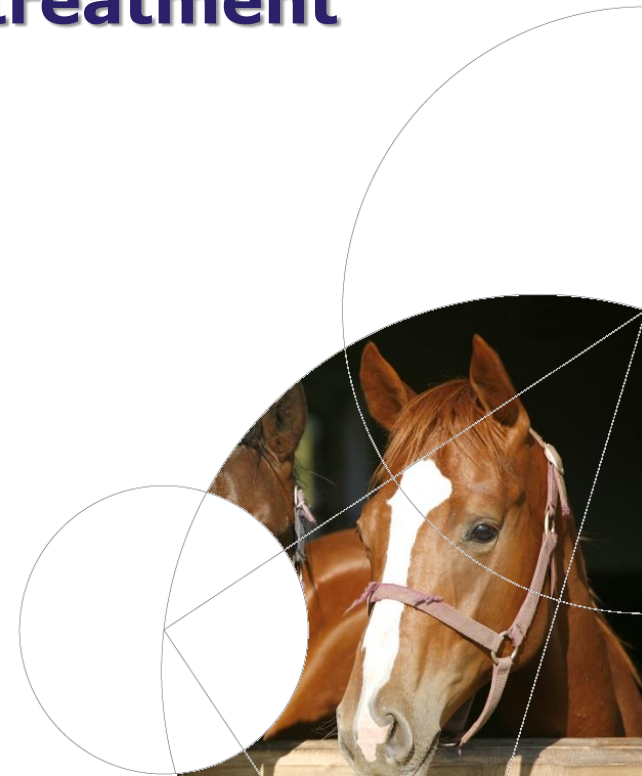




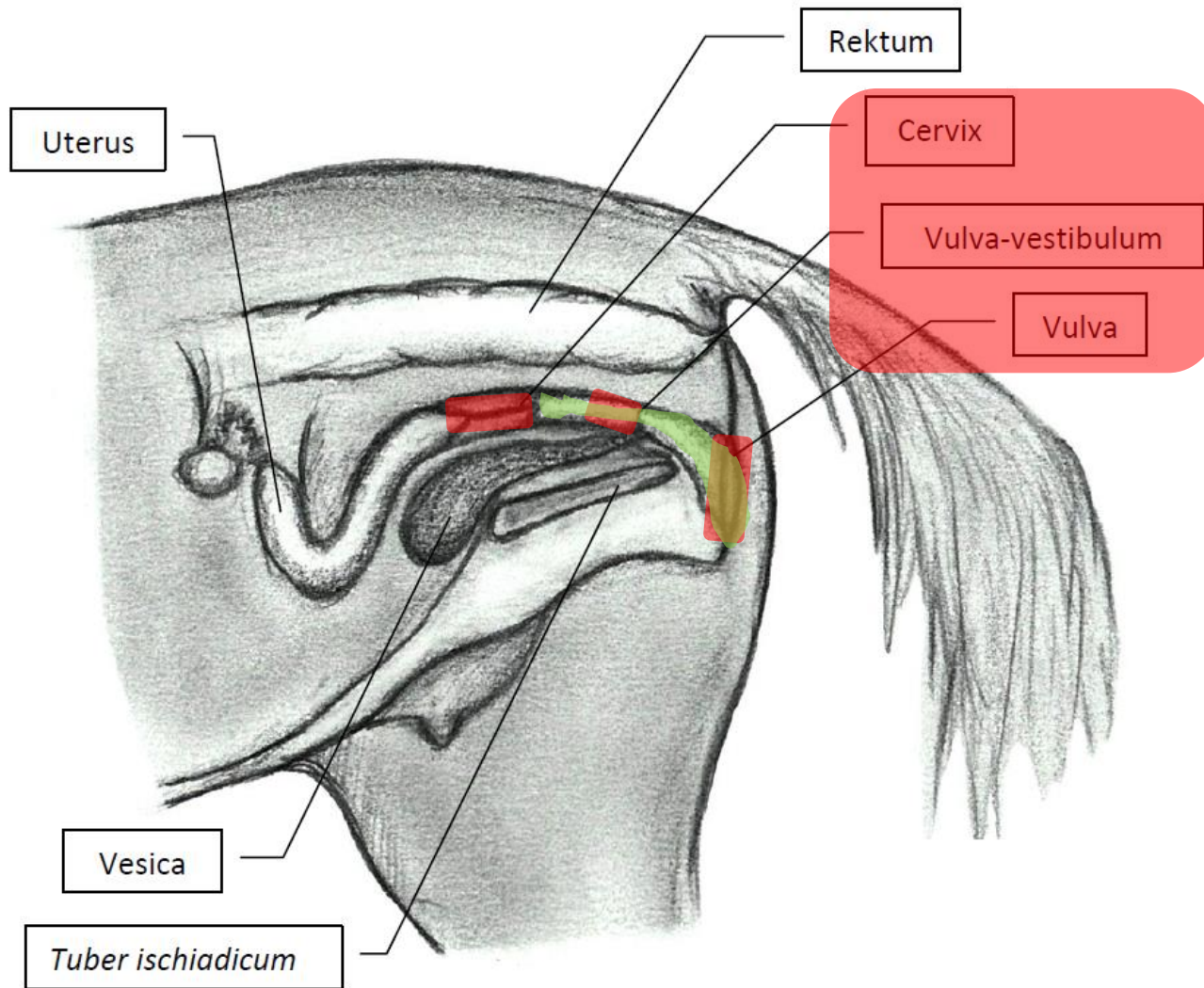
High prevalence of latent endometritis in problem mares – effect of activation and treatment

Anders Miki Bojesen, DVM, PhD, Dipl. ECPVS
Professor of Preventive Veterinary Microbiology
Veterinary Clinical Microbiology

