



Universiteit Utrecht

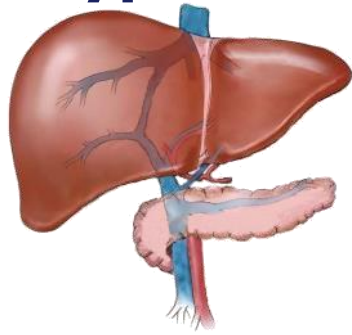
The background of the slide is a photograph of a bridge structure. On the bridge, there is a large, stylized sculpture of a lion and a human figure. The lion is standing on its hind legs, facing the human figure, who is also standing on its hind legs. They appear to be shaking hands or holding hands. The sculpture is made of metal and is painted white. The bridge has a metal railing and some structural beams. The sky is cloudy and grey.

Congenital portosystemic shunts: the clinical relevance of genomic alterations

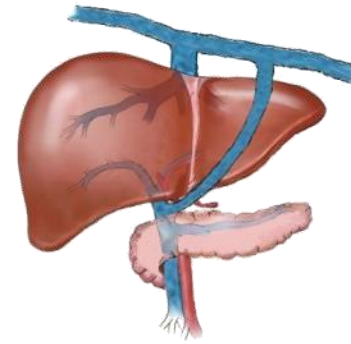
Frank van Steenbeek
Assistant Professor

Congenital Portosystemic Shunts

Main subtypes



Intrahepatic



Extrahepatic

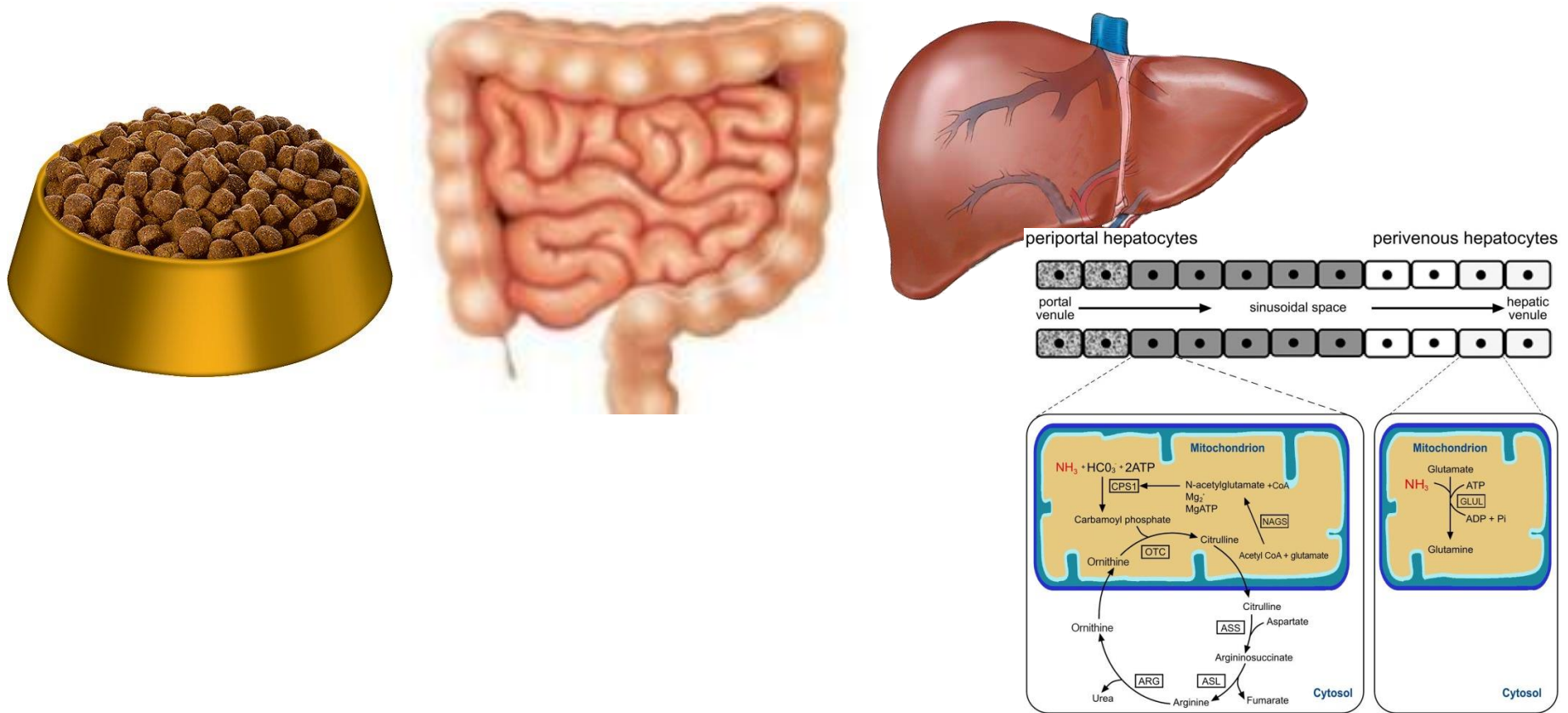


van Steenbeek et al., *Mamm. genome.*, 2012



Congenital Portosystemic Shunts

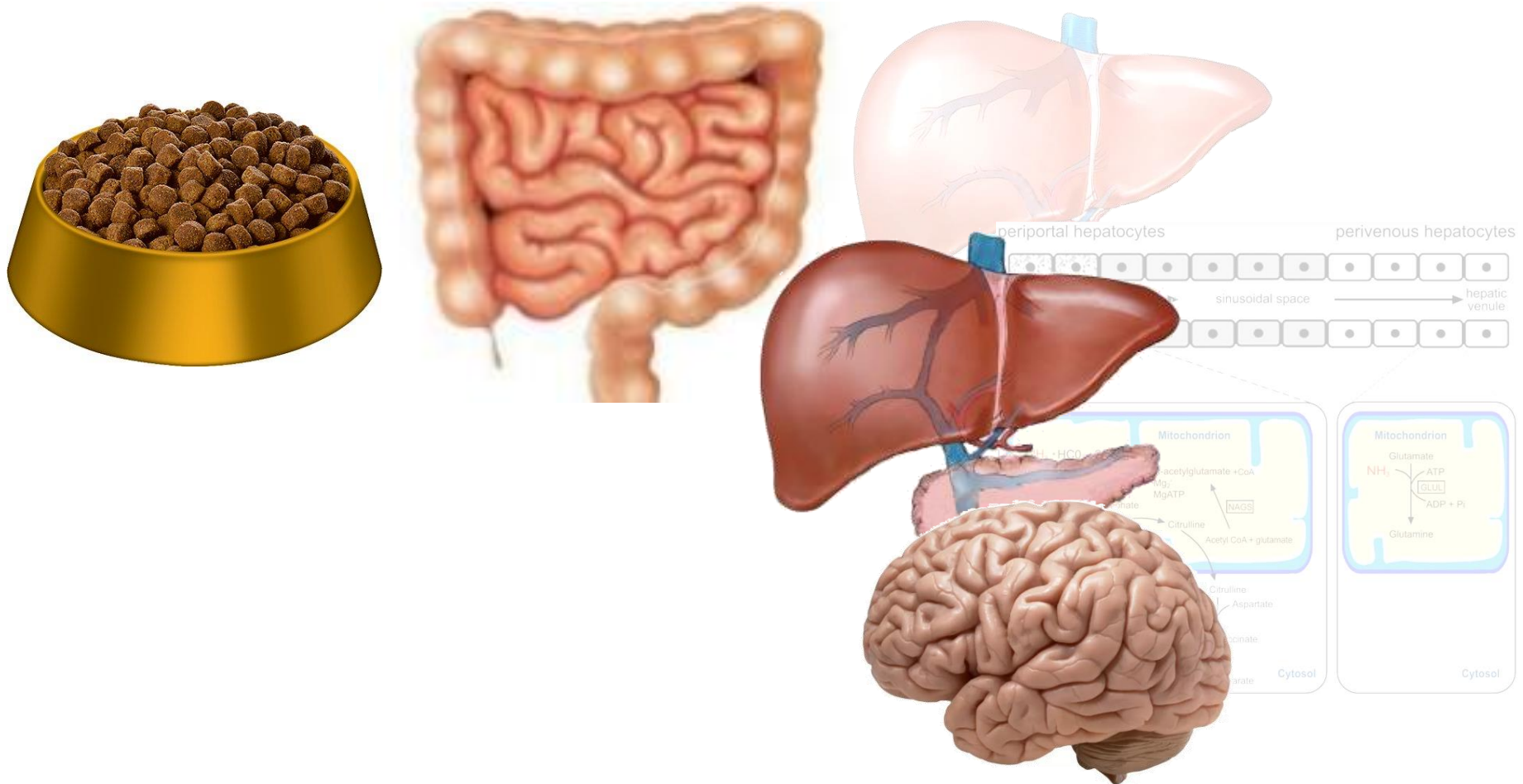
Healthy situation



Van Straten and van Steenbeek *et al.*, *Plos ONE*, 2014

Congenital Portosystemic Shunts

Consequence



Congenital Portosystemic Shunts

Symptoms

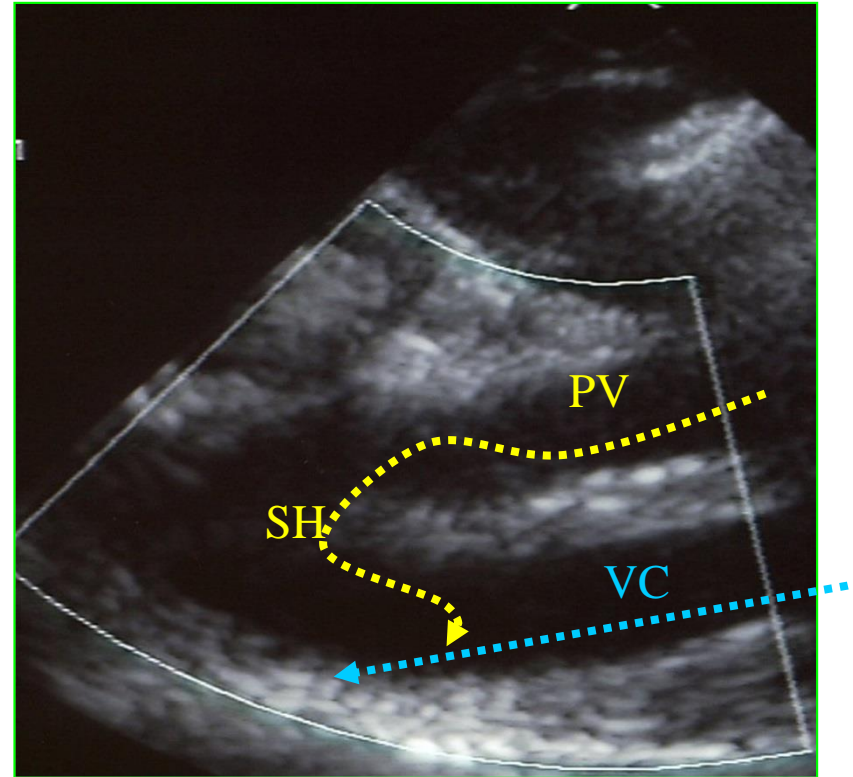
- rapid fatigue
- being slow
- excessive drinking and urinating
- retarded growth
- vomiting, sometimes also diarrhea
- inflammation of the bladder
- brain defects



Congenital Portosystemic Shunts

Diagnosis

- Ammonia/Bile Acids
- ATT
- Echo
- CT



Viktor Szatmari



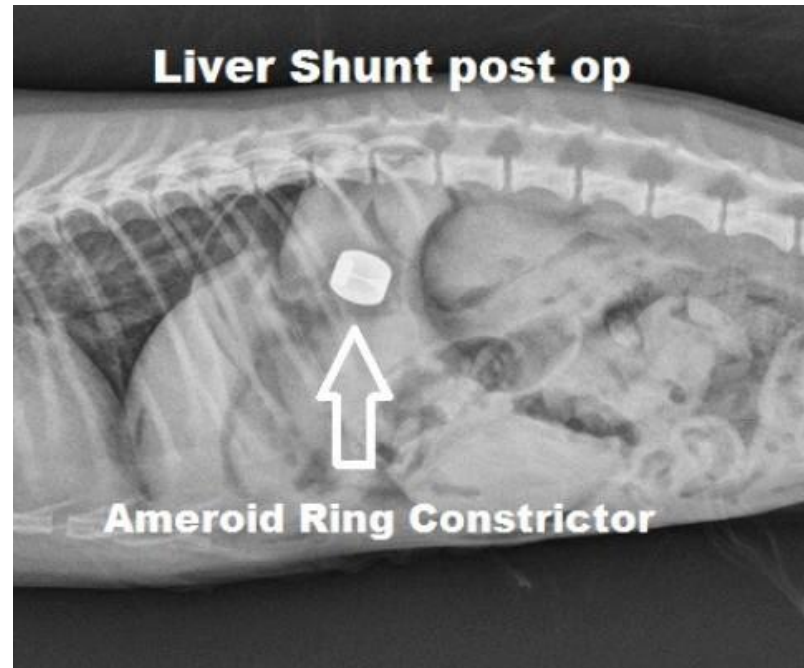
Treatment pre surgery

- Lactulose
 - disaccharide
 - metabolized by colonic bacteria
 - Acidification transforms NH_3 to NH_4^+
- Low protein diet



Shunt occlusion

- Suture ligation
 - Ameroid constrictors
 - Cellophane bands
-
- Success rate ~70%



Intrahepatic portosystemic shunts

Liver tissue

RESEARCH ARTICLE

Aberrant hepatic lipid storage and metabolism in canine portosystemic shunts

Lindsay Van den Bossche¹, Vivien A. C. Schoonenberg¹, Iwan A. Burgener¹, Louis C. Penning¹, Ingrid M. Schraal¹, Hedwig S. Kruitwagen¹, Monique E. van Wolferen¹, Guy C. M. Grinwis², Anne Kummeling¹, Jan Rothuizen¹, Jeroen F. van Velzen³, Nikolas Stathonikos⁴, Martijn R. Molenaar⁵, Bernd J. Helms⁵, Jos F. H. M. Brouwers⁵, Bart Spee^{1*}, Frank G. van Steenbeek^{1*}

OPEN ACCESS Freely available online



Altered Subcellular Localization of Heat Shock Protein 90 Is Associated with Impaired Expression of the Aryl Hydrocarbon Receptor Pathway in Dogs

