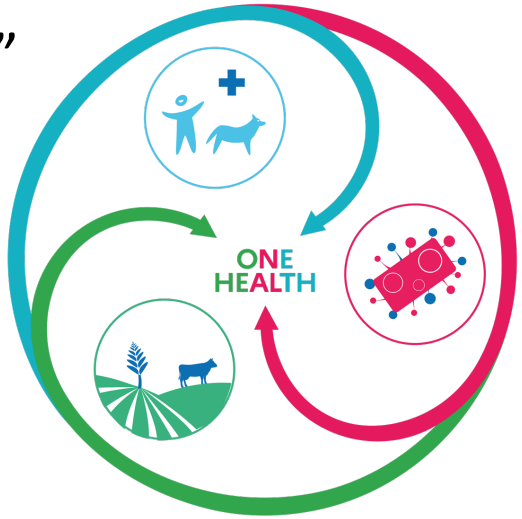
One Health: contribution of animals to the fight against sepsis.

"Essentially, all models are wrong, but some are useful"

George Box (British statistician)



Professor Vanessa Louzier
Doctor in Veterinary Medicine , PhD
APCSE : Pulmonary and Cardiovascular Agression in Sepsis
VetAgro Sup, Veterinary Campus of Lyon

Clinical Review & Education | The JAMA Network

Special Communication | CARING FOR THE CRITICALLY ILL PATIENT

The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3)

JAMA. 2016;315(8):801-810. doi:10.1001/jama.2016.0287

sepsis = life-threatening organ dysfunction caused by a dysregulated host response to infection

Septic shock = severe form of sepsis. Profound circulatory, cellular, and metabolic abnormalities are associated with a greater risk of mortality than with sepsis alone.

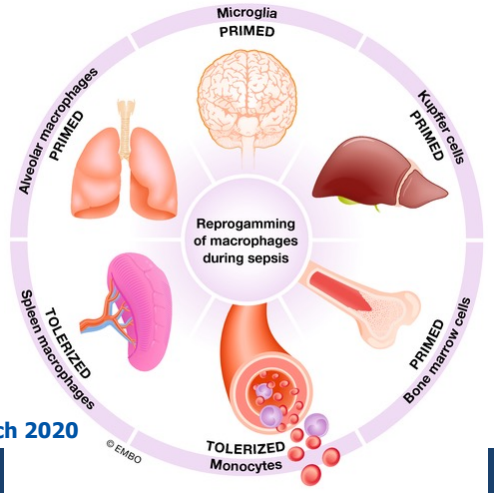
infection → sepsis → Septic shock

Infection

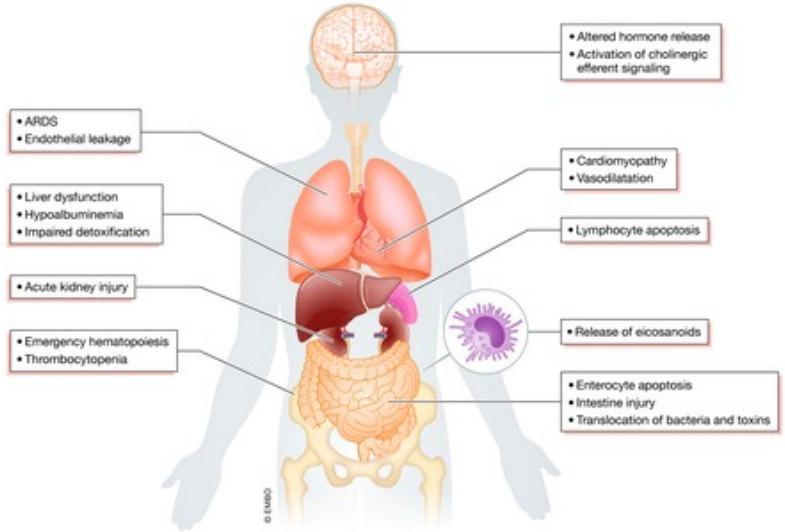
Bacteria, Fongi, Virus
(SARS-CoV-2)

Inappropriate host response

Macrophage reprogramming



Organ failure



Cavaillon *et col*, EMBO Mol Med, Volume: 12, Issue: 4, First published: 16 March 2020

Sepsis animal model definition

SHOCK, Vol. 50, No. 4, pp. 377–380, 2018

OPEN

International Expert Consensus for Pre-Clinical Sepsis Studies

MINIMUM QUALITY THRESHOLD IN PRE-CLINICAL SEPSIS STUDIES (MQTiPSS): AN INTERNATIONAL EXPERT CONSENSUS INITIATIVE FOR IMPROVEMENT OF ANIMAL MODELING IN SEPSIS

Marcin F. Osuchowski,[*] Alfred Ayala,[†] Soheyl Bahrami,[*] Michael Bauer,[‡] Mihaly Boros,[§] Jean-Marc Cavaillon,[‖] Irshad H. Chaudry,[¶] Craig M. Coopersmith,[**] Clifford S. Deutschman,[††] Susanne Drechsler,[*] Philip Efron,[‡‡] Claes Frostell,[§§] Gerhard Fritsch,[‖‖¶¶] Waldemar Gozdzik,[***] Judith Hellman,[†††] Markus Huber-Lang,[‡‡‡] Shigeaki Inoue,[§§§] Sylvia Knapp,[‖‖‖] Andrey V. Kozlov,[*] Claude Libert,[¶¶¶****] John C. Marshall,[††††] Lyle L. Moldawer,[‡‡] Peter Radermacher,[‡‡‡‡] Heinz Redl,[*] Daniel G. Remick,[§§§§] Mervyn Singer,[‖‖‖‖] Christoph Thiemermann,[¶¶¶¶] Ping Wang,[*****] W. Joost Wiersinga,[†††††] Xianzhong Xiao,[‡‡‡‡‡] and Basilia Zingarelli[§§§§§]

[*]Ludwig Boltzmann Institute for Experimental and Clinical Traumatology in the AUVA Research Center

“An experimental animal (mammal) model of sepsis should be defined as life-threatening organ dysfunction caused by a dysregulated host response to an infection.”