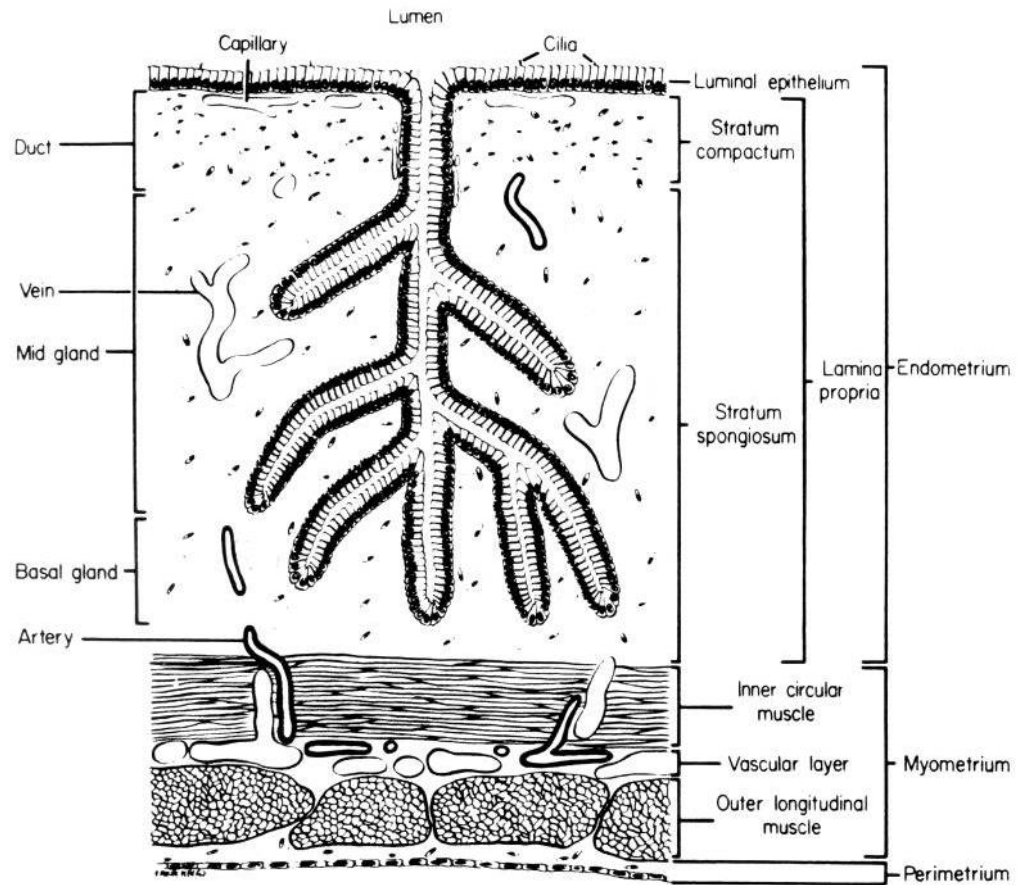
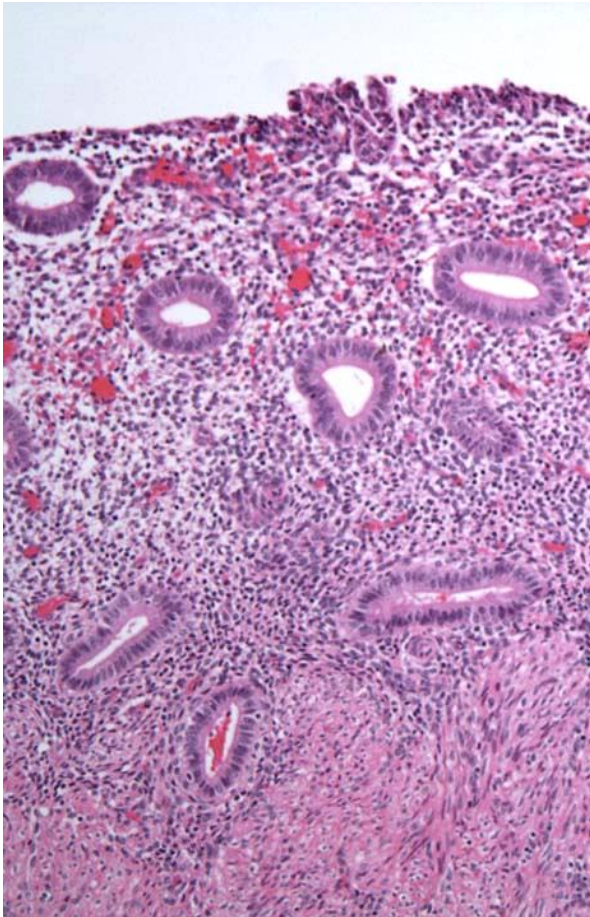
Department of Veterinary and Animal Sciences



The reproductive tract in mares

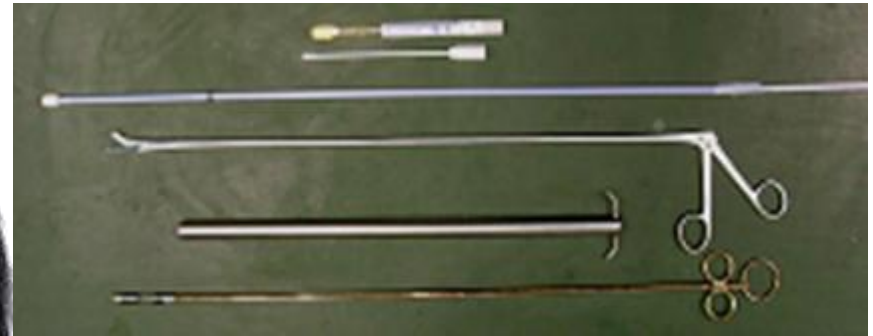
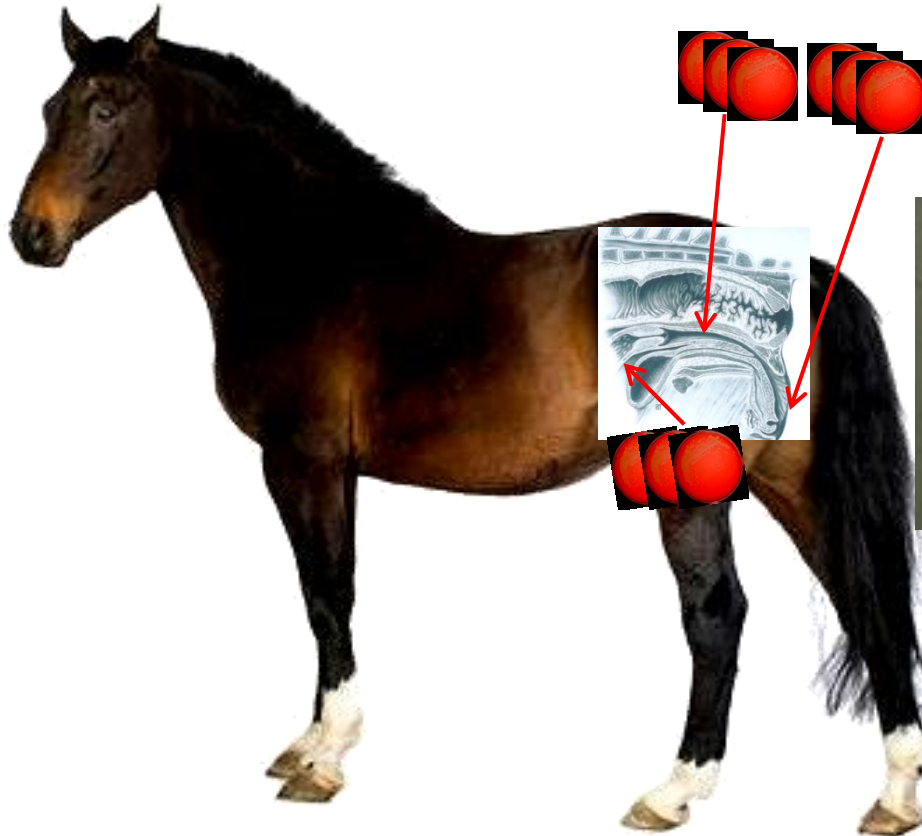


Streptococcus equi subsp. *zooepidemicus*



Most frequent cause of reproductive tract infection

Bacterial subpopulations in different anatomical s

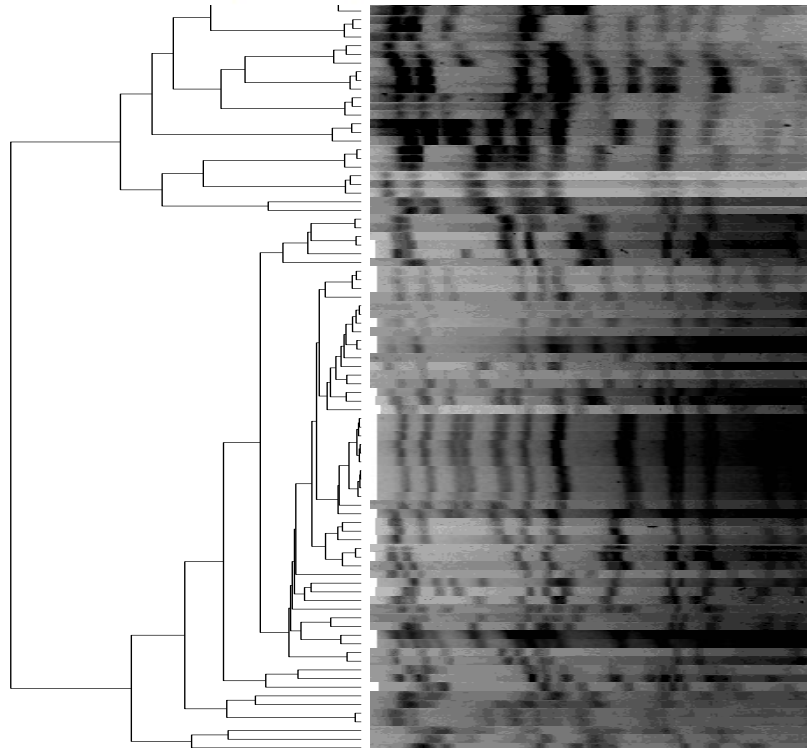


RESEARCH

Open Access

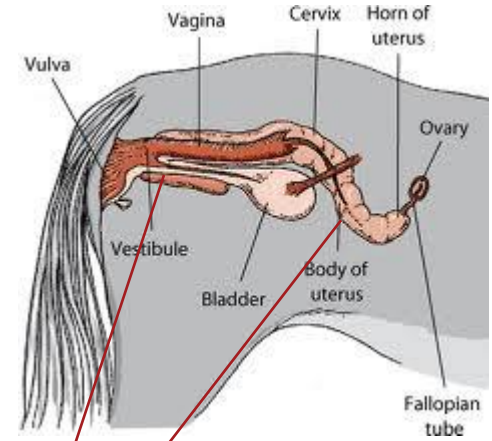
Streptococcus equi subsp. *zooepidemicus* isolates from equine infectious endometritis belong to a distinct genetic group

