Ceftazidime / avibactam

"We're gonna geed a bigger boat" Spellberg B, Bonomo RA

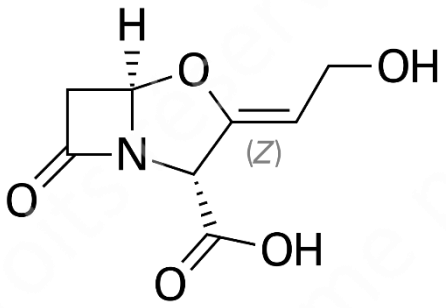
Michel Arthur
Laboratoire de Recherche Moléculaire sur les Antibiotiques
Centre de Recherches des Cordeliers
INSERM UMR S 1138 Equipe 12

A new generation of β-lactamase inhibitors: Structure

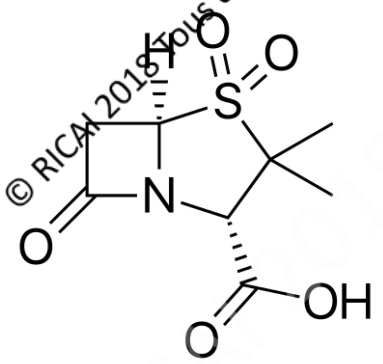
1st generation : β-lactam

2nd generation : Diazabicyclooctane

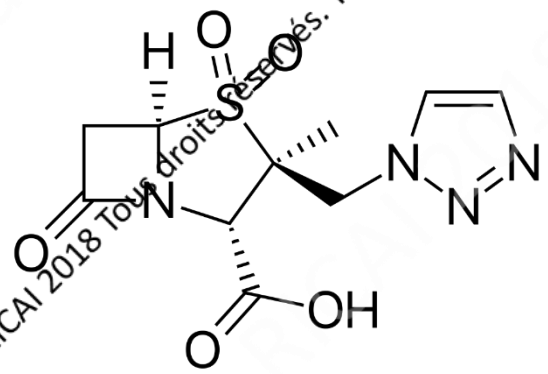
Clavulanate



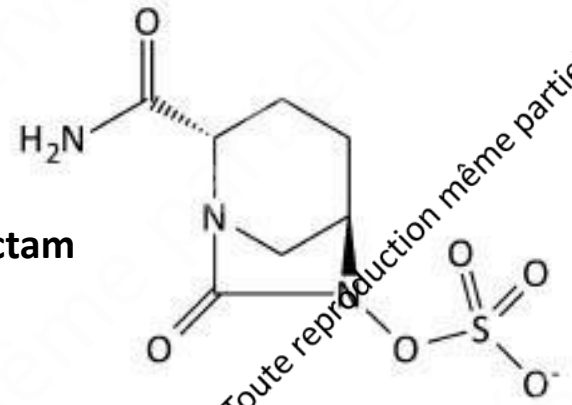
Sulbactam



Tazobactam



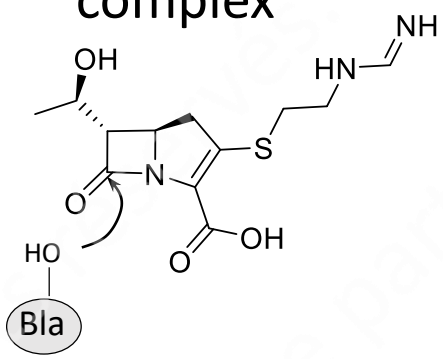
Avibactam

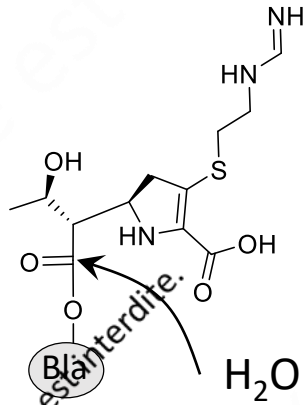


Different modes of action

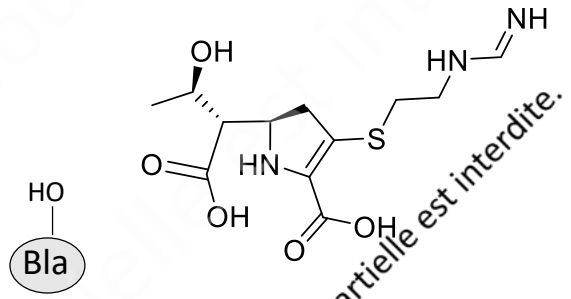
E β-lactamase + **S** Substrate β-lactam ⇌ **ES** Non-covalent complex → **ES*** Acylenzyme → **E** β-lactamase + **S*** Detoxified β-lactam

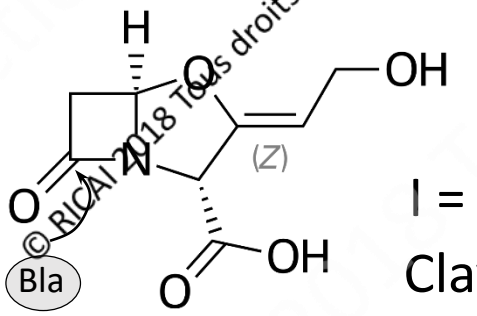
Imipenem





H_2O



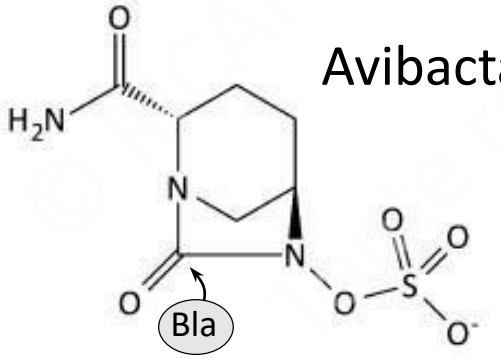


I = Inhibitor

Clavulanate

E + I ⇌ **EI** Non-covalent complex → **EI*** Acylenzyme + H_2O ⇏ **E** β-lactamase + **I*** Detoxified inhibitor

Secondary acylenzyme + CO_2



Avibactam

E + I ⇌ **EI** Non-covalent complex → **EI*** Acylenzyme + H_2O ⇏ **E** β-lactamase + **I*** Detoxified inhibitor

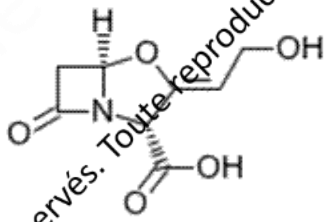
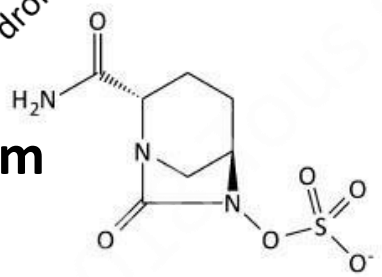
Inactivation spectrum of avibactam

β-lactamase	
Class A	Active
Class B	Inactive
Class C	Active
Class D	Variable

Questions to be addressed:

- Are there naturally-occurring "resistant" class A β-lactamases?
- Are variations in the efficacy of avibactam and clavulanate
  - positively correlated?
  - negatively correlated?
  - independent?
- Is acquisition of resistance to avibactam-β-lactam combinations possible following changes in the sequence of the β-lactamases under the selective pressure of the drugs?

Naturally occurring β-lactamases not inactivated by avibactam

Inhibitor	β-lactamase: BlaC *M. tuberculosis*	β-lactamase: Bla_{Mab} *M. abscessus*
Clavulanate 	Rapid inactivation k_2/K_i = 230,000 M^{-1} s^{-1} No deacylation k_{-2} = 0 Insignificant hydrolysis k_3 = 0.0001 s^{-1}	No inactivation, Hydrolysis k_{cat} / K_m = 210,000 M^{-1} s^{-1}
Avibactam 	Slow inactivation k_2 / K_i = 24 M^{-1} s^{-1}	Rapid inactivation k_2 / K_i = 480,000 M^{-1} s^{-1} k_{-2} = 0.00002 s^{-1}

Red: Full "irreversible" inactivation

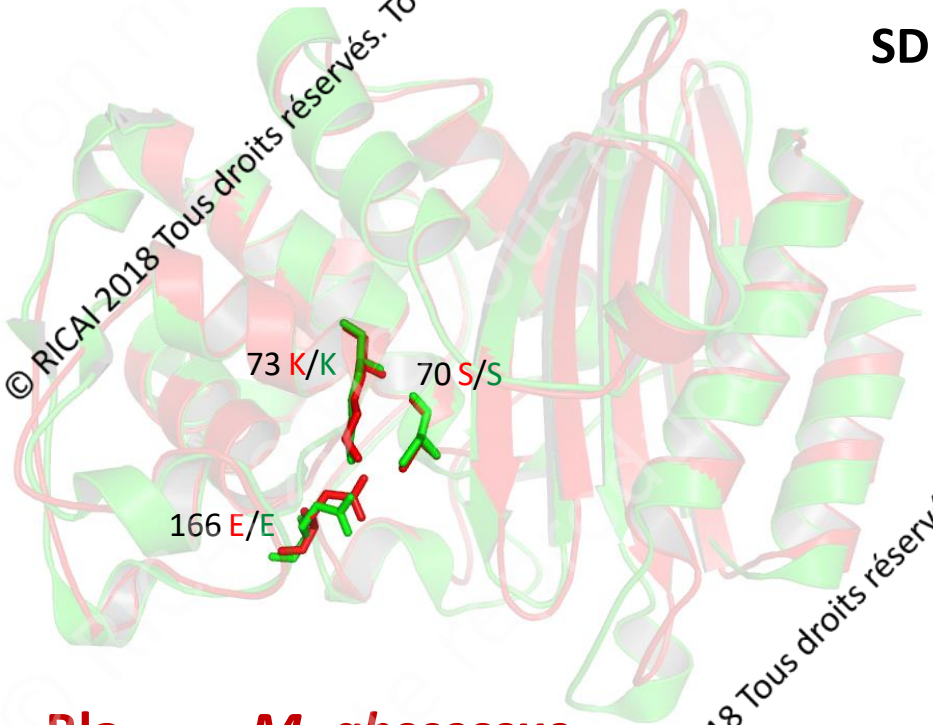
Blue : Partial or no inactivation

$$E + I \underset{\text{Dissociation } k_{-1}}{\overset{k_1 \text{ Association}}{\rightleftharpoons}} EI \underset{\text{Deacylation } k_{-2}}{\overset{k_2 \text{ Acylation}}{\rightleftharpoons}} EI^* \xrightarrow{k_3 \text{ Hydrolysis}} E + I^*$$

