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II CONGRESO INTERNACIONAL DEL COLEGIO NACIONAL DE BACTERIOLOGÍA
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Cartagena, Colombia 3 al 6 OCTUBRE 2024

THE OHIO STATE UNIVERSITY
COLLEGE OF VETERINARY MEDICINE

Actualización en Pruebas de Susceptibilidad Antimicrobiana en Patógenos Animales

Dubraska Diaz-Campos, DVM, PhD
Microbiólogo Clínico
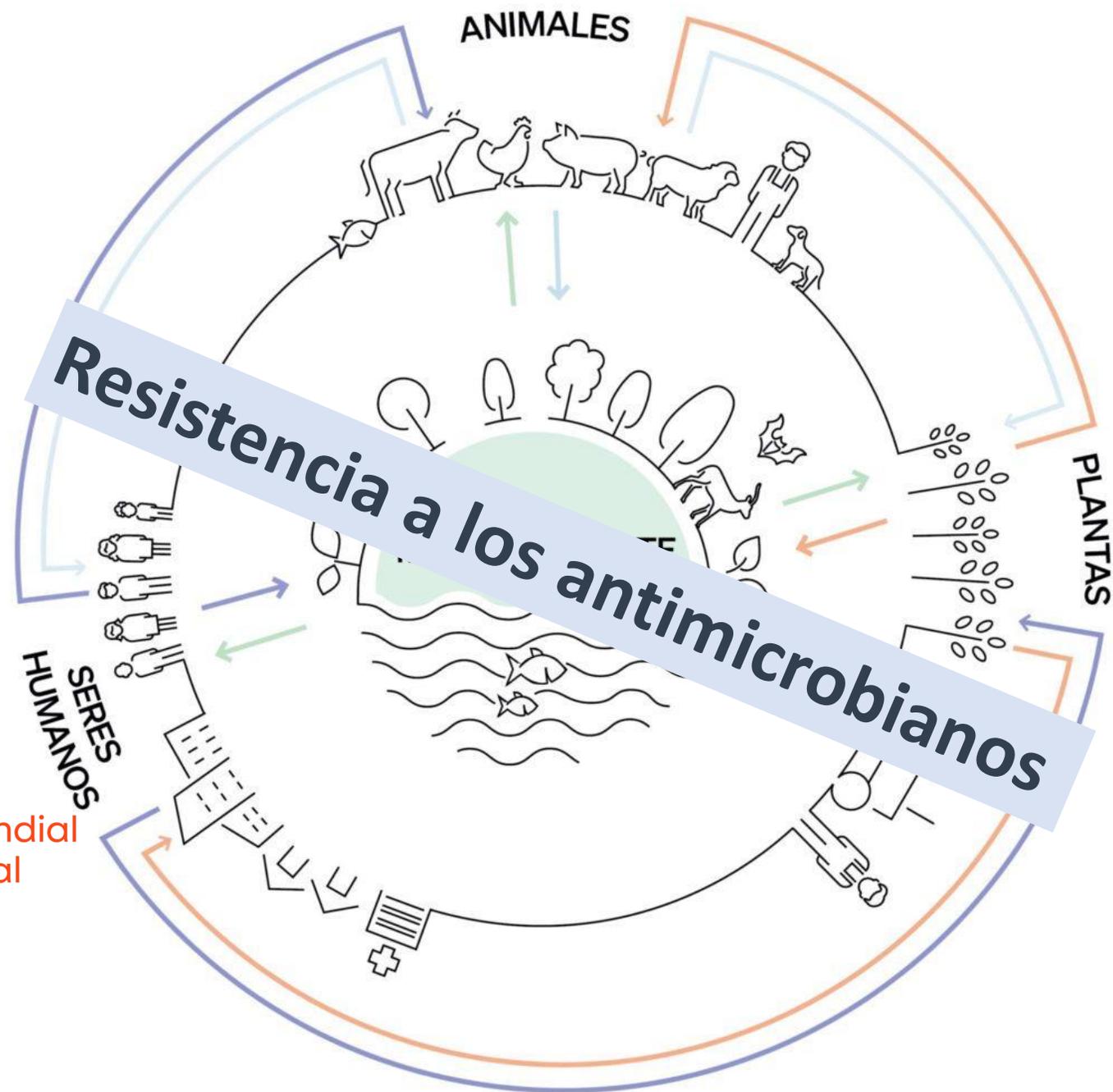Centro Médico Veterinario – Universidad Estadal de Ohio
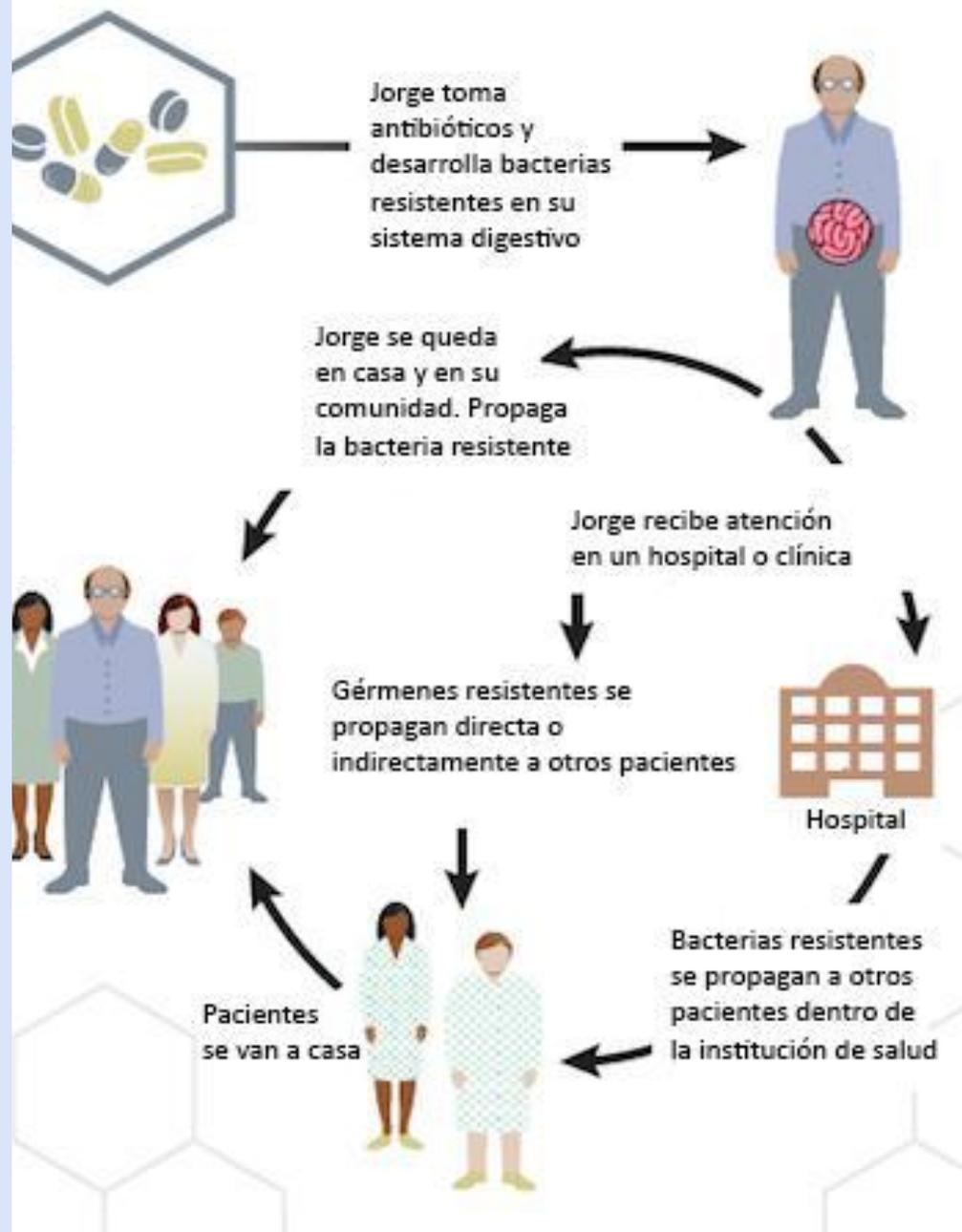Diaz-campos.1@osu.edu

¿Cómo se propaga la resistencia a los antibióticos?

