

Penicillinresistens hos streptokocker grupp C (Streptococcus dysgalactiae)

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Deaths attributable to antimicrobial resistance every year compared to other major causes of death



Source: Review on Antimicrobial Resistance 2014

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The Antibiotic Apocalypse: What You Need To Know

However, in spite of penicillin use for >70 years

- **No resistance toward penicillin for**
- Beta-haemolytic streptococci???
- Treponema-Pallidum??
- Borrelia burgdorferi??
- **But is it slowly coming!**

Description and characterization of a penicillin-resistant *Streptococcus dysgalactiae* subsp. *equisimilis* clone isolated from blood in three epidemiologically linked patients

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Background: During a 27 month period, we detected four incidents of penicillin-resistant (PR) *Streptococcus dysgalactiae* subsp. *equisimilis* (SDSE) isolated from blood cultures of three patients.

Methods: The 4 PR-SDSE were compared phenotypically and molecularly (using WGS) with 36 penicillin-susceptible SDSE from blood cultures obtained in the same catchment area and time period.

Results: Phylogenetic analysis showed that the four PR-SDSE belonged to a single clone and a possible epidemiological link between the three patients was identified to be a dermatology department. MICs of penicillin were determined to be 0.5–2 mg/L using Etest and 0.5 mg/L when tested by a broth microdilution method. The four PR-SDSE were unrelated to the 36 penicillin-susceptible isolates, which could suggest that they did not evolve locally from a susceptible clone, but have been introduced into the region. *In silico* genome-based resistome analysis revealed identical PBP mutations in all four isolates. We detected mutations in multiple PBPs, including two amino acid substitutions within the active sites of the transpeptidase domain of PBP2x (T341P and Q555E), which have also been detected in other PR streptococci. The remaining mutations were, however, all located outside the active-site motifs of the transpeptidase domain.

Conclusions: To the best of our knowledge, this is the first description and characterization of invasive PR-SDSE. The resistant isolates had several amino acid changes in various PBPs compared with penicillin-susceptible SDSE. The observation that SDSE also can become PR emphasizes the importance of performing antimicrobial susceptibility testing.

4 resistant SDSE and 36 susceptible SDSE: We wanted to know

- Was it a spreading clone?
- Where did it come from?
- Was what the penicillin mechanism behind?

How was it detected: Results from screening

- *Streptococcus dysgalactiae* subsp. *equisimilis* by MALDI-TOF
- Oxacillin: 6-9 mm (i.e. resistant)
- Penicillin G: 10-17 mm (i.e. resistant)
- Cephalosporins and carbapenems: (all susceptible)

Table 2. Characteristics of the 4 PR-SDSE compared with 40 penicillin-susceptible isolates (36 blood culture isolates and 4 published genome sequence isolates)