$K_i = k_{-1}/k_1$

Structural data provided a clue

β-lactamase	Motif	Avibactam	Clavulanate
Bla_{Mab}	SDN	Active	Inactive
BlaC	SDG	Inactive	Active

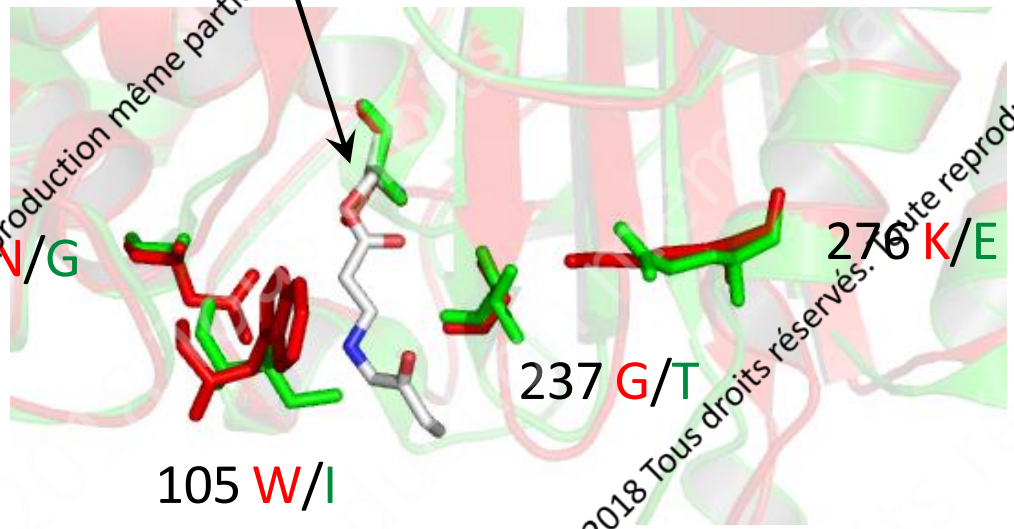


Bla_{Mab} *M. abscessus*

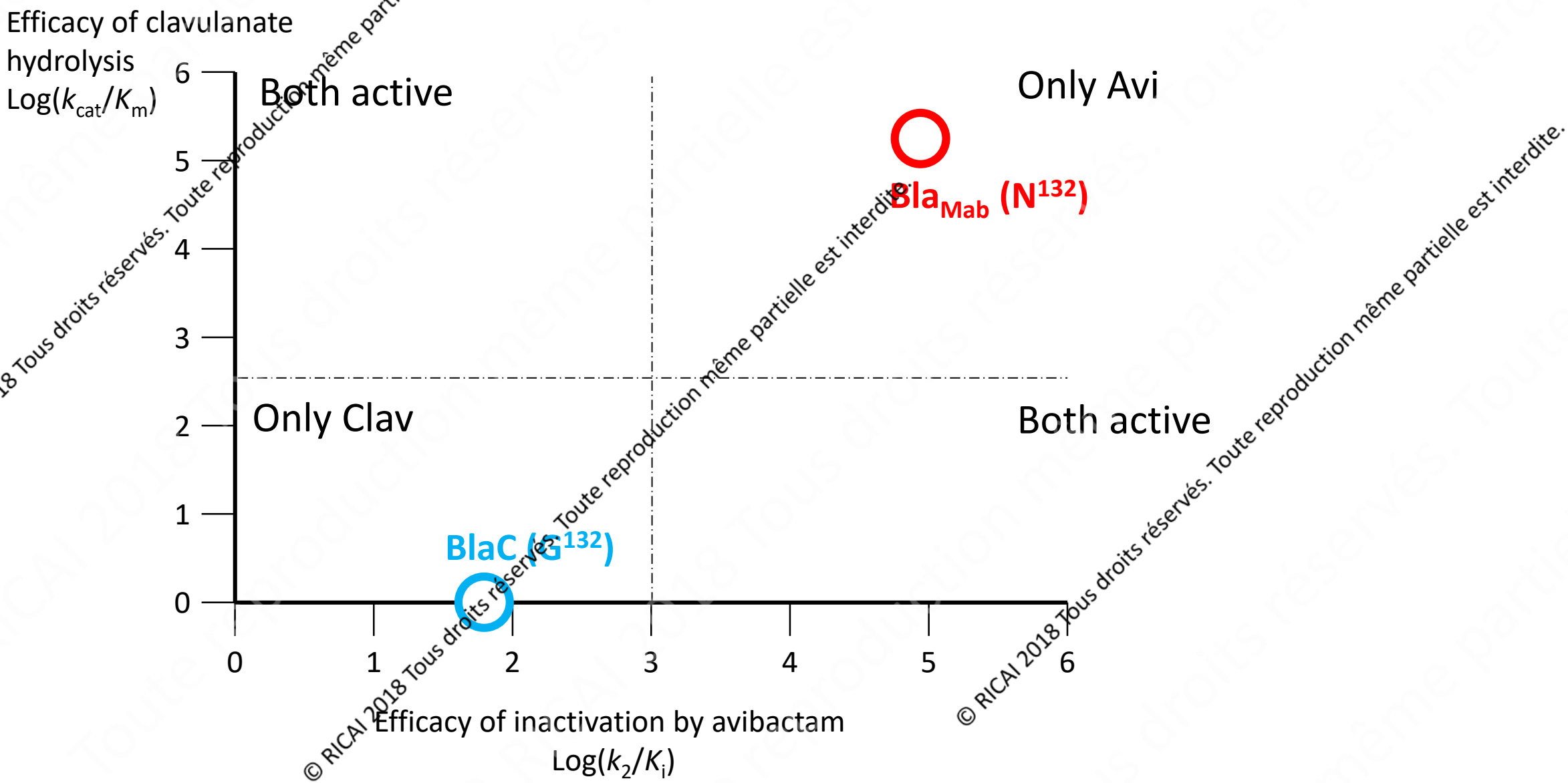
BlaC *M. tuberculosis*

SDN or SDG

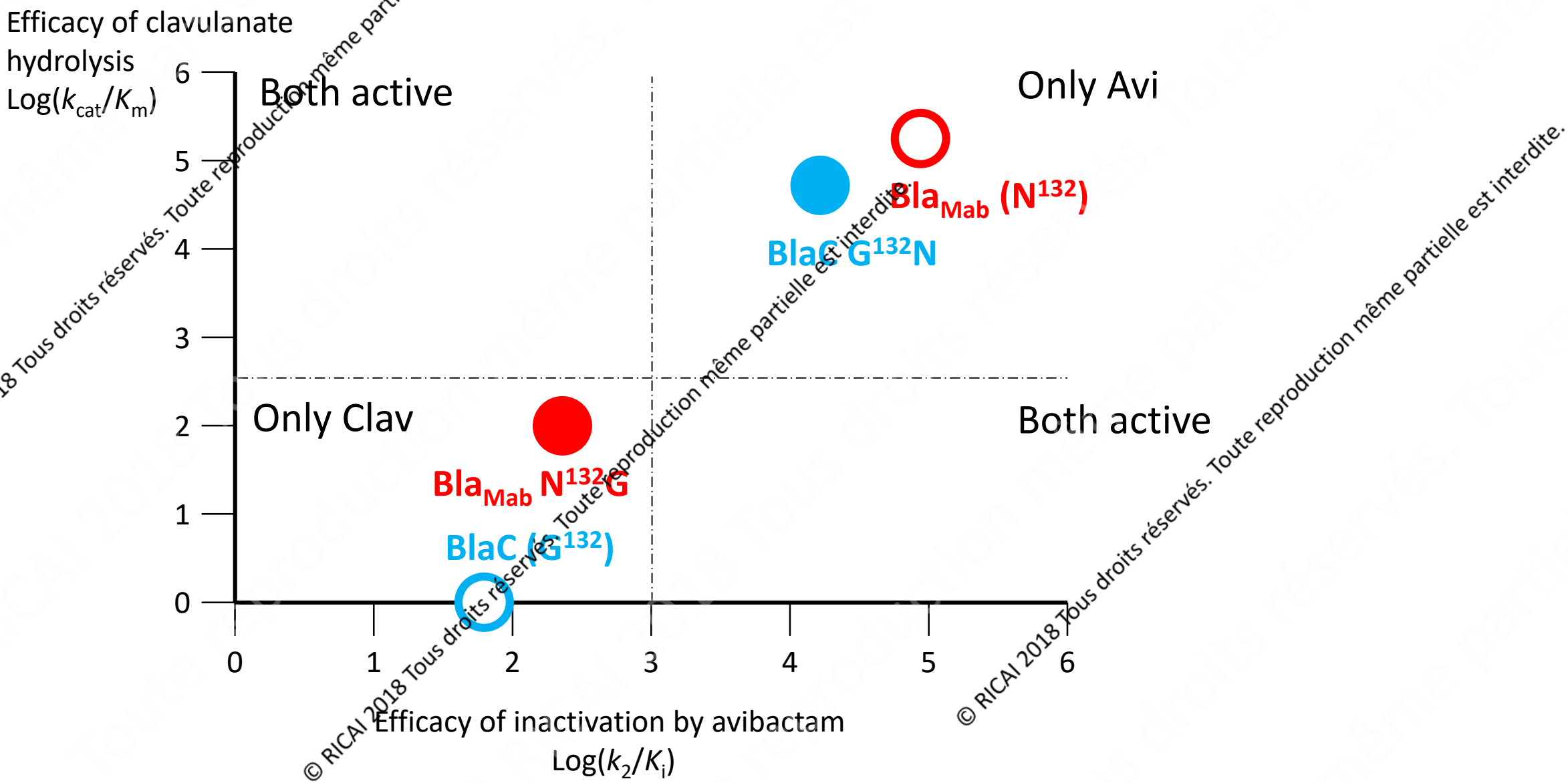
Catalytic serine (S70) acylated by clavulanate