Camilla Dooleweerd Rasmussen¹, Maria Mathilde Haugaard², Morten Roenn Petersen³, Jesper Møller Nielsen⁴, Hanne Gervi Pedersen¹ and Anders Miki Bojesen^{5*}

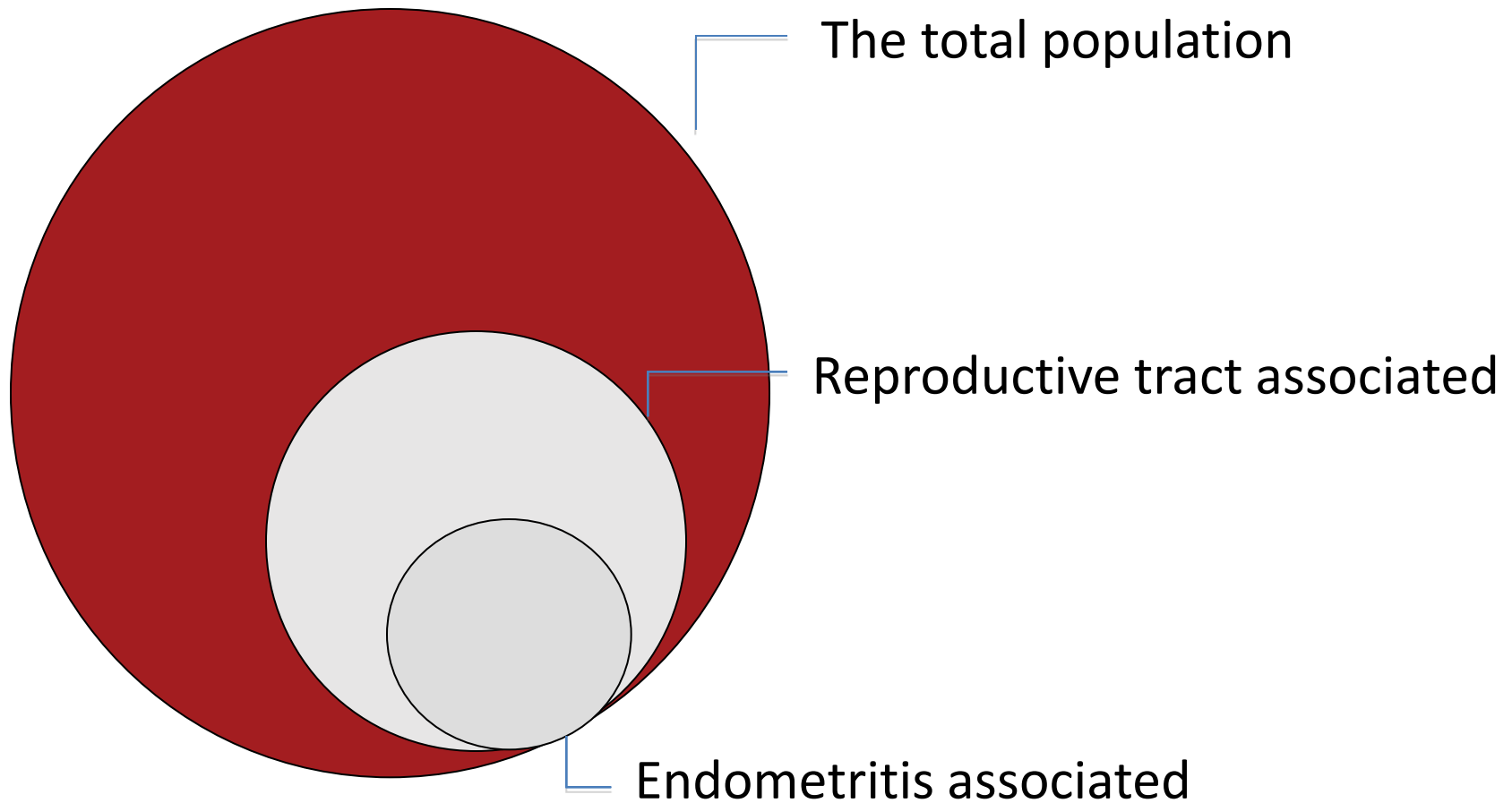


Endometritis isolates

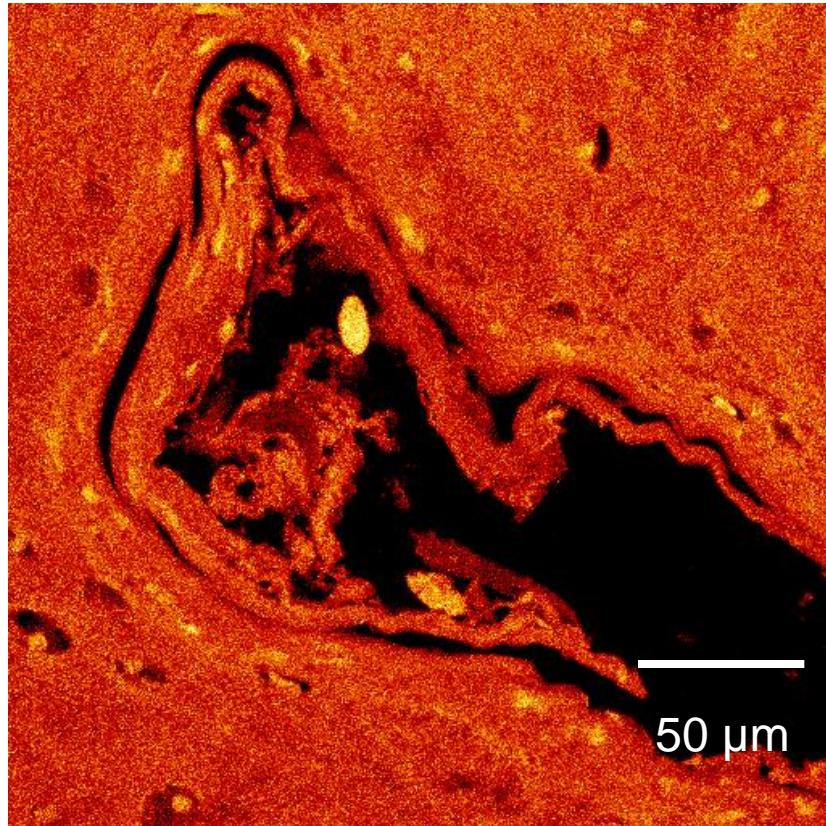
Lower reproductive tract



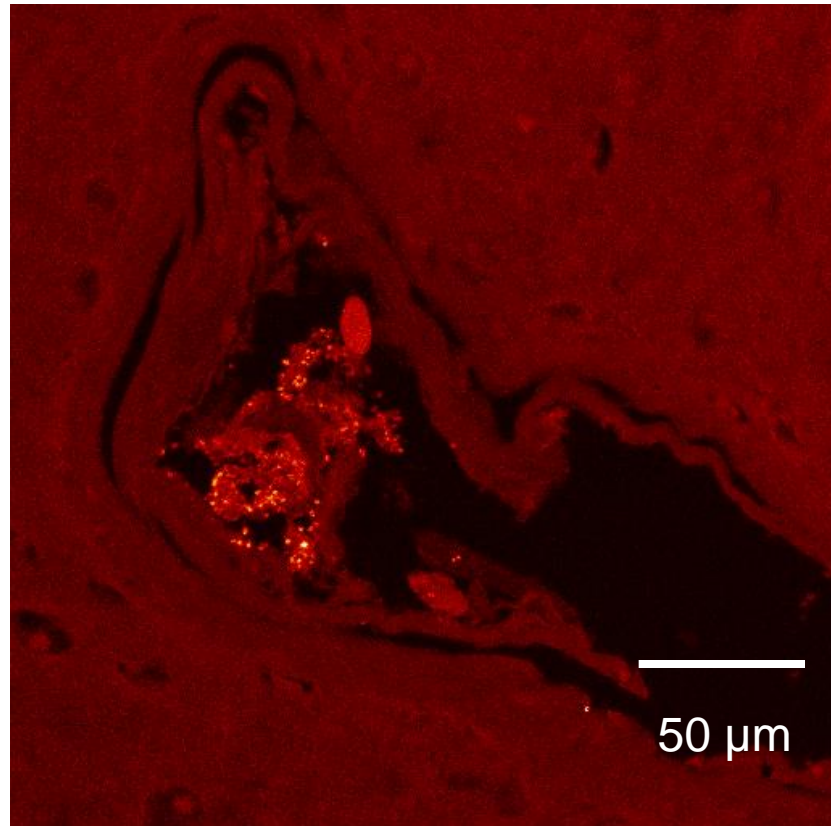
S. zooepidemicus subpopulations