Osuchowski ,SHOCK, 2018

Sepsis animal model definition

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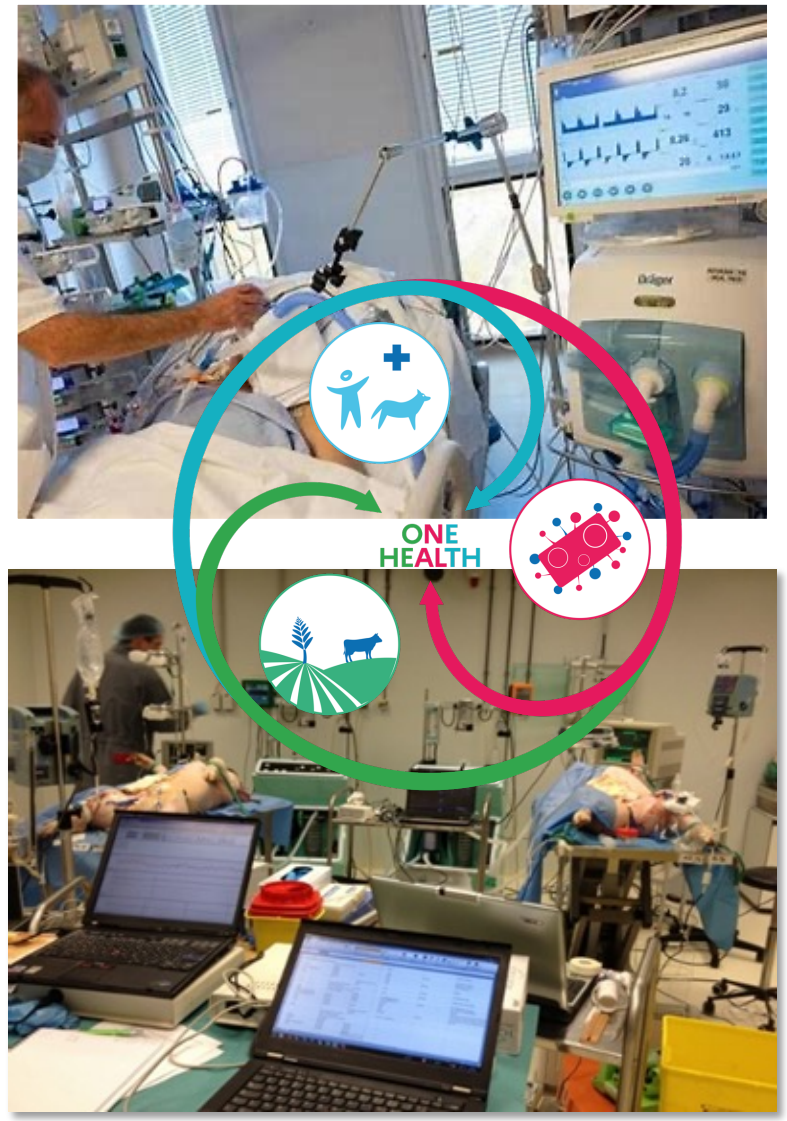

"An experimental animal (mammal) model of sepsis should be defined as life-threatening organ dysfunction caused by a dysregulated host response to an infection."

Osuchowski ,SHOCK, 2018

Animal model what for?

failed to lead to the development of one or more useful pharmacological therapies

BUT…

gain insights into pathophysiology

Agent	Species	Challenge	Design	Animal study result	References
IL1-RA	Mouse	Intraperitoneal LPS	First dose of IL1-RA administered 20 min after LPS challenge and continued every 4 h for 24 h	Benefit	114
Methylprednisolone sodium succinate (MPSS)	Baboon	Viable intravenous *E. coli*	Infusion of MPSS started 2 h after start of bacterial challenge	Benefit	65
IL1-RA	Baboon	Viable intravenous *E. coli*	Continuous infusion of IL1-RA started at same time as bacterial challenge	Benefit	115
Lenercept	Baboon	Viable intravenous *E. coli*	Pre-treatment 1 h prior to bacterial challenge	Benefit	116
CDP571	Baboon	Viable intravenous *E. coli*	Pre-treatment 2 h prior to bacterial challenge	Benefit	117
BN 5021	Mouse	Intravenous LPS	Pre-treatment 30–45 min prior to LPS challenge	Benefit	118
BB-882	Mouse	Intravenous LPS	Pre-treatment 5 min prior to LPS challenge	Benefit	119
TCV-309	Mouse	Intravenous LPS 24 h after intraperitoneal carrageenan	Pre-treatment 30 min prior to LPS challenge	Benefit	120
TAK-242	Mouse	Intraperitoneal viable *E. coli* injected 13 d after priming with intravenous viable *Mycobacterium bovis*	Treatment with ceftazidime and TAK-142 at 1 h after bacterial challenge	Benefit	121
TAK-242	Mouse	Intraperitoneal LPS	Pre-treatment with TAK-242 1 h before LPS *or* post-treatment TAK-242 up to 4 h after LPS challenge	Benefit	122
Tifacogin	Rabbit	Peritonitis caused by *E. coli* O18:K+	Treatment with gentamicin at tifacogin, beginning 4 h after onset of infection	Benefit	59
Tifacogin	Baboon	Viable intravenous *E. coli*	Treatment with tifacogin started at 30 min after bacterial challenge	Benefit	123
Tifacogin	Mouse	Cecal ligation and puncture (CLP)	Treatment with tifacogin started 30–60 min after CLP	Benefit	66