Impact of SDN ↔ SDG substitutions



Impact of SDN ↔ SDG substitutions



Conclusions

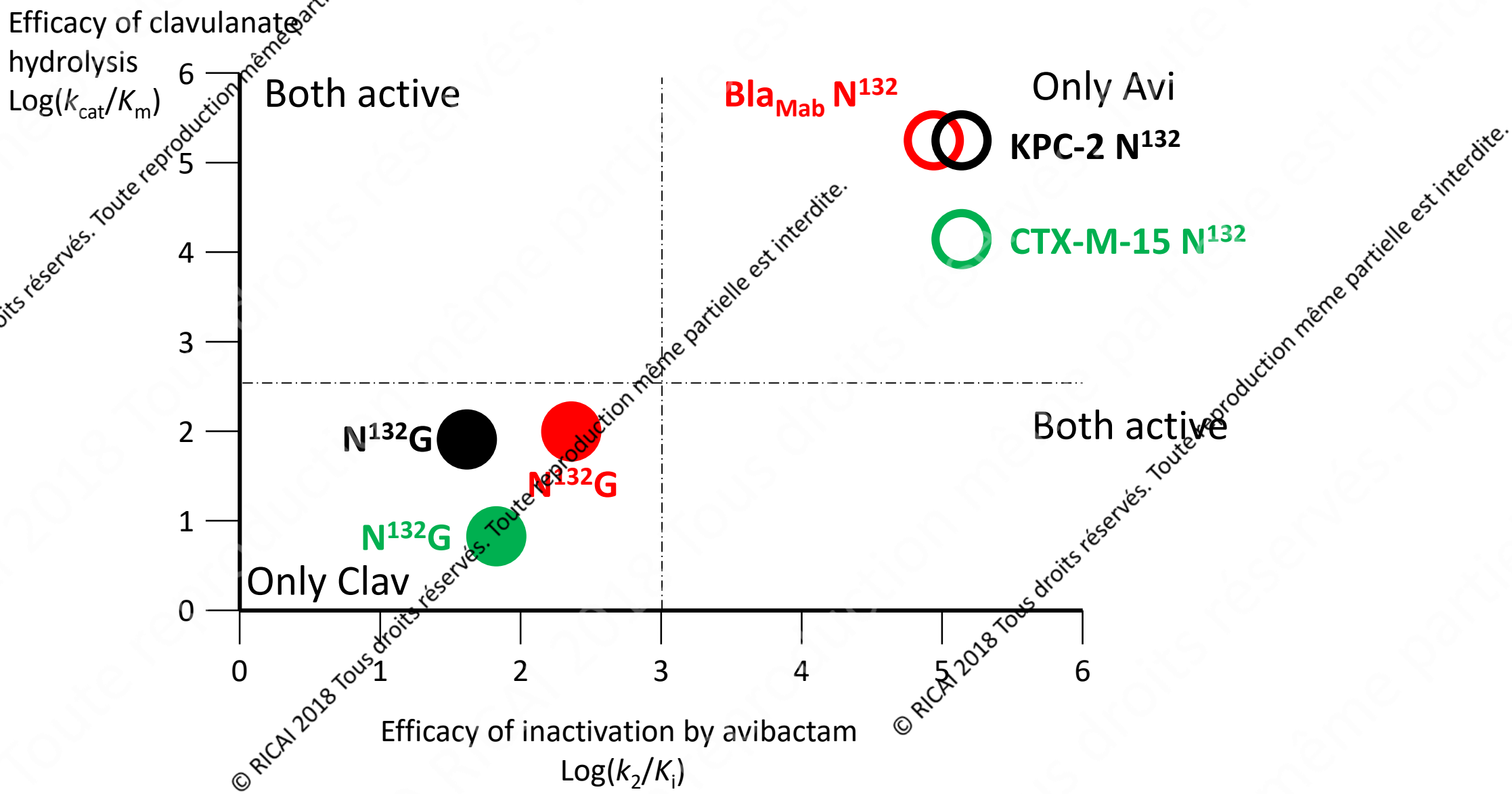
- BlaC is a naturally-occurring avibactam “resistant” class A β-lactamase

- Variations in the efficacy of avibactam and clavulanate are inversely correlated and determined by the SDN versus SDG motif

- SDN → SDG acquisition of avibactam resistance (but increased susceptibility to clavulanate)

→ Do these conclusions apply to β-lactamases from Enterobacteria?

Impact of SDN ↔ SDG substitutions



Conclusion

- The SDN → SDG substitution has similar impacts on the spectrum of inhibition of distantly related Class A β-lactamases from mycobacteria and enterobacteria

→ Does this substitution lead to resistance to β-lactam/inhibitor combinations?

MIC of amoxicillin (S) against *E. coli* producing Bla_{Mab} (E) with or without avibactam (I)

β-lactamase	MIC (µg/ml)	Active (black) and inactive (white) pathways
None	2	$S \rightarrow$ PBP inactivation
Bla_{Mab}	>512	$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation)
Bla_{Mab} + Avibactam	4	$E + S \rightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation); $E + I \leftrightarrows EI \leftrightarrows EI^*$
Bla_{Mab} $N^{132}G$	>512	$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation)
Bla_{Mab} $N^{132}G$ + Avi	64	$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation); $E + I \leftrightarrows EI \leftrightarrows EI^*$

Emergence of ceftazidime-avibactam resistance in enterobacteria

(Resistance: MIC of ceftazidime > 8 µg/ml in the presence of 4 µg/ml of avibactam)

- ***In vivo***: Emergence of resistance to ceftazidime-avibactam in 8% (3/37) of the patients infected with carbapenem-resistant Enterobacteriaceae and treated with the ceftazidime-avibactam combination

  $D^{179}Y$ $V^{240}G$ $D^{179}Y + T^{243}M$ *Shields RK, Clin Infect Dis 63:1615–8*
  Antimicrob Agents Chemother 61:e02097-16

- ***In vitro***: *ca*. 2 x 10^{-9} (ceftazidime 8 µg/ml + avibactam 4 µg/ml)

  $D^{179}Y$

Impact of D^{179}Y in KPC-2 on the :

MIC of β-lactams against *E. coli*

	Ceftazidime			Aztreonam	Meropenem	Imipenem	Ceftriaxone
β-Lactamase	None	+Avi	+Clav	None	None	None	None
None	0.25	0.25	0.25	0.25	<0.12	0.25	<0.12
KPC-2	>128	1	64	>128	>128	128	>128
KPC-2 D^{179}Y	>128	32	8	0.5	<0.12	0.25	32

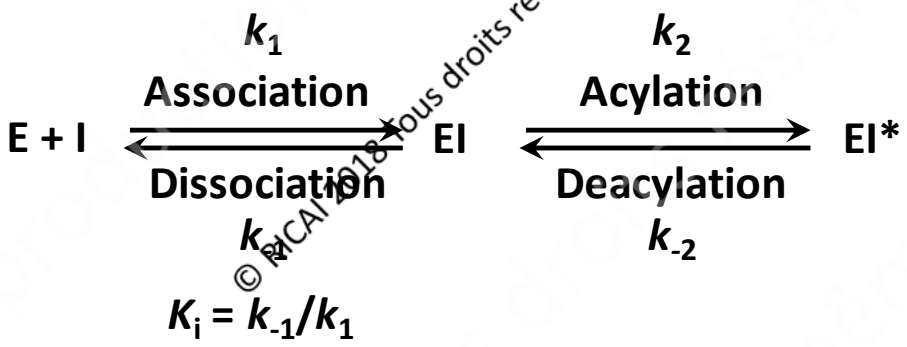
Efficacy* of β-lactam hydrolysis

β-lactam	KPC-2	KPC-2 D^{179}Y
Ceftazidime	3,700	70
Aztreonam	69,000	Not detected
Meropenem	67,000	Not detected
Imipenem	730,000	Not detected
Ceftriaxone	250,000	3,500
Clavulanate	140,000	Not detected

*k_{cat}/K_m ($M^{-1}s^{-1}$)

Efficacy of inactivation by avibactam

β-lactamase	k_2/K_i ($M^{-1}s^{-1}$)	k_2 (s^{-1})
KPC-2	290,000	0.001
KPC-2 D^{179}Y	0.4	0.00005



$K_i = k_{-1}/k_1$

Conclusions

- $D^{179}Y$ is sufficient for resistance to the combination since it enables the combination of: Sufficient residual ceftazidime hydrolase activity (2%)
  Very low acylation efficacy by avibactam (0,0001%)

- $D^{179}Y$: Abolishes resistance to aztreonam, imipenem, and meropenem
  Increases the efficacy of β-lactamase inactivation by clavulanate

→ Alternative therapies for isolates producing KPC $D^{179}Y$?
→ Combine therapies to prevent emergence of $D^{179}Y$?