Jorge toma antibióticos y desarrolla bacterias resistentes en su sistema digestivo

Jorge se queda en casa y en su comunidad. Propaga la bacteria resistente

Jorge recibe atención en un hospital o clínica

Gérmenes resistentes se propagan directa o indirectamente a otros pacientes

Hospital

Bacterias resistentes se propagan a otros pacientes dentro de la institución de salud

Pacientes se van a casa

Imagen: **Lic. María Angélica Rivera de Linton**http://afqfelsalvador.com/wp-content/uploads/2015/08/Resistencia-antimicrobianos.jpg

Medicina Veterinaria

Optimización del Diagnóstico - Fases

1 Pre-Analítica — Vet (Lab)

2 Analítica — El lab

3 Post- Analítica — El lab & Vet
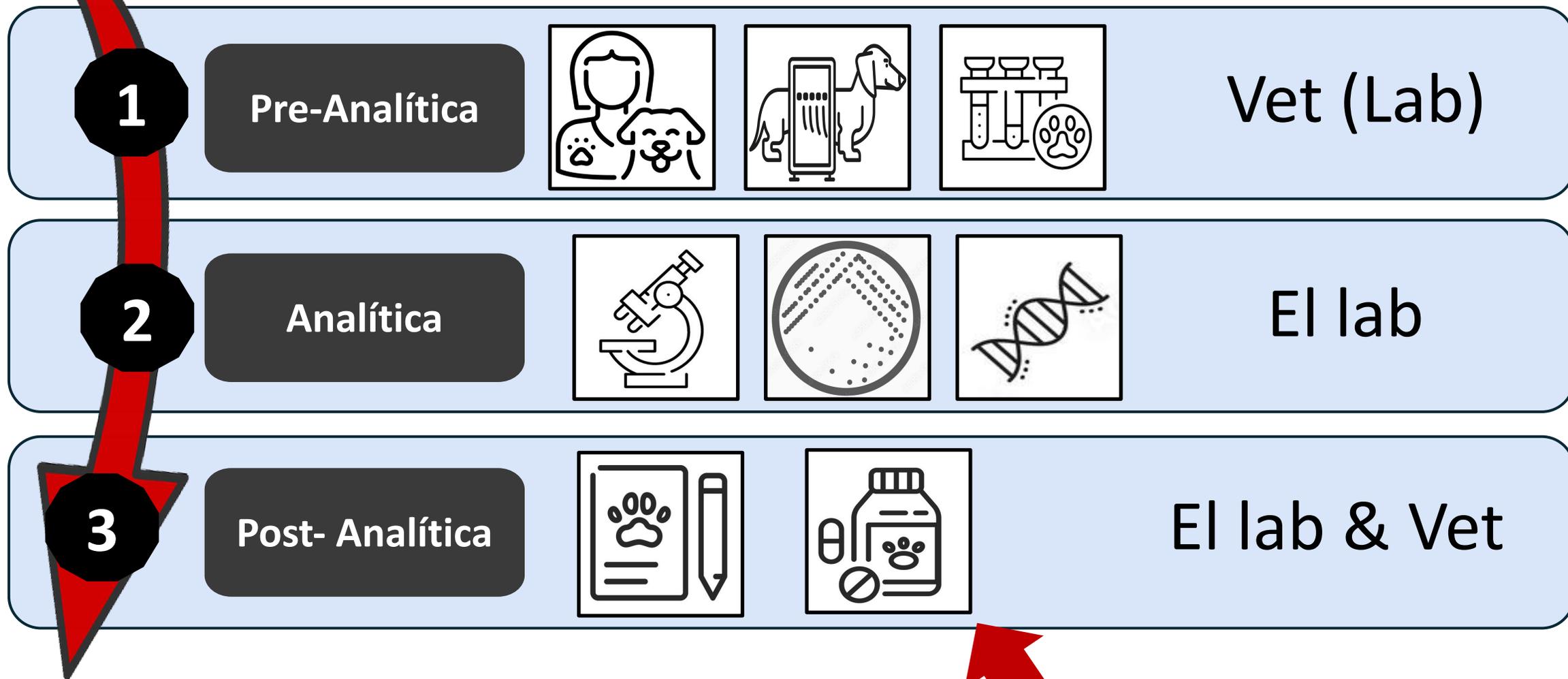
El Proceso de Diagnóstico

Fase Preclínica, algunos desafíos

¿Agente infeccioso o normal microbiota?

Staphylococcus pseudintermedius
Coagulasa-negativa *Staphylococcus* spp

E. coli
Proteus mirabilis
Enterococcus spp

Factores a considerar:
- Toma de muestra
- Síntomas clínicos
- Sitio de aislamiento Vs presentación clínica

El dilema…
¿reportamos o no?

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¿Normal microbiota?

Estrategias:

- No reportar

- Reportar como un comentario

Specimen:	Incision :: Swab		Collection Date:

BACTERIOLOGY

Test: MI. Culture-Aerobic w/Susceptibility + Anaerobe :: Aerobic Culture Results

Isolate #	Quantity	Organism	Comment
	Moderate growth	Mixed Bacterial Flora	Multiple colony types (2-3 morphologies) present in light to moderate numbers suggestive of normal microbiota overgrowth and/or contamination.
			If clinical improvement is not observed, histological examination and recollection of a new specimen for an additional culture are recommended to validate these results.

Estrategias al reportar ASTs en animales

Objetivo: **Promover prácticas de POAs**

- Resistencia intrínseca

Specimen:	Urine :: Cystocentesis		Collection Date:

BACTERIOLOGY

Test: MI. Culture-Aerobic w/Susceptibility :: Aerobic Culture Results

Isolate #	Quantity	Organism	Comment
1	>100,000 cfu/mL	Escherichia coli	
2	>100,000 cfu/mL	Enterococcus faecalis	This microorganism is intrinsically resistant to cephalosporins, aminoglycosides, clindamycin, trimethoprim and trimethoprim-sulfamethoxazole.