Frank G. van Steenbeek^{1*}, Bart Spee¹, Louis C. Penning¹, Anne Kummeling¹, Ingrid H. M. van Gils¹, Guy C. M. Grinwis², Dik Van Leenen³, Frank C. P. Holstege³, Manon Vos-Loohuis¹, Jan Rothuizen¹, Peter A. J. Leegwater¹

¹ Department of Clinical Sciences of Companion Animals, Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands, ² Department of Pathobiology, Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands, ³ Molecular Cancer Research, University Medical Centre Utrecht, Utrecht, The Netherlands

Liver INTERNATIONAL
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Liver International ISSN 1478-3223

BASIC STUDIES

Recombinant hepatocyte growth factor treatment in a canine model of congenital liver hypoplasia

Hedwig S. Kruitwagen^{1*}, Brigitte Arends^{1*}, Bart Spee¹, Bas Brinkhof¹, Ted S.G.A.M. van den Ingh², Victor P.M.G. Rutten^{3,4}, Louis C. Penning¹, Tania Roskams⁵ and Jan Rothuizen¹

OPEN ACCESS Freely available online



Aberrant Expression and Distribution of Enzymes of the Urea Cycle and Other Ammonia Metabolizing Pathways in Dogs with Congenital Portosystemic Shunts

Giora van Straten^{1*}, Frank G. van Steenbeek¹, Guy C. M. Grinwis², Robert P. Favier¹, Anne Kummeling¹, Ingrid H. van Gils¹, Hille Fieten¹, Marian J. A. Groot Koerkamp³, Frank C. P. Holstege³, Jan Rothuizen¹, Bart Spee¹

OPEN ACCESS Freely available online



Aberrant Gene Expression in Dogs with Portosystemic Shunts

Frank G. van Steenbeek^{1,2*}, Lindsay Van den Bossche^{1,3}, Guy C. M. Grinwis², Anne Kummeling¹, Ingrid H. M. van Gils¹, Marian J. A. Groot Koerkamp³, Dik van Leenen³, Frank C. P. Holstege³, Louis C. Penning¹, Jan Rothuizen¹, Peter A. J. Leegwater¹, Bart Spee¹

The Veterinary Journal 204 (2015) 226–228

Contents lists available at ScienceDirect



The Veterinary Journal

journal homepage: www.elsevier.com/locate/tvj



Short Communication

Increased bone morphogenetic protein 7 signalling in the kidneys of dogs affected with a congenital portosystemic shunt

Astrid M. van Dongen, Susanne M. Heuving, Marianna A. Tryfonidou, Frank G. van Steenbeek, Jan Rothuizen, Louis C. Penning *



Pre-surgical prediction

	microarray		qPCR	
	recovered	Not recovered	recovered	Not recovered
EHPSS	19	13	21	14
IHPSS	4	10	10	17
total	23	23	31	31



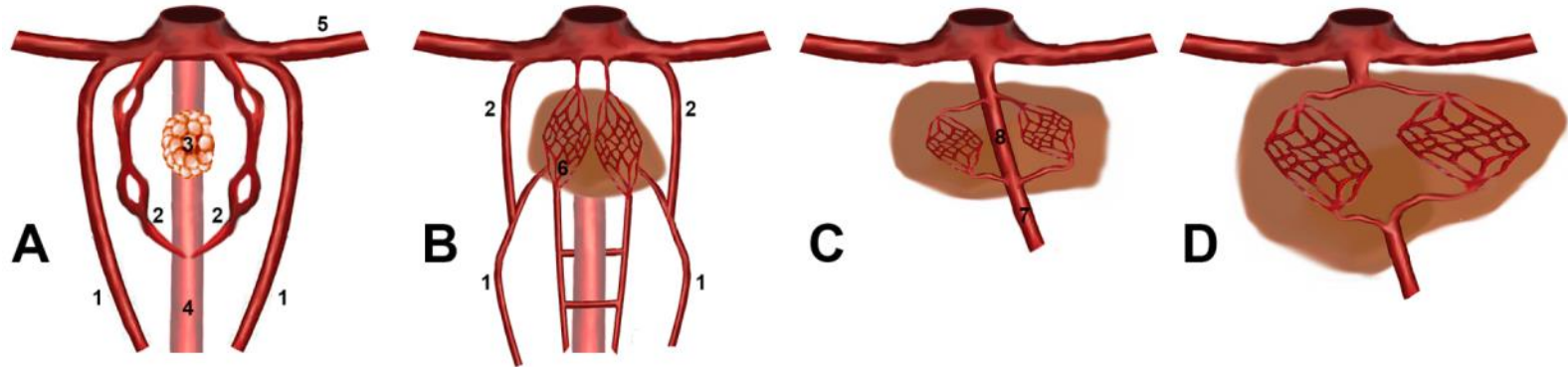
Pre-surgical prediction

$$\text{Logit } (\pi) = 0.187 + 1.78(\text{b albumin}) + 1.07(\text{b}\Delta\text{CqDHDH}) \\ - 1.59(\text{b}\Delta\text{CqERLEC1}) - 1.53(\text{b}\Delta\text{CqLYSMD2}),$$