→ Is emergence of resistance to β-lactam/inhibitor combinations possible in other Class A β-lactamases?

CTX-M β-lactamases are refractory to gain of ceftazidime-avibactam resistance

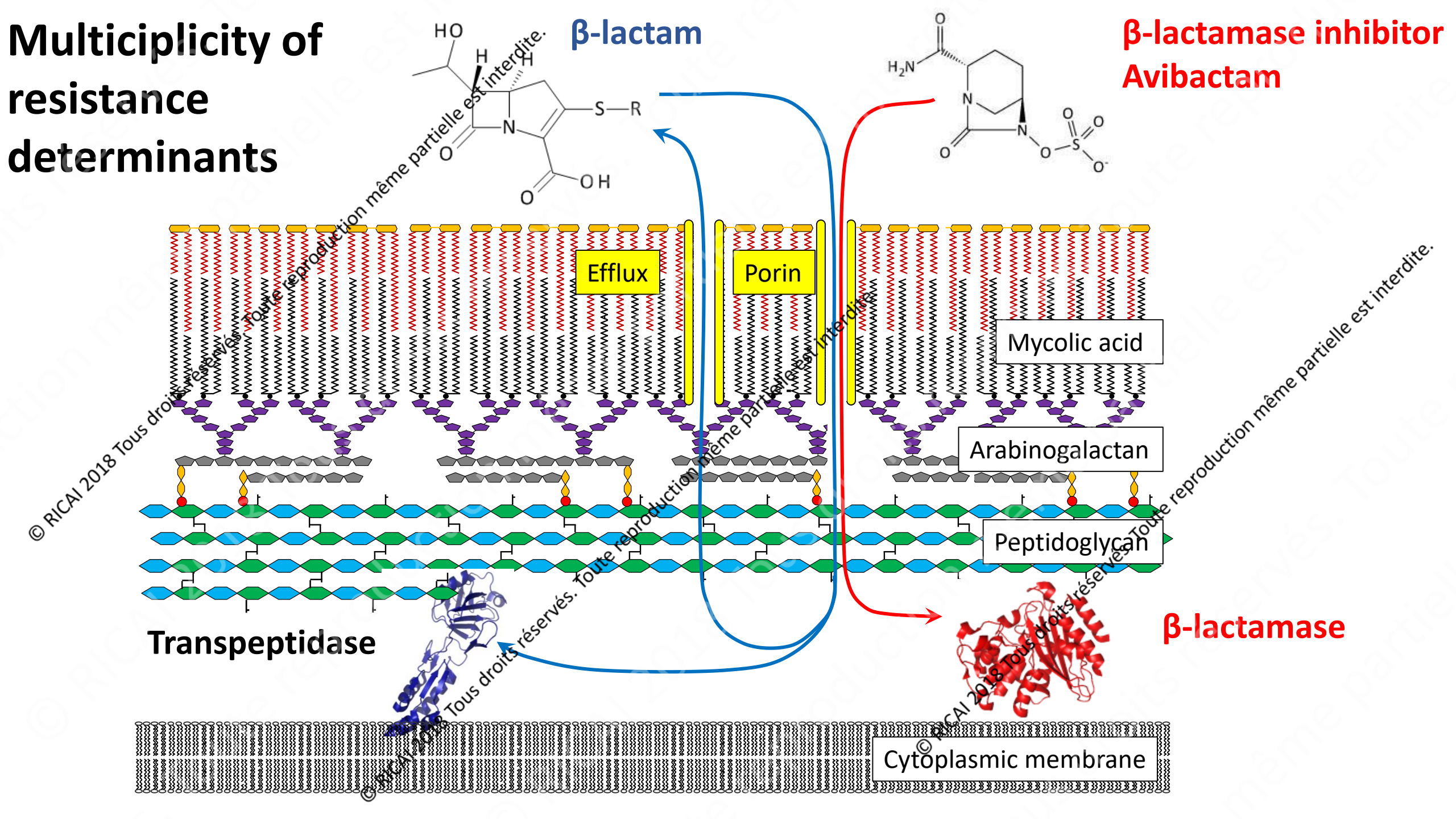
- Pre-existing polymorphisms: 9 single amino acid variations in the Ω loop of 172 CTX-M sequences

- None was associated with ceftazidime-avibactam resistance but

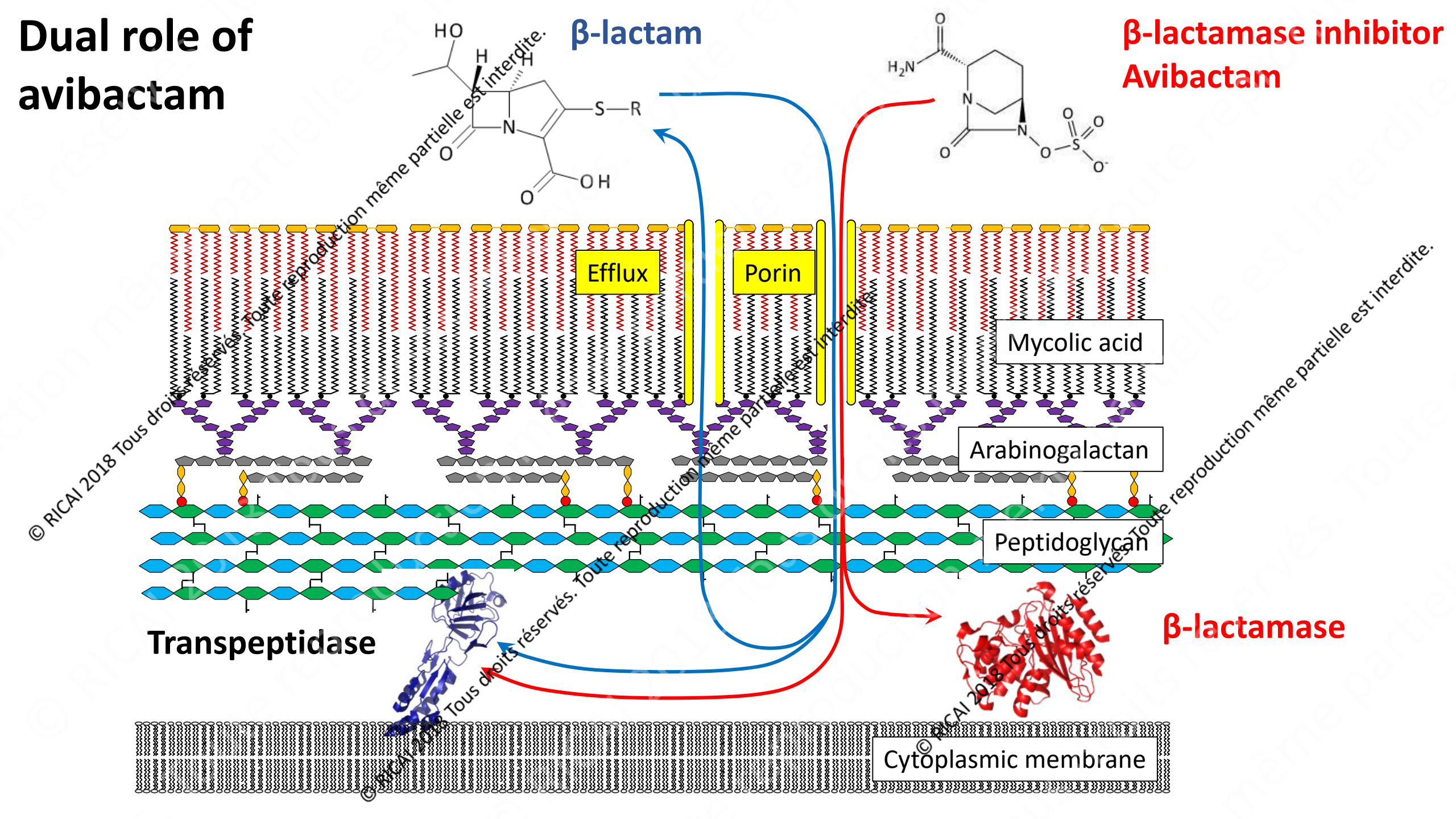
  Two of them increased the MIC of ceftazidime (in the absence of avibactam) when introduced in CTX-M-15: $P^{167}S$ (4 fold) and $L^{169}Q$ (16 fold)

- $L^{169}Q$ (rare) and $D^{240}G$ (only in CTX-M-15 and derivatives) are prerequisites for the emergence of ceftazidime-avibactam resistance