examples of some pharmacological agents, which have been evaluated in an animal model of sepsis and yielded negative results in one or more human clinical trials (Fink, virulence, 2014)

toll-like receptor 4 (TLR4) was initially characterized in mice as the pattern recognition receptor (PRR) for LPS *(Poltorak, Science, 1998)*

characterization of immunosuppressive cells (regulatory T-cells, myeloid-derived suppressor cells) occurred initially in mouse models, before further verification of their importance in human sepsis

Assessment of the roles of tissue and organ immunology in sepsis in animal model vs blood sample in human
→ characterization of compartment-specific immunopathy in sepsis

how to develop a suitable animal model for the study of sepsis?

SHOCK, Vol. 51, No. 1, pp. 23–32, 2019

Review Article

PART II: MINIMUM QUALITY THRESHOLD IN PRECLINICAL SEPSIS STUDIES (MQTiPSS) FOR TYPES OF INFECTIONS AND ORGAN DYSFUNCTION ENDPOINTS

Claude Libert,[*†] Alfred Ayala,[‡] Michael Bauer,[§] Jean-Marc Cavaillon,[‖] Clifford Deutschman,[¶] Claes Frostell,[#] Sylvia Knapp,[**] Andrey V. Kozlov,[††] Ping Wang,[‡‡] Marcin F. Osuchowski,[††] and Daniel G. Remick[§§]

[*]Center for Inflammation Research, VIB, Ghent, Belgium; [†]Ghent University, Ghent, Belgium; [‡]Rhode Island Hospital & Alpert School of Medicine at Brown University, Providence, Rhode Island; [§]Jena University Hospital, Jena, Germany; [‖]Institut Pasteur, Paris, France; [¶]Feinstein Institute for Medical Research, Northwell Health, Manhasset, New York; [#]Karolinska Institutet, Danderyd Hospital, Stockholm, Sweden; [**]Medical University Vienna, Vienna, Austria; [††]Ludwig Boltzmann Institute for Experimental and Clinical Traumatology in the AUVA Research Center, Vienna, Austria; [‡‡]Feinstein Institute for Medical Research, Manhasset, New York; and [§§]Boston University School of Medicine, Boston, Massachusetts

Received 19 Apr 2018; first review completed 18 May 2018; accepted in final form 26 Jul 2018

how to develop a suitable animal model for the study of sepsis? choose the protocol

types of infections:
including the pathogens

the site of infection

measurements of organ injury

SHOCK, Vol. 51, No. 1, pp. 23–32, 2019

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how to develop a suitable animal model for the study of sepsis? choose the protocol

types of infections:
including the pathogens

~~LPS~~

microorganisms => replicate those commonly found in human sepsis

BACTERIA
>Viral > fungus

the site of infection

measurements of organ injury

how to develop a suitable animal model for the study of sepsis? choose the protocol

types of infections:
including the pathogens

~~LPS~~

microorganisms => replicate those commonly found in human sepsis

Response to bacterial strain depend on animal species

BACTERIA
>Viral > fungus

Found similar « diseases » between human and animal

the site of infection

measurements of organ injury

how to develop a suitable animal model for the study of sepsis? choose the protocol

types of infections: including the pathogens

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~~LPS~~

microorganisms => replicate those commonly found in human sepsis

BACTERIA

>Viral > fungus

Use clinical isolate= pathogenic bacteria

strains subcultured long term → lose important patho-physiological characteristics → fail to reflect ‘‘real world’’ pathogenesis

similarities between human neonatal sepsis and the foal sepsis
interest of veterinary spontaneous "*models*" in sepsis

types of infections:
including the pathogens

Received: 25 October 2017 | Revised: 18 December 2017 | Accepted: 31 January 2018
DOI: 10.1111/jvim.15087

STANDARD ARTICLE

Journal of Veterinary Internal Medicine ACVIM Open Access
American College of Veterinary Internal Medicine

Evaluation of updated sepsis scoring systems and systemic inflammatory response syndrome criteria and their association with sepsis in equine neonates

D. M. Wong[1] | R. E. Ruby[1] | K. A. Dembek[1] | B. S. Barr[2] | S. M. Reuss[3] | K. G. Magdesian[4] | E. Olsen[5] | T. Burns[6] | N. M. Slovis[7] | P. A. Wilkins[8]

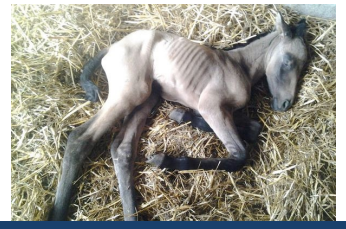


TABLE 1 Blood culture results from 273 neonatal foals presented to 7 referral hospitals during the 2016 foaling season

