Epidemiology of Plasmid Mediated Quinolone Resistance Mechanisms (PMQRs) in Argentina: Differential Distribution in Enterobacteria

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INTRODUCTION

Since the first report of *qnrA1* (formerly *qnrA*), the spreading of PMQRs is of major concern in quinolone resistance.

We previously found *qnrB10*, *qnrS1* and *aac(6')-Ib-cr* in clinical isolates from Argentina. Quiroga et al, *AAC* 51: 4466-70, 2007; Andres et al, *49th ICAAC* (C2-716), 2009

OBJECTIVES

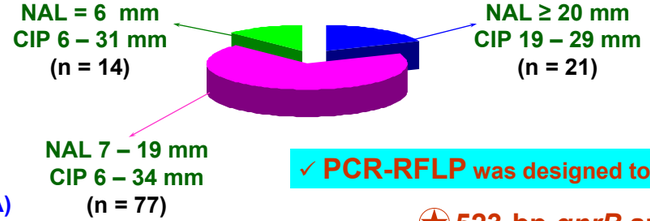
To search for PMQRs in *Enterobacteriaceae* with decreased quinolone susceptibility routinely recovered at hospital level and to characterize their main genetic platforms.

MATERIALS & METHODS

CLINICAL ENTEROBACTERIAL ISOLATES Submitted to the National Reference Laboratory due to decreased quinolone susceptibility

Distribution by species ← N = 112 → Distribution by disk difussion inhibition zones

N	
38	*Klebsiella* spp. (KLB)
27	*Escherichia coli* (ECO)
23	*Salmonella* spp. (SAL)
10	*Enterobacter* spp. (ENT)
8	*Serratia marcescens* (SMA)
6	Others



MOLECULAR ASSAYS

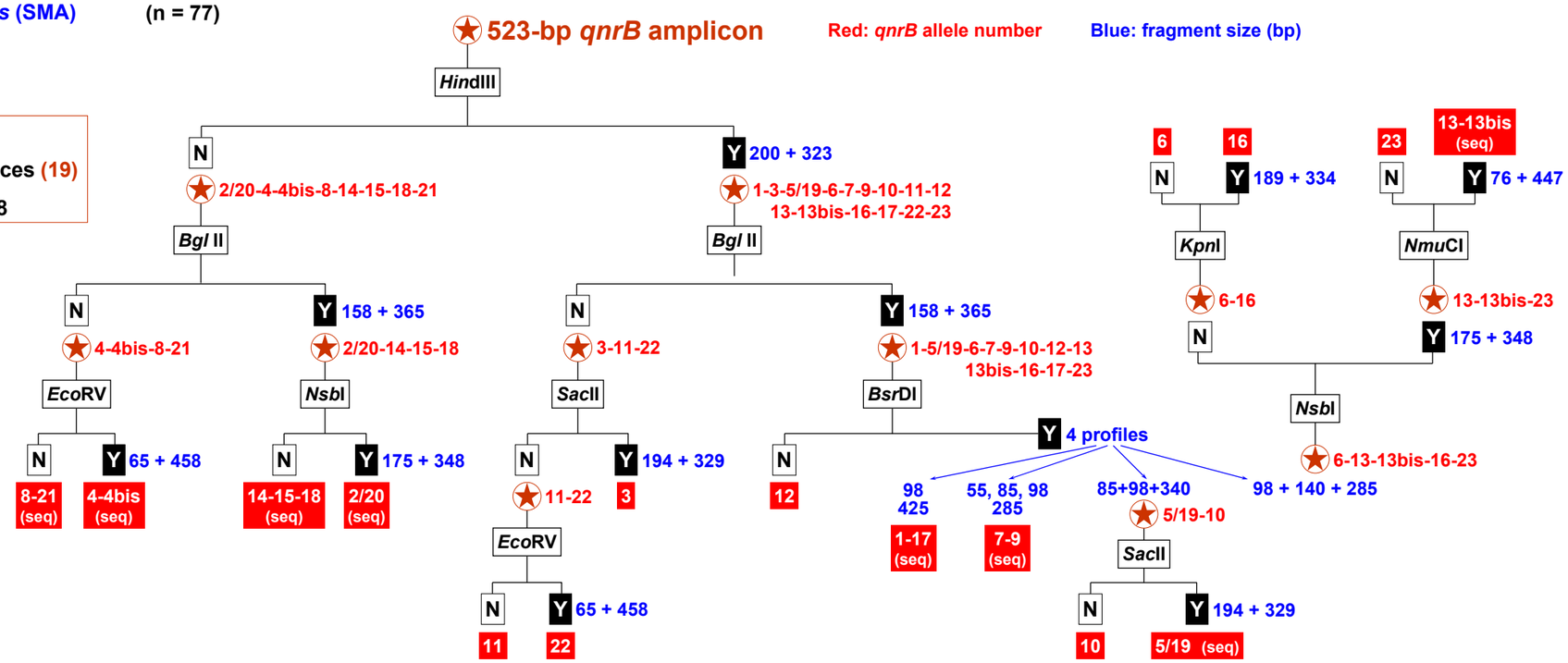
- ✓ PCR for *qnrA*, *B* and *S* and DNA sequencing were done by standard methods.
- ✓ *aac(6')-Ib-cr* was identified by allele-specific PCR (P. Andres *et al*, ICAAC 2009).
- ✓ Genetic platforms were analyzed by PCR cartography and sequencing.
- ✓ Plasmids were extracted and sequencing by standard techniques.