Tratamiento empírico

COLABIOCLI Confederación Latinoamericana de Bioquímica Clínica

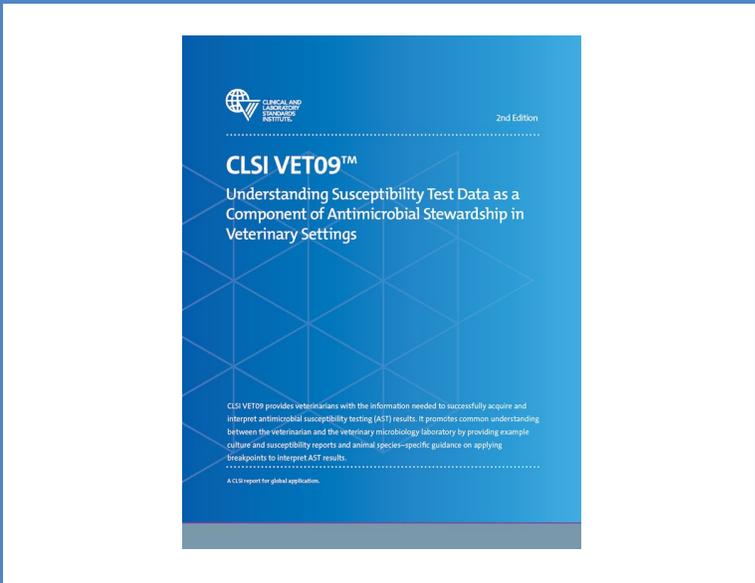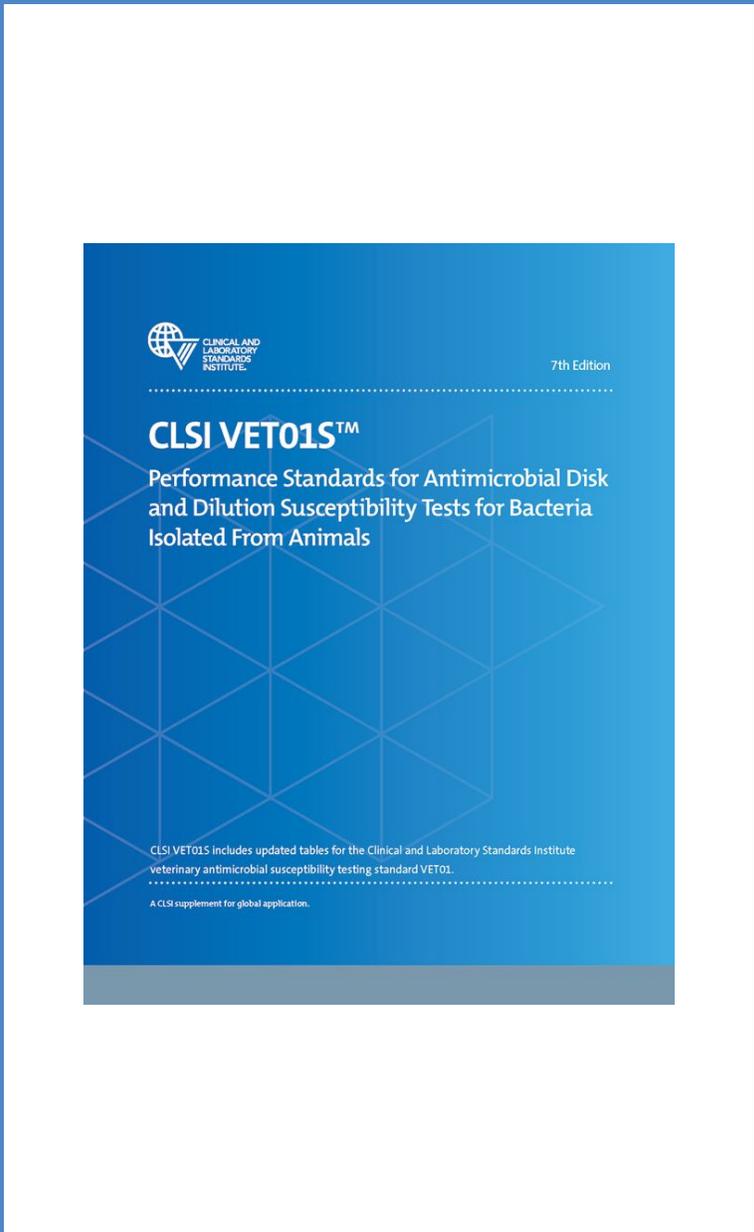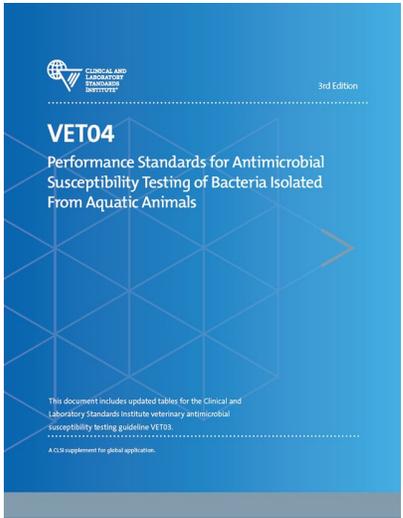CnB Colegio Nacional de Bacteriología®

El CLSI Subcomité VAST

Los voluntarios del Subcomité de Pruebas de Susceptibilidad Antimicrobiana Veterinaria (VAST) del CLSI colaboran para desarrollar estándares y pautas que promuevan pruebas de susceptibilidad antimicrobiana precisas y reportes apropiados.

Miembros:

- Laboratorios de microbiología veterinaria
- Agencias gubernamentales
- Proveedores de atención veterinaria y educadores
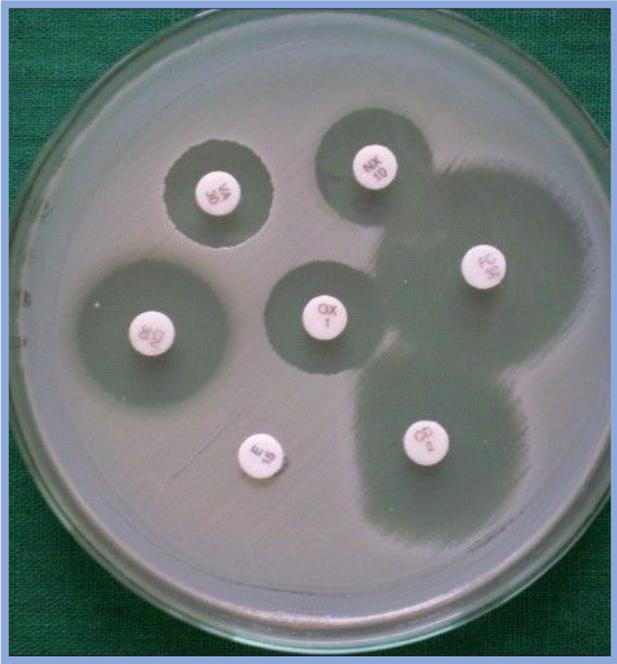- Laboratorios de microbiología farmacéutica y de diagnóstico

CLINICAL AND LABORATORY STANDARDS INSTITUTE

3rd Edition

VET04

Performance Standards for Antimicrobial Susceptibility Testing of Bacteria Isolated From Aquatic Animals

This document includes updated tables for the Clinical and Laboratory Standards Institute veterinary antimicrobial susceptibility testing guideline VET03.

A CLSI supplement for global application.