Spatial distribution of *S. zooepidemicus* -confocal microscopy

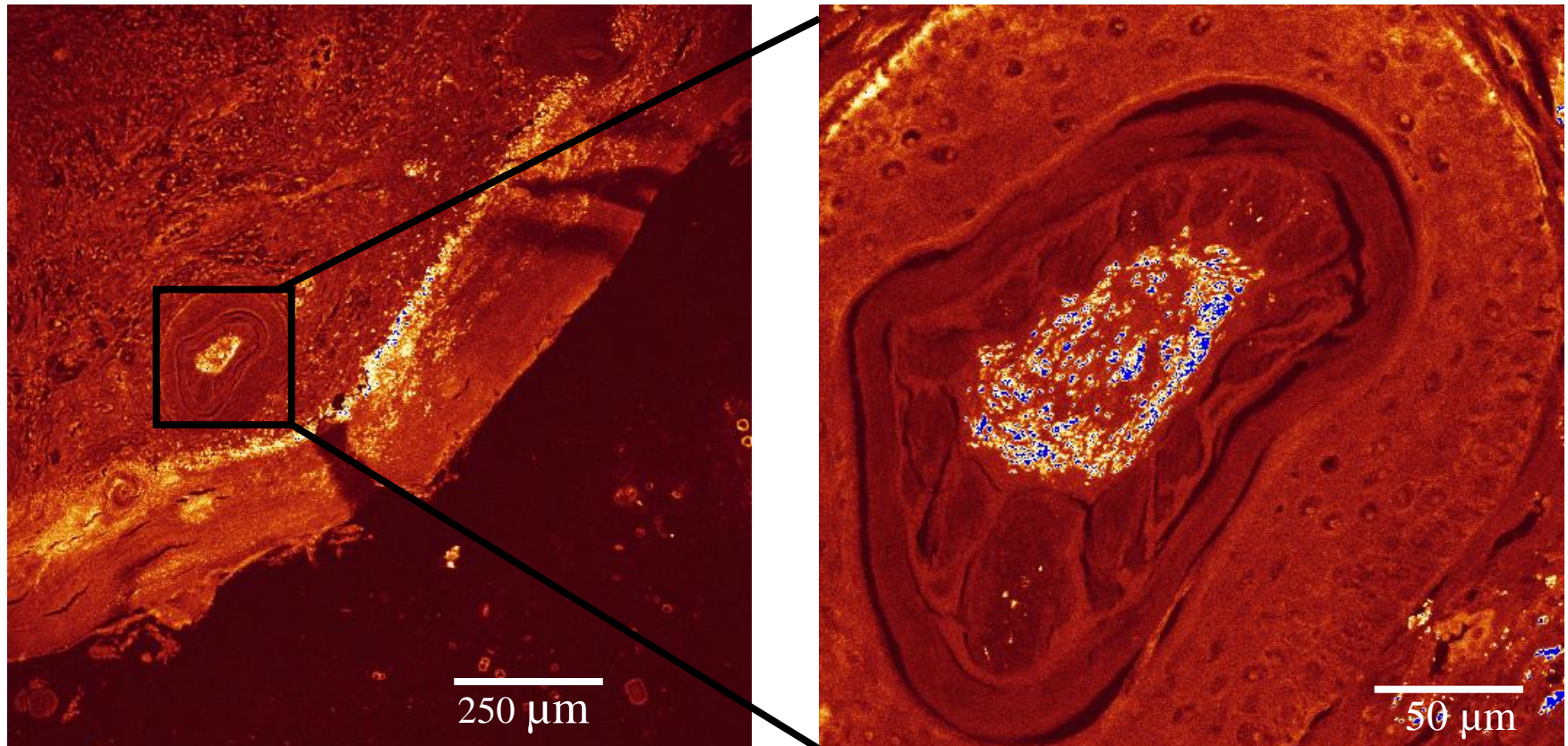


Endometrial gland, background, (15 sections)



Specific Streptococcal Probe CY3, (15 sections)

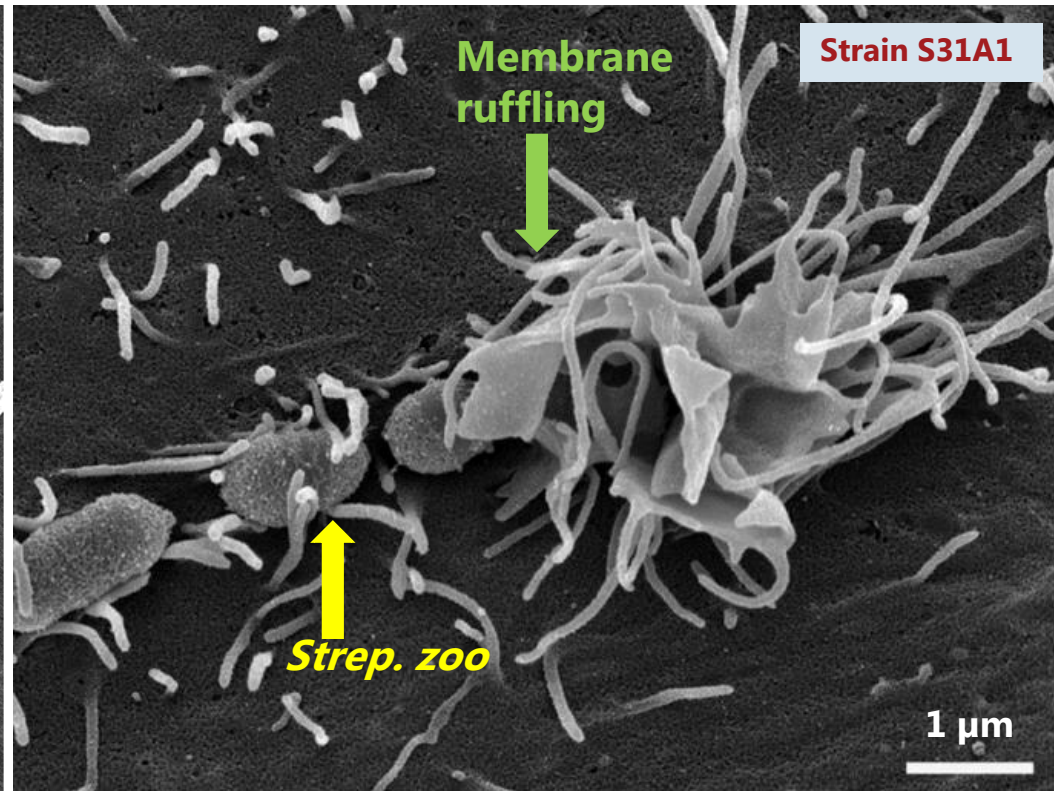
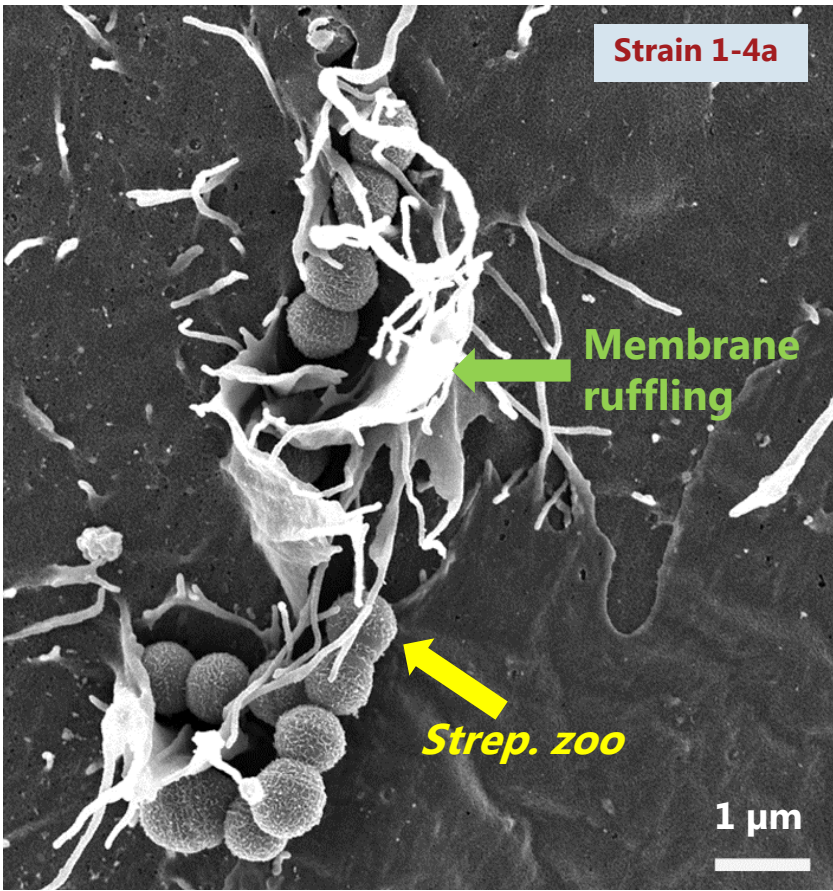
Clinical cases – localization below luminal epithelia and in uterine crypt



