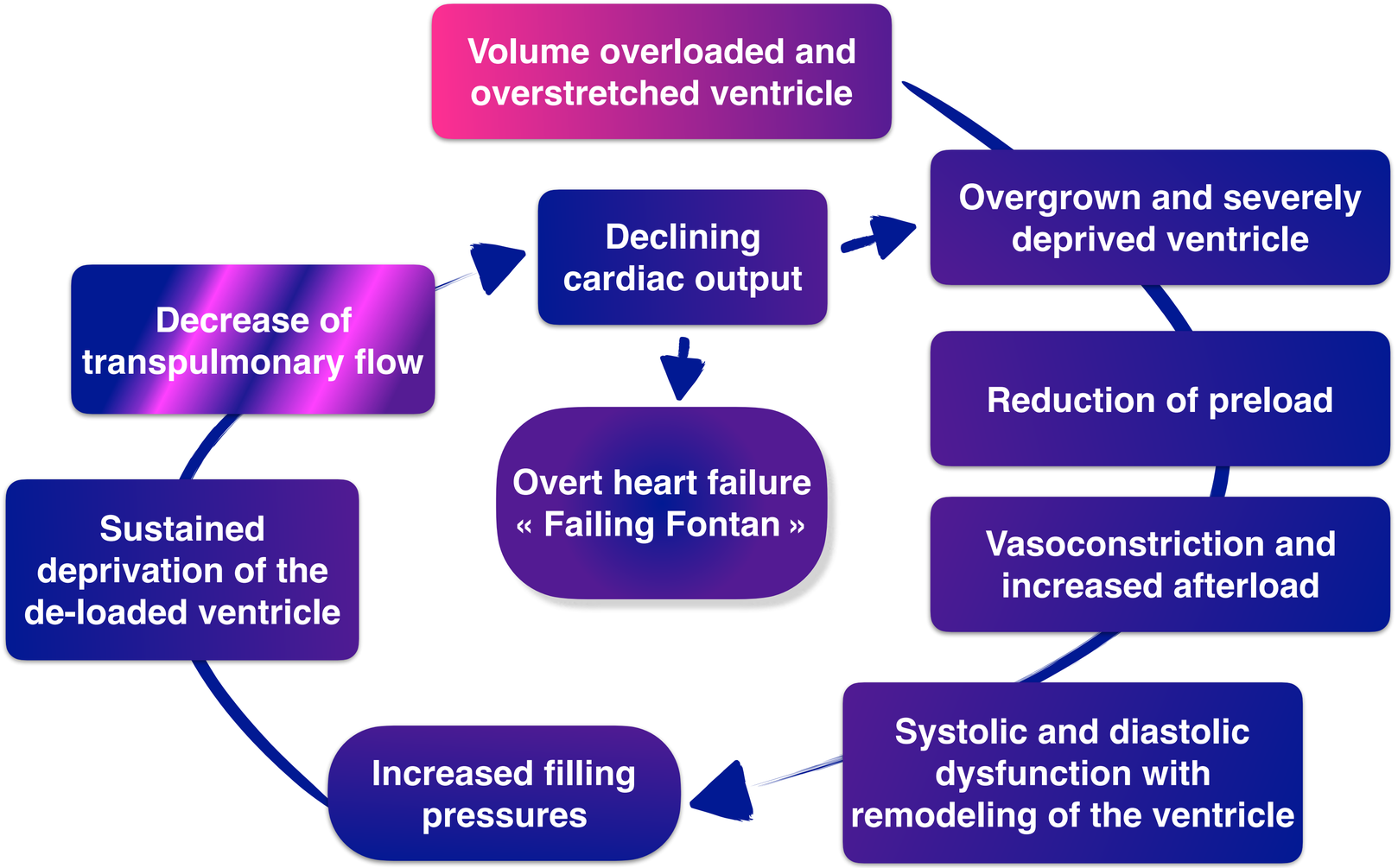
Increased PVR in the Fontan circulation

Pulsatile flow is important for shear stress-mediated release of endothelium-derived nitric oxide and for the lowering of the PVR by the passive recruitment of capillaries

Fontan patients do not fulfil the hemodynamic characteristics for PH but have increased PVR and exhibit pulmonary vascular remodelling « resembling » that of other forms of PH