CLINICAL AND LABORATORY STANDARDS INSTITUTE

6th Edition

CLSI VET01™

Performance Standards for Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria Isolated From Animals

This standard covers the current recommended methods for disk diffusion susceptibility testing and the reference methods for determining minimal inhibitory concentrations for aerobic bacteria by broth macrodilution, broth microdilution, and agar dilution for veterinary use.

A standard for global application developed through the Clinical and Laboratory Standards Institute consensus process.

CLINICAL AND LABORATORY STANDARDS INSTITUTE

7th Edition

CLSI VET01S™

Performance Standards for Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria Isolated From Animals

CLSI VET01S includes updated tables for the Clinical and Laboratory Standards Institute veterinary antimicrobial susceptibility testing standard VET01.

A CLSI supplement for global application.

CLINICAL AND LABORATORY STANDARDS INSTITUTE

4th Edition

VET02

Development of Quality Control Ranges, Breakpoints, and Interpretive Categories for Antimicrobial Agents Used in Veterinary Medicine

This guideline discusses the necessary and recommended data for selecting appropriate quality control ranges, breakpoints, and interpretive categories for antimicrobial agents for veterinary use.

A guideline for global application developed through the Clinical and Laboratory Standards Institute consensus process.

CLINICAL AND LABORATORY STANDARDS INSTITUTE

2nd Edition

CLSI VET09™

Understanding Susceptibility Test Data as a Component of Antimicrobial Stewardship in Veterinary Settings

CLSI VET09 provides veterinarians with the information needed to successfully acquire and interpret antimicrobial susceptibility testing (AST) results. It promotes common understanding between the veterinarian and the veterinary microbiology laboratory by providing example culture and susceptibility reports and animal species–specific guidance on applying breakpoints to interpret AST results.

A CLSI report for global application.

Pruebas de Susceptibilidad a los antimicrobianos (AST)

1 Difusión de disco (KB)

2 Dilución en caldo

3 E-test

Concentración mínima inhibitoria (CIM)

VET01s

Acceso gratuito: http://clsivet.org/Login.aspx

Tabla 1. Agentes antimicrobianos aconsiderarse para pruebas de rutina en los laboratorios de microbiología veterinaria

Test/Report Group	Animal Species						
	Swine	Cattle[a]	Bovine Mastitis[b]	Poultry[c]	Horses	Dogs	Cats[d]
Group A – Veterinary-Specific Breakpoints					Amikacin	Amikacin	
						Amoxicillin-clavulanate	Amoxicillin-clavulanate
	Ampicillin[e]	Ampicillin[e]			Ampicillin[e]	Ampicillin[e]	Ampicillin[e]
					Cefazolin	Cefazolin	
						Cefovecin	Cefovecin
			Cefoperazone				
						Cefpodoxime	
	Ceftiofur	Ceftiofur	Ceftiofur		Ceftiofur		
						Cephalexin	
						Cephalothin	
						Clindamycin[f]	
		Danofloxacin					
						Difloxacin	
					Doxycycline	Doxycycline	
	Enrofloxacin	Enrofloxacin			Enrofloxacin	Enrofloxacin	Enrofloxacin
	Florfenicol	Florfenicol					
		Gamithromycin					
					Gentamicin	Gentamicin	
			Kanamycin-cephalexin				
						Marbofloxacin	Marbofloxacin
					Minocycline	Minocycline	
						Orbifloxacin	Orbifloxacin
	Penicillin G	Penicillin G			Penicillin G		

Tabla 2E. Diámetro de zona y puntos de corte de CMI para *Streptococcus* spp.

Test/ Report Group	Body Site	Antimicrobial Agent	Antimicrobial Agent Class or Subclass	Organism	Disk Content	Interpretive Categories and Zone Diameter Breakpoints, nearest whole mm			Interpretive Categories and MIC Breakpoints, µg/mL			Comments
						S	I	R	S	I	R	
Horses (Continued)												
A	Resp	Ampicillin	Penicillinase-labile penicillins	*S. equi* subsp. *equi* and subsp. *zooepidemicus*	–	–	–	–	≤ 0.25	–	–	**See comment (7).**
A	Gen, resp	Cefazolin	Cephalosporin I	*Streptococcus* spp. β-hemolytic group	–	–	–	–	≤ 2	4	≥ 8	
A	Resp	Ceftiofur	Cephalosporin III	*S. equi* subsp. *zooepidemicus*	30 µg	≥ 22	–	–	≤ 0.25	–	–	**See comment (7).**
A		Doxycycline	Tetracyclines	*S. equi* subsp. *equi* and subsp. *zooepidemicus*	–	–	–	–	≤ 0.12	0.25	≥ 0.5	**(8)** Do not test tetracycline as a surrogate for doxycycline and minocycline in horses.
A	Resp, SST	Enrofloxacin	Fluoroquinolones[c]	*S. equi* subsp. *equi* and subsp. *zooepidemicus*	–	–	–	–	≤ 0.12	0.25	≥ 0.5	
A	Resp, SST	Minocycline	Tetracyclines	*Streptococcus* spp.	–	–	–	–	≤ 0.12	0.25	≥ 0.5	See comment (8).
A	Resp, soft tissue	Penicillin G	Penicillinase-labile penicillins	*Streptococcus* spp.	–	–	–	–	≤ 0.5	1	≥ 2	

Interpretación de AST

¿Qué debemos reportar?

Ejemplo:

UTIs no complicada

Enterobacterales

1st opción abx

✔ Ampicilina

✔ Amox

✔ Amox-Clav

Specimen Type	Urine		
Comment	Ampicillin, amoxicillin, and amoxicillin-clavulanate are effective only for cases of uncomplicated (lower) UTIs caused by E. coli and other Enterobacterales (e.g. Enterobacter spp., Klebsiella spp., Proteus spp., among others). In the case of complicated (upper) UTIs or other medical conditions caused by these microorganisms, these drugs aren't effective because they do not reach therapeutic target.		
Organism	Escherichia coli		
Antibiotics		Category	MIC (mcg/ml)
Amikacin		S	<= 4
Amoxicillin-clavulanate		S	8
Ampicillin		S	4
Cefazolin		S	2
Cefovecin		S	1
Cefpodoxime		S	<= 1
Ceftazidime		S	<= 4
Cephalexin		S	8
Chloramphenicol		S	8
Enrofloxacin		S	<= 0.125
Gentamicin		S	<= 0.25
Marbofloxacin		S	<= 0.125
Piperacillin-tazobactam		S	<= 8
Pradofloxacin		S	<= 0.25
Trimethoprim-sulfamethoxazole		S	<= 0.5

Predice amoxicilina

BP nuevos, en 2024 es R

Suprimimos:
Carbapenémico
Nitrofurantoína

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Interpretación de AST

Proveer información adicional → Promueve los POAs
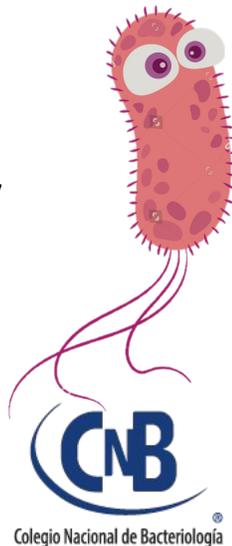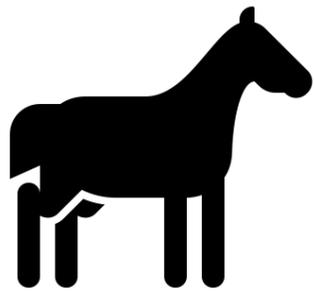
Ejemplo:

UTIs **no complicada**

Enterobacterales

1st opción abx

- ✔ Ampicilina
- ✔ Amox
- ✔ Amox-Clav

Ampicillin Breakpoints – CLSI Enterobacterales, DOGS

Body site	Susceptible	Intermediate	Resistant
Urine	≤ 8	-	-
Systemic	≤ 0.25	0.5	≥ 1
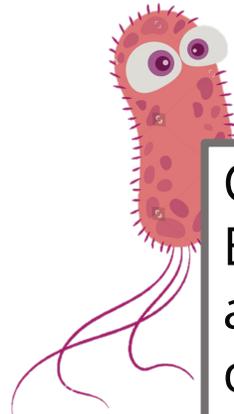
Con excepción de los aislamientos de infecciones urinarias, los Enterobacterales deben informarse como resistentes a la ampicilina, amoxicilina y amoxicilina-clavulanato porque las concentraciones del fármaco alcanzadas según el régimen de dosificación utilizado para establecer los puntos de corte no son lo suficientemente altas para alcanzar el objetivo terapéutico.

Proveer información adicional → Promueve los POAs

Ejemplo:

UTIs **no complicada**

Enterobacterales

1st opción abx

✔ Ampicilina

✔ Amox

✔ Amox-Clav

Specimen Type	Urine
Comment	Ampicillin, amoxicillin, and amoxicillin-clavulanate are effective only for cases of uncomplicated (lower) UTIs caused by E. coli and other Enterobacterales (e.g. Enterobacter spp., Klebsiella spp., Proteus spp., among others). In the case of complicated (upper) UTIs or other medical conditions caused by these microorganisms, these drugs aren't effective because they do not reach therapeutic target.

Organism		Escherichia coli
Antibiotics	Category	MIC (mcg/ml)
Amikacin	S	<= 4
Amoxicillin-clavulanate	S	8
Ampicillin	S	4
Cefazolin	S	2
Cefovecin	S	1
Cefpodoxime	S	<= 1
Ceftazidime	S	<= 4
Cephalexin	S	8
Chloramphenicol	S	8
Enrofloxacin	S	<= 0.125
Gentamicin	S	<= 0.25
Marbofloxacin	S	<= 0.125
Piperacillin-tazobactam	S	<= 8
Pradofloxacin	S	<= 0.25
Trimethoprim-sulfamethoxazole	S	<= 0.5

Interpretación de AST

Potrillo con pneumonía

Bordetella bronchiseptica
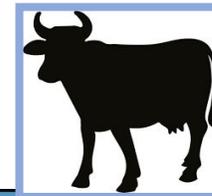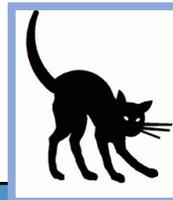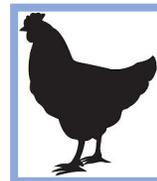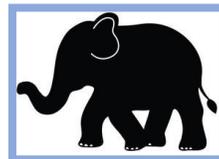
Test/ Report Group	Body Site	Antimicrobial Agent	Antimicrobial Agent Class or Subclass	Disk Content	Interpretive Categories and Zone Diameter Breakpoints, nearest whole mm			Interpretive Categories and MIC Breakpoints, µg/mL			Comments
					S	I	R	S	I	R	
Swine											
A	Resp	Ampicillin	Penicillinase-labile penicillins		–	–	–	≤ 0.5	1	≥ 2	**(3)** Results of ampicillin testing can be used to predict results for amoxicillin.
A	Resp	Florfenicol	Phenicols	30 µg	≥ 22	19–21	≤ 18	≤ 2	4	≥ 8	
A	Resp	Tildipirosin	Macrolides	60 µg	≥ 18	–	–	≤ 8	–	–	
A	Resp	Tulathromycin	Macrolides	30 µg	≥ 18	15–17	≤ 14	≤ 16	32	≥ 64	

Interpretación de AST

- Puntos de corte nuevos
- ¡Actualizaciones!

VET01S

Performance Standards for Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria Isolated From Animals

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Chloramphenicol Breakpoints *Staphylococcus* spp			
	Susceptible	Intermediate	Resistant
Current BP	≤ 8	16	≥ 32
Updated BP	≤ 2	4	≥ 8

¿Solución? Riesgo de incrementar el uso de antibióticos de importancia critica (ej., linezolid)

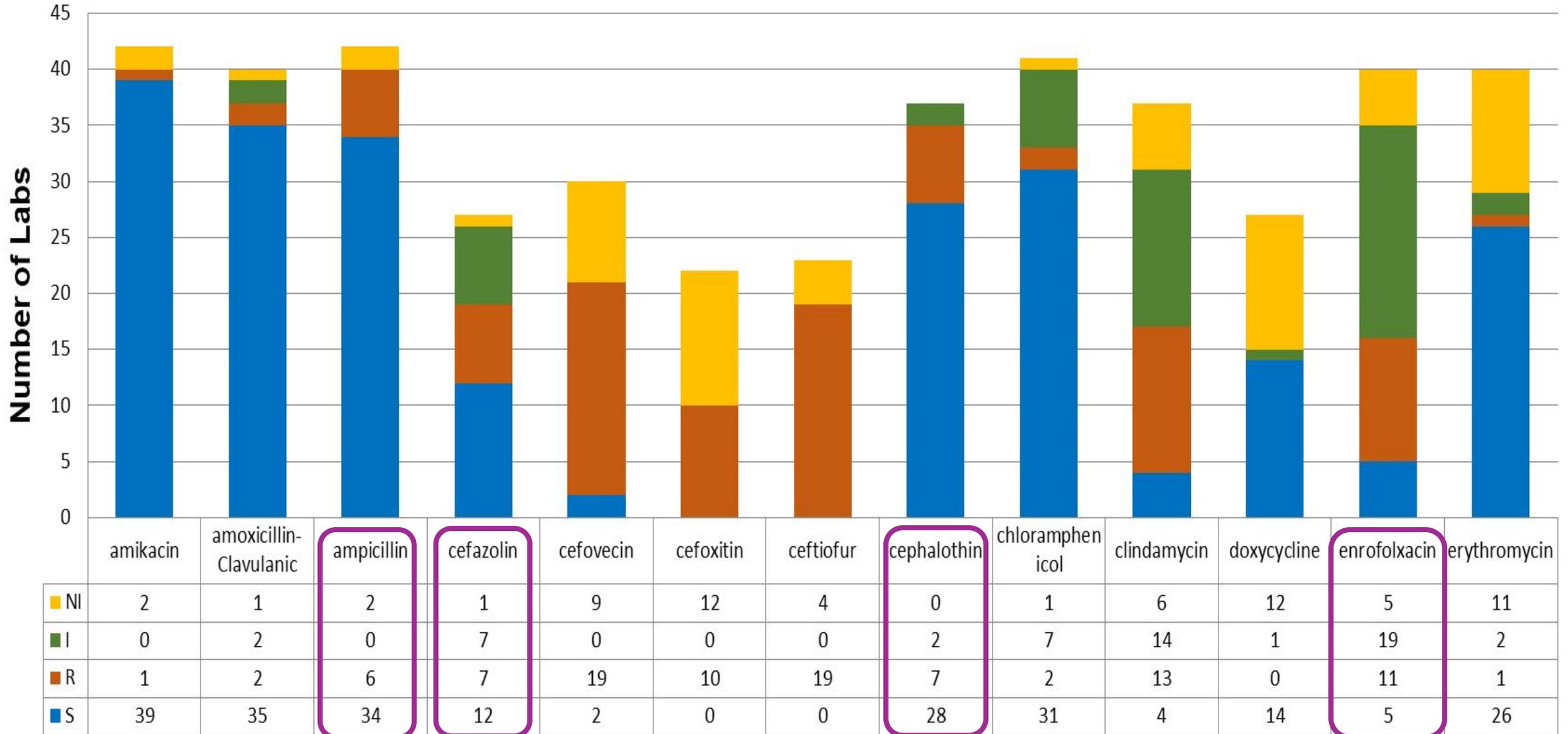| Source	: Skin		
Isolate	: 1		
Organism	: MRSP		
Offline Tests	: Beta-lacta mase	+	
Requires Expert Review	: Yes		
Antimicrobic			
Amikacin		<= 16	NI
Amoxicillin/Clavulanic Acid			R
Ampicillin			R
Cefazolin			R
Cefovecin			R
Cefpodoxime			R
Cephalothin			R
Chloramphenicol		<= 8	S
Clindamycin		> 4	R
Doxycycline		> 0.5	R
Enrofloxacin		> 4	R
Erythromycin		> 4	R
Gentamicin		> 16	R
Marbofloxacin		> 4	R
Minocycline		> 2	R
Oxacillin + 2% NaCl			R
Penicillin			R
Pradofloxacin		2	R
Rifampin		<= 1	S
Tetracycline		> 1	R
Trimethoprim/Sulfamethoxazole		> 4	R