(Petersen *et al.*, 2009, *Clin. Theriogenology*)

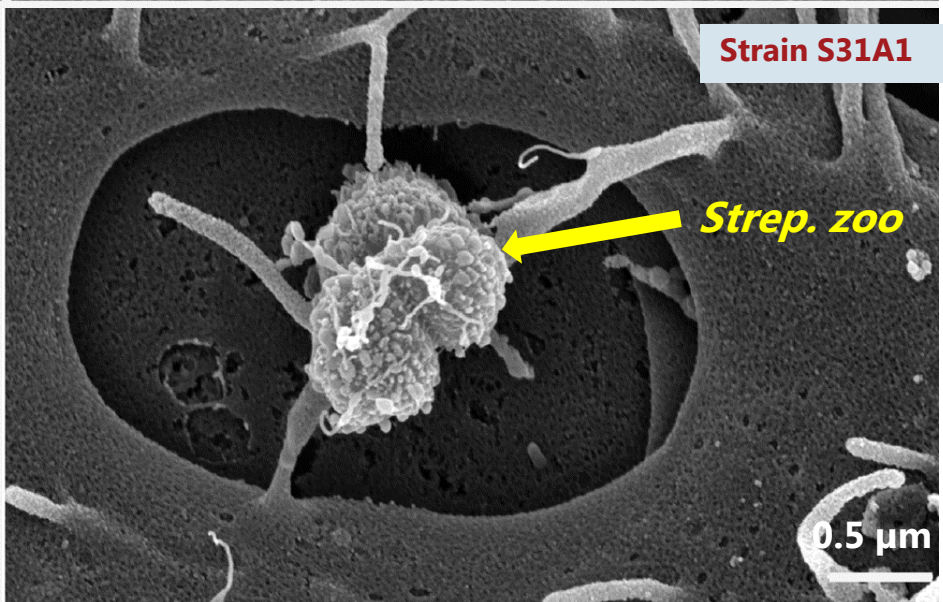
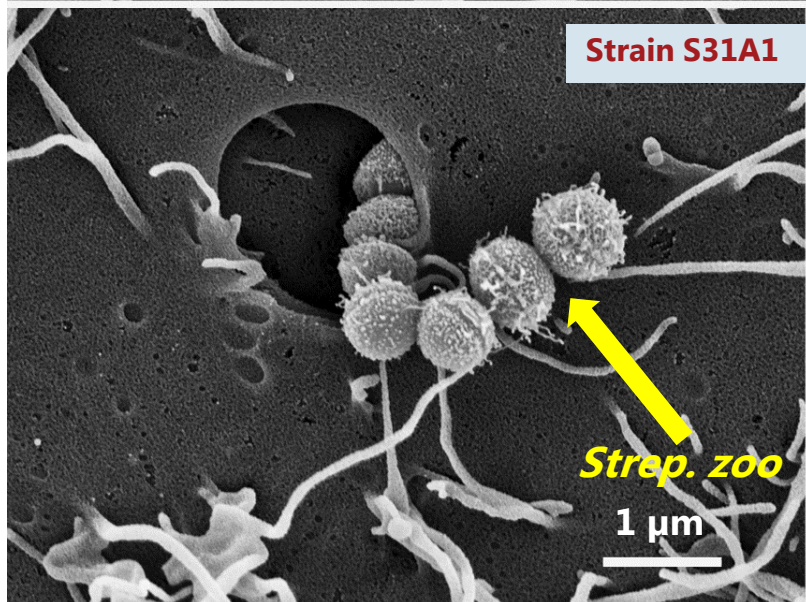
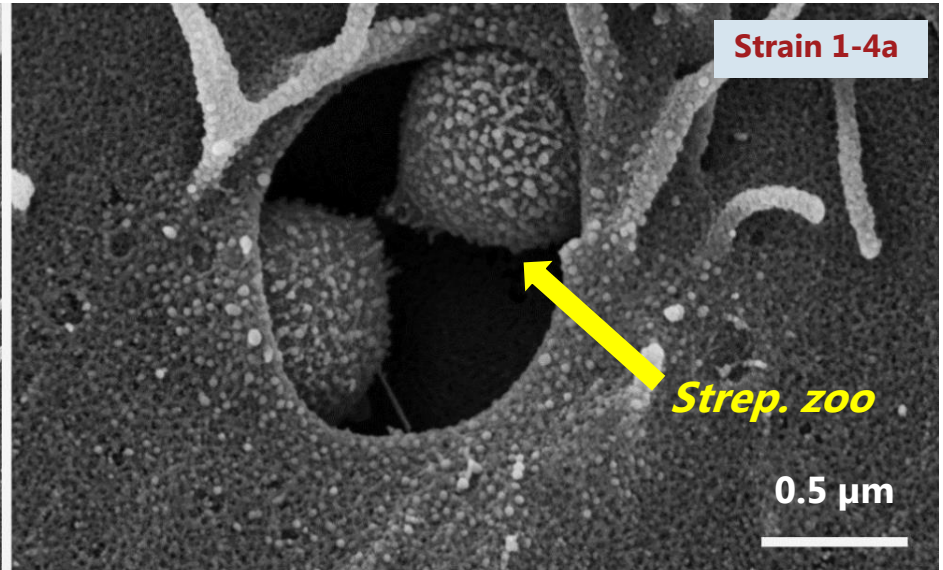
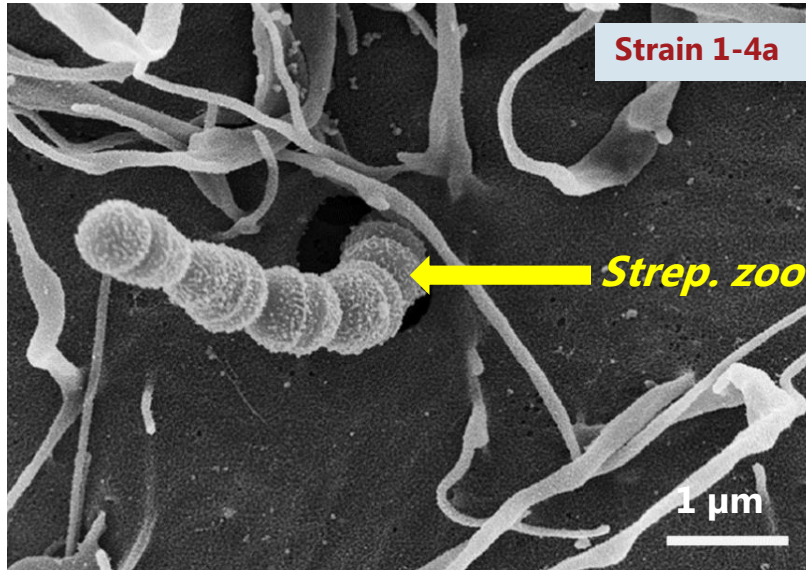
Epithelial adherence and invasion

