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Faculty of Veterinary Science

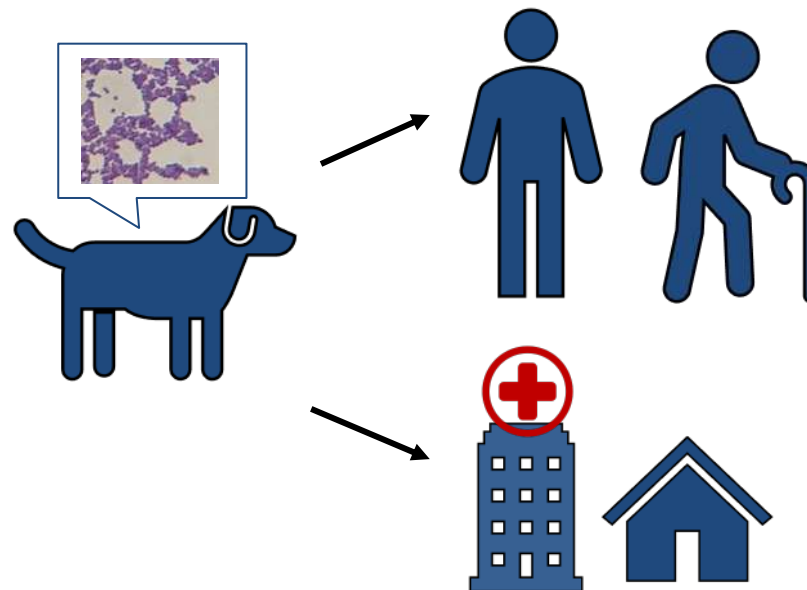
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# Antimicrobial resistance and genomic analysis of bacterial zoonosis in animals, One-health approach

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# *Staphylococcus pseudintermedius*

- Coagulase-positive staphylococci (CoPS)
- Commonly found in companion dogs
- Can cause opportunistic infections
- Methicillin-resistance *S. pseudintermedius* (MRSP)
- Zoonotic pathogen
- Contaminates the environment around dog areas



# *Staphylococcus pseudintermedius*

- The **surface proteins** involved in host binding and colonization
- Developing rapid multiplex PCR tools for detecting surface proteins (**sps**) genes that responsible for colonization and infection in different hosts
- spsP** and **spsQ** found more frequently in canine clinical isolates than in human or environmental isolates
  - May play a role in pathogenicity

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Development of a set of multiplex PCRs for detection of genes encoding cell wall-associated proteins in *Staphylococcus pseudintermedius* isolates from dogs, humans and the environment