	Case-year			
	1-2010	2-2010	2-2012	3-2012
MLST	novel	novel	novel	novel
Allele profile ^a	18-2-3-4-2-7-3	18-2-3-4-2-7-3	18-2-3-4-2-7-3	18-2-3-4-2-7-3
Mutations in PBP2x (337–549 ^b)	T341P^e , Y392N, Q555E	T341P , Y392N, Q555E	T341P , A373V, Y392N, Q555E	T341P , A373V, Y392N, Q555E
Mutations in PBP1a (370–557)	L411F, T487I, A489V	L411F, T487I, A489V	L411F, T487I, A489V	L411F, T487I, A489V
Mutations in PBP1b (337–782)	S662L	S662L	S662L	S662L
Mutations in PBP2a (264–678)	none	none	none	none
Mutations in PBP2b (386–617)	outside the domain	outside the domain	outside the domain	outside the domain
Penicillin MIC ^c : Etest/BMD (SIR ^d)	0.5/0.5 (R)	0.5/0.5 (R)	2/0.5 (R)	2/0.5 (R)
Oxacillin MIC: Etest	8	8	8	8
Cefuroxime MIC	≤0.5	≤0.5	≤0.5	≤0.5
Ceftriaxone MIC	0.25	0.25	≤0.125	0.25
Cefotaxime MIC	≤0.125	≤0.125	≤0.125	≤0.125
Cefepime MIC	≤0.5	≤0.5	≤0.5	≤0.5
Meropenem MIC	≤0.25	≤0.25	≤0.25	≤0.25
Levofloxacin MIC (SIR)	2 (I)	2 (I)	2 (I)	1 (S)
Linezolid MIC (SIR)	2 (S)	1 (S)	1 (S)	1 (S)
Vancomycin MIC (SIR)	≤0.5 (S)	≤0.5 (S)	≤0.5 (S)	≤0.5 (S)
Co-trimoxazole MIC (SIR)	≤0.5 (S)	≤0.5 (S)	≤0.5 (S)	≤0.5 (S)
Daptomycin MIC (SIR)	0.125 (S)	0.125 (S)	0.125 (S)	≤0.06 (S)
Erythromycin MIC (SIR)	≤0.25 (S)	≤0.25 (S)	>2 (R)	>2 (R)
Clindamycin MIC (SIR)	≤0.125 (S)	≤0.125 (S)	>1 (constitutive R)	>1 (inducible R)
Tetracycline MIC (SIR)	≤1 (S)	≤1 (S)	≤1 (S)	≤1 (S)
Resistance genes detected with ResFinder 2.1	none	none	<i>erm(A)</i>	<i>erm(A)</i> and <i>erm(B)</i>

^a*atob-gki-gtr-muri-muts-recp-xpt*.

^bCodons of the transpeptidase domain.

^cMIC testing was done by broth microdilution (BMD), unless otherwise stated.

^dS=susceptible, I=intermediate resistant and R=resistant according to EUCAST using the breakpoints for β-haemolytic streptococci groups A, B, C and G.

^eAmino acid substitutions in bold designate mutations within the active-site motifs of the transpeptidase domain