- Membrane ruffling/trigger mechanism



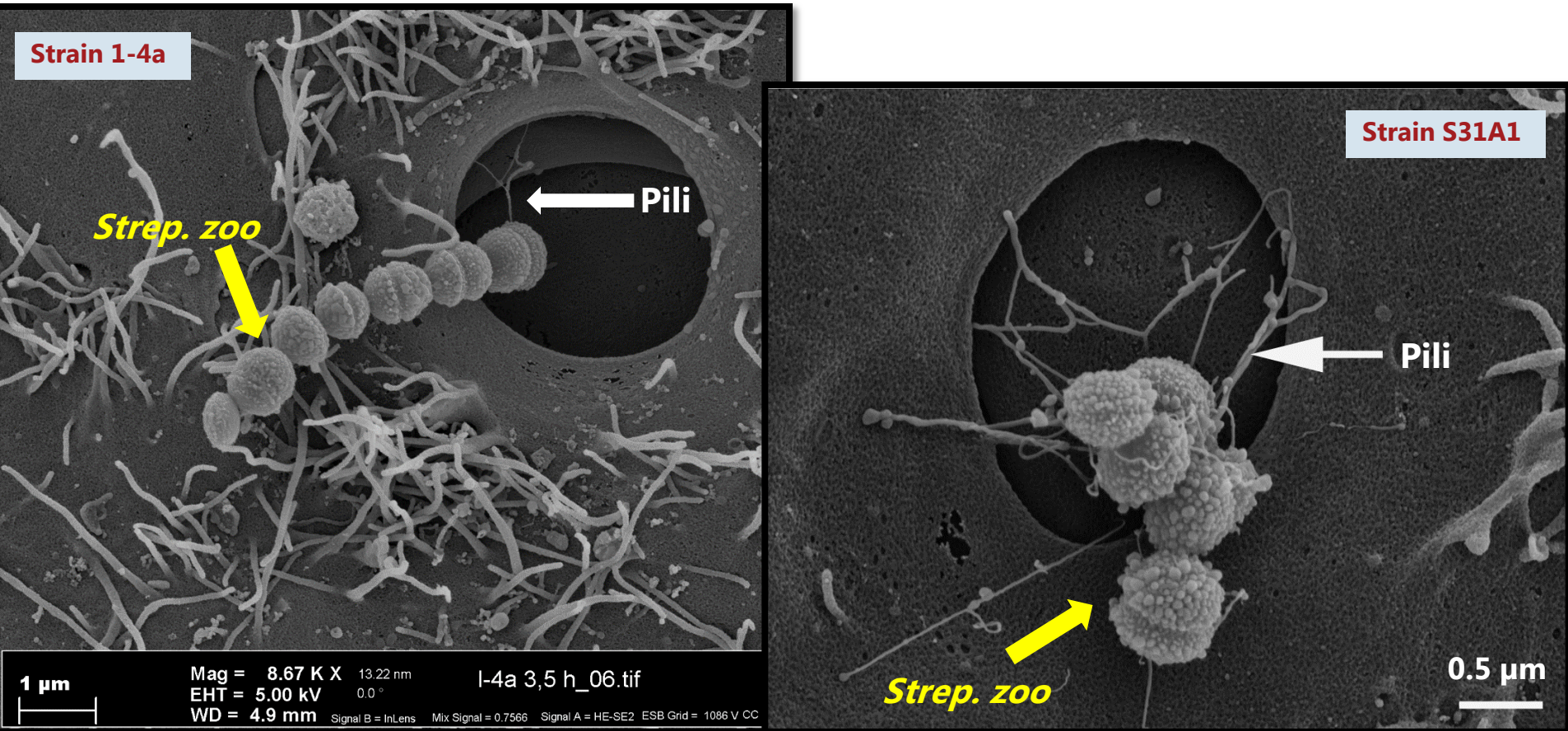
Epithelial adherence and invasion

- Large invaginations in host cell membrane/caveolae-like uptake

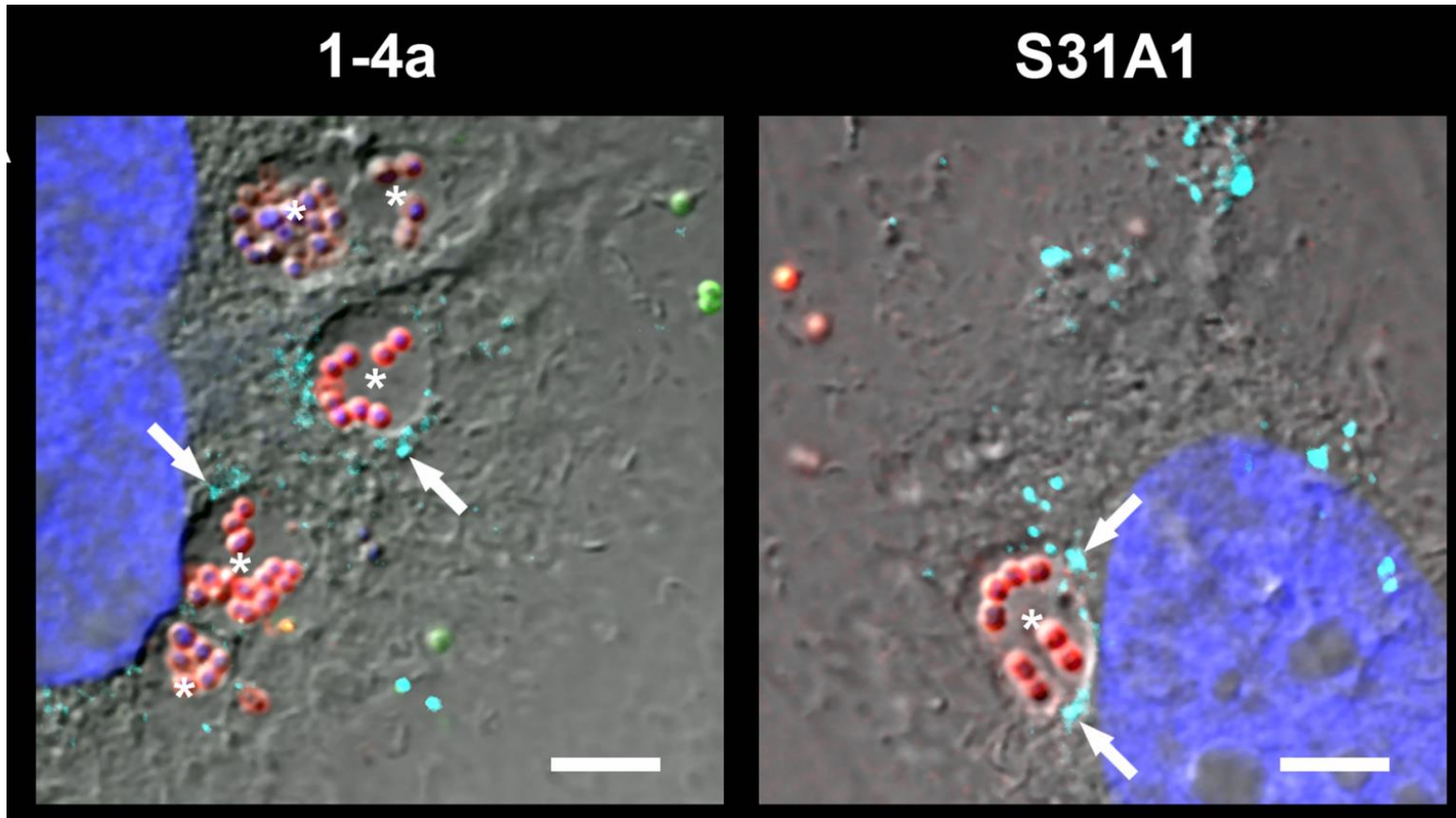


Epithelial adherence and invasion

- Pili-like appendages

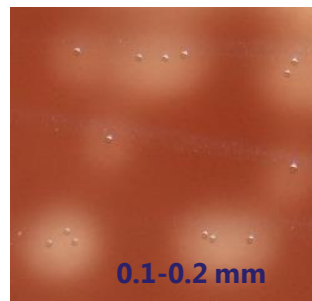