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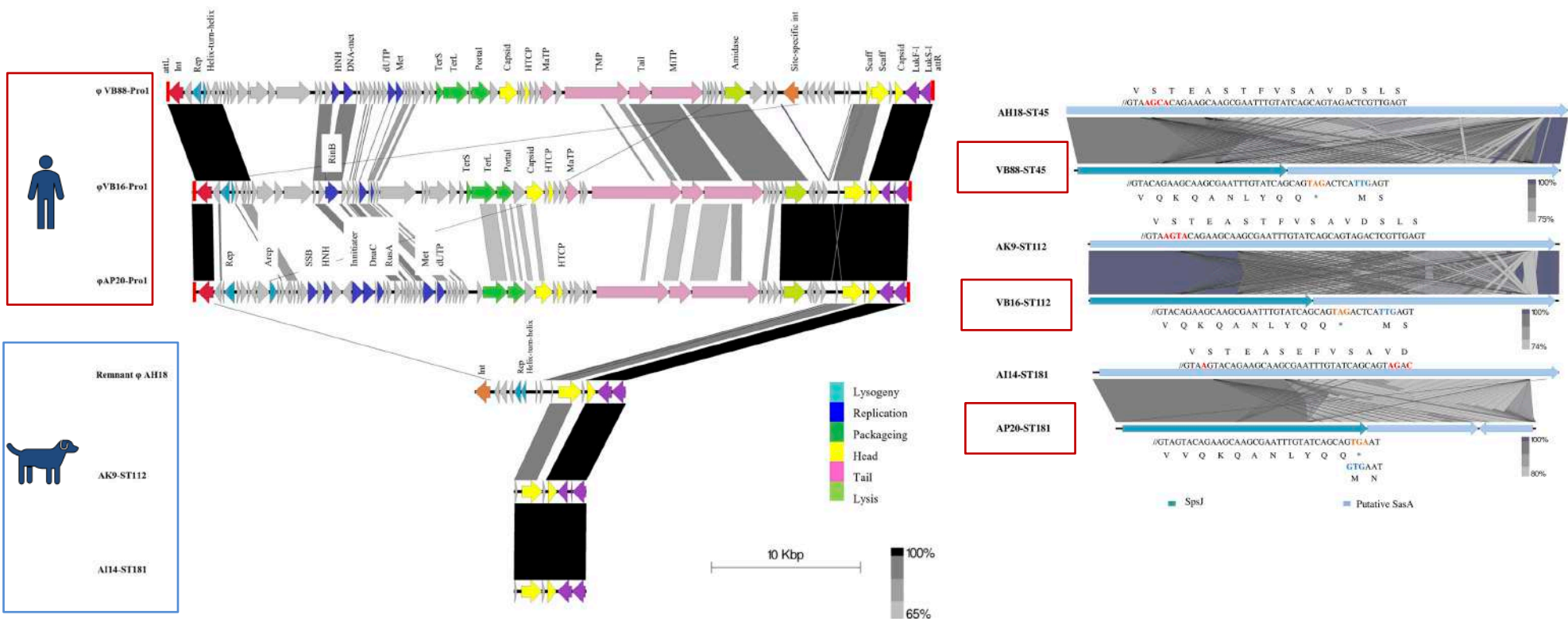
Percentage and numbers in parentheses of variable genes detected among *S. pseudintermedius* isolated from dogs, humans and the environment.

Genes	Dogs			Humans (n = 25)	Environment (n = 40)
	Carriage (n = 35)	Clinical (n = 35)	Total (n = 70)		
<i>spsD</i>	28.6 (10)	17.1 (6)	24.3 (16)	20.0 (5)	25.0 (10)
<i>spsF</i>	14.3 (5)	14.3 (5)	14.3 (10)	12.0 (3)	27.5 (11)
<i>spsI</i>	80.0 (28)	80.0 (28)	80.0 (56)	76.0 (19)	87.5 (35)
<i>spsO</i>	42.9 (15)	34.3 (12)	38.6 (27)	24.0 (6)	52.5 (21)
<i>spsP</i>	25.7 (9)	48.6 (17)*	37.1 (26)	48.0 (12)	40.0 (16)
<i>spsQ</i>	25.7 (9)	48.6 (17)*	37.1 (26)	48.0 (12)	40.0 (16)

\* Statistically significant by  $\chi^2$ -test ( $p < 0.05$ ) between carriage and clinical group of isolates.

# Staphylococcus pseudintermedius

- Genomic comparative analyses of MRSP isolates from dogs and humans



The differences were mainly associated with the presence of two additional **prophages** as well as truncation of some of the **CWA protein genes** in the human ST45 isolate.