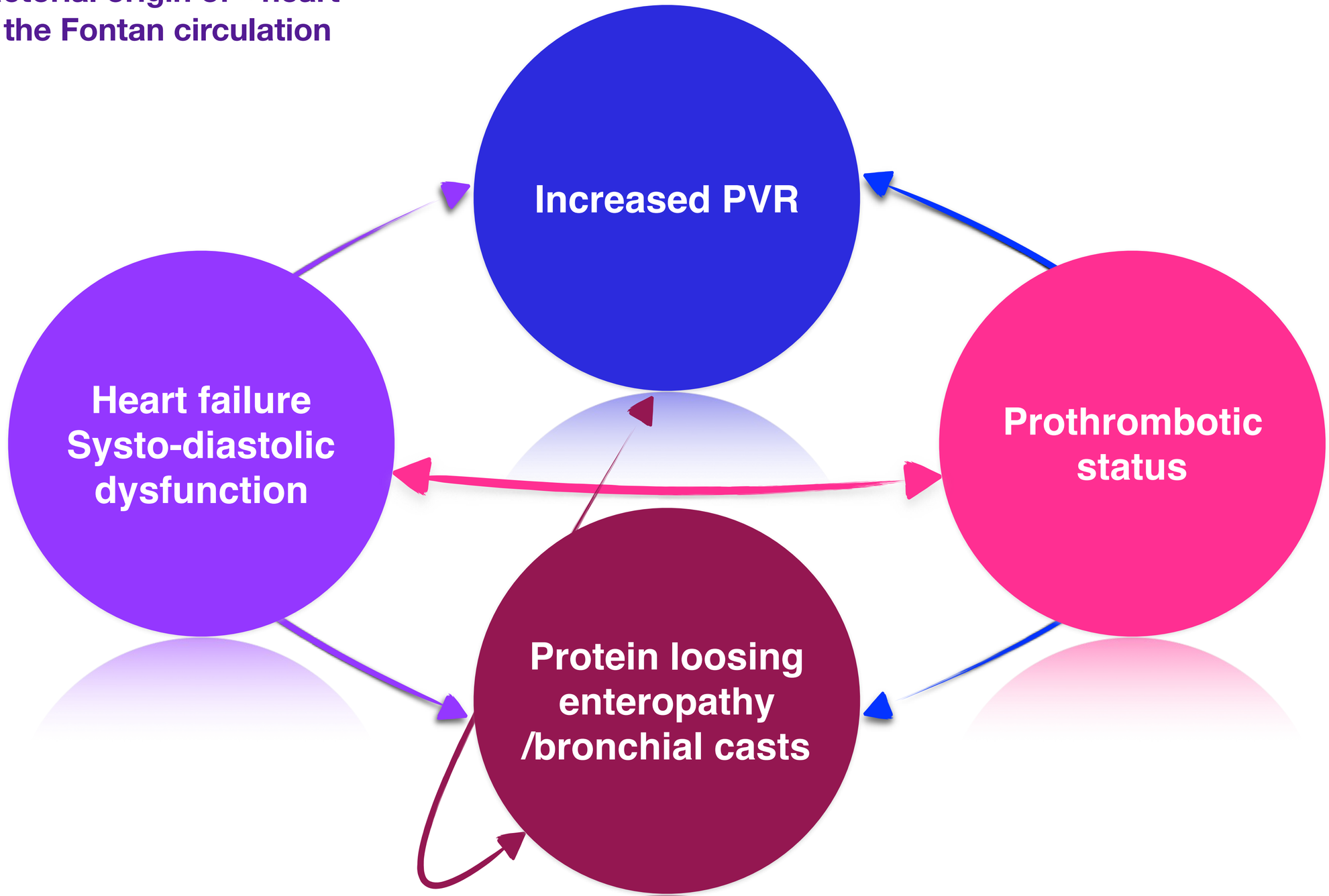
The Fontan circulation - a new portal system

The vicious circle to failing Fontan



Adapted from Marc Gewillig-Heart 2016

The multifactorial origin of « heart failure » in the Fontan circulation



Endothelial function in TCPC

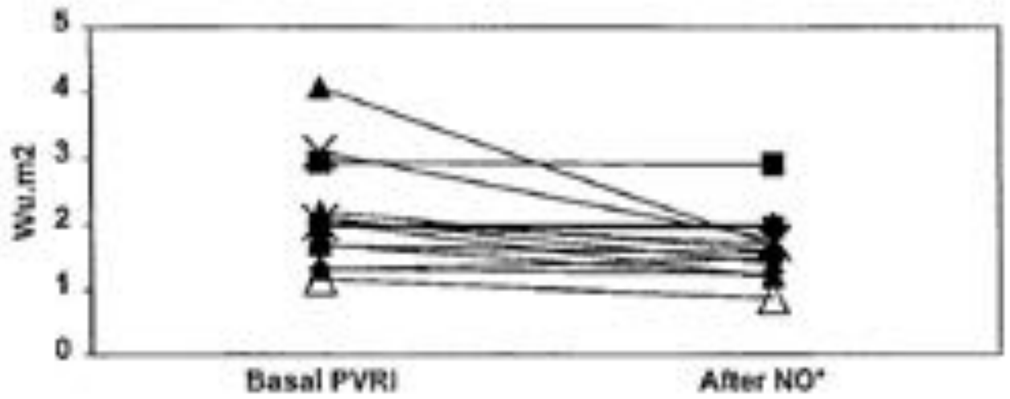


Figure 4. Effect of exogenous NO on PVRI late after Fontan operation. NO caused a significant drop of mean PVRI in the study group (*P=0.016).