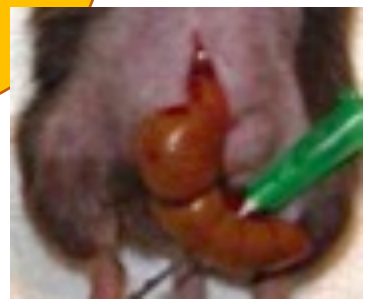
Gram-negative organisms	Number of positive results	Percent of Gram-negative	Percent total positive	Gram-positive organisms	Number of positive results	Percent of gram-positive	Percent total positive
Escherichia coli	15	21.1	11.2	*Staphylococcus* spp	12	19	9
Pantoea agglomerans	12	16.9	9	*Enterococcus* spp	6	9.5	4.5
Actinobacillus spp	9	12.7	6.7	*Staphylococcus spp (coag. neg)*	6	9.5	4.5
Enterobacter spp	7	9.9	5.2	*Streptococcus* (α-hemolytic)	6	9.5	4.5
Klebsiella pneumonia	7	9.9	5.2	*Bacillus spp*	5	7.9	3.7
Agrobacterium spp	4	5.6	3	*Diptheroids*	5	7.9	3.7
Acintobacter spp	3	4.2	2.2	*Streptococcus* Group A	4	6.3	3
Gram-Negative rod	3	4.2	2.2	*Streptococcus* spp	4	6.3	3
Pseudomonas spp.	3	4.2	2.2	*Corynebacterium spp*	3	4.8	2.2
Salmonella spp	3	4.2	2.2	Gram-Positive rod	3	4.8	2.2
Gram-Negative bacilli	2	2.8	1.5	*Leifsonia aquatic*	3	4.8	2.2
Aeromonas	1	1.4	0.7	*Clostridium* spp	2	3.2	1.5
Campylobacter fetus	1	1.4	0.7	*Curtobacterium flaccumfaciens*	1	1.6	0.7
Neisseria	1	1.4	0.7	*Gemella morbillorum*	1	1.6	0.7
Saccharomyces cerevisiae[a]	1	–	–	*Kytococcus sedantarius*	1	1.6	0.7
				Okibacterium fritillariae	1	1.6	0.7
Total	71	100	53		63	100	47

how to develop a suitable animal model for the study of sepsis? choose the protocol

types of infections: including the pathogens

the site of infection

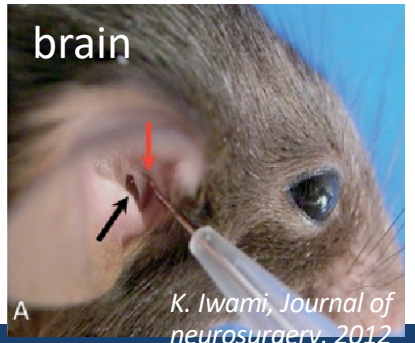
Consider modeling sepsis syndromes that are initiated at sites other than the peritoneal cavity



lung

measurements of organ injury

urinary tract



K. Iwami, Journal of neurosurgery, 2012

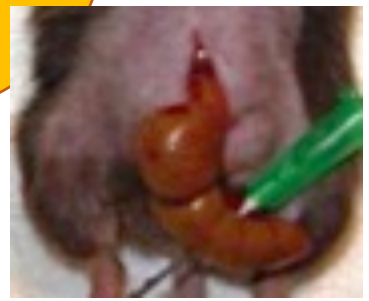
how to develop a suitable animal model for the study of sepsis? choose the protocol

mice were used in 79% of the 2003 to 2012 papers

horse colic a good model of sepsis related to enterotoxemia ?

types of infections: including the pathogens

the site of infection



measurement

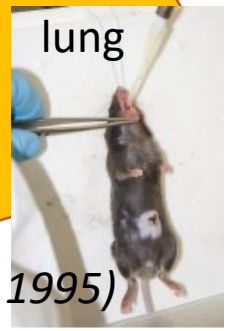
injury

how to develop a suitable animal model for the study of sepsis? choose the protocol

types of infections:
including the pathogens

IL-10

the site of infection



→ deterioration of mice with pulmonary sepsis : Klebsiella pneumonia *(Greenberger, J Immunol 1995)*

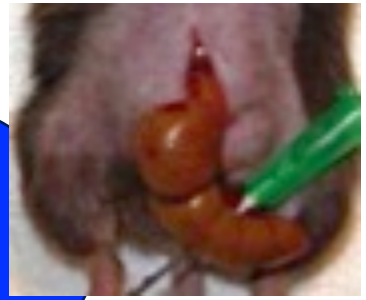
→ Protection abdominal sepsis : CLP *(van der Poll, J Immunol 1995)*

Better targeting of animal models to the questioning

But also

Better targeting of patients for clinical trials

measurements of organ injury



neurosurgery, 2012

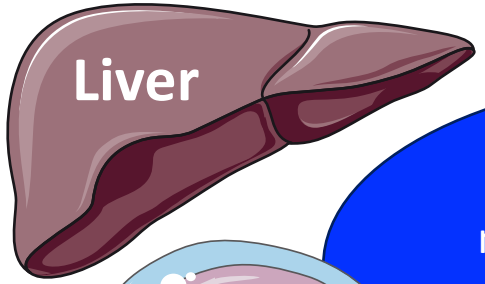
how to develop a suitable animal model for the study of sepsis? choose the protocol

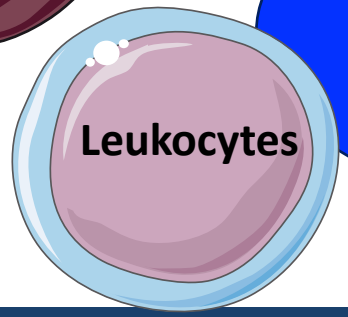
types of infections:
including the pathogens