# Coagulase-negative staphylococci (CoNS)

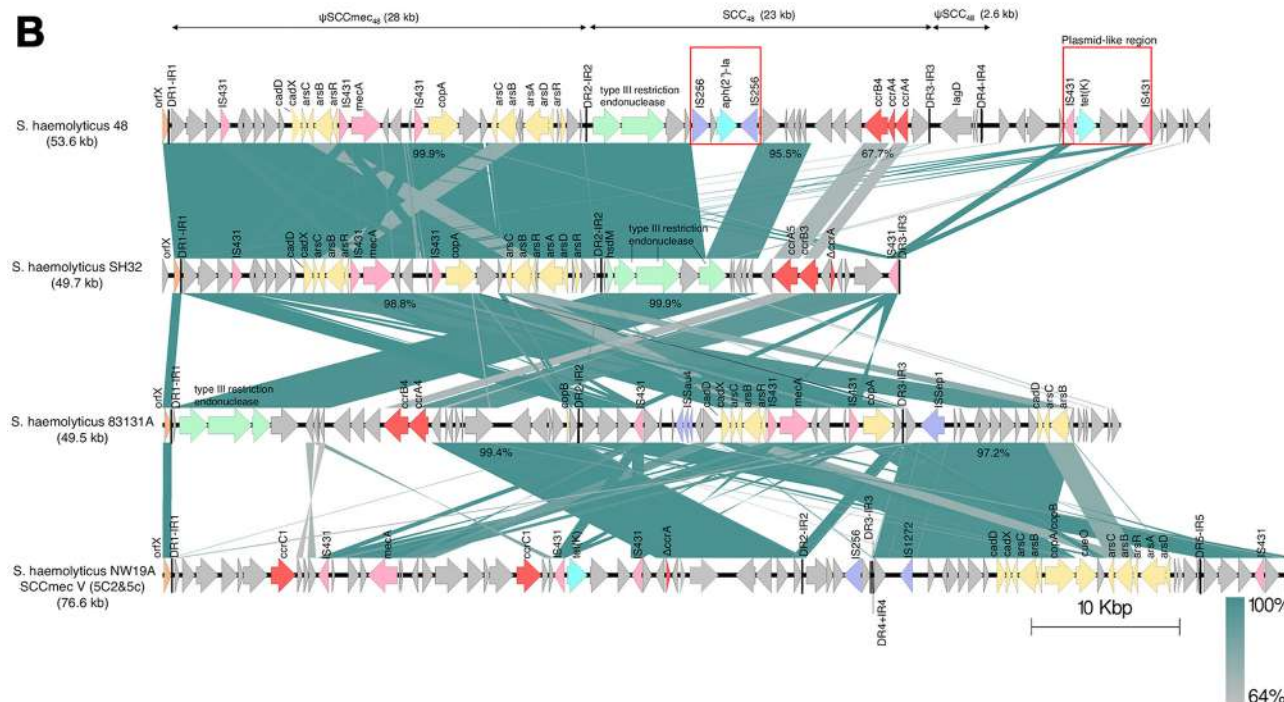
- There are fewer studies on CoNS compared with CoPS in pets.
- Zoonotic transmission of CoNS has been observed in pet owners.
- 185 CoNS (17.5%) were obtained and included 18 species from dogs (n = 116) and 14 species from cats (n = 69).

TABLE 1. SCCmec TYPES AND ANTIMICROBIAL RESISTANCE OF METHICILLIN-RESISTANT COAGULASE-NEGATIVE STAPHYLOCOCCI FOUND IN THIS STUDY