Congenital Portosystemic Shunts

Normal liver development

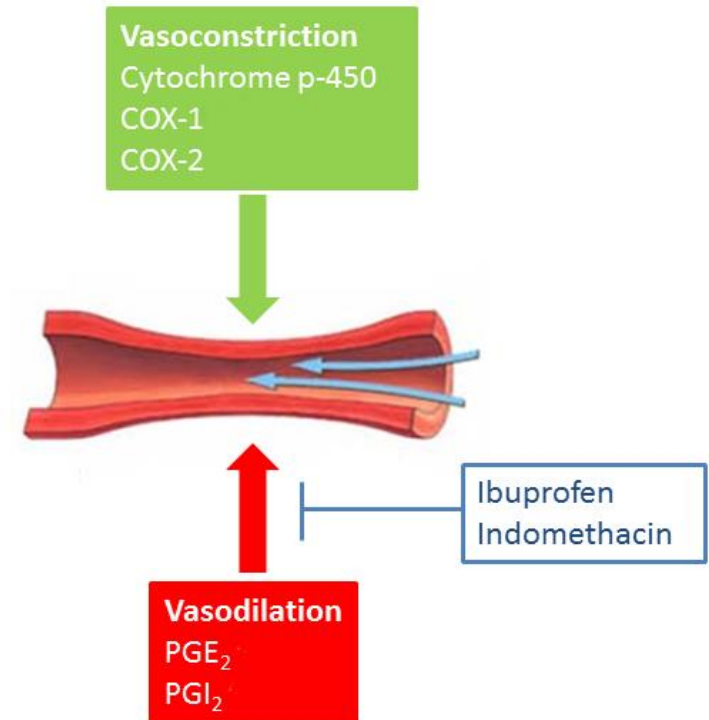
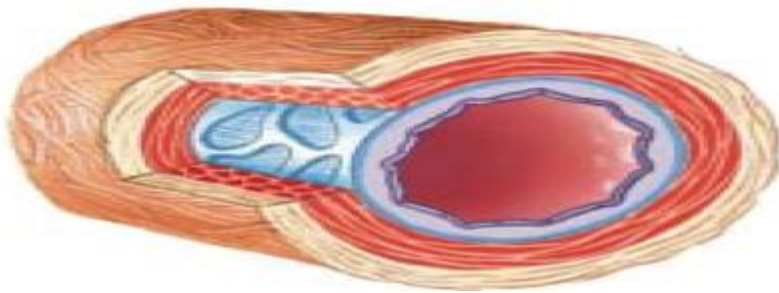