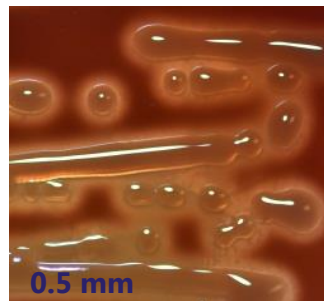
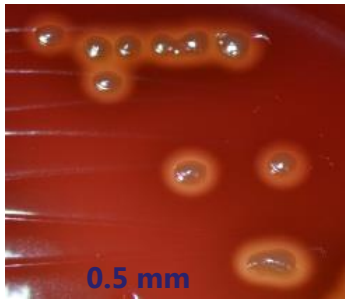
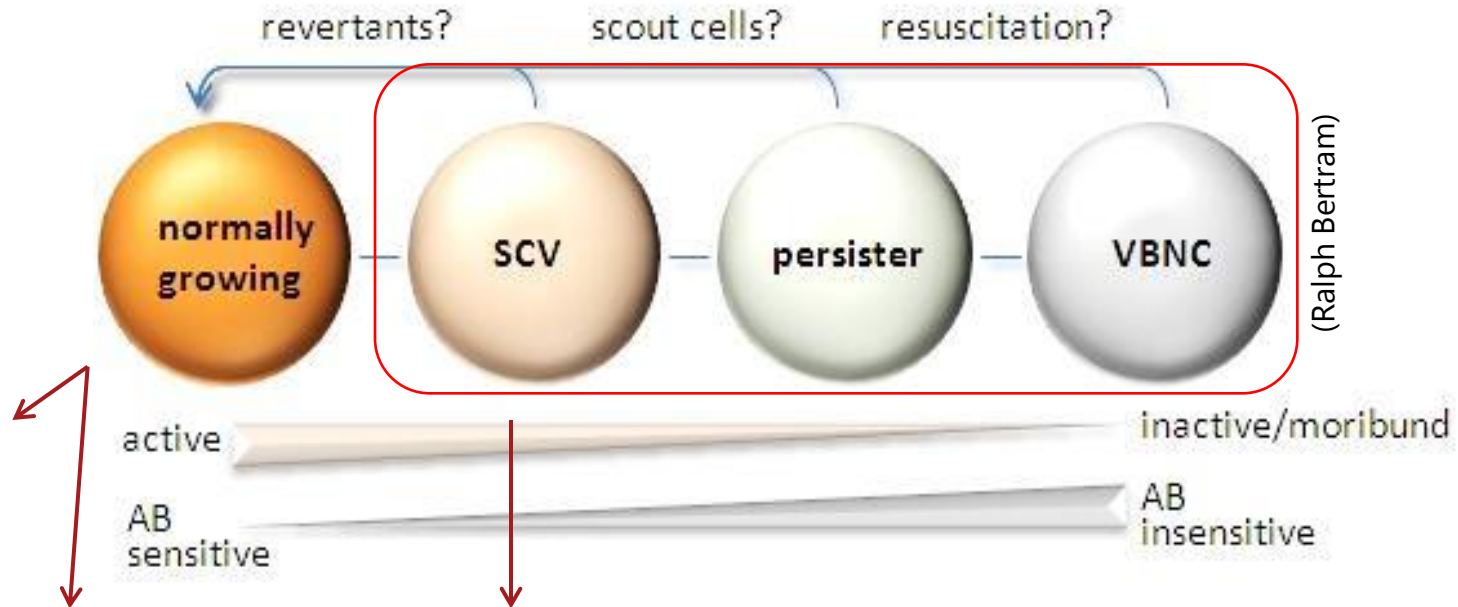


Intracellular trafficking



Lysosome Associated Membrane Protein-1 (LAMP-1)

Phenotypic variants of *S. zooepidemicus*

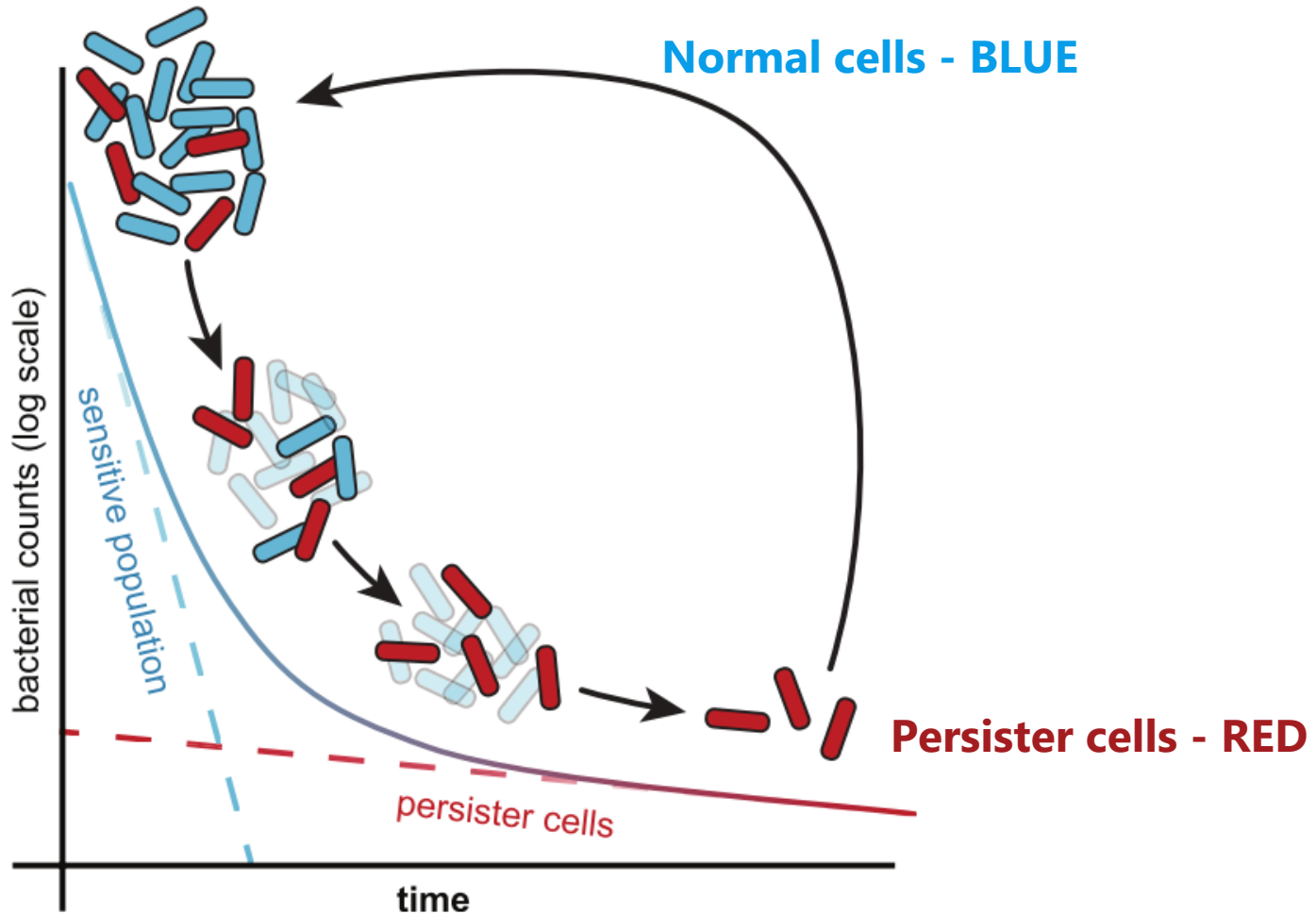


Parkins, Nature, 457, 1083

(Egle Kudirkiene)

What are persister cells?