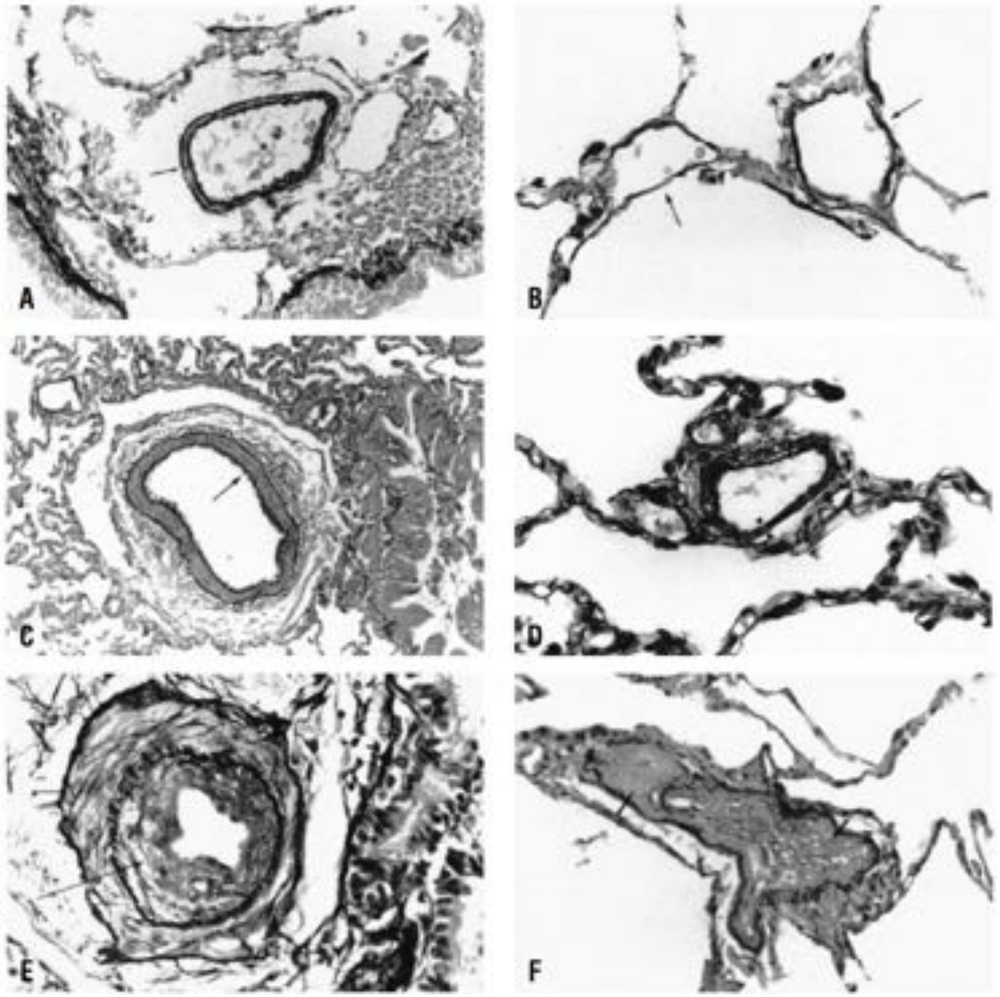
- Fontan patients have elevated PVRi
- Patients in NYHA 1 have a significantly lower mean PVRI (1.72+-0.38 WU.m2) compared with patients in NYHA 2 and 3 (2.82+/-0.88) (P=0.05)
- Significant drop in PVRi with NO°

Pulmonary endothelial dysfunction is related, at least in some part, to lack of pulsatility in the pulmonary circulation because of altered flow characteristics after Fontan operation