Penicillin resistance: PBP changes in PBP2x

Spreading clone

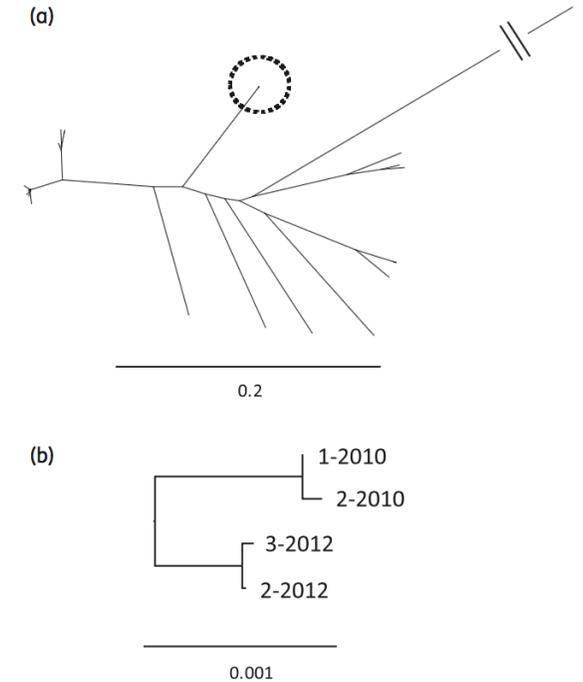
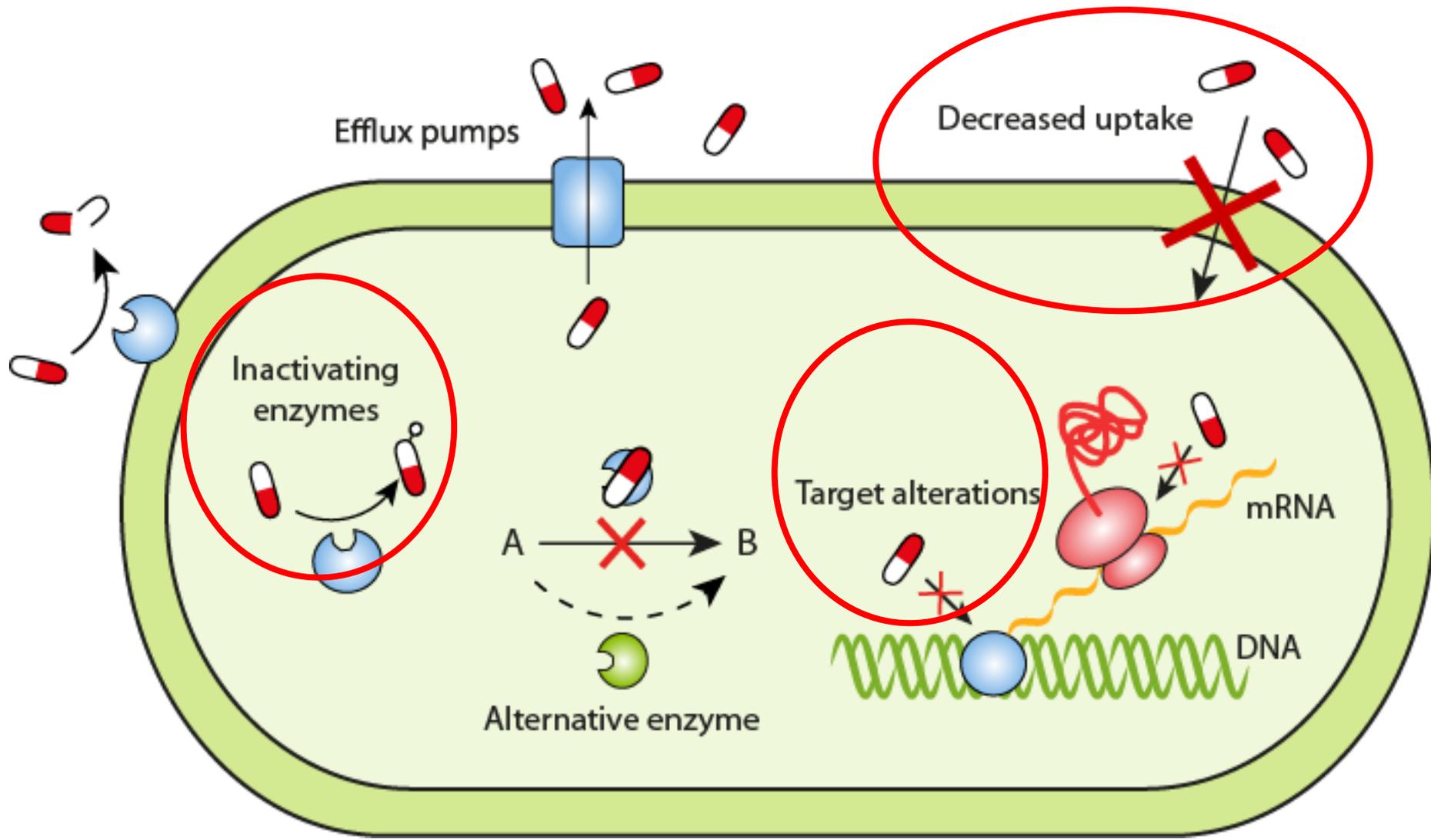


Figure 1. (a) Maximum-likelihood phylogenetic analysis of the 4 PR isolates and 36 penicillin-susceptible invasive isolates of SDSE collected between 2009 and 2013. (b) Zoomed-in view of the four PR isolates encircled in (a). Scales indicate substitutions per site.