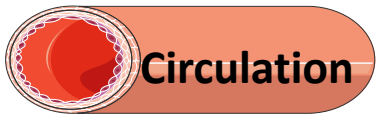
the site of infection

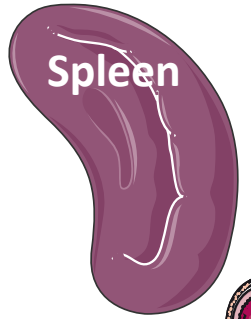
Score SOFA
score Sequential Organ Failure Assessment

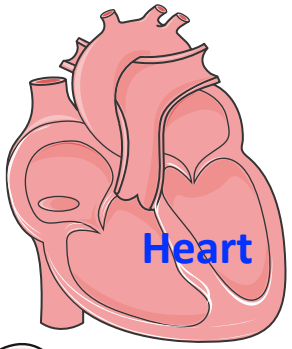
measurements of organ injury

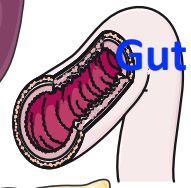


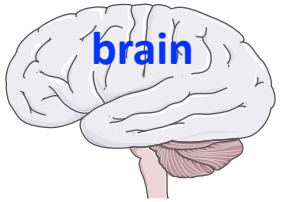




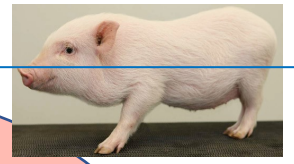








how to develop a suitable animal model for the study of sepsis?
Individual Animal status



farm animals raised in the presence of many germs or animals raised in "clean" laboratories

Age
young animals
vs old patient

Living environment
pre-exposure to specific pathogenic or antigenic stimuli → immune memory

Gender
female more resistant

affect the responsiveness in sepsis
(eg immunity responsiveness)

Immunological status
↙ Sepsis if already immunodeficient or develop early immunosuppression post-infection.

health
healthy vs co-morbidities

pre-existing diseases often produce more heterogenous experimental outcomes, thereby demanding more standardized and elaborate modeling procedures.

Genetic background
Humanised animal model (mouse pig)

Wang, Frontiers in Immunology, 2022

how to develop a suitable animal model for the study of sepsis?
Choice of species

Small animal

→ **low cost, ease to breed, numerous assays**
→ less compound required (€)
→ **biological /immunological tools**
→ technical limitations : invasive procedures very challenging
→ biology, physiology # human

Large animal

→ **resuscitation / positive pressure ventilation / antimicrobial therapy ≈ human**
→ physiologic et biological parameters ≈ human
→ blood volume → serial sampling of blood, tissues (serial biopsies)
→ mirror the thermogenesis response to stress
→ easy surgery and monitoring (existing clinical monitoring equipment)
→ study in serial fashion: haemodynamic, organ function evaluations, haemostasis…
→ test and develop tools to explore organ failure in models close to the clinical situations encountered in patients (haemodynamics, microcirculation, ...)
→ ethical, social challenges / dog
→ less biological /immunological tools
→ high cost (pig << dog)


Jacquet-Lagrèze *et al. Critical Care* 2014, **18**:R153
http://ccforum.com/content/18/4/R153

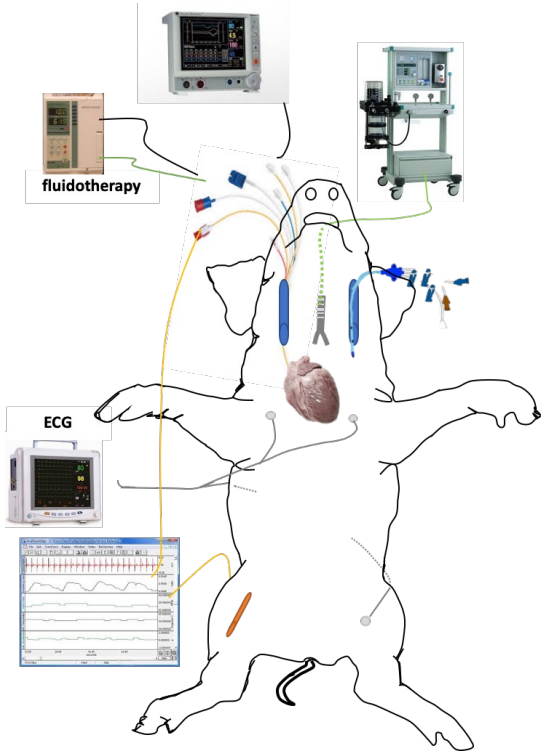
CRITICAL CARE

RESEARCH Open Access

A new device for continuous assessment of gut perfusion: proof of concept on a porcine model of septic shock

Matthias Jacquet-Lagrèze[1*], Jeanne-Marie Bonnet-Garin[2], Bernard Allaouchiche[1,3], Olivia Vassal[1], Damien Restagno[2], Christian Paquet[2], Jean-Yves Ayoub[2], Jérôme Etienne[3,4], François Vandenesch[3,4], Olivier Daulwader[4] and Stéphane Junot[2]

how to develop a suitable animal model for the study of sepsis?
Choice of species: PIG