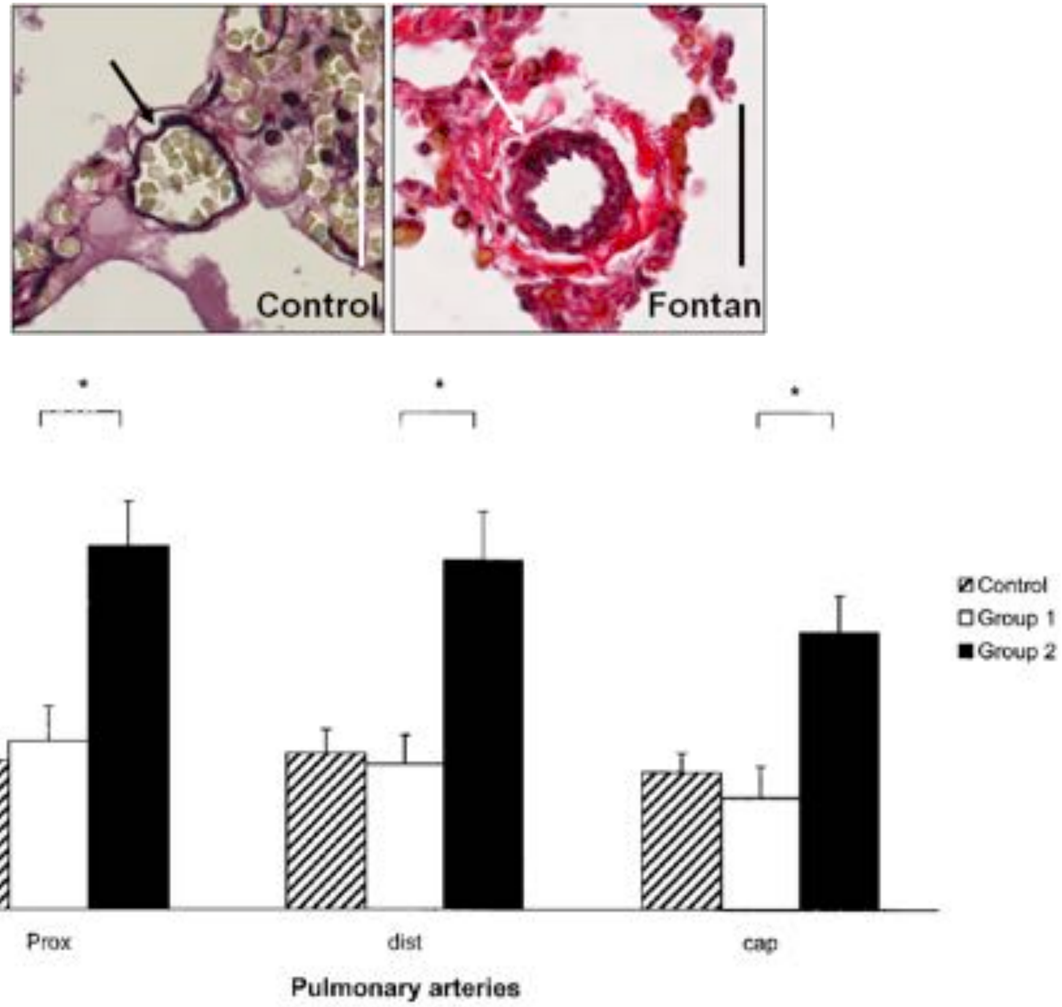
M3C

Khambadkone S et al. Circulation 2003;107:3204-8
Henaine R et al. J Thorac Cardiovasc Surg 2013;146:522-9

Vascular remodelling in TCPC



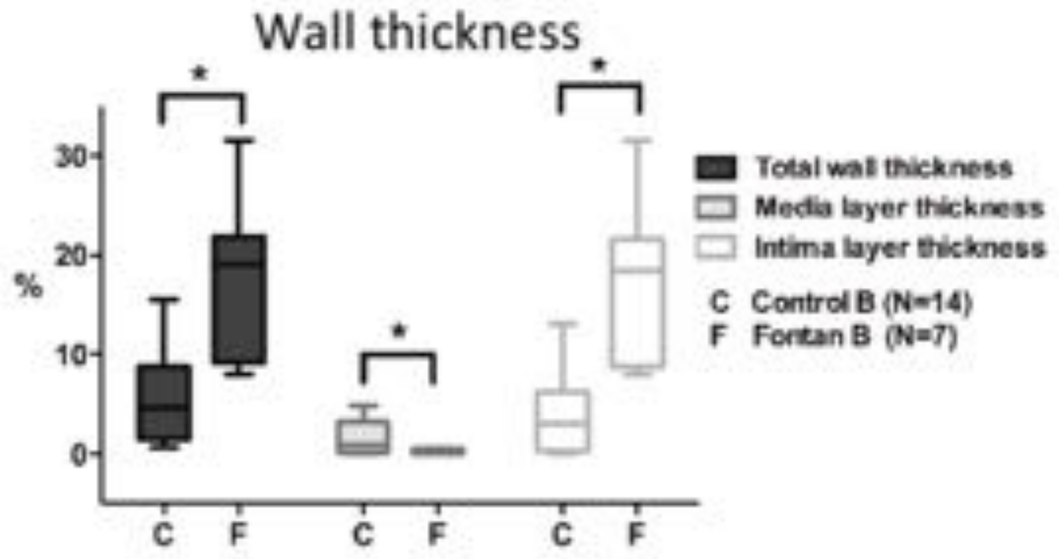
Remodeling of pulmonary arteries is present in half of patients with favorable hemodynamic at surgery and predicts outcome