Multiciplicity of resistance determinants



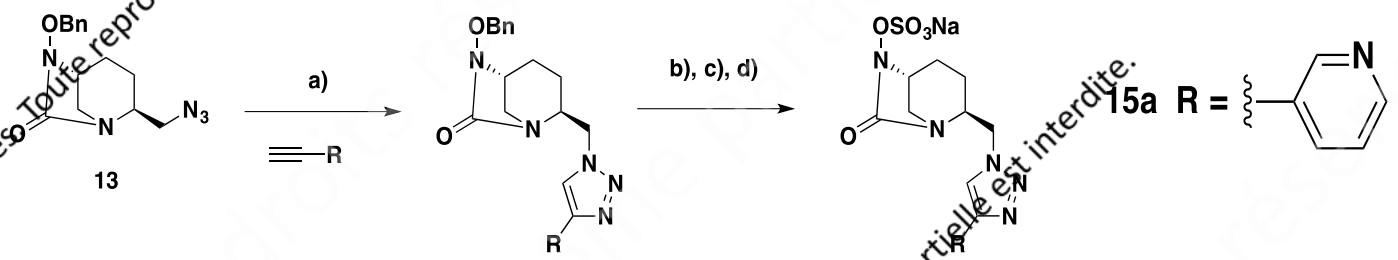
Dual role of avibactam

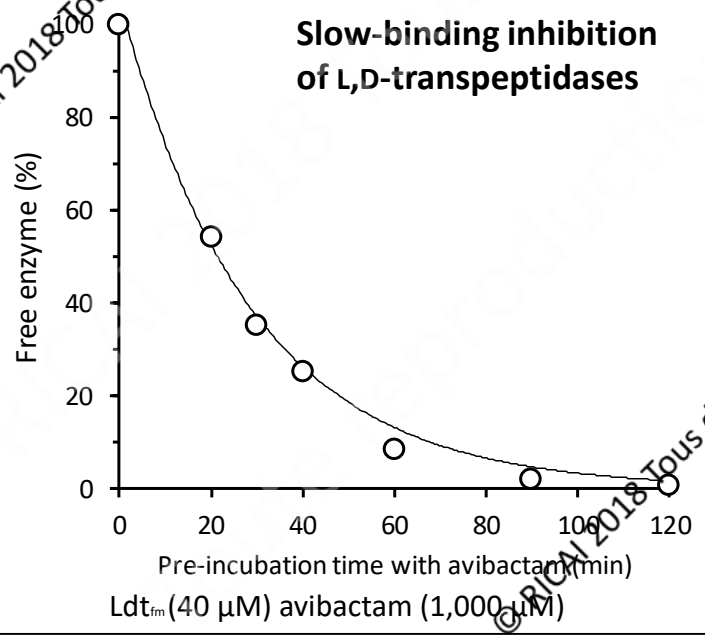


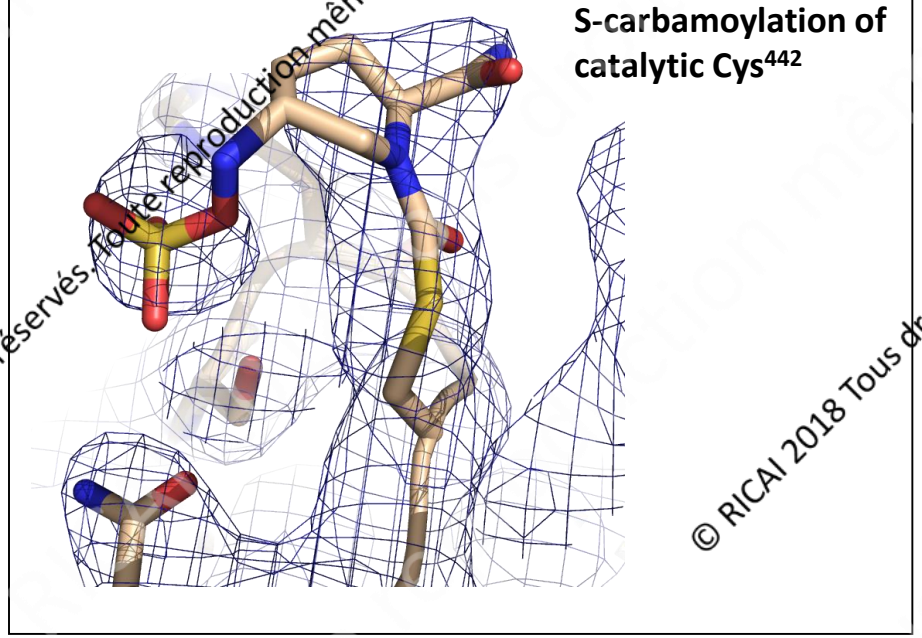
Additional target(s) for avibactam and derivatives

Strain	β-lactamase	MIC of amoxicillin (against Δ*bla*)			Inhibition efficacy (k_2/K_i $M^{-1}s^{-1}$)	
		None	Avibactam	15a	Avibactam	15a
M. abscessus	Bla_{MAb}	>256 (4)	16 (4)	16 (4)	170,000	22,000
M. tuberculosis	BlaC	128 (1)	8 (1)	16 (1)	24	< 5

Organic synthesis of avibactam derivatives







“We're gonna need a bigger boat” Spellberg B, Bonomo RA

- A more potent “irresistible” inhibitor

and/or

- Diverse inhibitors (pan resistance to all β-lactam-inhibitor combinations is at the very least uncommon)

- Formulation of inhibitors independently from β-lactams
  → access to combinations for therapy (and research!)

- Chemistry
  L Iannazzo, M Ethève-Quelquejeu, M Fonvielle, F Bochet

- Crystallography
  I Galley, H van Tilbeurgh, M Fonvielle

- Microbiology
  F Compain, JL Mainardi, E Le Run

- Enzymology
  Z Edoo, F Compain, JE Hugonnet

Agence Nationale de la Recherche ANR
MycWall project

FONDATION RECHERCHE MÉDICALE
PhD fellowship to ZE

VAINCRE LA MUCOVISCIDOSE
PhD fellowship to ELR

MIC of amoxicillin against *E. coli* strains producing BlaC from *M. tuberculosis*

E = BlaC
S = Amoxicillin
I = Clavulanate

	β-lactamase	MIC (µg/ml)
$S \xrightarrow{\text{PBP inactivation}}$	None	2
$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation)	BlaC	512
$E + S \rightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation) $E + I \rightleftharpoons EI \rightarrow EI^* + H_2O \rightarrow E + I^*$	BlaC + Clav	8
$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation)	BlaC $G^{132}N$	512
$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation) $E + I \rightleftharpoons EI \rightarrow EI^* + H_2O \rightarrow E + I^*$	BlaC $G^{132}N$ + Clav	64

MIC of amoxicillin against *E. coli* strains producing BlaC from *M. tuberculosis*

E = Bla_{Mab}
S = Amoxicillin
I = Clavulanate

	β-lactamase	MIC (µg/ml)
S → PBP inactivation	None	2
$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation)	Bla_{Mab}	>512
$E + S \rightleftharpoons ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation); $E + I \rightleftharpoons EI \rightarrow EI^* + H_2O \rightarrow E + I^*$	Bla_{Mab} + Clav	>512
$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation)	Bla_{Mab} $N^{132}G$	>512
$E + S \leftrightarrow ES \rightarrow ES^* + H_2O \rightarrow E + S^*$ (PBP inactivation); $E + I \rightleftharpoons EI \rightarrow EI^* + H_2O \rightarrow E + I^*$	Bla_{Mab} $N^{132}G$ + Clav	32

MIC of amoxicillin against *E. coli* strains producing BlaC from *M. tuberculosis*

E = BlaC
S = Amoxicillin
I = Avibactam

	β-lactamase	MIC (µg/ml)
S → PBP inactivation	None	2
E + S ⟷ ES → ES* + H_2O → E + S* (PBP inactivation)	BlaC	512
E + S ⟷ ES → ES* + H_2O → E + S* (PBP inactivation); E + I ⇆ EI ⇆ EI*	BlaC + Avibactam	64
E + S ⟷ ES → ES* + H_2O → E + S* (PBP inactivation)	BlaC $G^{132}N$	512
E + S ⟷ ES → ES* + H_2O → E + S* (PBP inactivation); E + I ⇆ EI ⇆ EI*	BlaC $G^{132}N$ + Avi	2

TABLE 3 Impact of avibactam on the MICs of β-lactams for *E. coli* strains producing various β-lactamases

β-Lactamase	MIC (μg/ml) of indicated β-lactam with or without Avi or Clav[a]											
	Amoxicillin			Ceftazidime			Ceftaroline			Aztreonam		
	None	Avi	Clav	None	Avi	Clav	None	Avi	Clav	None	Avi	Clav
None[b]	2	2	2	0.25	0.25	0.25	0.06	0.06	0.06	0.25	0.25	0.25
KPC-2	>4,096	256	2,048	128	1	32	>1,024	1	128	>1,024	0.25	128
KPC-2 $N^{132}G$	2,048	2,048	8	4	1	0.5	32	0.5	0.06	1	0.125	0.25
CTX-M-15	4,096	2	16	16	0.5	0.5	>2,048	0.06	0.06	128	0.25	0.25
CTX-M-15 $N^{132}G$	128	8	4	0.5	0.5	0.25	0.5	0.06	0.06	0.25	0.25	0.125

[a] Values are the medians of results from three to five independent determinations. None, absence of avibactam and clavulanate; Avi, presence of avibactam (4 μg/ml);

© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.