CoNS spp.	Strain	Animal	Sample	SCCmec type ( <i>ccr</i> and <i>mec</i> complex)	Phenotypic resistance profiles <sup>a</sup>	Genotypic resistance profiles
<i>Staphylococcus haemolyticus</i> (n=8)	195	Dog	Skin (wound)	V ( <i>ccrC</i> , C)	β-lactams, ERY, CLI, TET, FUS, SMX-TMP	<i>blaZ</i> , <i>erm</i> (C), <i>tet</i> (L), <i>fusB</i> , <i>dfrG</i>
	625	Cat	Skin (wound)	V ( <i>ccrC</i> , C)	β-lactams, TET	<i>blaZ</i> , <i>tet</i> (K)
	1283	Dog	Skin (wound)	V ( <i>ccrC</i> , C)	β-lactams, ENR, MAR, CLI, TET	<i>blaZ</i> , <i>gyrA</i> (S84L) <sup>b</sup> , <i>erm</i> (C), <i>tet</i> (K)
	521	Dog	Urine	V ( <i>ccrC</i> , C)	β-lactams, ERY, CLI, CHL	<i>blaZ</i> , <i>erm</i> (C), <i>cat</i> <sub>Pc221</sub>
	50	Dog	Skin (crust)	V ( <i>ccrC</i> and <i>ccrAB</i> , C)	β-lactams, ERY, CLI, CHL	<i>blaZ</i> , <i>mph</i> (C), <i>cat</i> <sub>Pc221</sub>
	48	Dog	Skin (wound)	NT (–/–)	β-lactams, GEN, ENR, MAR, ERY, CLI, TET, MUP, SMX-TMP	<i>blaZ</i> , <i>aph</i> (3')-IIIa, <i>gyrA</i> (S84L) <sup>b</sup> , <i>erm</i> (C), <i>tet</i> (K), <i>mupA</i> , <i>dfrG</i>
	289	Dog	Skin (wound)	NT (–/–)	β-lactams, ENR, MAR, CLI, TET, MUP, RIF	<i>blaZ</i> , <i>gyrA</i> (S84L) <sup>b</sup> , <i>erm</i> (C), <i>mph</i> (C), <i>msr</i> (A), <i>tet</i> (K), <i>mupA</i>
<i>Staphylococcus epidermidis</i> (n=6)	1864	Dog	Urine	NT (–/–)	β-lactams, ENR, MAR, ERY, CLI, TET, MUP, SMX-TMP, RIF	<i>blaZ</i> , <i>gyrA</i> (S84L) <sup>b</sup> , <i>erm</i> (C), <i>mph</i> (C), <i>msr</i> (A), <i>tet</i> (K), <i>mupA</i> , <i>dfrG</i>
	187	Dog	Pleural effusion	IV ( <i>ccrAB2</i> , B)	β-lactams, ERY, CLI	<i>blaZ</i> , <i>msr</i> (A), <i>inu</i> (A)