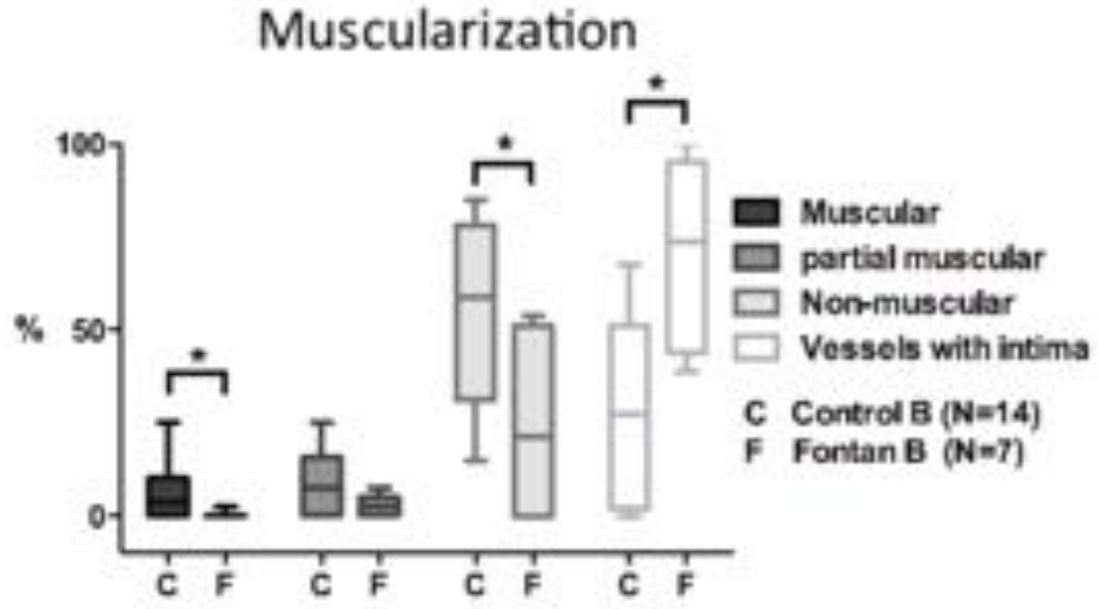


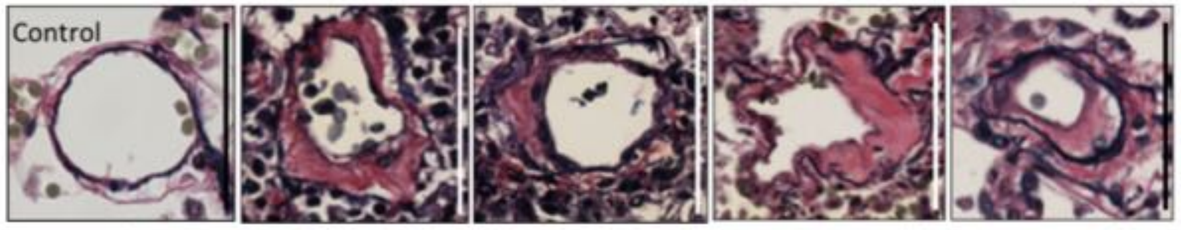
eNOS and ET1 expression is increased from baseline in « failed » Fontan procedures

Juaneda E, Hawaorth S. Br Heart J 1984
Levy M et al. J Thorac Cardiovas Surg 2002;123:263
Levy M et al. J Thorac Cardiovasc Surg 2003;125:1083

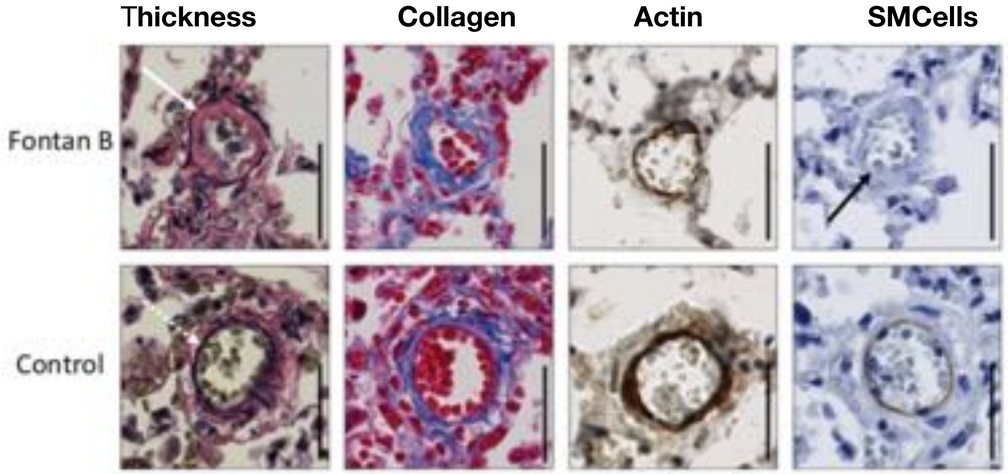
The unusual remodelling of intra-acinar pulmonary vessels in TCPC