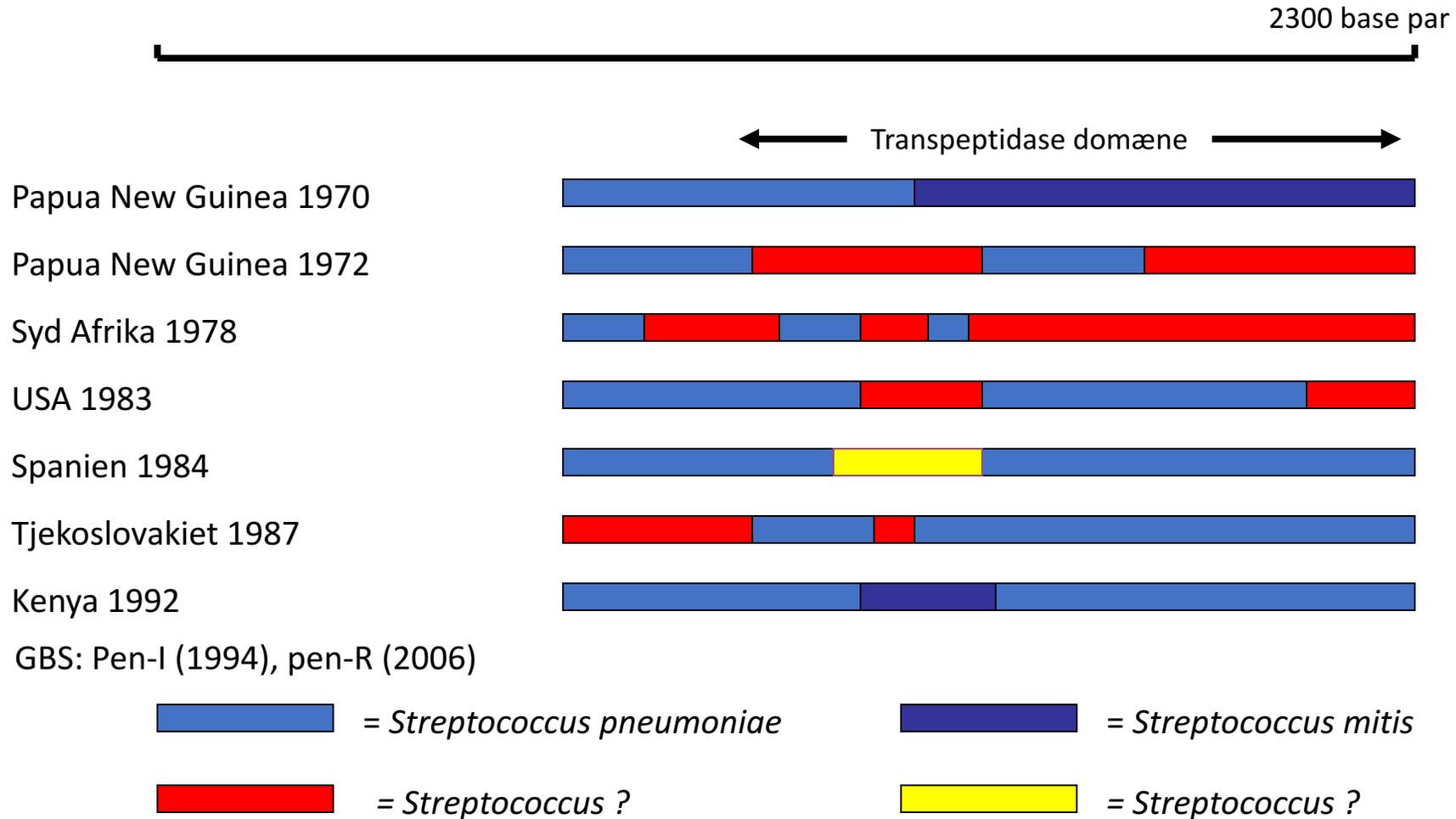
Probably introduced and evolved from a susceptible clone

Penicillin resistance mechanisms



History of penicillin resistance (PBP) in streptococci

Penicillin-resistente pneumokokker: Mosaik PBP gener (2B)



Penicillin resistance mechanism: PBP changes:

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Well known mutations



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