van den Bossche and van Steenbeek, Vet J, 2016



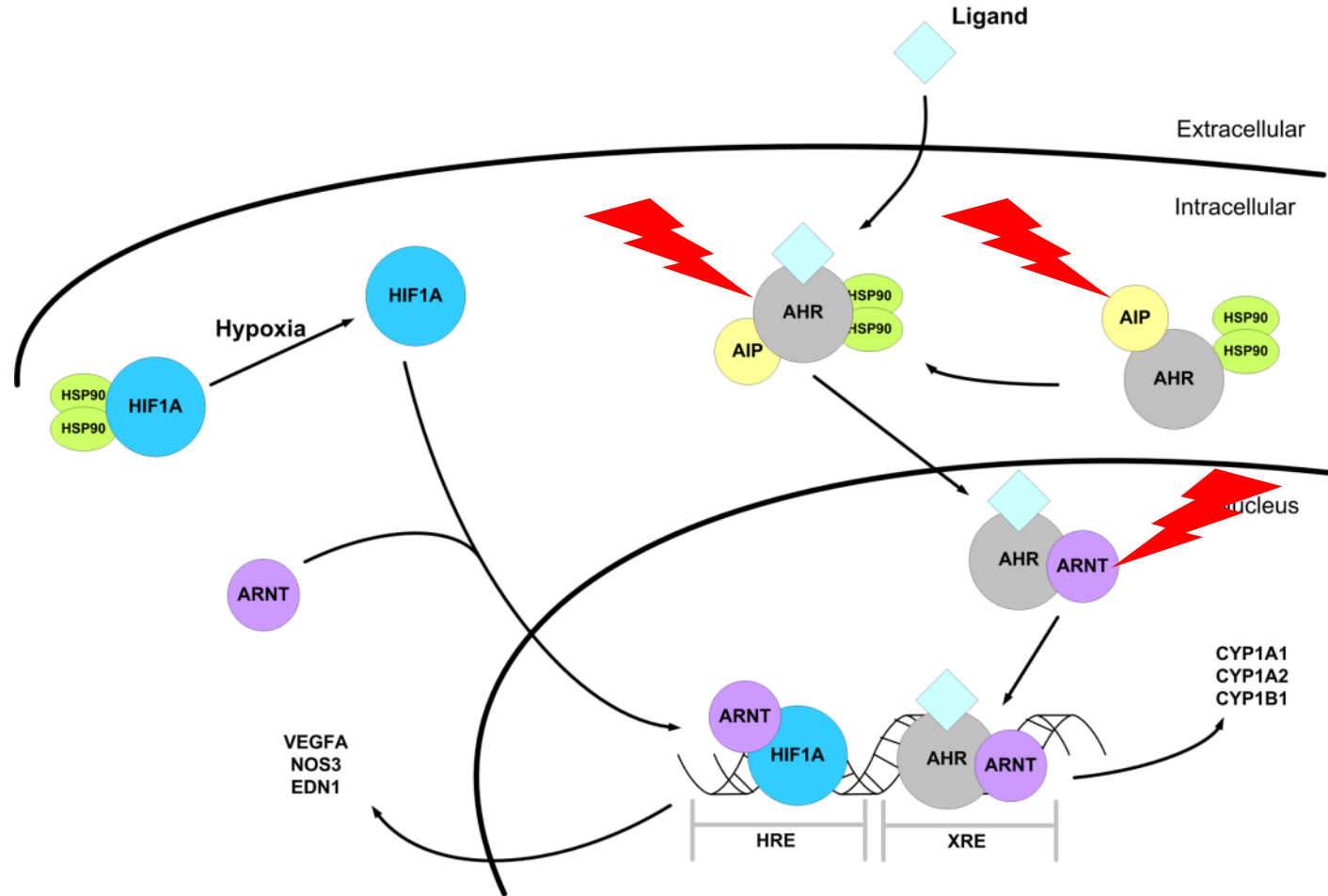
Intrahepatic portosystemic shunts

Mechanism



Intrahepatic portosystemic shunts

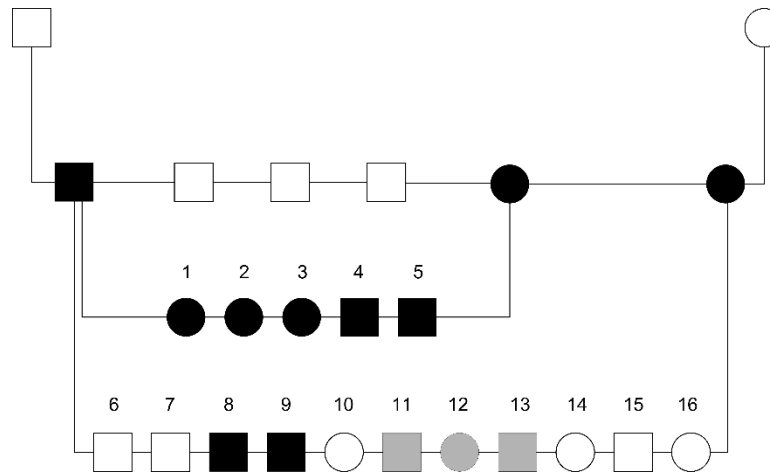
Candidate gene



van Steenbeek, *PLoS ONE*, 2013b

Intrahepatic portosystemic shunts

Inheritance



Complex inheritance
(digenic, tri-allelic)
At least 2 genes involved

van Steenbeek et al., JVIM 2009



Intrahepatic portosystemic shunts

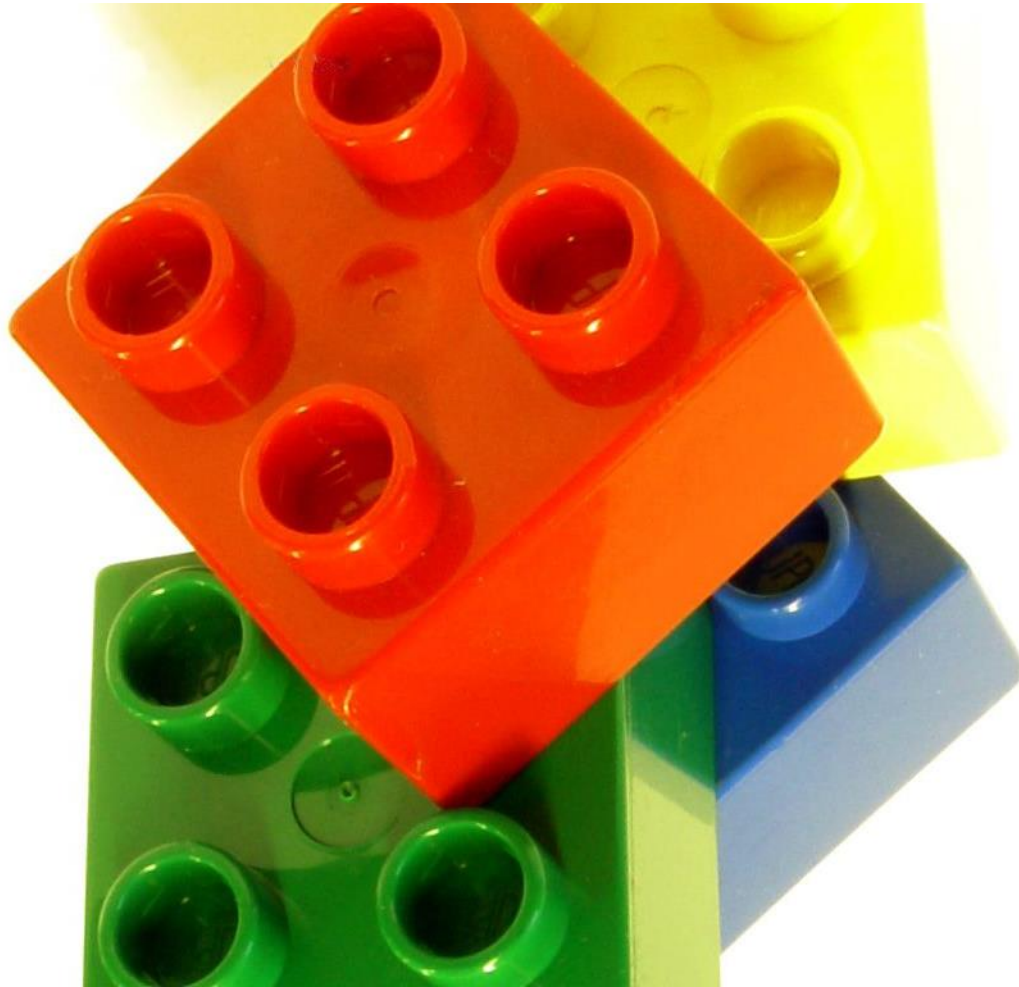
Inheritance

- Both parents are carrier in case of an affected dog
- Combining two carriers does not have to result in affected dogs