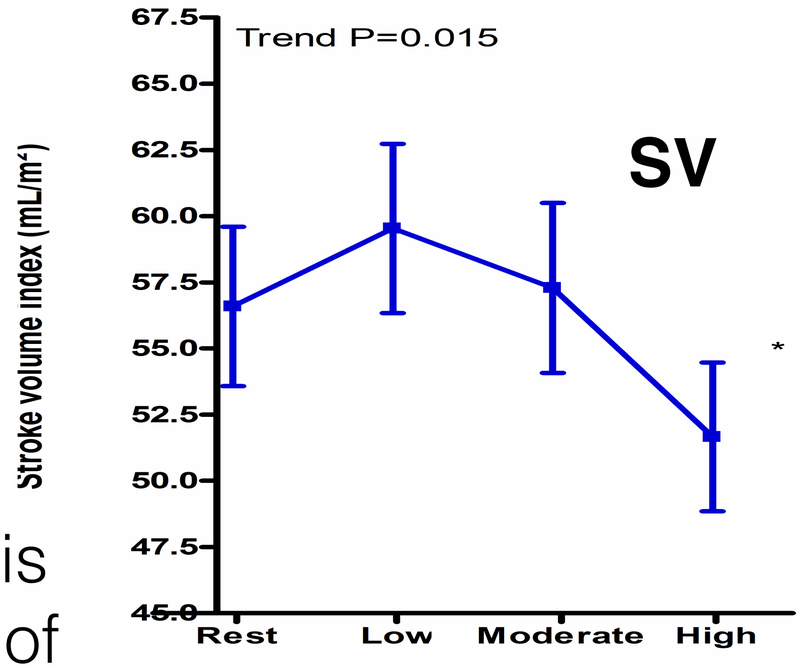
eccentric acellular intima fibrosis in the intra-acinar pulmonary vessels

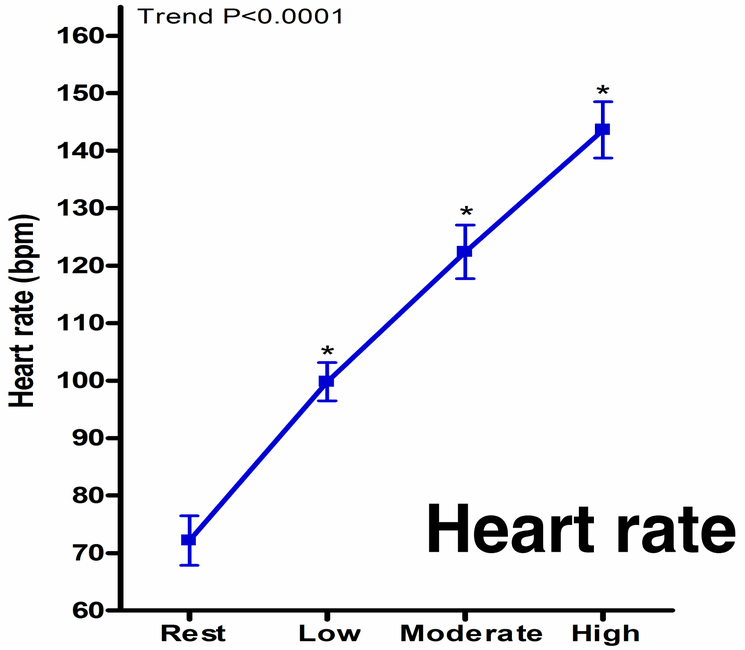


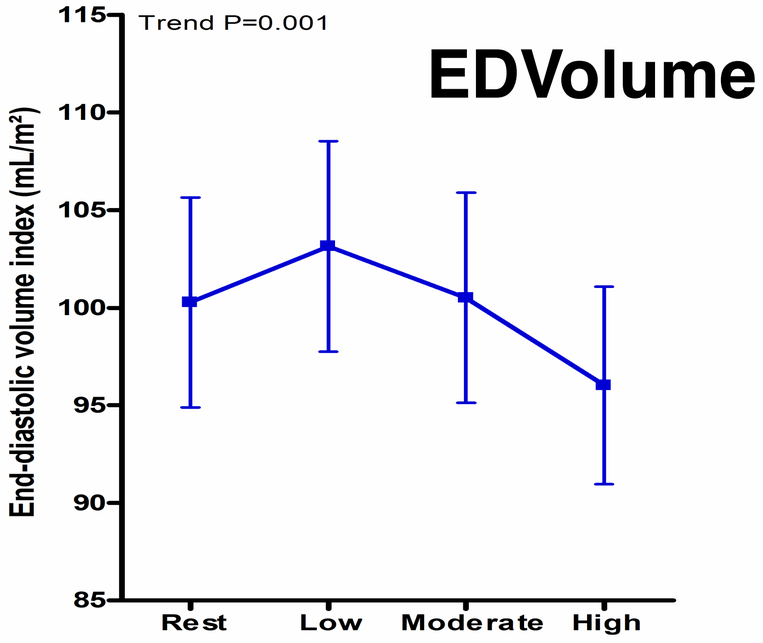
Ridderbos FJS et al.