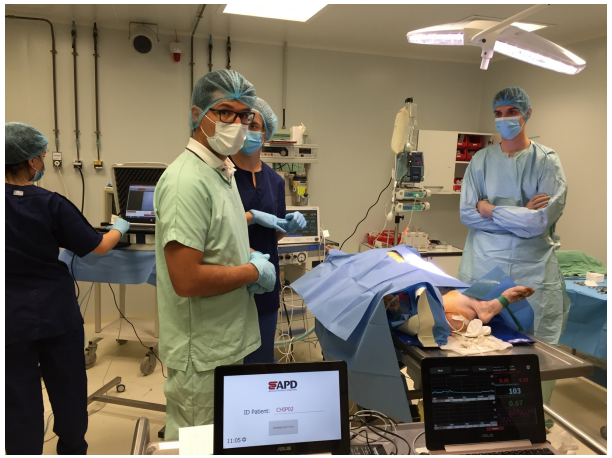
- circulation of the pig has been shown to be most similar to that of humans (except primates)

- lung vascular smooth muscle is very sensitive to live bacteria and endotoxin → increase in pulmonary vascular resistance

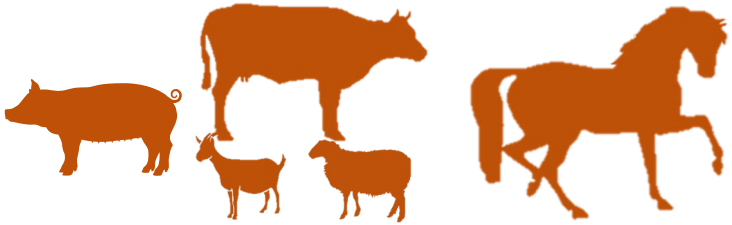
- Increased pulmonary vascular resistance leads to hypodynamic circulation in porcine sepsis models, unlike the hyper dynamic circulation seen in human septic shock.

- Hyperdynamic porcine models are characterized by long observation periods with the first cardiac output measurements after six hours.

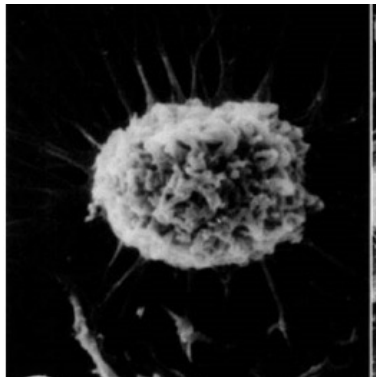
- risk of malignant hyperthermia during long sedation with halogenated drugs



Example of comparative pathophysiology



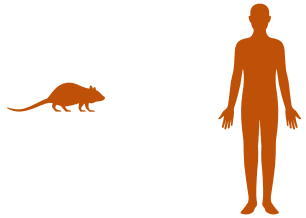
Constitutive



Porcine
(Morton, J Leukocyte Biol 1988)

Pulmonary intravascular macrophages (PIMs)

PIMs are pro-inflammatory



Induced in response to endotoxins and bacteria

how to develop a suitable animal model for the study of sepsis?
Choice of species: SHEEP

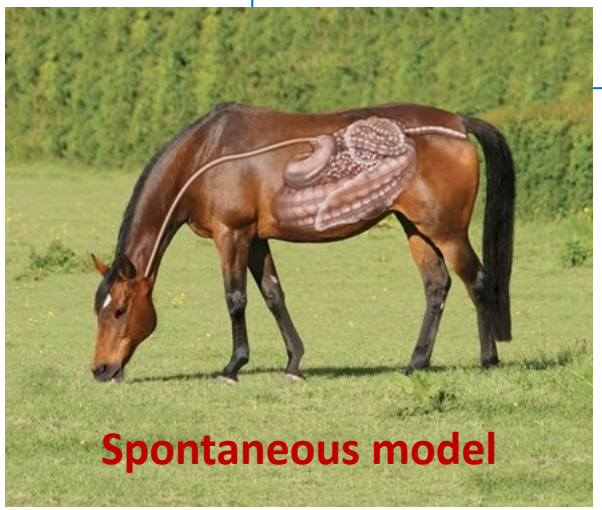
- docile large animals.

- extremely sensitive to LPS like humans: continuous infusion of LPS at a rate as low as 9 ng/kg per h → changes in pulmonary arterial pressure, cardiac output, and lung microvascular permeability

- True sepsis is induced in sheep in a variety of ways: (e.g: infusing viable *Pseudomonas aeruginosa* i.v)

- Ruminant : herbivorous + risk of free gas bloat during long anaesthesia in dorsal decubitus

how to develop a suitable animal model for the study of sepsis?
Choice of species: HORSE



although horse also lives with microorganisms they are very sensitive to endotoxins which makes it very susceptible to sepsis.

Moore, Vet Clin North Am Equine Pract, 2014.