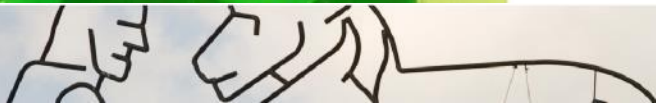
DNA

Trigger your imagination



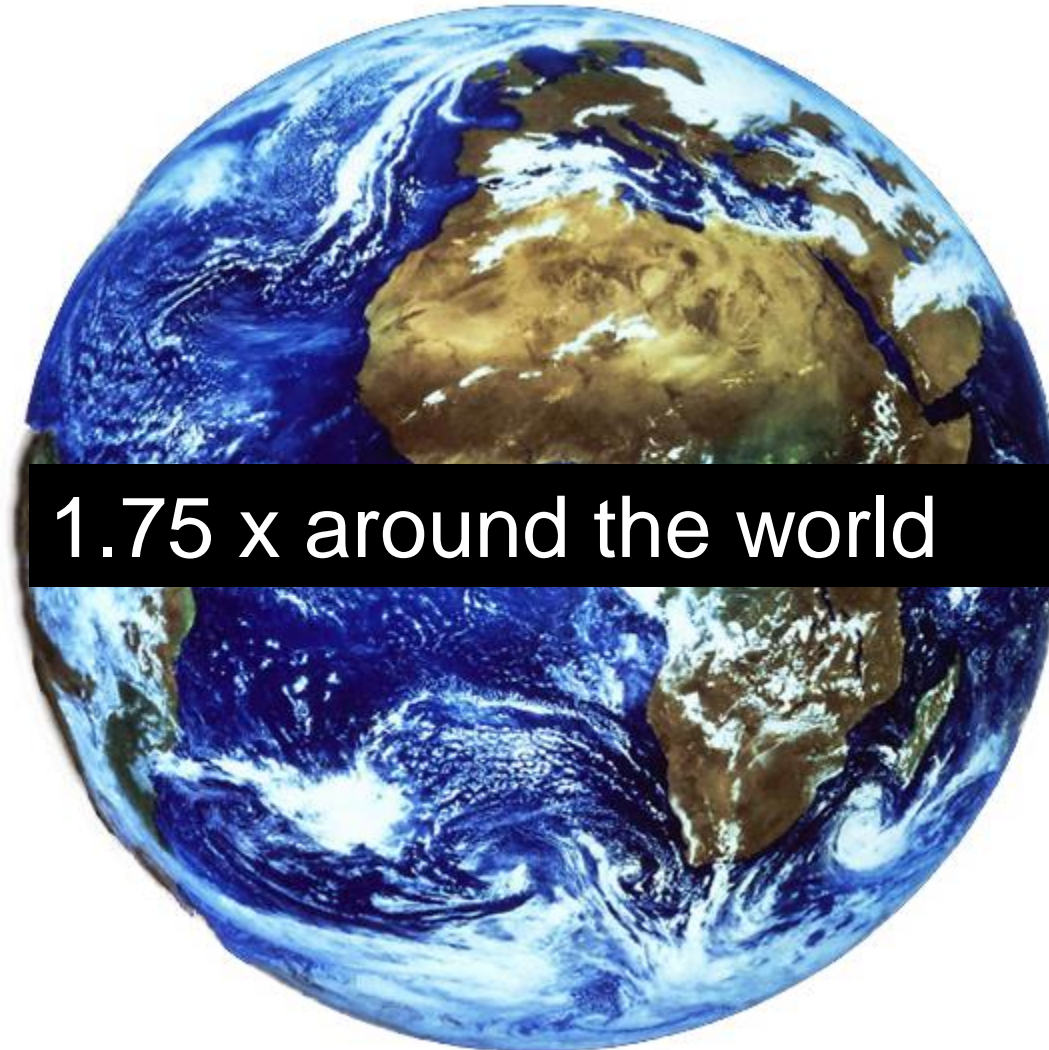
2,384,996,543 blocks
4 colors

= sequence



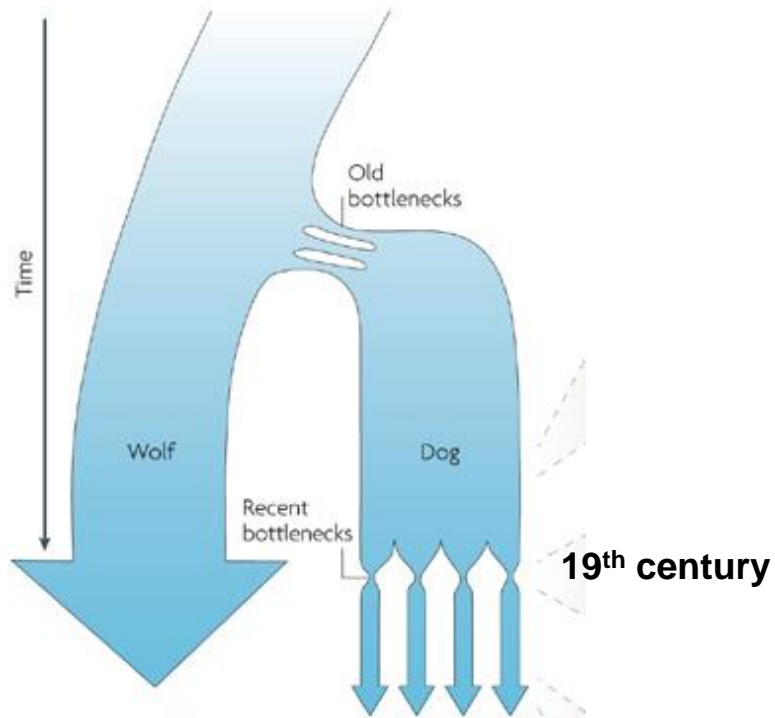
DNA

Trigger your imagination



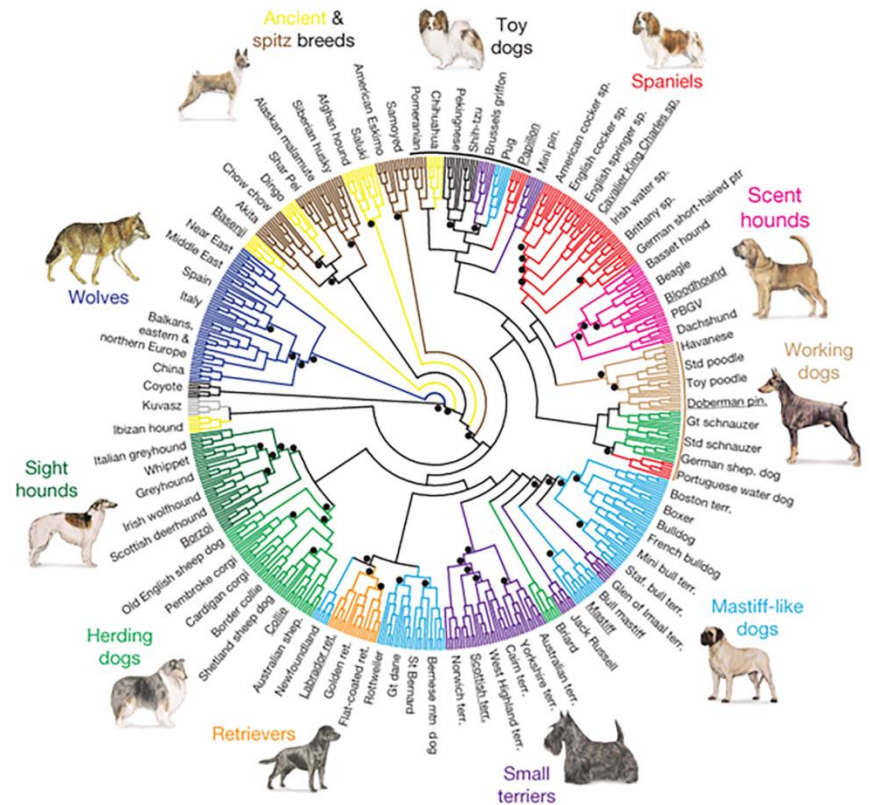
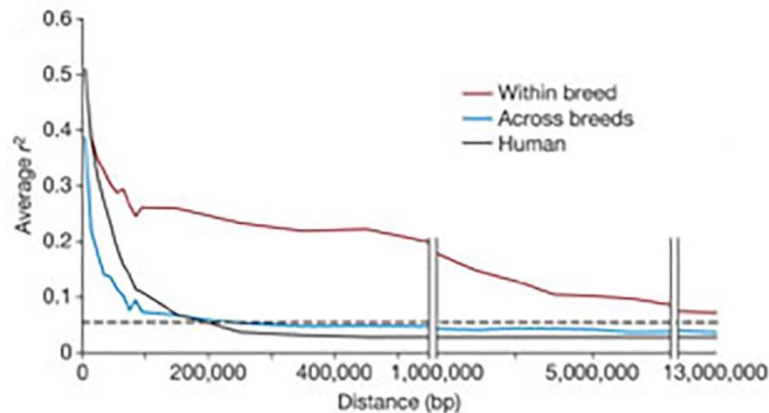
DNA

Variation in the dog



DNA Variation in the dog

~6M known SNPs

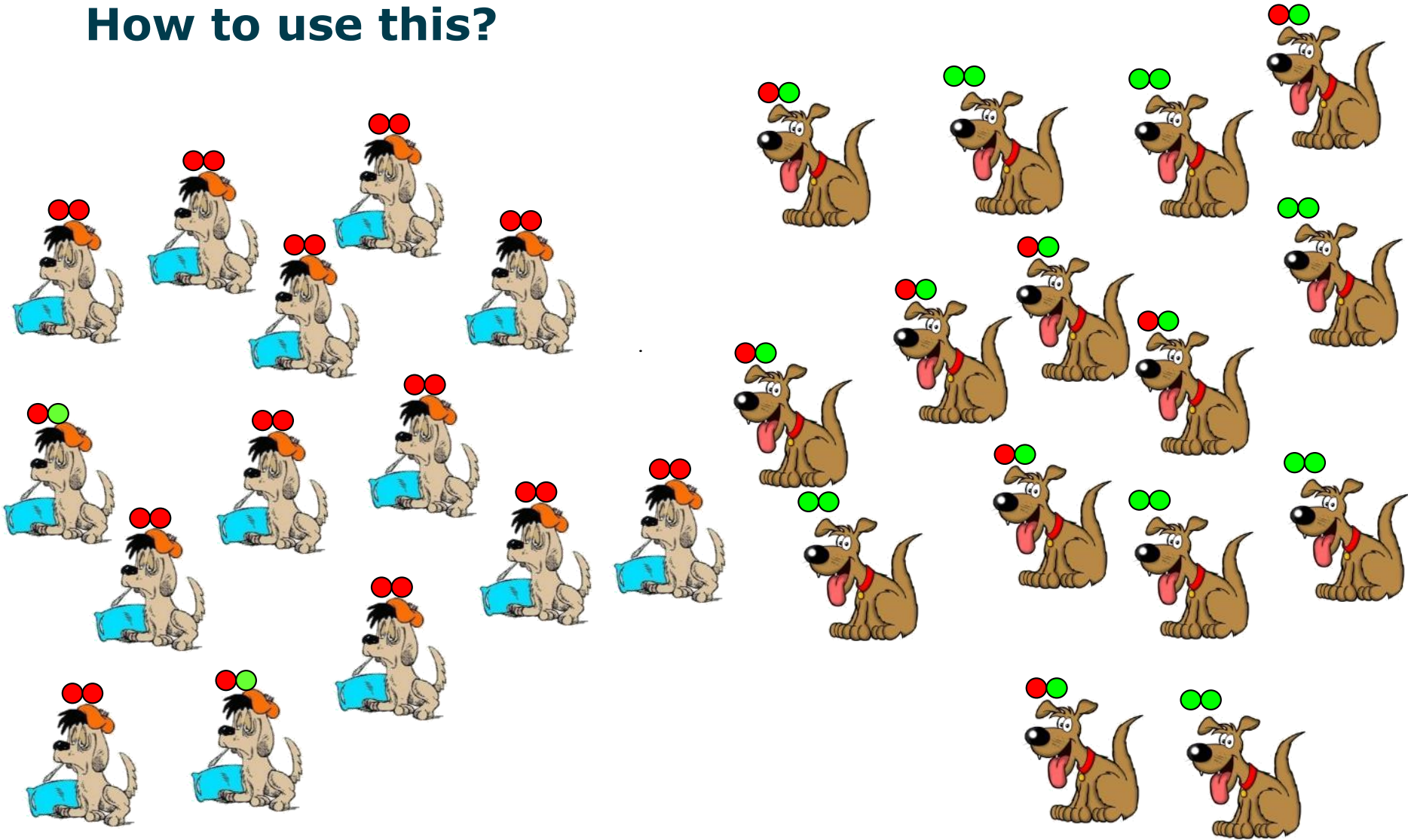


Genome sequence, comparative analysis and haplotype structure of the domestic dog. Lindblad-Toh K 2005 Nature

Genome-wide SNP and haplotype analyses reveal a rich history underlying dog domestication, vonHoldt BM 2010 Nature

DNA variation

How to use this?



DNA variation

Usage in IHPSS research

Genom-wide analysis



419.737 bp

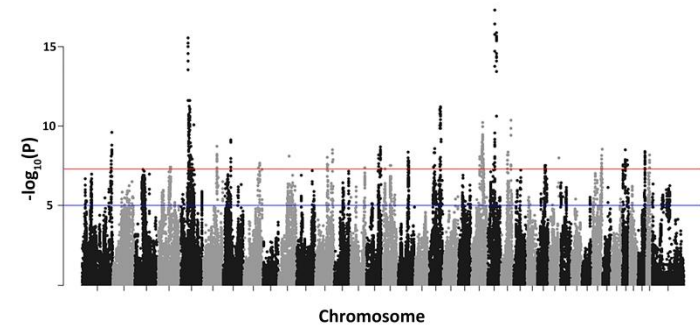
Sequence analysis



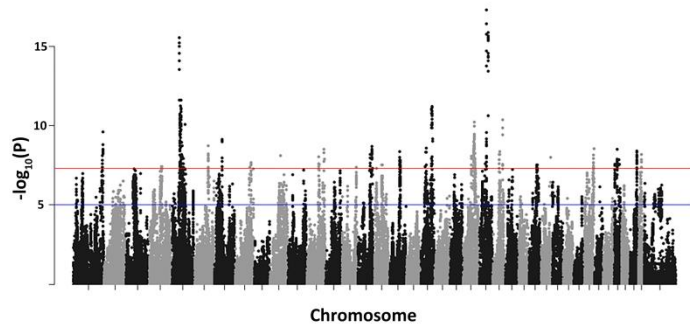
1.046 bp

Validation

4 bp

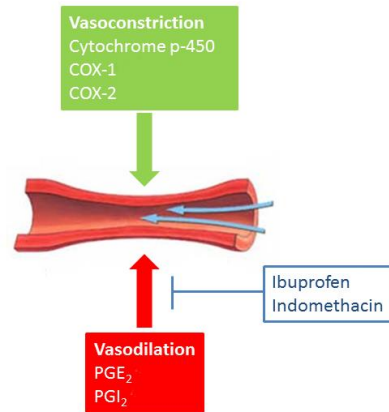
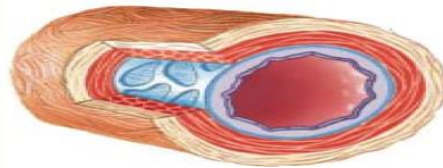


Intrahepatic portosystemic shunts overlap



+ PDA (48 Stabyhouns (24 cases versus 24 controls))

?