Rhodococcus equi: AST

- Las interpretaciones de los puntos de corte (PB) son extrapolaciones de *Staphylococcus* spp (documento de PB humanos)

- Estos no son PB "reales"

Antimicrobial Class	Antimicrobial Agent	MIC (µg/mL) Interpretive Categories and Breakpoints			Comments
		S	I	R	
MACROLIDES					
	Azithromycin	≤2	4	≥8	
	Clarithromycin	≤2	4	≥8	
	Erythromycin	≤0.5	1–4	≥8	
TETRACYCLINES					
	Tetracycline	≤4	8	≥16	
FOLATE PATHWAY INHIBITORS					
	Trimethoprim-sulfamethoxazole	≤2/38	–	≥4/76	(4) This drug is not approved in the United States and Canada, or this drug is prohibited from certain extra-label uses in the United States, but may be approved in other countries. Check country and local regulations. See Subchapter 3.1 for additional information. (5) The breakpoints for trimethoprim-sulfamethoxazole are human-derived breakpoints and may or may not apply to animals.
FOLATE PATHWAY INHIBITORS (Continued)					
	Sulfonamides	≤256	–	≥512	(6) Sulfisoxazole can be used to represent any of the currently available sulfonamide preparations.
PHENICOLS					
	Chloramphenicol	≤8	16	≥32	See comment (4).
ANSAMYCINS					
	Rifampin	≤1	2	≥4	(7) *Rx:* Rifampin should not be used alone for antimicrobial therapy.

AAVLD - IBQAS RESULTS

Listeria monocitogenes

	amikacin	amoxicillin-Clavulanic	ampicillin	cefazolin	cefovecin	cefoxitin	ceftiofur	cephalothin	chloramphenicol	clindamycin	doxycycline	enrofolxacin	erythromycin
NI	2	1	2	1	9	12	4	0	1	6	12	5	11
I	0	2	0	7	0	0	0	2	7	14	1	19	2
R	1	2	6	7	19	10	19	7	2	13	0	11	1
S	39	35	34	12	2	0	0	28	31	4	14	5	26

Interpretación y ejecución de AST

- Publicación: MDR β-hemolytic Streptococcus en medicina veteranaria

- 2023- programas nacionales identificaron inconsistencias en los métodos para β-streps

Lenguaje universal β-hemolytic Streptococcus

¿No es un MDR?

- β-lactámico R en medicina humana: casi inexistente, a pesar del uso de penicilina de primera línea durante décadas

- Hasta un 8 % en medicina veterinaria

¿Por qué?

- Discrepancias en los métodos de prueba

- Brechas de conocimiento entre el personal

- ¿AST en todos los aislamientos?

Cortesía: Dr. Kelli Maddox (editado por DDC).

Viewpoint

Viewpoint articles represent the opinions of the authors and do not represent AVMA endorsement of such statements.

β-Lactam resistance in veterinary β-hemolytic Streptococcus species: Are we experiencing a public health or test method crisis?

Kelli J. Maddock, MD[1]*, Sarah J. Gerken, DO[2], Claire R. Burbick, DVM, PhD, DACVM[3]

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Washington Animal Disease Diagnostic Laboratory, Pullman, WA

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Nuevas técnicas

- Divertidas, pero…¿son útiles?

- Canino. Tipo de muestra: hisopado de la piel, axilas y patas, abdomen.

1.Bacteria

Species Detected	Percentage	Cells per Sample	Normal Range	Significance
Staphylococcus schleiferi [1]	46.4 %	300,000	0-0	🔴 High
Staphylococcus pseudintermedius [2]	23.6 %	150,000	0-65,000	⚫ Intermediate
Lawsonella clevelandensis [3]	7.2 %	46,000	0-2,100	🔴 High
Corynebacterium auriscanis [4]	4.8 %	31,000	0-2,600	🔴 High
Enterococcus faecalis [5]	0.5 %	3,400	0-670	⚫ Intermediate

2.Fungi

| Species Detected	Percentage	Cells per Sample	Normal Range	Significance
Malassezia pachydermatis [6]	4.2 %	78	0-22	⚫ Intermediate

Abbreviation Key:

🟢 **Normal.** Species detected within the reference range of clinically healthy dogs.
⚫ **Intermediate.** Species detected outside the reference range of clinically healthy dogs.
🔴 **High.** Species detected significantly higher than the reference range of clinically healthy dogs.

Charts below depict the relative abundance of all detected bacterial species. Each color represents a different bacterial species. The larger the colored segment is, the more abundant that species is in the specimen.