TABLE 2 Impact of $D^{179}Y$ substitution on kinetic parameters of KPC-2 for hydrolysis of β-lactams

Kinetic parameter by β-lactam	Results[b]	
	KPC-2	KPC-2 $D^{179}Y$
Ampicillin		
k_{cat} (s^{-1})	200 ± 10	NA[a]
K_m (μM)	200 ± 40	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(1.0 ± 0.2) × 10^6	NA
CENTA		
k_{cat} (s^{-1})	100 ± 10	0.11 ± 0.01
K_m (μM)	32 ± 8	23 ± 3
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(3.1 ± 0.3) × 10^6	(4.8 ± 0.6) × 10^3
Ceftazidime		
k_{cat} (s^{-1})	1.4	(1.3 ± 0.2) × 10^{-3}
K_m (μM)	>600	19 ± 4
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(3.7 ± 0.1) × 10^3	70 ± 20
Ceftriaxone		
k_{cat} (s^{-1})	>25	(7.0 ± 0.1) × 10^{-4}
K_m (μM)	>100	0.20 ± 0.03
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(2.5 ± 0.1) × 10^5	(3.5 ± 0.5) × 10^3
Aztreonam		
k_{cat} (s^{-1})	>350	NA
K_m (μM)	>5 × 10^3	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(6.9 ± 0.3) × 10^4	NA
Meropenem		
k_{cat} (s^{-1})	1.8 ± 0.2	NA
K_m (μM)	27 ± 9	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(6.7 ± 2.3) × 10^4	NA
Imipenem		
k_{cat} (s^{-1})	48 ± 5	NA
K_m (μM)	66 ± 21	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(7.3 ± 2.4) × 10^5	NA
Clavulanate		
k_{cat} (s^{-1})	5.2 ± 0.7	NA
K_m (μM)	36 ± 4	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(1.4 ± 0.3) × 10^5	NA

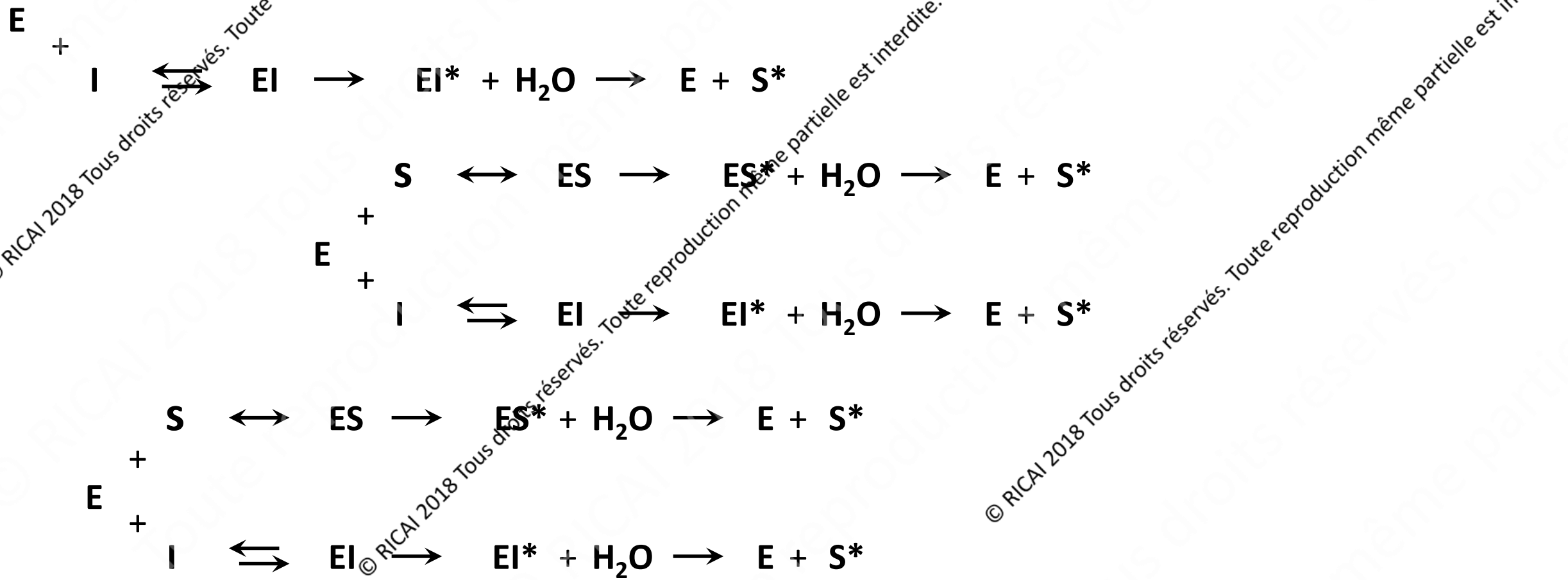
[a]NA, not applicable, as hydrolysis of β-lactams (100 μM, except for aztreonam at 1,000 μM) was not detected at the highest β-lactamase concentration tested (10 μM). Under these conditions, the lower limits of detection correspond to a turnover of $<2 \times 10^{-3} \cdot s^{-1}$, $<2.6 \times 10^{-3} \cdot s^{-1}$, $<1.5 \times 10^{-4} \cdot s^{-1}$, $<1.5 \times 10^{-4} \cdot s^{-1}$, and $<4 \times 10^{-4} \cdot s^{-1}$ for ampicillin, aztreonam, meropenem, imipenem, and clavulanate, respectively.

[b]Data are means ± standard errors of the mean (SEM).

© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.

Kinetic parameter by β-lactam	Results[b]	
	KPC-2	KPC-2 D[179]Y
Ampicillin		
k_{cat} (s^{-1})	200 ± 10	NA[a]
K_m (μM)	200 ± 40	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(1.0 ± 0.2) × 10^6	NA
CENTA		
k_{cat} (s^{-1})	100 ± 10	0.11 ± 0.01
K_m (μM)	32 ± 8	23 ± 3
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(3.1 ± 0.8) × 10^6	(4.8 ± 0.6) × 10^3
Ceftazidime		
k_{cat} (s^{-1})	>1.4	(1.3 ± 0.2) × 10^{-3}
K_m (μM)	>600	19 ± 4
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(3.7 ± 0.1) × 10^3	70 ± 20
Ceftriaxone		
k_{cat} (s^{-1})	>25	(7.0 ± 0.1) × 10^{-4}
K_m (μM)	>100	0.20 ± 0.03
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(2.5 ± 0.1) × 10^5	(3.5 ± 0.5) × 10^3
Aztreonam		
k_{cat} (s^{-1})	>350	NA
K_m (μM)	>5 × 10^3	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(6.9 ± 0.3) × 10^4	NA
Meropenem		
k_{cat} (s^{-1})	1.8 ± 0.2	NA
K_m (μM)	27 ± 9	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(6.7 ± 2.3) × 10^4	NA
Imipenem		
k_{cat} (s^{-1})	48 ± 5	NA
K_m (μM)	66 ± 21	NA
k_{cat}/K_m ($M^{-1} \cdot s^{-1}$)	(7.3 ± 2.4) × 10^5	NA
Clavulanate		
k_{cat} (s^{-1})	5.2 ± 0.7	NA

E = BlaC
S = Amoxicillin
I = Clavulanate



β-Lactamase	Residue 132	MIC (mg/L) of amoxicillin in the presence of the indicated inhibitor: none	avibactam (4 mg/L)	clavulanate (4 mg/L)
None	NA	2	2	2
Bla_{Mab}	N	>512	4	>512
BlaC	G	512	64	8
Bla_{Mab} $N^{132}G$	G	>512	64	32
BlaC $G^{132}N$	N	512	2	64

NA, not applicable.

TABLE 2 Impact of the $N^{132}G$ substitution in KPC-2 and CTX-M-15 on the kinetic parameters of β-lactams and their hydrolysis

β-Lactamase	Amoxicillin k_{cat} (s^{-1})	Amoxicillin K_m (μM)	Amoxicillin k_{cat}/K_m (M^{-1} s^{-1})	Nitrocefin k_{cat} (s^{-1})	Nitrocefin K_m (μM)	Nitrocefin k_{cat}/K_m (M^{-1} s^{-1})
KPC-2	150 ± 10	190 ± 50	(7.9 ± 0.2) × 10^5	42 ± 2	11 ± 3	(3.9 ± 1.0) × 10^6
KPC-2 $N^{132}G$	7.0 ± 0.4	46 ± 9	(1.5 ± 0.3) × 10^5	35 ± 1	27 ± 3	(1.3 ± 0.1) × 10^6
CTX-M-15	40 ± 4	19 ± 1	(4.8 ± 0.5) × 10^5	190 ± 30	47 ± 17	(4.0 ± 1.5) × 10[illegible]
CTX-M-15 $N^{132}G$	0.3 ± 0.01	<50	>6.2 × 10^3	8.3 ± 0.3	3.3 ± 0.7	(2.5 ± 0.5) × [illegible]

β-Lactamase	Ceftazidime k_{cat} (s^{-1})	Ceftazidime K_m (μM)	Ceftazidime k_{cat}/K_m (M^{-1} s^{-1})	Ceftaroline k_{cat} (s^{-1})	Ceftaroline K_m (μM)	Ceftaroline k_{cat}/K_m (M^{-1} s^{-1})	Aztreonam k_{cat} (s^{-1})	Aztreonam K_m (μM)	Aztreonam k_{cat}/K_m (M^{-1} s^{-1})
KPC-2	>1.4	>600	(3.7 ± 0.1) × 10^3	>40	>600	(5.1 ± 2.2) × 10^4	>350	[illegible]5,000	(6.9 ± 0.3) × 10^4
KPC-2 $N^{132}G$	[illegible]0.008	>600	14 ± 3	>2.5	>600	(4.5 ± 0.2) × 10^3	>0.6	>5,000	(9.8 ± 1.9) × 10^1
CTX-M-15	>1.4	>600	(2.2 ± 0.2) × 10^3	90 ± 6	51 ± 12	(1.8 ± 0.4) × 10^6	1.2 ± 0.[illegible]	<150	>8.0 × 10^3
CTX-M-15 $N^{132}G$	>0.01	>600	13 ± 1	4.7 ± 0.5	240 ± 50	(1.9 ± 0.4) × 10^4	0.33 ± 0.06	3,600 ± 1,100	(9.0 ± 3.0) × 10^1