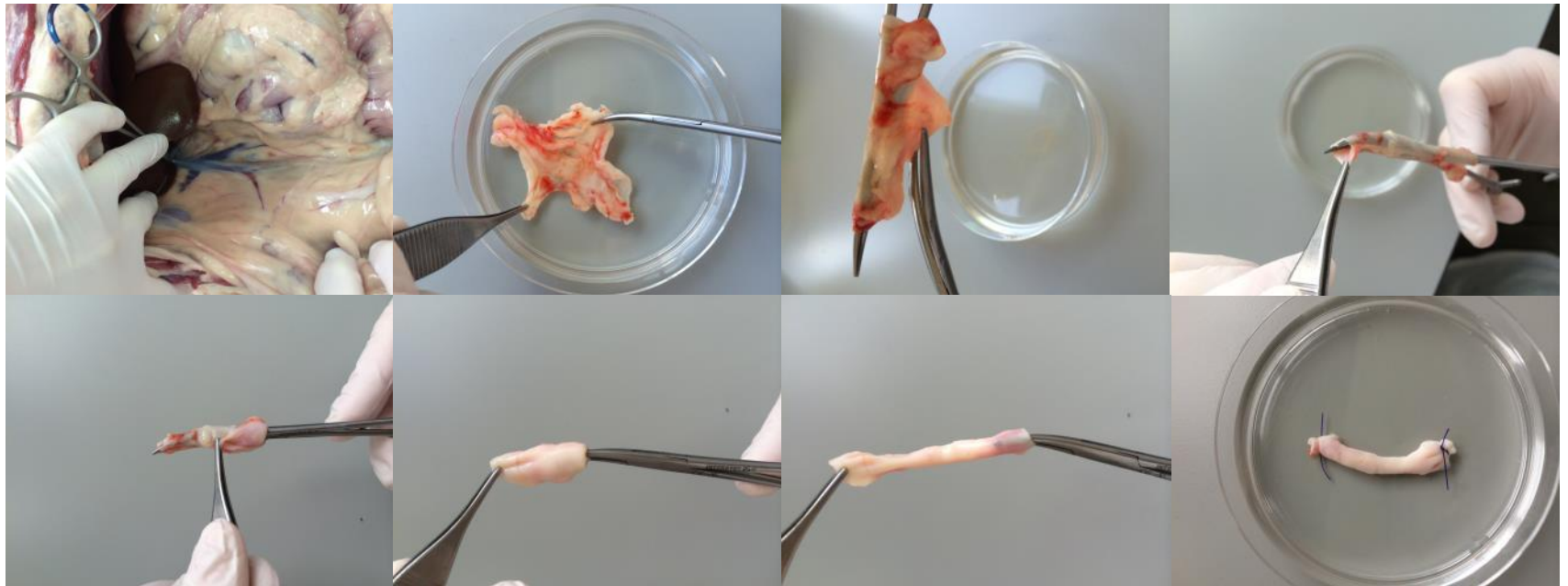


Vascular cell systems

primary endothelial cells

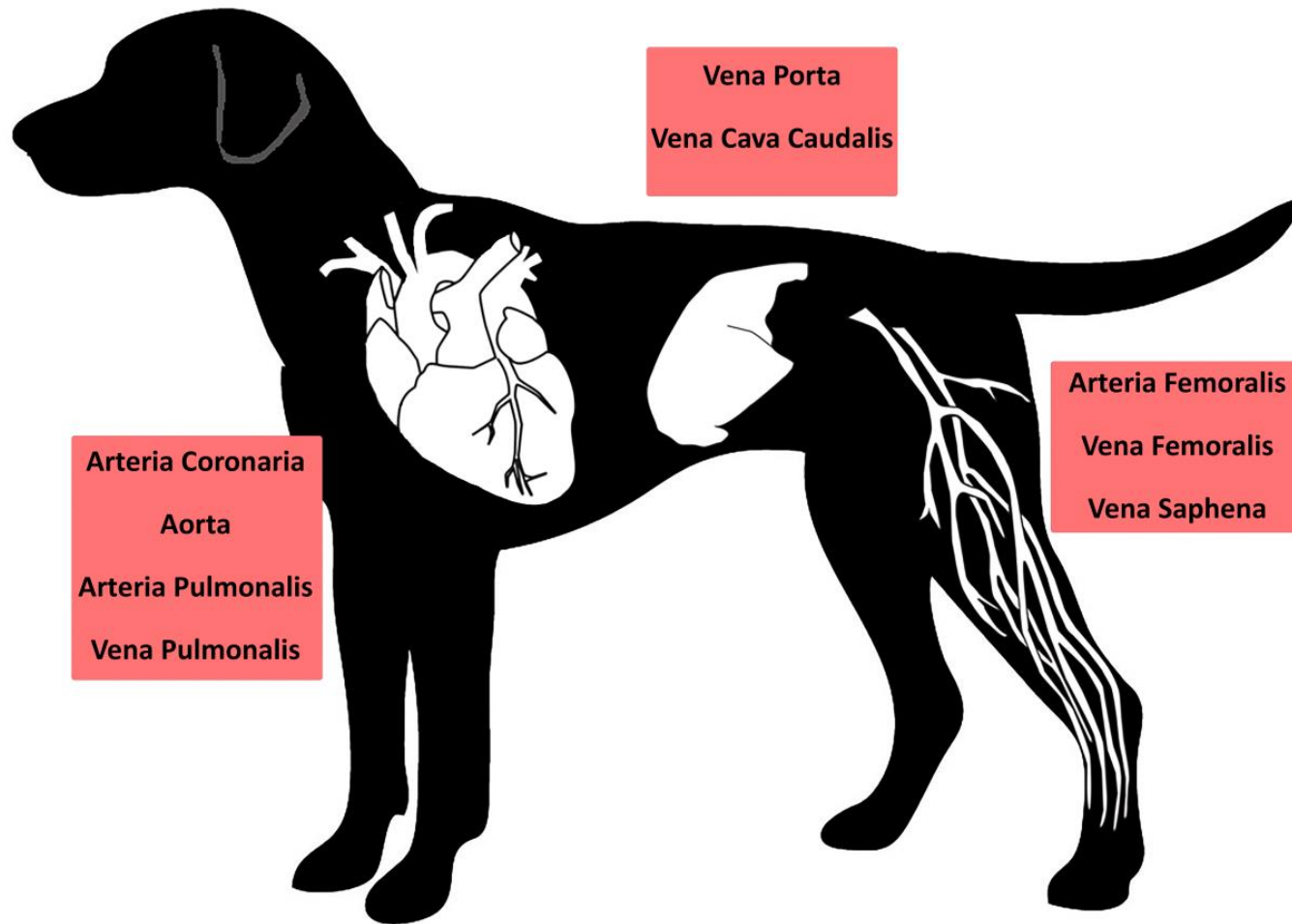
Isolation and Culture of Primary Endothelial Cells from Canine Arteries and Veins