The sample was screened for the presence of antibiotic resistance genes and intrinsic resistances of clinically relevant microorganisms. For this analysis more than 90 antibiotic resistance genes were screeened. The cautious use of any antibiotic drug is highly recommended. Please follow the guidelines for antimicrobial
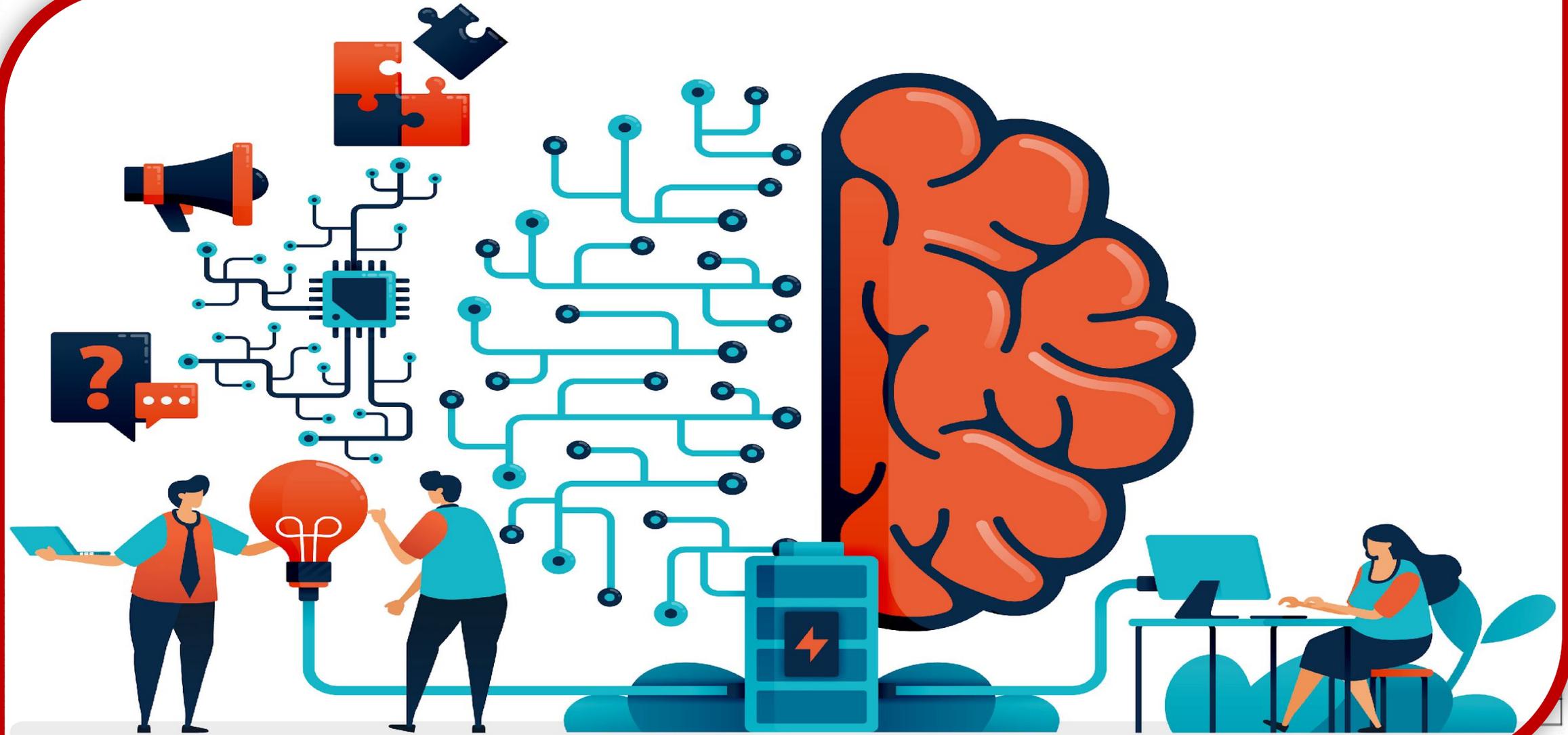
Abbreviation Key:

- 🟢 **Normal.** Species detected within the reference range of clinically healthy dogs.
- ⚪ **Intermediate.** Species detected outside the reference range of clinically healthy dogs.
- 🔴 **High.** Species detected significantly higher than the reference range of clinically healthy dogs.

P	Poor Performance (< 50% Effectiveness in Antibiogram Studies)
F	Fair Performance (< 75% Effectiveness in Antibiogram Studies)
G	Good Performance (> 75% Effectiveness in Antibiogram Studies)
NRD	No Antibiotic Resistance Detected Based on the MiDOG Antibiotic Target Panel

IV	Intravenous Injection
SC	Subcutaneous Injection
TU	Topical Use
--	No Info

Referencias
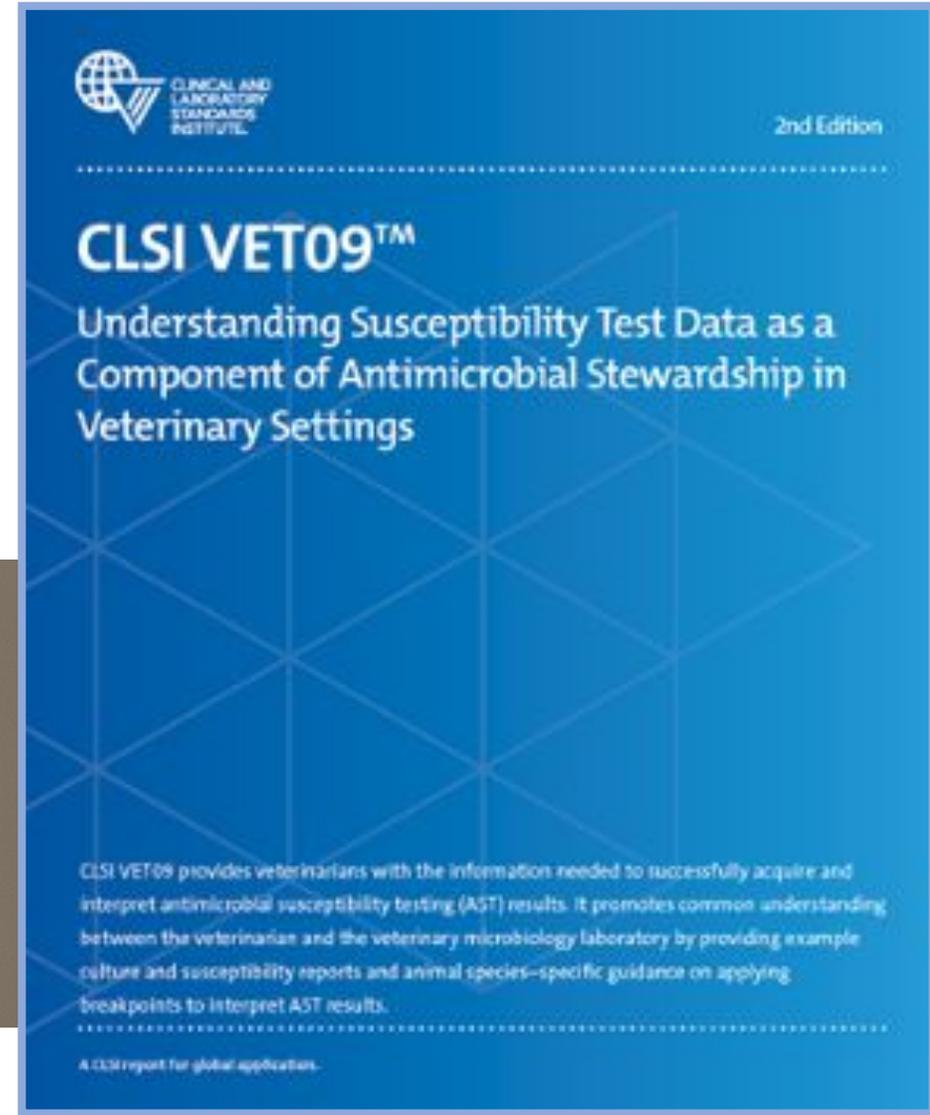
AVMA Publications

< Previous Article   Next Article >

A One Health perspective on the use of genotypic methods for antimicrobial resistance prediction