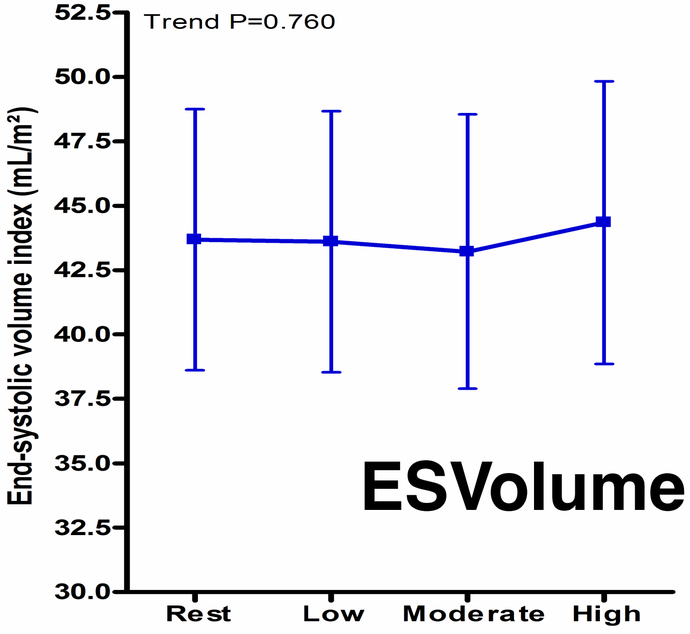
Fontan Ergo MRI

Stroke volume decrease is related to impaired filling of the single ventricle at exercise









Post-transplantation increased PVR in TCPC

Post-transplantation PVR is elevated (2.0 Wood units · m2) in the majority of survivors past initial hospitalization (mean 3.3+/-1.7 Wood units · m2).

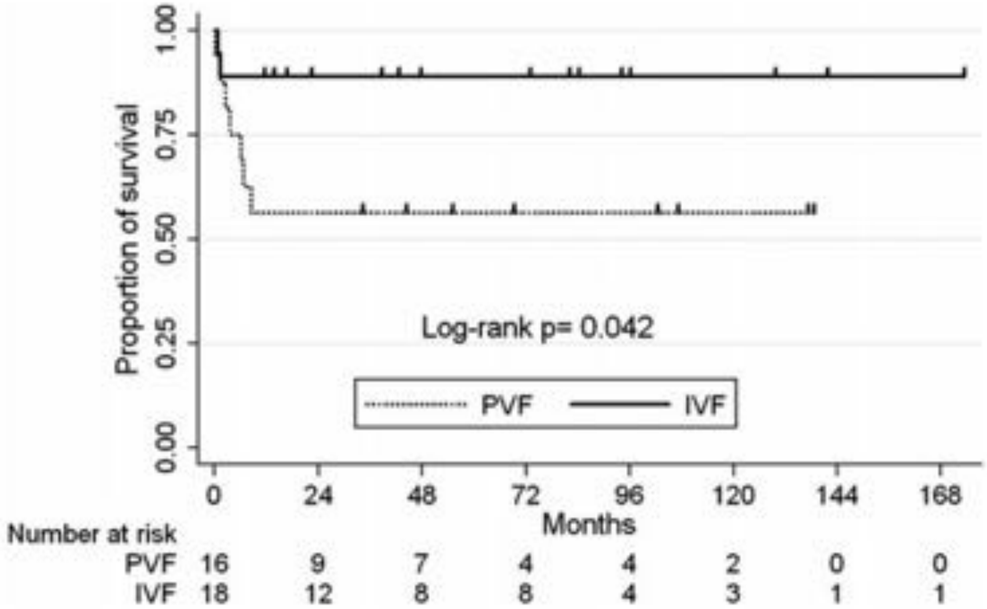
Only patients with early Fontan failures (<1 year) had normal post-transplantation PVR.

In paired comparisons, post-transplantation transpulmonary gradient was increased by a mean of 6.8 mm Hg (P=0.0001) relative to pretransplantation value.