Loes A. Oosterhoff, Hedwig S. Kruitwagen, Bart Spee, Frank G. van Steenbeek

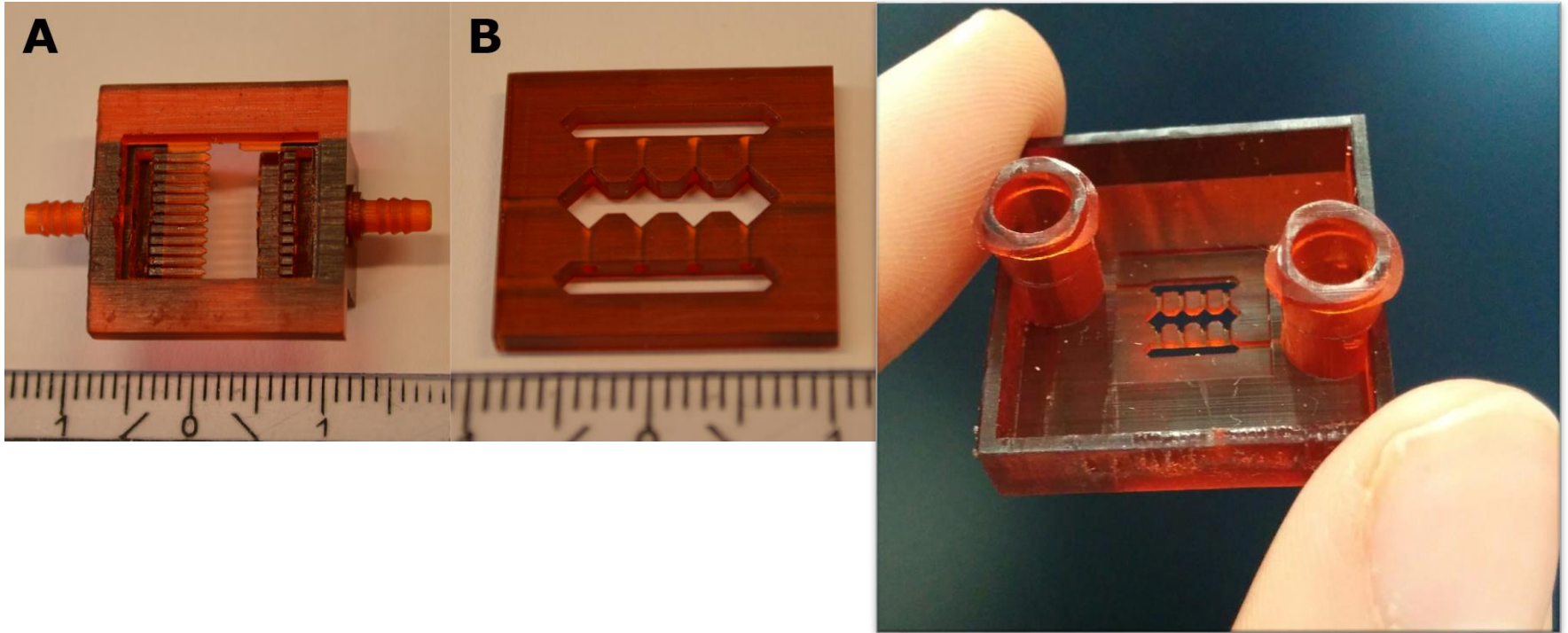


Vascular cell systems

primary endothelial cells

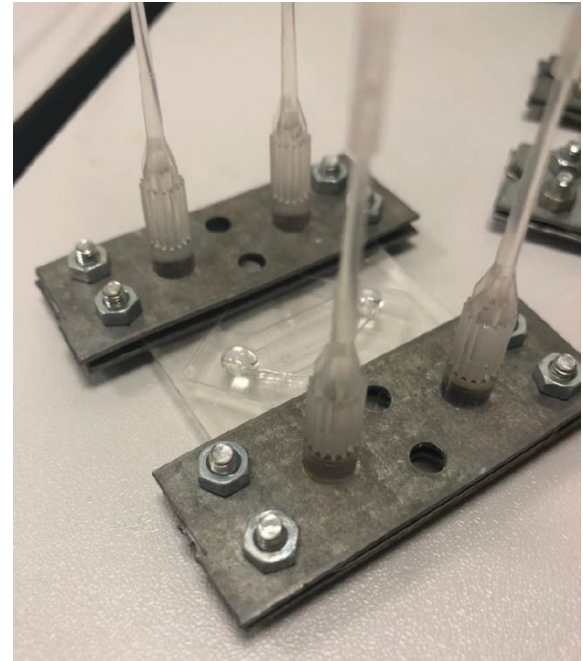


Angiogenesis bioreactor

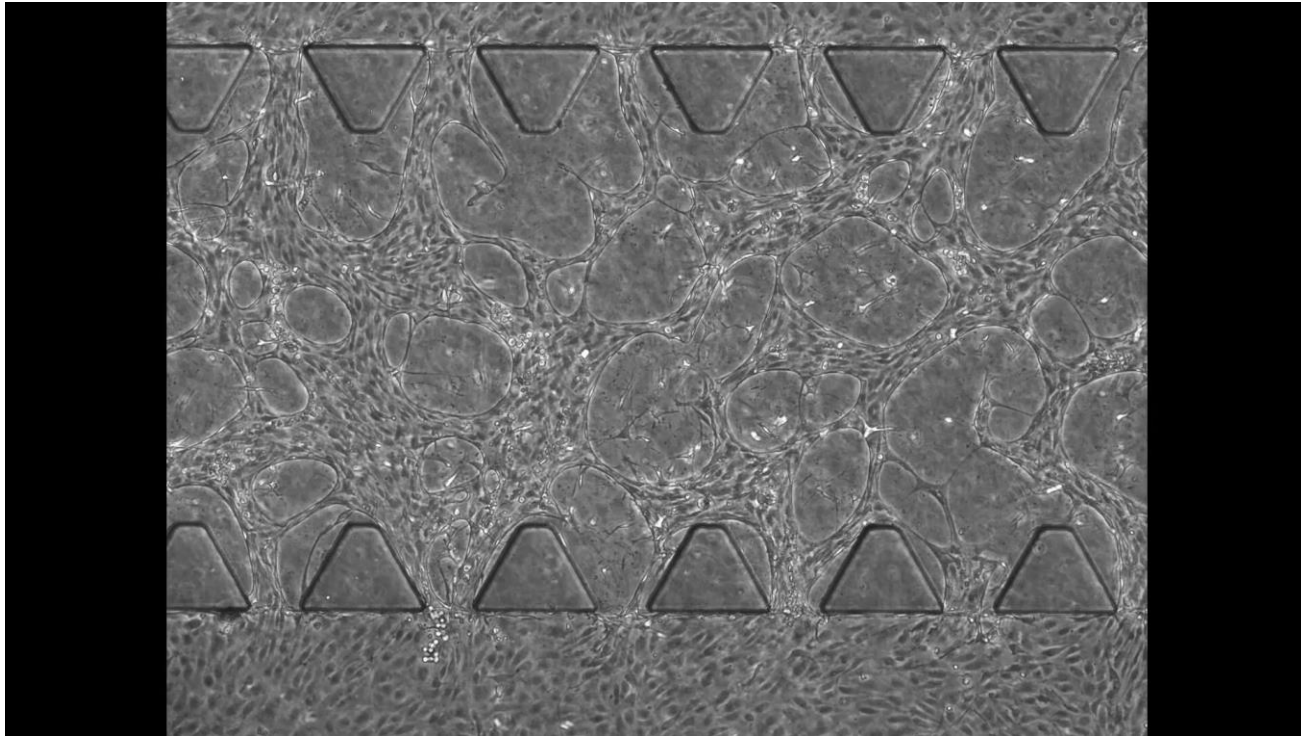


Vascular cell systems

primary endothelial cells



Extrahepatic portosystemic shunts angiogenesis bioreactor



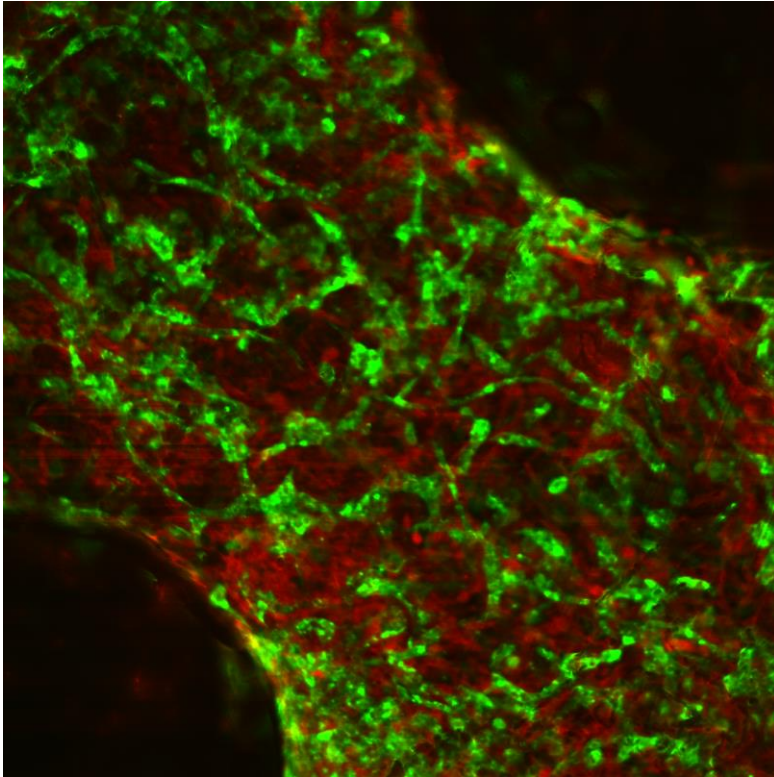
“vasculature” = HUVEC + pericytes
Dextranbeads