	236	Dog	Skin (wound)	IV ( <i>ccrAB2</i> , B)	β-lactams, GEN, TET, SMX-TMP	<i>blaZ</i> , <i>aacA-aphD</i> , <i>tet</i> (K), <i>dfrG</i>
	459	Dog	Lung biopsy	IV ( <i>ccrAB2</i> , B)	β-lactams, ERY, TET, FUS	<i>blaZ</i> , <i>msr</i> (A), <i>tet</i> (K), <i>fusB</i>
	1536	Cat	Nasal discharge	IV ( <i>ccrAB2</i> , B)	β-lactams, ENR, MAR, TET	<i>blaZ</i> , <i>gyrA</i> (S84Y) <sup>b</sup> , <i>glrA</i> (S80Y), <i>tet</i> (K)
	1574	Dog	Pleural effusion	IV ( <i>ccrAB2</i> , B)	β-lactams, ERY, CLI, TET, FUS, MUP	<i>blaZ</i> , <i>msr</i> (A), <i>tet</i> (K), <i>fusB</i> , <i>mupA</i>
	1076	Dog	Urine	NT ( <i>ccrAB2</i> /–)	β-lactams, MAR, CLI, TET	<i>blaZ</i> , <i>gyrA</i> (S84F) <sup>b</sup> , <i>glrA</i> (S80F), <i>erm</i> (C), <i>tet</i> (K)
<i>Staphylococcus hominis</i> ssp. <i>hominis</i> (n=5)	1231	Dog	Cerebrospinal fluid	III ( <i>ccrAB3</i> , A)	β-lactams, ERY, CLI, TET	<i>blaZ</i> , <i>erm</i> (C), <i>msr</i> (A)
	385	Cat	Skin (pustules)	V ( <i>ccrC</i> , C)	β-lactams, GEN, ENR, MAR, ERY, CLI, TET, MUP, SMX-TMP	<i>blaZ</i> , <i>aacA-aphD</i> , <i>aph</i> (3')-IIIa, <i>gyrA</i> (S84L) <sup>b</sup> , <i>erm</i> (B), <i>tet</i> (M), <i>mupA</i> , <i>dfrG</i>
	371	Dog	Peritoneal effusion	NT (–/A)	β-lactams, CLI, SMX-TMP	<i>blaZ</i> , <i>dfrG</i>
	384	Dog	Skin (wound)	NT (–/A)	β-lactams, ERY, CLI	<i>blaZ</i> , <i>mph</i> (C), <i>msr</i> (A), <i>inu</i> (A)
	464	Dog	Skin	NT (–/–)	β-lactams, GEN, ENR, MAR, ERY, CLI, TET, CHL, SMX-TMP	<i>blaZ</i> , <i>aacA-aphD</i> , <i>aph</i> (3')-IIIa, <i>erm</i> (B), <i>tet</i> (M), <i>dfrG</i>