Kelli J. Maddock MS ✉ , Claire R. Burbick DVM, PhD, DACVM, Step...   View More +

DOI: https://doi.org/10.2460/javma.23.12.0687

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https://doi.org/10.1186/s44280-023-00024-w

One Health Advances

REVIEW   Open Access

Antimicrobial susceptibility testing in veterinary medicine: performance, interpretation of results, best practices and pitfalls

Referencias

Currents in One Health
Leading at the intersection of animal, human, and environmental health

AJVR · AVMA

Current state and future directions for veterinary antimicrobial resistance research

Kelli J. Maddock, MS[1]*; Robert Bowden, BS[2]; Stephen D. Cole, VMD, MS, DACVM[3]; Dubraska Diaz-Campos, DVM, PhD[4]; Joshua B. Daniels, DVM, PhD, DACVM[5]; Tessa E. LeCuyer, DVM, PhD, DACVM[6]; Xian-Zhi Li, MD, PhD[7]; John Dustin Loy, DVM, PhD, DACVM[8]; Susan Sanchez, PhD[9]; Brianna L. S. Stenger, PhD[1]; Claire R. Burbick, DVM, PhD, DACVM[10]

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Brote en PennVet: CRE

One Health, wicked problems, and carbapenem-resistant E. Coli: Penn Vet weathers the perfect storm

Tony McReynolds - 1/30/2020

US veterinary hospital faces rare antibiotic-resistant E coli

Chris Dall | News Reporter | CIDRAP News, January 10, 2020
Topics: *Antimicrobial Stewardship*

On Apr 1, 2019, Shelley Rankin, PhD, the chief of clinical microbiology at the University of Pennsylvania's School of Veterinary Medicine, got a surprising notification from the US Food and Drug Administration (FDA).

Four times a year, Rankin and her colleagues at PennVet ship off bacterial isolates from the school's veterinary hospitals to the

The Inquirer

HEALTH

Drug-resistant bacteria identified in 15 animal patients at Penn Vet Hospital

The veterinary hospital is working with the Philadelphia Department of Health and the Centers for Disease Control and Prevention.

Infection Control & Hospital Epidemiology
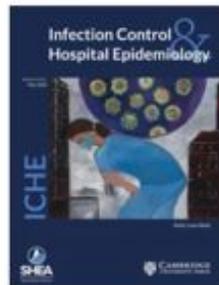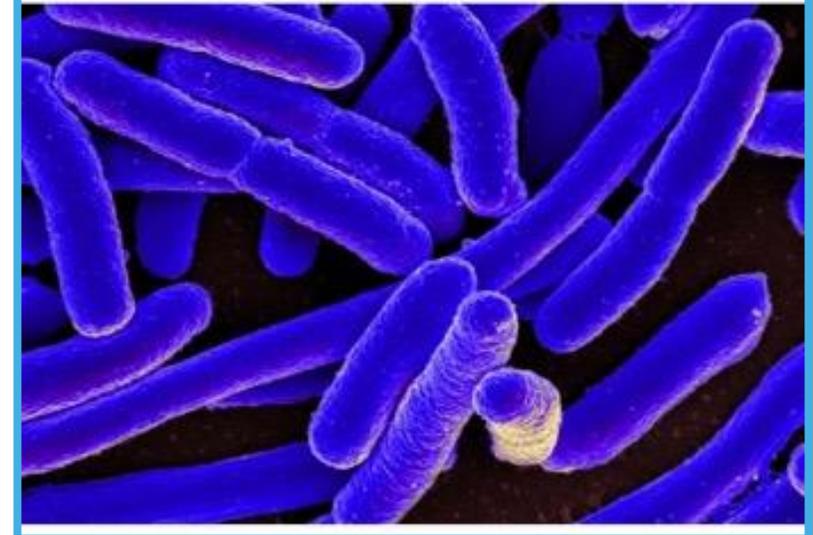
An Outbreak of New Delhi Metallo--Lactamase-5 (blaNDM-5)–Producing *Escherichia coli* in Companion Animals in the United States

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Shelley C. Rankin and Stephen D. Cole
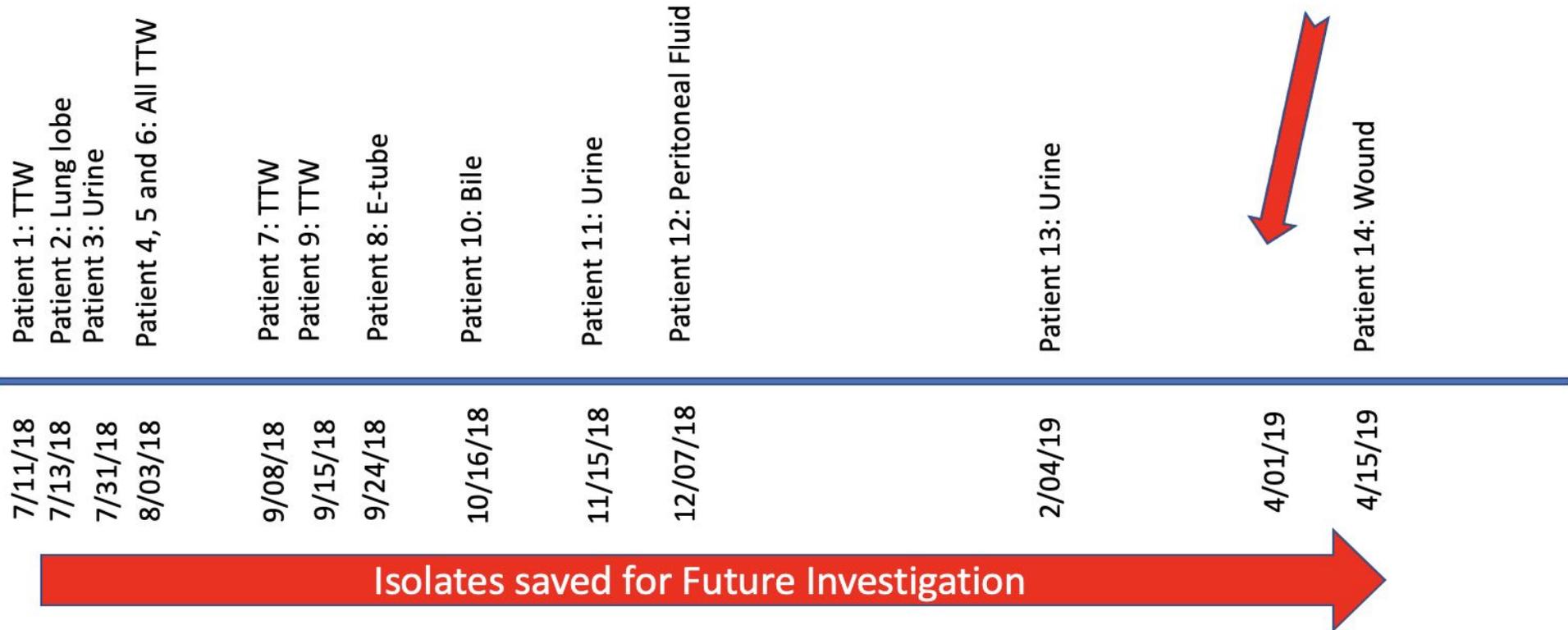
Infection Control & Hospital Epidemiology

PennVet

Cortesía: Dr. Stephen Cole

Programas de Vigilancia

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