M3C

Mitchell M et al. J Thorac Cardiovas Surg 2004;128:693

Fontan patients with reduced EF are different from those with preserved EF



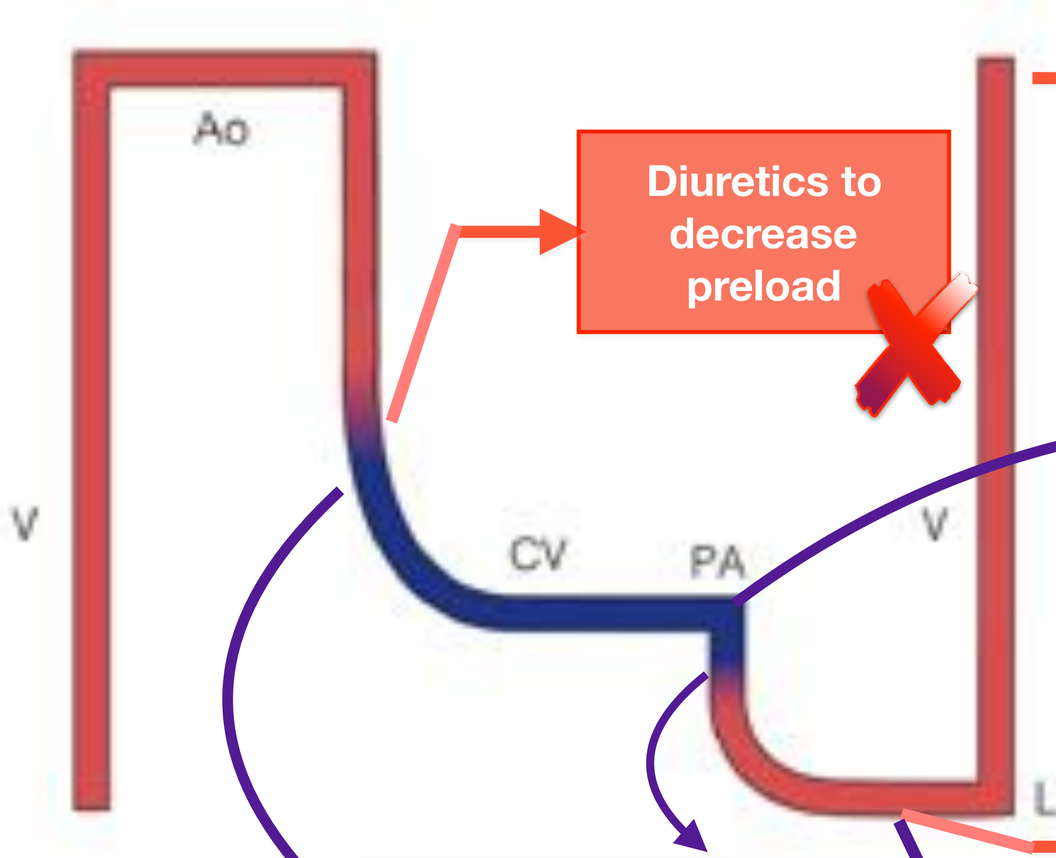
- In a group of Fontan patients undergoing transplantation, **patients with preserved EF had significantly worse outcomes than those with reduced EF** suggesting that important mechanisms other than systolic dysfunction contributed to heart failure in the former group.

- This also suggests that **preventive treatment with heart failure drugs** aiming to prevent deleterious remodeling of the SV **might not be beneficial**.

Griffiths ER, Kaza AK, Wyler von Ballmoos MC, Loyola H, Valente AM, Blume ED, del Nido P. Evaluating failing Fontans for heart transplantation: predictors of death. Ann Thorac Surg. 2009;88:558–563.

Heart failure drugs in Fontan circulation

Potentially a wrong reasoning and a predictable minimal effect



ACE Inhibitors to decrease after load

Diuretics to decrease preload

Pulmonary vasodilatation to increase preload

Beta-blockers to lengthen diastole and ventricular filling

Exercise to increase preload and decrease PVR

Lusitropic drugs sGC stimulators

Conclusion

- **Altered pulmonary blood flow is the trigger for pulmonary vascular remodelling in shunt lesions**
- **PAH-CHD is one of the most interesting model to examine the mechanisms or reversibility in PH**
- **The mechanisms leading to irreversibility are multiple (anti-apoptotic, inflammation, altered signalling, DNA damage) and are key to identify future therapeutic pathways in PH**
- **Mechanisms for increased PVR and vascular remodelling in pulmonary venous hypertension remain unclear**
- **Lack of pulsatility is also a trigger for pulmonary vascular remodelling but with reduced involvement of SMC and higher role of intimal remodelling suggesting that alternative pathways should be explored to manipulate PVR in the Fontan circulation**

Thank you

Collective ignorance is the motivation
Curiosity is the strength
Research is the path

Individual experience is the brake
Indifference is the weakness
Authority argument is the threat