# Coagulase-negative staphylococci (CoNS)

- Short- and long-read nucleotide sequencing was used to characterize nontypeable (NT)-SCCmec elements in *S. haemolyticus* and *S. hominis*.
- Several **antimicrobial resistance** and **heavy metal resistance genes** were identified on the SCCmec element.

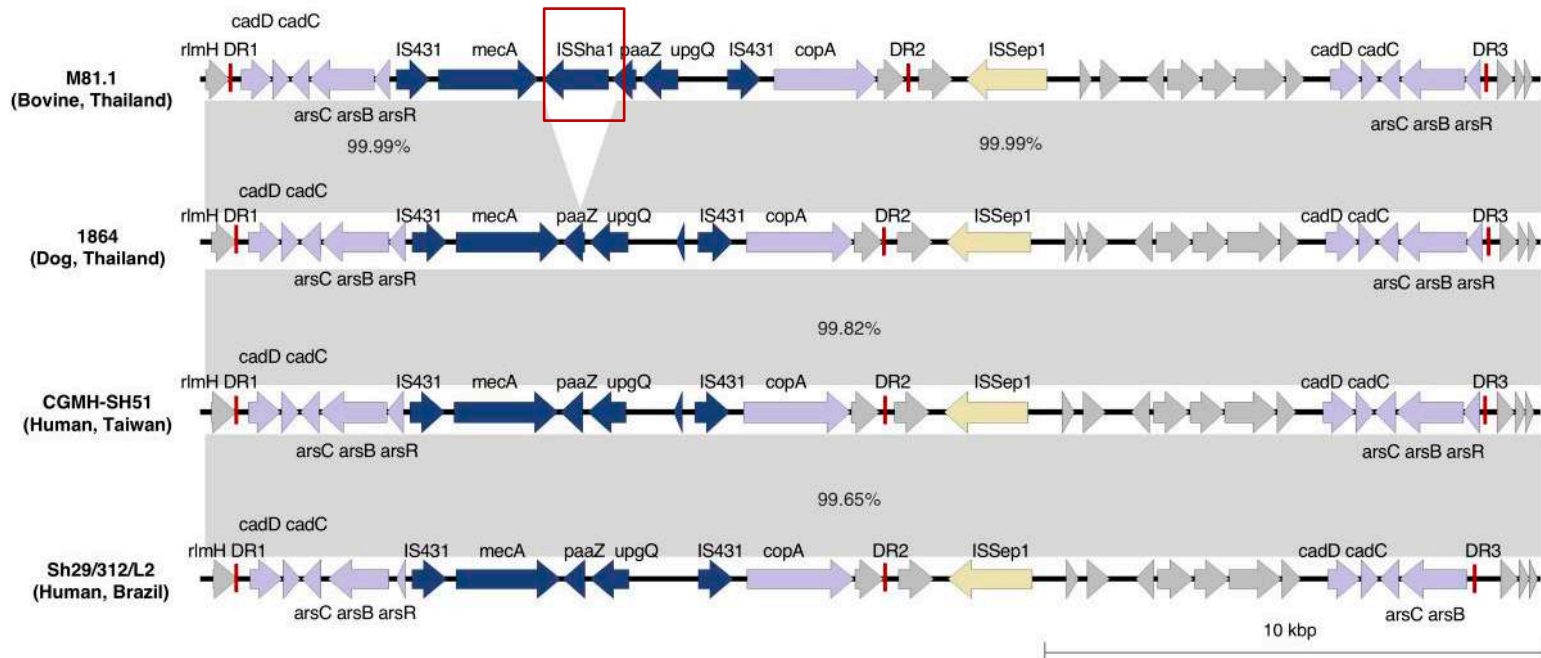




# Coagulase-negative staphylococci (CoNS)

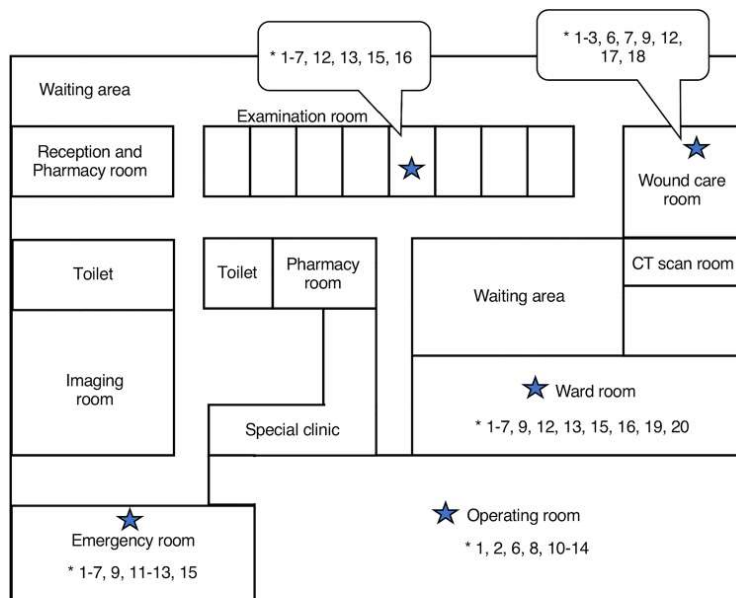
- *S. haemolyticus* from bovine mastitis milk
- MRSH-ST42 strain M81.1 contained a **novel variant of C1 *mec* complex**
- Distinct evolution of the MRSH lineage through *ISSha1* insertion, highlighting its role in SCCmec diversification

(b)



# *Pseudomonas aeruginosa*

- Major nosocomial pathogen causing healthcare-associated infections
- Moist and water-rich hospital environments as key sources of infection
- Molecular epidemiology of *P. aeruginosa* from a **veterinary teaching hospital environment**