β-Lactamase	MIC (μg/ml) of indicated β-lactam with or without Avi or Clav[a]: Amoxicillin None	Amoxicillin Avi	Amoxicillin Clav	Ceftazidime None	Ceftazidime Avi	Ceftazidime Clav	Ceftaroline None	Ceftaroline Avi	Ceftaroline Clav	Aztreonam None	Aztreonam Avi	Aztreonam Clav
None[b]	2	2	2	0.25	0.25	0.25	0.06	0.06	0.06	0.25	0.25	0.25
KPC-2	>4,096	256	2,048	128	1	32	>1,024	1	128	>1,024	0.25	128
KPC-2 $N^{132}G$	2,048	2,048	8	4	1	0.5	32	0.5	0.06	1	0.125	0.25
CTX-M-15	4,096	2	16	16	0.5	0.5	>2,048	0.06	0.06	128	0.25	0.25
CTX-M-15 $N^{132}G$	128	8	4	0.5	0.5	0.25	0.5	0.06	0.06	0.25	0.25	0.125

[a]Values are the medians of results from three to five independent determinations. None, absence of avibactam and clavulanate; Avi, presence of avibactam (4 μg/ml);

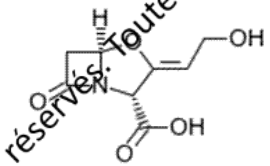
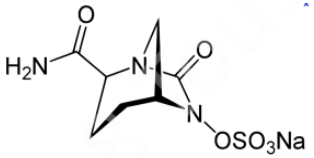
BlaC	Km	Kcat	kcat/Km, M/s tableau	kcat/Km, µM/s	Kcat/Km, M/s
Amoxicilline	44	5.8	130000	0.131818182	131818.1818
Nitrocéfine	70	35	490000	0.5	500000
Céfoxitine	140	1.1	7900	0.007857143	7857.142857
Céphalotine	260	23	88000	0.088461538	88461.53846
Céftazidime	>200	>0.004	270	0.00002	20
Céftaroline	>300	> 4.5	16000	0.015	15000
Imipénème	142	0.13	830	0.000915493	915.4929577
Méropénème	1	0.0003	330	0.0003	300
Faropénème	140	0.3	2100	0.002142857	2142.857143
Aztréonam	1600	0.18	120	0.0001125	112.5
Clavulanate			1E-20	#DIV/0!	#DIV/0!
Avibactam			63	#DIV/0!	#DIV/0!
BlaC G132N	Km	Kcat	kcat/Km, M/s tableau	kcat/Km, µM/s	Kcat/Km, M/s
Amoxicilline	220	26	120000	0.118181818	118181.8182
Nitrocéfine	32	54	1700000	1.6875	1687500
Céfoxitine	44	0.013	300	0.000295455	295.4545455
Céphalotine	>350	>32	110000	0.091428571	91428.57143
Céftazidime	3850	>0.02	59	5.71429E-05	57.14285714
Céftaroline	160	1.4	8700	0.00875	8750
Imipénème	10	0.12	12000	0.012	12000
Méropénème	0.9	0.0004	440	0.000444444	444.4444444
Faropénème	18	0.06	3300	0.003333333	3333.333333
Aztréonam	>1600	>4	2100	0.0025	2500
Clavulanate			21000	#DIV/0!	#DIV/0!
Avibactam			8900	#DIV/0!	#DIV/0!

BlaMab	Km	Kcat	kcat/Km, M/s tableau
Amoxicillin	890	780	880000
Nitrocefin	24	1000	43000000
Cefoxitin	500	0.003	6.7
Cephalotin	17	6.7	410000
Ceftazidime	>200	>0.03	83
Ceftaroline	>400	>22	55000
Imipenem	90	2.7	30000
Meropenem	120	1.8	15000
Faropenem	120	1.2	10000
Aztreonam	2900	1.8	620
Clavulanate			210000
Avibactam			110000

© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.

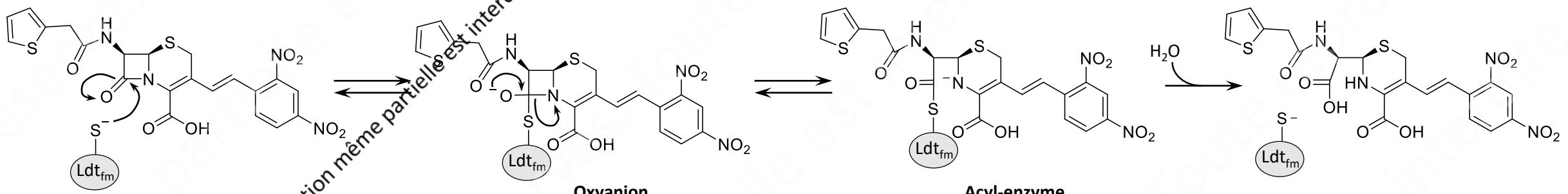
Table . Kinetic parameters for hydrolysis of β-lactams by β-lactamases from mycobacteria and inhibition of their activity

		β-lactamases (conserved motif)						
β-lactams	Parameter	BlaC (SDG)	BlaC $G^{132}N$	Bla_{Mab}	Bla_{Mab} $N^{132}G$	Bla_{Mch}	Bla_{Mma}	BlaS
Amoxicillin	K_m (μM)	44 ± 6.0	217 ± 44	890 ± 200	> 1150	710 ± 90	45 ± 16	63 ± 12
	k_{cat} (s^{-1})	5.8 ± 0.2	26 ± 3.0	780 ± 50	> 80	540 ± 30	12.4 ± 1.3	170 ± 10
	k_{cat}/K_m (M^{-1} s^{-1})	1.3 ± 0.2×10^5	1.2 ± 0.3×10^5	8.8 ± 2.0×10^5	7.3 ± 1.0 ×10^4	7.6 ± 1×10^5	2.8 ± 1.0×10^5	2.7 ± 0.5×10^6
Nitrocefin	K_m (μM)	71 ± 11	32 ± 8.0	24±7	26 ± 4.8	65±20	25±14	160±40
	k_{cat} (s^{-1})	35 ± 1.7	54 ± 6.0	1000±70	120 ± 51	930±70	2.7±0.3	640±50
	k_{cat}/K_m (M^{-1} s^{-1})	4.9±0.8×10^5	1.7 ± 0.5×10^6	4.3±1.2×10^7	2.2 ± 1.0 ×10^5	1.4±0.5×10^7	1.0±0.1×10^5	4.0±1.0×10^6
Cefoxitin	K_m (μM)	140±30	44 ±28	500 ± 270	350 ± 25	>800	210±45	>700
	k_{cat} (s^{-1})	1.1±0.07	0.013 ± 0.002	0.003±0.001	0.0016 ± 0.000075	>0.01	0.064±0.004	>0.001
	k_{cat}/K_m (M^{-1} s^{-1})	7.9±1.8×10^3	3.0 ± 2.0×10^2	6.7±3.8	4.5 ± 0.39	15±1	300±70	1.5±0.2
Cephalotin	K_m (μM)	260±50	> 350	17±1	220 ± 44	50±20	170±30	9.3±2.2
	k_{cat} (s^{-1})	23±2.2	> 32	6.7±0.1	0.39 ± 0.04	120±10	0.32±0.02	22±1
	k_{cat}/K_m (M^{-1} s^{-1})	8.8±1.9×10^4	1.1 ± 0.1×10^5	4.1 ± 0.2×10^5	1.8 ± 0.4 ×10^3	2.3±1×10^6	1.9±0.3×10^3	2.4±0.6×10^6
Ceftazidime	K_m (μM)	>200	> 350	>200	89 ± 35	>300	170±30	>300
	k_{cat} (s^{-1})	>0.004	> 0,02	>0.03	0.00048 ± 0.00008	>0.0004	0.32±0.02	>0.03
	k_{cat}/K_m (M^{-1} s^{-1})	2.7 ±0.62×10^2	5.9 ± 0.3×10^1	8.3 ±1.2 ×10^1	5.4 ± 2.3	1.1±0.05	1.9±0.3×10^3	110±10
Ceftaroline	K_m (μM)	>300	162±46	>400	> 300	33±6	>300	29±11
	k_{cat} (s^{-1})	>4.5	1.4±0.22	>22	> 0.0028	110±50	>0.02	250±20
	k_{cat}/K_m (M^{-1} s^{-1})	1.6 ± 0.05 ×10^4	8.7±2.8×10^4	5.5 ± 0.2 ×10^4	1.0 ± 0.2×10^1	3.0±1.5×10^5	50±7	8.3±2.9×10^6
Imipenem	K_m (μM)	142±29	10 ± 1.0	90±40	> 700	70±10	43±10	650±130
	k_{cat} (s^{-1})	0.13±0.02	0.12±0.002	2.7±0.3	>0.1	1.3±0.05	0.0031±0.0002	0.10±0.01
	k_{cat}/K_m (M^{-1} s^{-1})	8.3±2.3×10^2	1.2 ±0.1 ×10^4	3.0±1.4×10^4	9.5 ± 0.7 ×10^2	1.9±0.3×10^4	71±17	1.5±0.3×10^2
Meropenem	K_m (μM)	1±0.2	0.9 ± 0.5	120±20	> 450	7±2	10±6	22±5
	k_{cat} (s^{-1})	0.00030±0.00002	0.004 ±0 .0003	1.0±0.2	> 0.6	0.36±0.02	0.0018±0.0001	0.0095±0.0005
	k_{cat}/K_m (M^{-1} s^{-1})	3.3±0.6×10^2	4.4 ± 2.4 × 10^3	1.5±0.3×10^4	1.4 ± 0.03 ×10^3	5.5±1×10^4	1.7±1.0×10^2	4.3±1.0×10^2
Faropenem	K_m (μM)	141±74	18 ± 4.0	120±26	> 250	43±8	4.6±0.9	180±50
	k_{cat} (s^{-1})	0.30±0.08	0.06 ±0.002	1.26±0.12	> 0.3	1.1±0.06	0.0014±0.0001	0.23±0.02
	k_{cat}/K_m (M^{-1} s^{-1})	2.1±1.2×10^3	3.3 ± 0.8 × 10^3	1.0±0.2×10^4	1.0 ± 0.1 ×10^2	2.5±0.3×10^4	3.0±0.6×10^2	1.2±0.3×10^3
Aztreonam	K_m (μM)	1600±730	> 1600	2900±300	> 2000	>3000	>2500	>2500
	k_{cat} (s^{-1})	0.18±0.05	> 4.0	1.8±0.2	> 0.0008	>0.3	>0.03	>13
	k_{cat}/K_m (M^{-1} s^{-1})	1.2±0.6×10^2	2.1 ± 0.32 ×10^3	6.2±0.9×10^2	0.26 ± 0.09	100±10	7±1	5.4×10^3±220