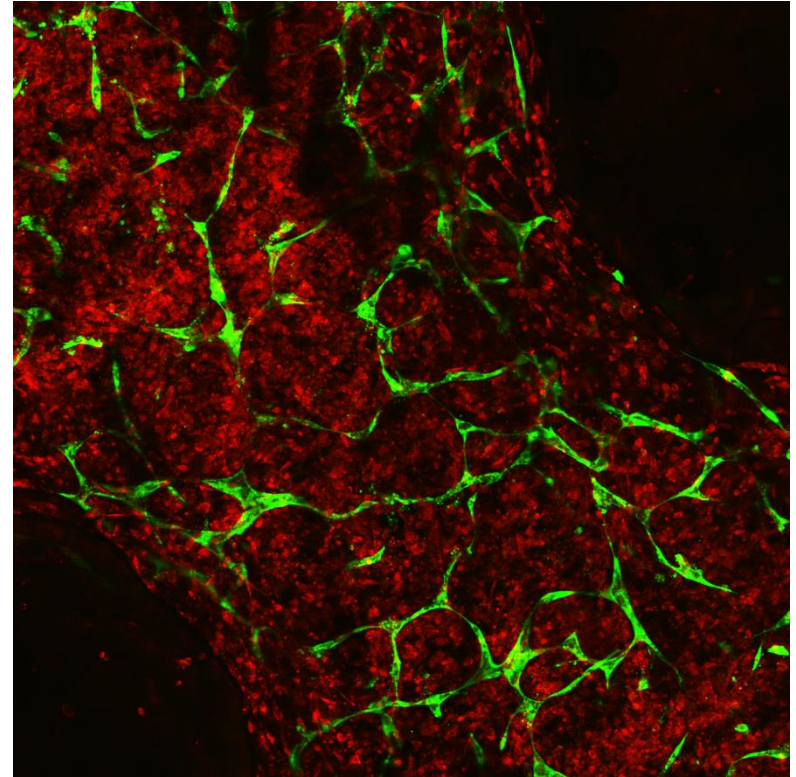
Vascular cell systems

cell culture

preFlow



postFlow



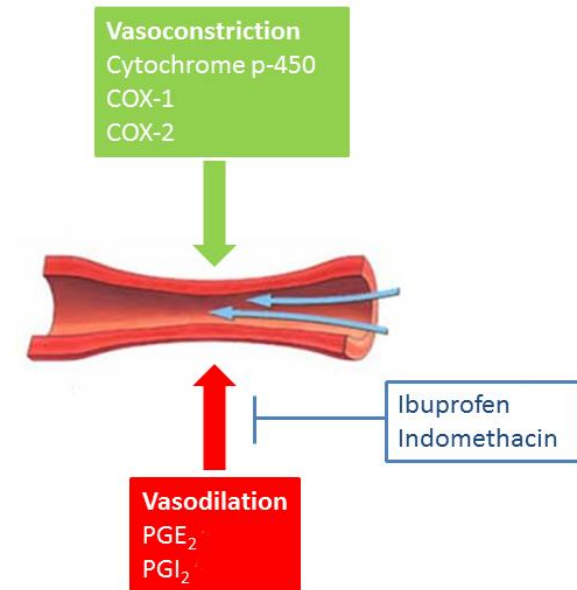
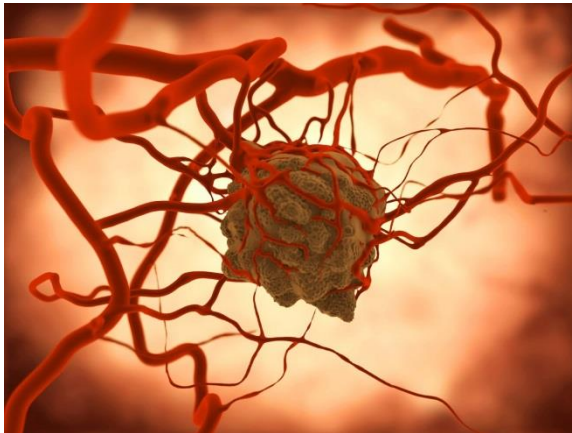
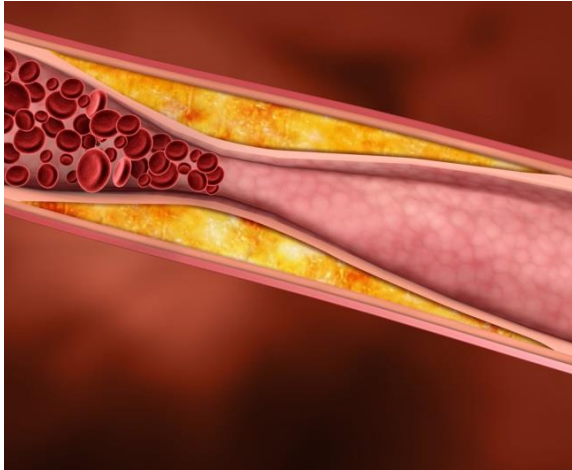
Endothelial cells (HUVEC-GFP)

Mesenchymal cells (hBMSCs-dsRed)



Vascular cell system

Future implications



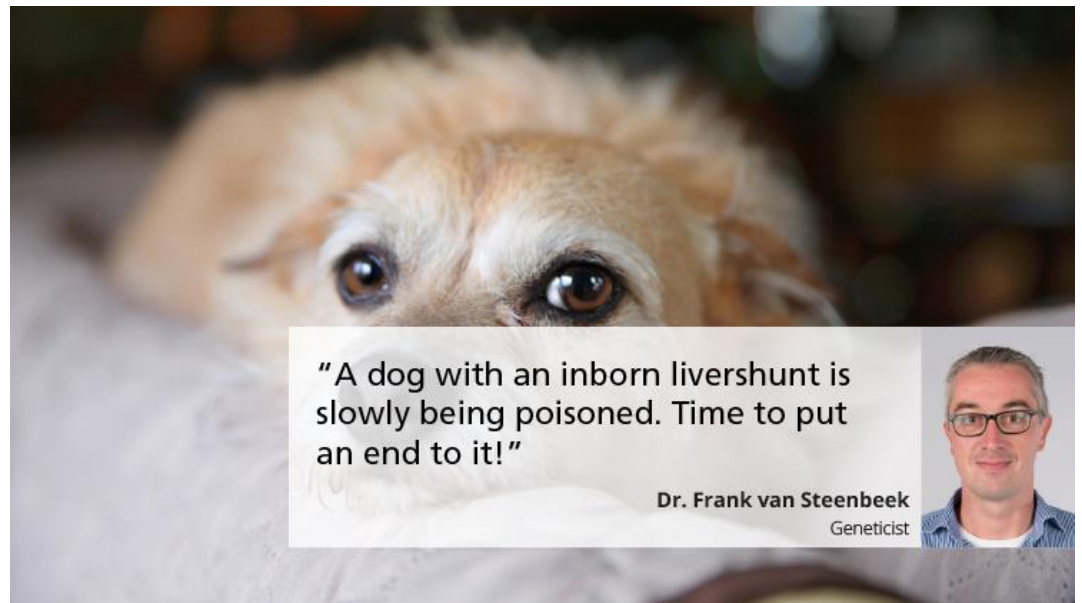
What is next?

- Genome analysis 2.0
- Understand the population



What is next?

- Genome analysis 2.0
 - Collaboration with Davis (USA) and Bern (Switzerland)
 - Full genome sequence



<https://www.vriendendiergeneeskunde.nl/project/levershunts>



What is next?

- Understand the population

- Frankness

An affected pup doesn't make a bad breeder

- Know the prevalence

What percentage of the population is affected

- Early recognition

Detect affecteds before leaving the breeder



Understand the population

Field example

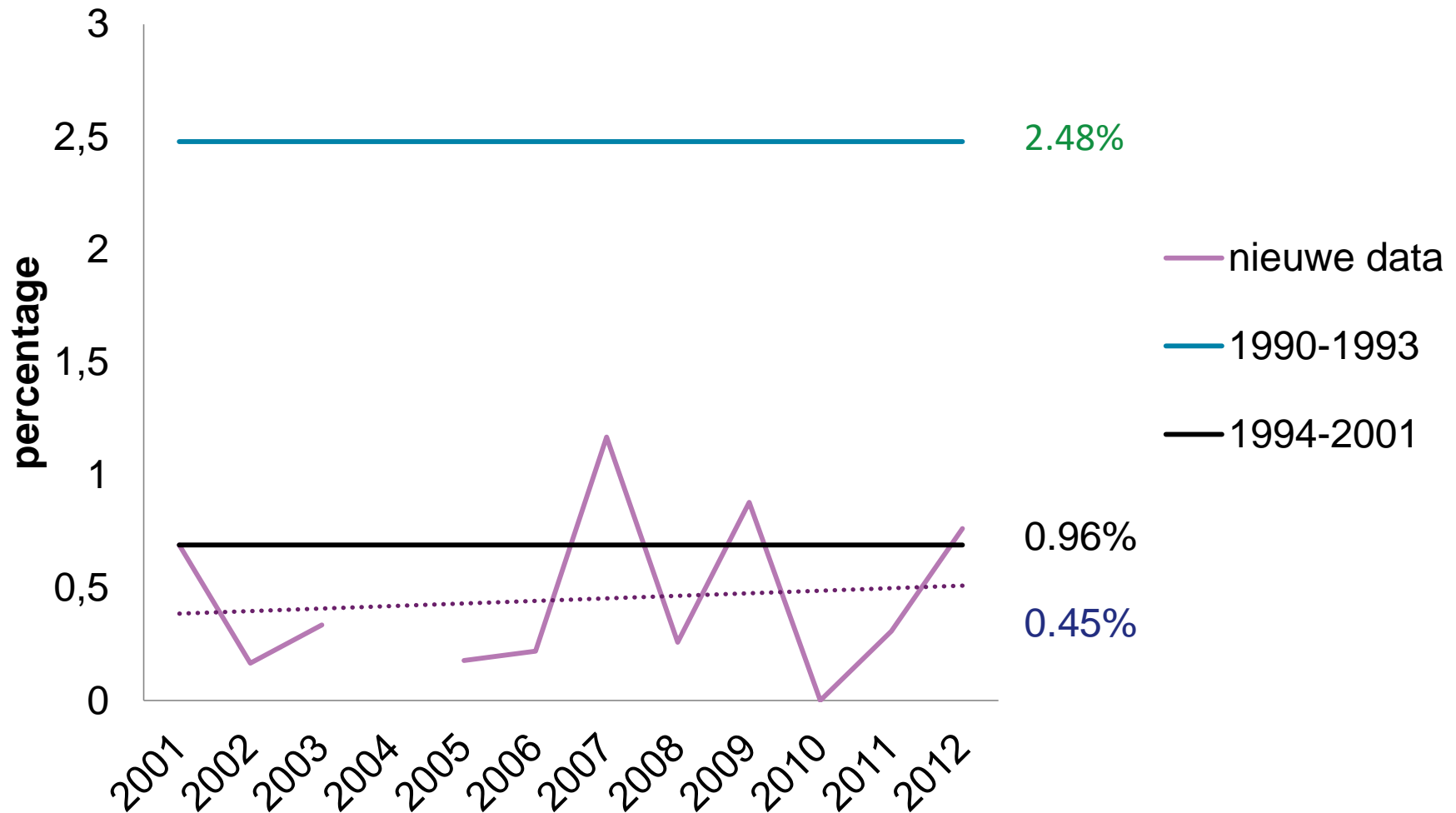
- EHPSS in Cairn terrier
- “breed at risk”
- Recognition 90` s

“Breeding a dog with a shunt doesn` t make you a bad breeder, keeping it as a secret DOES.”



Successful testing

Prevalence



Now what?

- Registration?
- Test?
- Openness?



Acknowledgements

Utrecht University

Faculty of Veterinary Medicine

Clinical Sciences of Companion Animals

- Jan Rothuizen
- Bart Spee
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Hubrecht institute

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- Michael Tivers
- Daniel Brockman

University of Bern

- Tosso Leeb
- Elisabeth Dietschi



Morris Animal
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Landbouw en Innovatie