Collected 3 times every 2 months

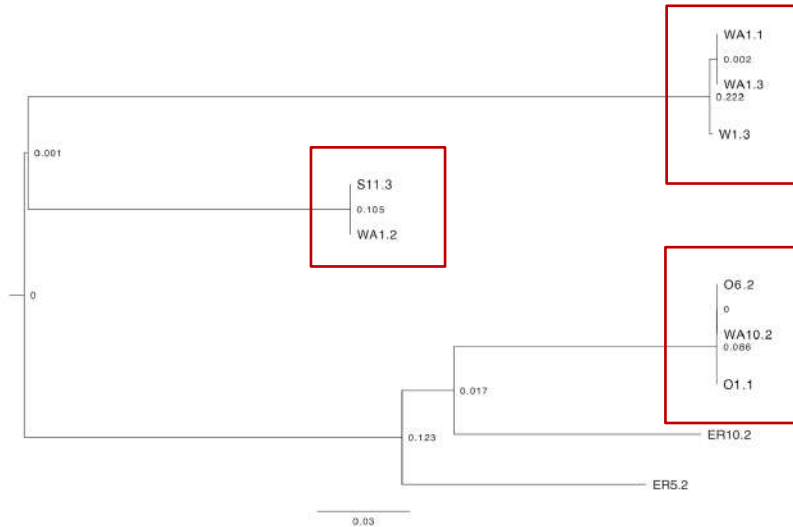
\* Sample collection sites

- |                                  |                                 |
|----------------------------------|---------------------------------|
| 1. Sink                          | 11. Endotracheal tube           |
| 2. Faucet                        | 12. Tap water                   |
| 3. Examination table             | 13. Thermometer jar             |
| 4. Equipment holder              | 14. Chlorhexidine solution      |
| 5. Work bench                    | 15. Keyboard                    |
| 6. Liquid hand soap dispenser    | 16. Detergent solution          |
| 7. Normal saline for irrigation  | 17. Nebulizer machine           |
| 8. Povidone iodine solution      | 18. Water for nebulizer machine |
| 9. Rubber tube                   | 19. Dog cage floor              |
| 10. Anesthesia-breathing circuit | 20. Cat cage floor              |



# *Pseudomonas aeruginosa*

- **Sinks** in exam and ward rooms consistently tested positive over 5 months
- Two **carbapenem-resistant** strains carried ***bla*<sub>VEB-1</sub>**
- Low-SNP, same-ST strains in different rooms indicate transmission link to the **ward room**
- Findings support improved management of *P. aeruginosa* in veterinary hospitals



**Table 1** Positive sampling sites of *P. aeruginosa* (n=19) found in this study<sup>a</sup>

Sampling sites <sup>b</sup>	Examination room (n=5)				Wound care room (n=4)			Ward room (n=4)			Emergency room (n=4)			Operating room (n=2)		
Collection time	1	2	3		1	2	3	1	2	3	1	2	3	1	2	3
Sink (n=8)	+(O1.1)	+(O1.2)	+(O1.3)	-	-	-	+(W1.3)	+(WA1.1)	+(WA1.2)	+(WA1.3)	-	-	-	-	-	+(S1.3)
Rubber tube (n=4)	-	-	-	-	-	+(W10.1)	-	-	+(WA 10.2)	-	+(ER10.1)	+(ER10.2)	-	-	-	-
Liquid hand soap dispensers (n=3)	-	+(O6.2)	+(O6.3)	-	-	+(W6.2)	-	-	-	-	-	-	-	-	-	-
Normal saline for irrigate (n=1)	-	-	-	-	+(W7.1)	-	-	-	-	-	-	-	-	-	-	-
Work bench (n=1)	-	-	-	-	-	-	-	-	-	-	-	+(ER5.2)	-	-	-	-
Equipment holder (n=1)	-	-	-	-	-	-	-	-	-	-	-	-	+(ER4.3)	-	-	-
Anesthesia- breathing circuit (n=1)	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+(S11.3)

<sup>a</sup> + and – represent positive and negative sampling sites, respectively