Tolerant – transient - not resistant



(Harms *et al.*, 2016, *Science*)

Activation of dormant uterine *S. zooepidemicus*

- Instillation of 10 ml PBS or 10 ml bActivate (resuscitation medium)
- Culture 48 h post instillation



	<i>S. zooepidemicus</i> positive	<i>S. zooepidemicus</i> negative	
bActivate	17	17] p < 0.02
PBS	1	34	
	18		



- **One of the first studies to show resuscitation of persisters *in vivo***

Activation of persistent *Streptococcus equi* subspecies *zooepidemicus* in mares with subclinical endometritis
 M.R. Petersen^{a,1,*}, B. Skive^{b,1}, M. Christoffersen^c, K. Lu^d, J.M. Nielsen^e,
 M.H.T. Troedsson^f, A.M. Bojesen^b



Do latent infections affect fertility?

Baseline data from a large Irish stud farm

- 3743 estrous periods of 2385 mares

Risk factors associated

- **Pregnancy rate per breeding cycle**
 - Old age ($p < 0.001$)
 - Cover within 20 days post-partum ($p < 0.003$)
- **Pregnancy loss**
 - High mare age ($p < 0.05$)
 - Barren, slipped or rested reproductive status ($p = 0.05$) increased the likelihood of pregnancy loss.

Reproduction in Domestic Animals

Reprod Dom Anim 51, 181–187 (2016); doi: 10.1111/rda.12655
ISSN 0936-6768

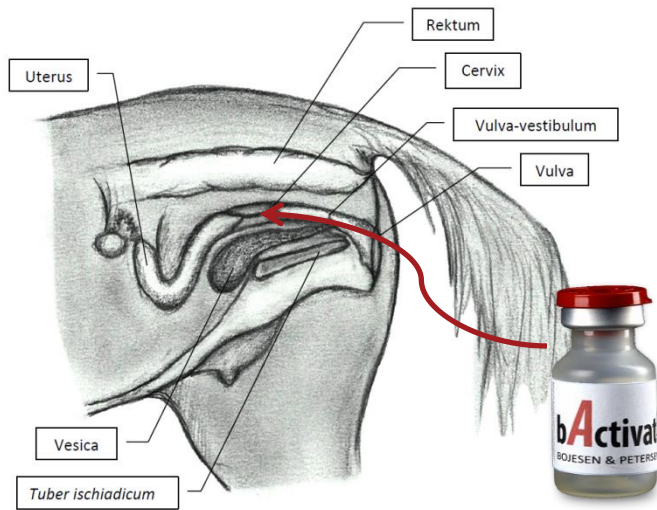
Key Factors Affecting Reproductive Success of Thoroughbred Mares and Stallions on a Commercial Stud Farm

EA Lane¹, MLJ Bijnen², M Osborne³, SJ More^{1,2}, ISF Henderson⁴, P Duffy⁵ and MA Crowe²

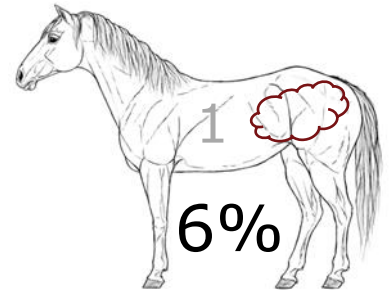
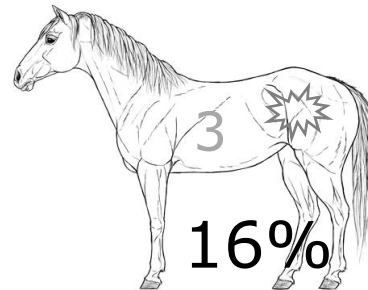
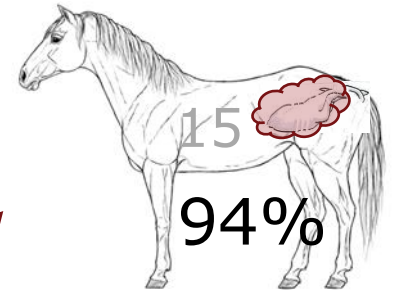
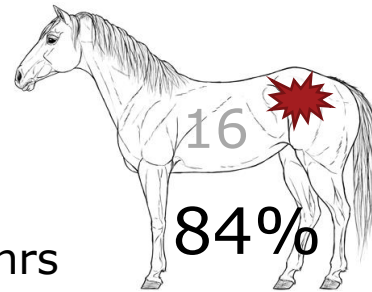
Latent infections and fertility



- 19 problem mares were included



48 hrs



Effect of latent infections on fertility

No. mares	Age	Barren Years/Cycle	Activation (+/-)	Covers post- activation	Pregnant/ Foaled
19	12.7 yrs	Years barren ^a : 1.4 Cycles barren ^b : 6.3	16/19= 84%	1.1 ^c	15/13= 87%

No. mares		Pregnancy rate per cycle	Featal loss rate
19		16/18 = 89%	15/13 = 13%
888	Lane <i>et al.</i> Reproduction in Domestic Animals 2016;51:181-187	61%	19%

S. zooepidemicus

- is highly adapted to the equine reproductive tract
- can survive intracellularly
- dormant cells can persist for extended periods of time
- Negatively affect establishment and maintenance of pregnancy
- bActivate allows detection and treatment



Do latent infections affect fertility?

Year	Mare ID	Age	Barren Years/Cycle	Activation (+/-)	Covers post-activation	Pregnant/ Foaled
2015	1	9	1 yr/ 9 cycls	+	1	+/+
	2	8	1 yr/ 6 cycls	+	1	+/+
	3	17	2 yrs	+	Not bred	Not bred
	4	6	3 yrs	+	1	+/-
	5	11	2 yrs	+	1	+/?
	6	9	1 yr	+	2	+/+
	7	11	2 yrs	+	1	+/-
	8	?	1 yr	+	1	-/-
	9	19	2 yrs	-	3	-/-
2016	1	13	2 yrs /8 cycls	+	2	+/+
	2	14	2 yrs/8 cycls	+	1	+/+
	3	12	1 yr	+	1	+/+
	4	9	1 yr/5 cycls	+	1	+/+
	5	13	1 yr/5 cycls	+	1	+/+
	6	19	2 yrs/7 cycls	+	1	+/+
	7	21	1 yr/3 cycls	+	1	+/+
	8	14	1 yr/5 cycls	-	1	+/+
	9	14	4 cycls	-	1	+/+
	10	9	1 yr/4 cycls	+	2	+/+

Total / average

19 mares

12.7 yrs

**Years barren^a: 1.4
Cycles barren^b: 6.3**

**16/19=
84%**

1.1^c

16/18= 89%

^a27/19=1.4 years. ^b(1y=4 cycles) 120/19=6.3 cycles.

^c19 covers post activ / 17 pregnancies: 19/17=1.1. ^d2016 season:preg at +150 days