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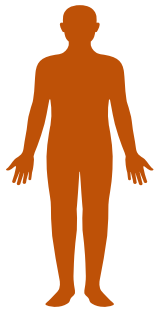
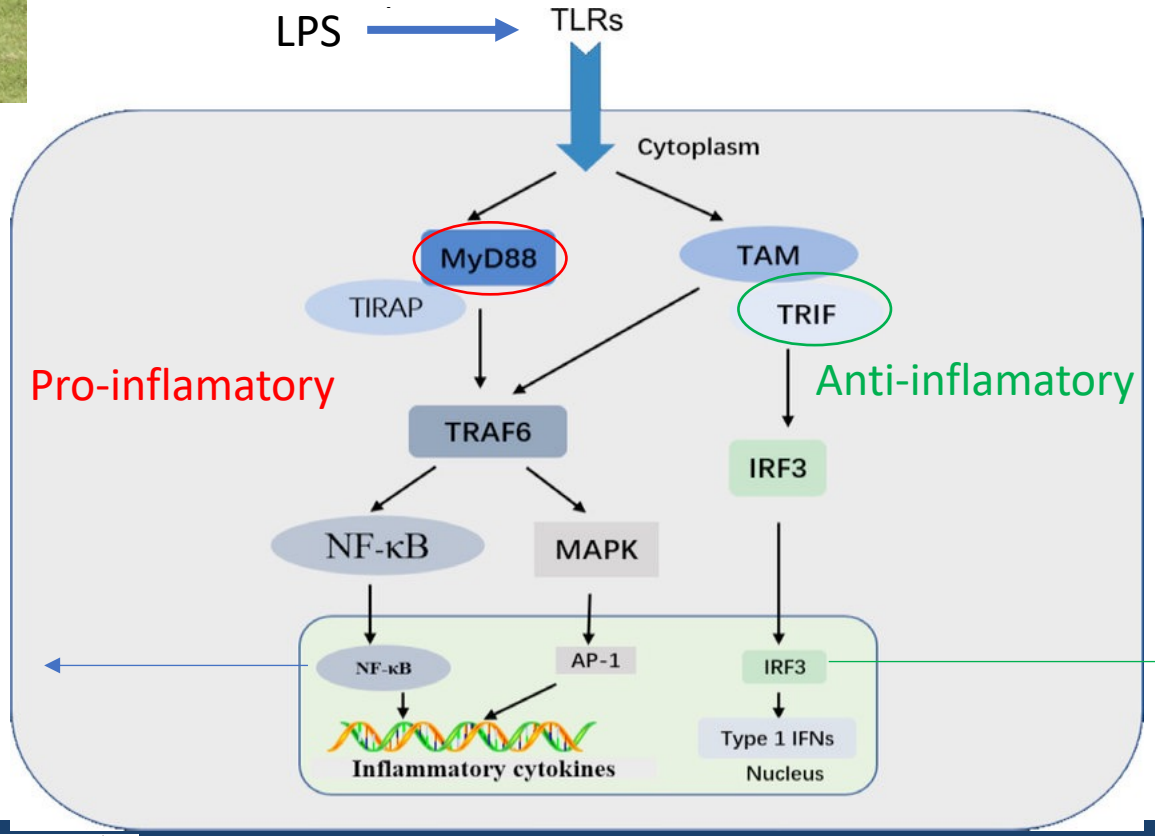
HUMAN





TNFα, IL-1β, IL-6

IL-10 and IL-1Rα

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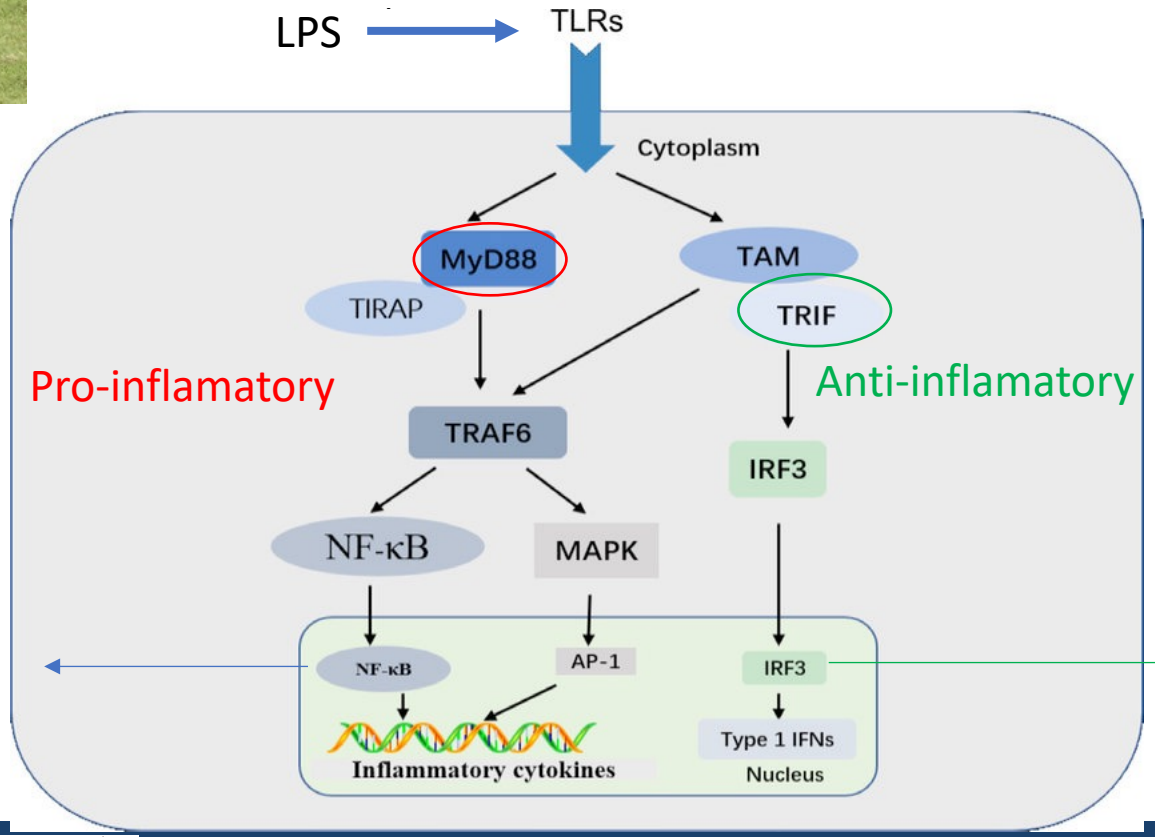

TNFα, IL-1β, IL-6

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In other species, PI(3)Kδ regulates the switch in TLR4 signaling between MyD88 and TRIF, but this event does not seem to occur in equine monocytes.