✓ PCR-RFLP was designed to discriminate among 23 known *qnrB* alleles (results were validated by DNA sequencing).

● 33 HOSPITALS

Bs As (14) and 12 Provinces (19)

4 years, 2005-2008



RESULTS

DISTRIBUTION AND OCURRENCE OF PMQRs

55% (62/112) isolates had at least 1 PMQR

9/14 hospitals from Bs As
14/19 hospitals from 10/12 Provinces

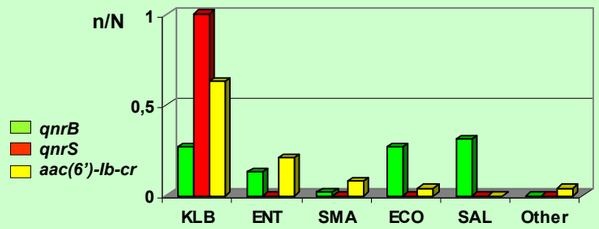
92% (57/62) *qnr* (+)

- 91% (52/57): *qnrB* → *qnrB* alleles, n = 52
  - 6% *qnrB2* (n=3)
  - 2% *qnrB1* (n=1)
  - 38% *qnrB10* (n=20)
  - 54% *qnrB19* (n=28)
- 9% (5/57): *qnrS* → All *qnrS1* (KLB)
- No *qnrA*

39% (24/62) *aac(6')-Ib-cr* (+)

75% (18/24) coexpressed *qnrB*
4% (1/24) coexpressed *qnrS*
21% (5/24) as unique PMQR

Ocurrence of PMQRs among species



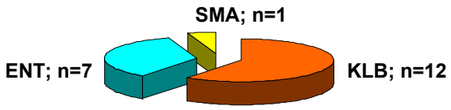
In each specie, the ratio n/N = Nº of isolates with the indicated PMQR / total Nº of isolates with that PMQR in the sample set.

TWO MAJOR SCENARIOS

LINKED TO IS*CR1* (complex class 1 integron): *qnrB10* and *qnrB2*

A) Distribution by species

qnrB10, n = 20

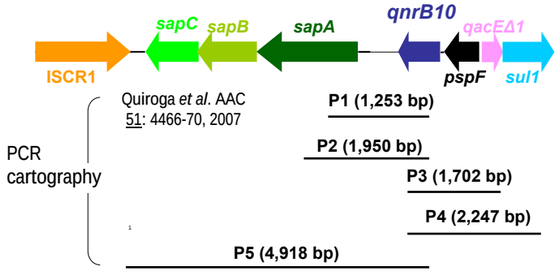


qnrB2, n = 3

KLB ECO SAL

16/20 *qnrB10* and 1/3 *qnrB2* were coexpressed with *aac(6')-Ib-cr*

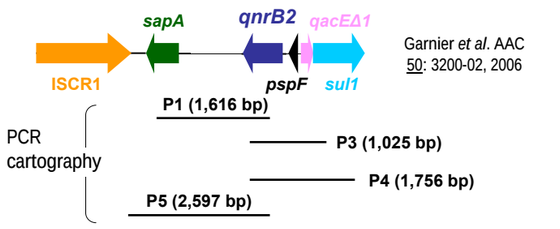
B) Genetic Platforms



qnrB10

✓ All isolates presented the same narrow environment [P1, P2, P3 and P4 (+)]

✓ IS*CR1* linkage was tested in 5 selected isolates [P5 (+)]: 3 KLB, 1 ENT and 1 SMA



qnrB2

✓ All the isolates presented the same narrow environment [P1, P3, P4 (+) and P2 (-)]

✓ IS*CR1* linkage [P5 (+)]: all isolates

NOT LINKED TO IS*CR1*: *qnrB19*

A) Distribution by species

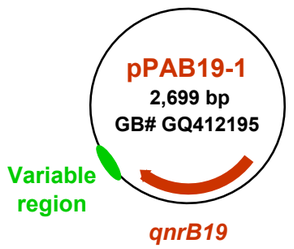
qnrB19, n = 28

Isolates DID NOT HARBOR *aac(6')-Ib-cr*

B) Genetic Platforms

→ PCR against *orf513*: NEGATIVE

26/28 *qnrB19* were located in 4 small ColE1-type plasmids → Plasmids only diverge in the VARIABLE REGION



1. pPAB19-1: 2,699 bp; 100% identical pECY6-7 (Pallecchi *et al*, AAC 2009) and pSGI15 (Hammerl *et al*, JAC 2010)
2. pPAB19-2: 3,082 bp; similar to pEC14-9 (Pallecchi *et al*, AAC 2009)
3. pPAB19-3: 2,989 bp; similar to pPAB19-2
4. pPAB19-4: 2,702 bp; similar to pPAB19-1

CONCLUDING REMARKS

A) *qnrB19*, *qnrB10* and *aac(6')-Ib-cr* were the prevalent PMQRs in these clinical bacterial set.

B) *qnrB10* and *aac(6')-Ib-cr* were mainly present in *Klebsiella* spp. and *Enterobacter* spp.; *qnrB10* was linked to IS*CR1* and it was highly related to *acc(6')-Ib-cr*.

C) *qnrB19* was only harbored by *E. coli* and *Salmonella* spp.; *qnrB19* was linked to 4 small ColE1 plasmids but not to IS*CR1*.