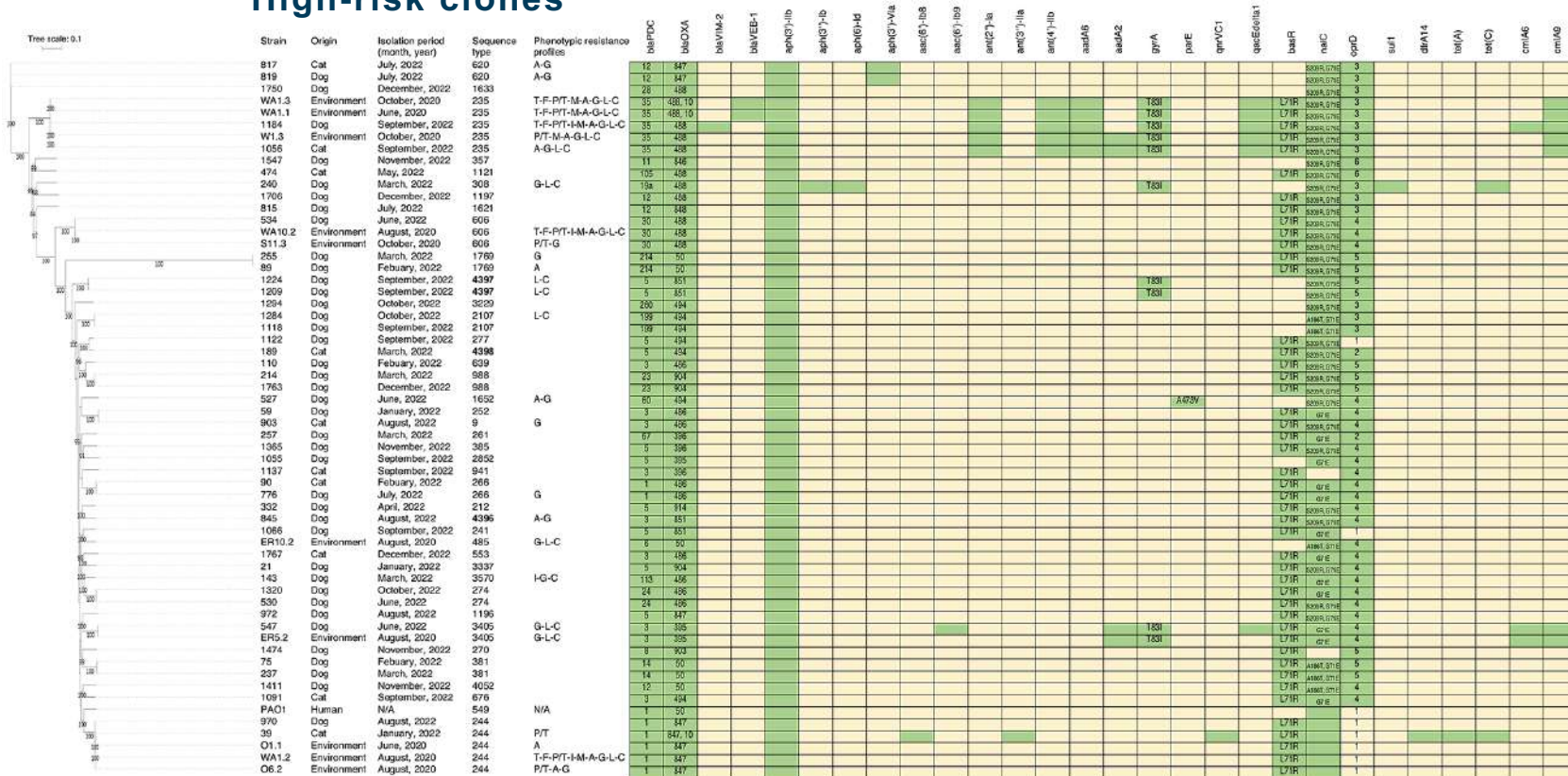
<sup>b</sup> The number of positive *P. aeruginosa* is presented in parenthesis



# *Pseudomonas aeruginosa*

Investigate the sequence typing and clones, and antimicrobial resistance of clinical *P. aeruginosa* isolates from the dogs and cats visiting a veterinary teaching hospital in Thailand within a 1-year period (2022).

## High-risk clones





# Current research

## **AMR and genomic analysis**

- Genomic characterization of AMR in clinical bacterial isolates from ornamental fish over a 10-year period
- Investigation of antimicrobial resistance profiles of *Enterococcus* species isolated from dogs and cats

## **Metagenomics**

- Application of metagenomic approaches to explore microbial communities and resistance gene diversity in animal hosts



# Acknowledgement



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**Thank you for your attention**