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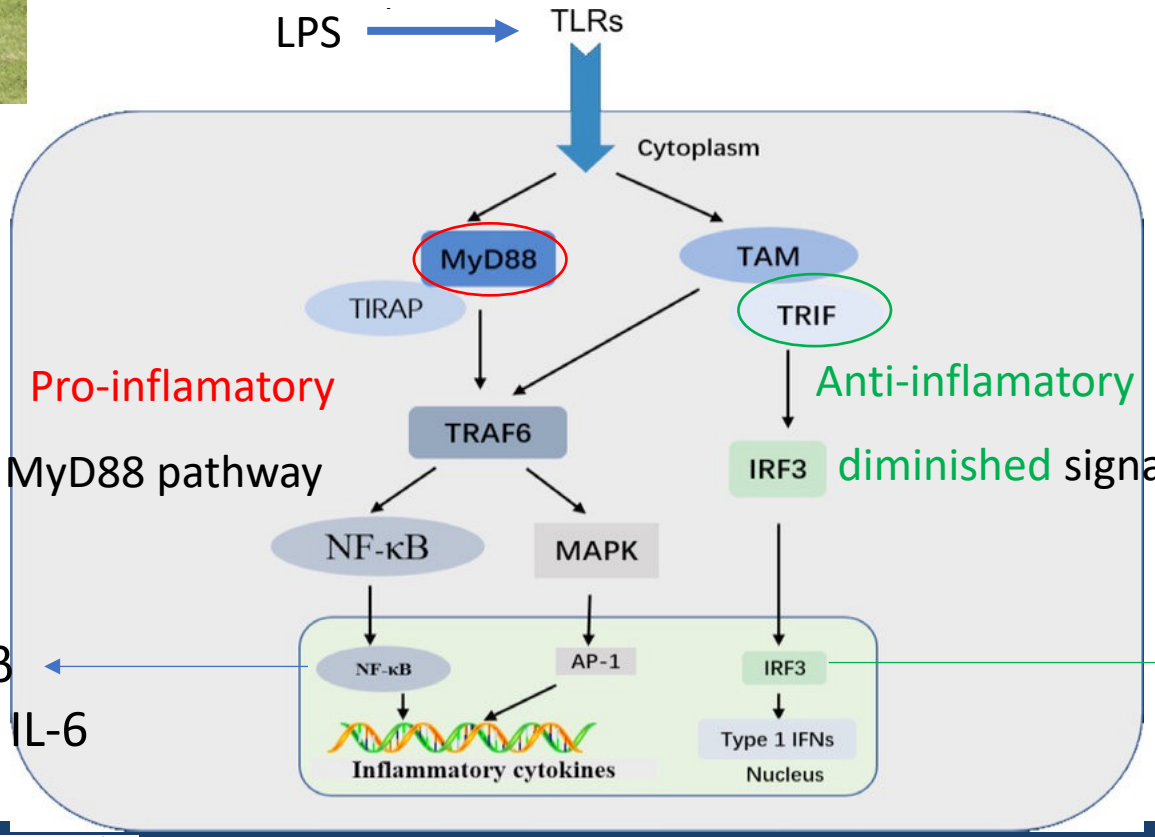

stronger signaling from the MyD88 pathway

diminished signaling from the TRIF pathway

short-lived TNFα

rapid and more sustained IL-1β

slower but sustained induction IL-6

IL-10 induced, but low expression

Moore, Vet Clin North Am Equine Pract, 2014.

how to develop a suitable animal model for the study of sepsis?
Choice of the structure→ ICU for Preclinical septic shock research what for ?

Current models do not mimic the monitoring of septic shock in intensive care units for humans

Need for mid-longer term understanding of the pathophysiology of sepsis

Study the onset and progression of organ dysfunction

current equipment and protocols make maintaining animals in an ICU setting challenging due to complications related to mechanical ventilation

pigs being particularly susceptible to develop impaired lung function
+ pronounced acute pulmonary hypertension

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high cost of 24/24 structures

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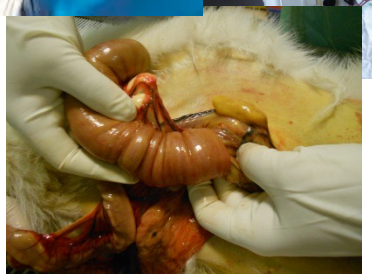
Conclusion: share the skills of professionals from different fields



Animal Sepsis

questions
???????

Exchanges of practices

Translational research

Canine
Feline
equine

Answers

Human Sepsis

To explore alterations + develop new therapeutics

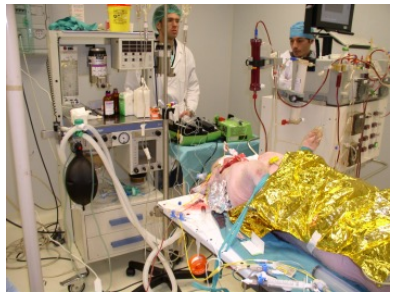
doctors and veterinarians

experimental and spontaneous models

ovine

murine

swine



Conclusion

As there is not one sepsis but many sepsis, there are also many animal models.

Animal model must be chosen to be adapted to the scientific questioning

Choice:

Pre-clinical models : large animal / murine models
Veterinary spontaneous clinical models

Species

For the same species the individual status (strain, gender, age, comorbidity, living environment,...)

Double-blind

The procedure: pathogens, route of entry, organ damage assessment and resuscitation / antimicrobial therapy