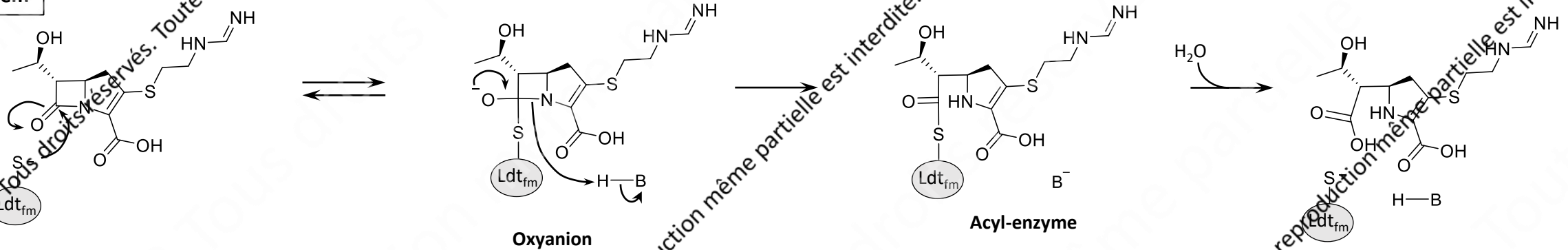
β-lactam	BlaC *M. tuberculosis*	Bla_{Mab} *M. abscessus*
Clavulanate 	Irreversible inactivation	Hydrolysis $k_{cat} / K_m = 2.1 \times 10^5\ M^{-1}\ s^{-1}$
Avibactam 	Slow inhibition $k_2 / K_i = 24\ M^{-1}\ s^{-1}$	Rapid inhibition $k_2 / K_i = 480{,}000\ M^{-1}\ s^{-1}$

© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.
© RICAI 2018 Tous droits réservés. Toute reproduction même partielle est interdite.

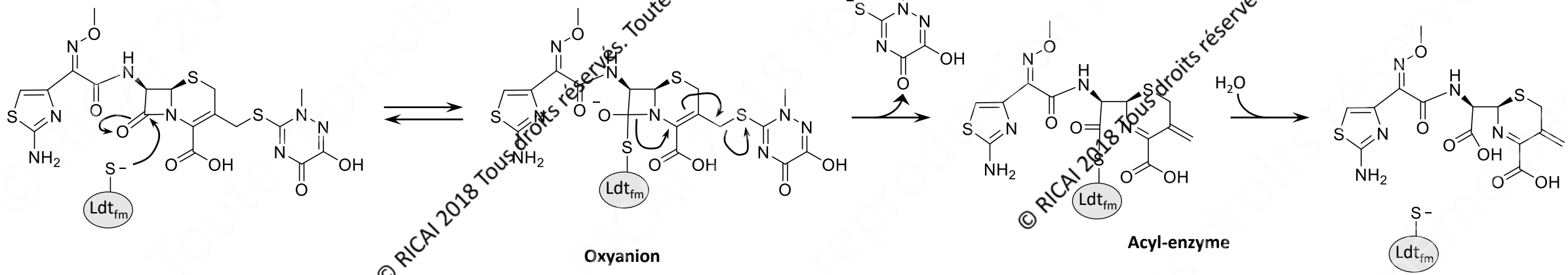
Nitrocefin

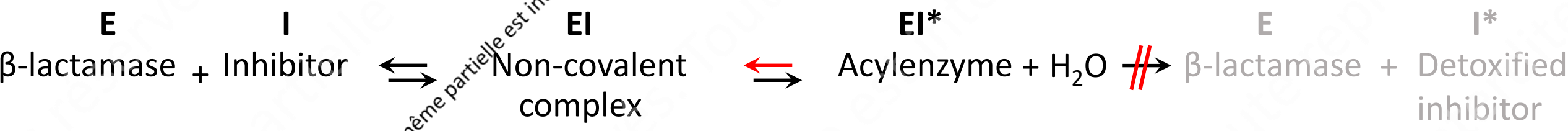


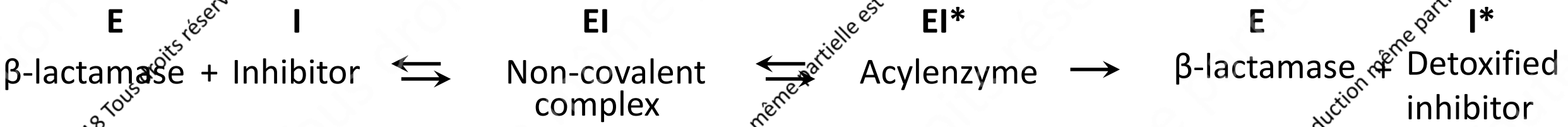
Imipenem

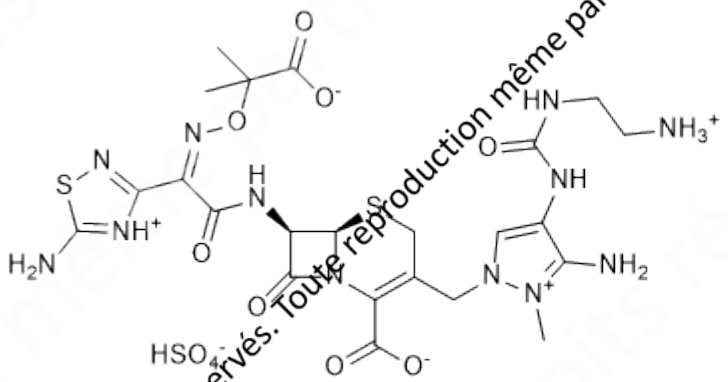


Ceftriaxone

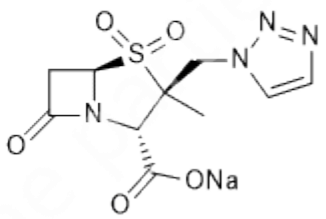




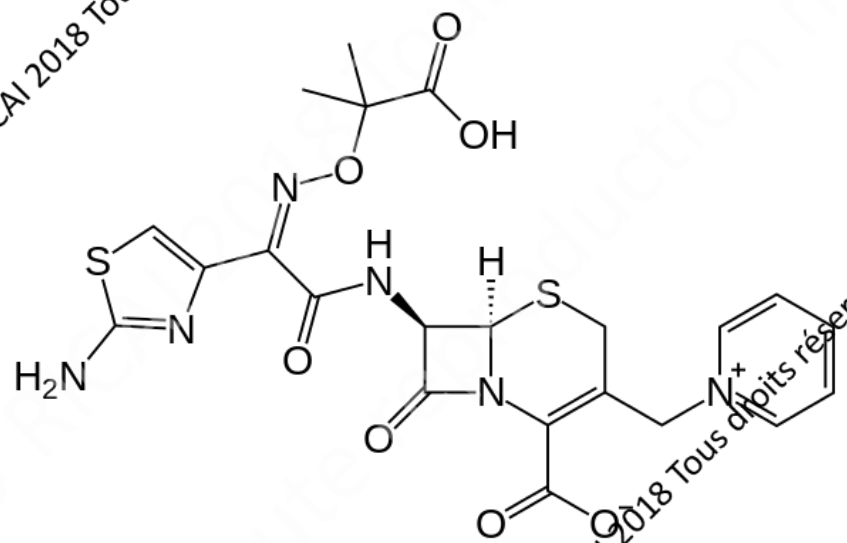




Ceftolozane sulfate
(CXA-101, FR264205



Tazobactam sodium salt



caftazidime