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## INTRODUCTION

*E. faecalis* and *E. faecium* are common gastrointestinal commensal organisms acquiring resistance through the transfer of plasmids and transposons and recombination or mutation events. Infection with vancomycin-resistant enterococci (VRE) is a growing problem. Currently, there are not many data available on the epidemiology of Enterococci and VRE in Belgium. A survey was initiated by Belgian Reference Centre for Enterococci (NRC) to assess the epidemiology of enterococci in Belgium on strains received from hospital laboratories between 01/01/2009 and 31/12/2015.

## MATERIALS & METHODS

Species identification was confirmed by conventional diagnostics, by Malditof Mass Spectrometry and by sod/ddl/16S rDNA-PCR and sequencing. Antibiotic susceptibility was determined by using disk diffusion and E-test and interpretation according to CLSI (till 2012) and EUCAST from January 1, 2012 onwards. The following antibiotics were tested: ampicillin, vancomycin, teicoplanin, linezolid (LZ) and tigecycline. PCR was applied to confirm VRE. Typing was done by PFGE and MLST (from 2014 onwards).

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**Acknowledgement/Disclosures:** The National Reference Centre is partially supported by the Belgian Ministry of Social affairs through a fund within the Health Insurance System.

## RESULTS

Since 2009, the Belgian NRC receives an increased number of enterococcal strains from hospital laboratories: n=32, 27, 81, 131, 178, 300 and 578 in respectively 2009, 2010, 2011, 2012, 2013, 2014 and 2015 (Table 1).

- The *E. faecium* % increased from 56.2% to 77.3%.
- The VRE% increased from 56.3% to 74.6%, in the same period.
- VanA increased from 27.8% to 90.5%, vanB decreased from 61.1% to 5.4%.
- All outbreaks, except 1, were caused by *E. faecium*, with an increase in number from 1-22 between 2009 and 2015;
- The majority of the outbreak strains were vanA positive.
- Overall, the most prevalent STs were 80, 117 and 203, each representing more than 1 PFGE-type (Table 2).
- A low level of LZ resistance was detected over the last 2.5 years (2.3%, MIC 8-64 mg/L) in both *E. faecium* and *E. faecalis*.
- None of the LZ R *E. faecalis* strains were VRE, 9/11 *E. faecium* were VRE.
- All, except 1 *E. faecium* and 1 *E. faecalis* strain (MIC 4.0 mg/L), were sensitive to tigecycline.
- Rare species are also more often submitted for confirmation eg. *E. avium* (n=5), *E. raffinosus* (n=5), *E. durans* (n=1), *E. gilvus* (n=1), *E. mundtii* (n=1).

	ST203	ST117	ST80	ST18	ST17	ST78	OTHER ST
<i>E. faecium</i> analysed/ total <i>E. faecium</i> 2014 (186)	59	33	42	8	2	1	9
<i>E. faecium</i> analysed/ total <i>E. faecium</i> 2015 (417)	233	71	53	4	34	5	15

Table 2. Overall STs *E. faecium*

Year	2009	2010	2011	2012	2013	2014	2015
Nr of strains received	32	27	81	131	178	300	578
other	0	4	2	2	0	2	0
<i>E. faecium</i>	18	19	47	104	95	215	447
<i>E. faecalis</i>	12	3	18	15	67	68	111
<i>E. cass/gal</i>	2	1	13	9	12	12	14
other Ent. Spp	0	0	1	1	4	3	4
Nr of VRE (%)	18 (56,3)	21 (77,8)	61 (75,3)	103 (77,4)	85 (47,8)	198 (65,8)	444 (76,1)
vanA (%)	5 (27,8)	8 (38,1)	24 (39,3)	84 (81,6)	61 (71,8)	163 (82,3)	402 (90,5)
vanB (%)	11 (61,1)	12 (57,1)	24 (39,3)	11 (10,7)	12 (14,1)	20 (10,1)	24 (5,4)
vanC (%)	2 (11,1)	1 (4,8)	13 (21,3)	8 (7,7)	12 (14,1)	10 (5,1)	13 (2,9)
other (vanAB, vanBC, vanAC, vanD) (%)	0	0	0	0	0	3 (1,5)	5 (1,1)
Nr of outbreaks*	1 (n=6)	1 (n=3)	3 (n=4, 12, 16)	4 (n=3, 4, 6, 39)	1 (n=36)	n=10 (2, 3, 3, 3, 6, 6, 8, 14, 35, 46)	n=22** (2, 3, 3, 3, 3, 3, 4, 4, 5, 5, 6, 6, 9, 11, 12, 12, 15, 18, 20, 37, 54, 76)

\*all *E. faecium*, except 1; \*\*1 clone in several hospitals in the same region

Table 1. Surveillance data 2009-2015

## CONCLUSIONS

Since Belgian laboratories for clinical microbiology are not legally bound to submit their VRE strains to the NRC, it is difficult to conclude on the exact epidemiology of VRE and on the number of outbreaks. However, based on the strain submissions in the last 6 years, the NRC received an increased number of *E. faecium* isolates. ST80, 117 and 203 are detected in the majority of outbreak, non-outbreak or outbreak-unknown *E. faecium* strains.

Due to the application of newer identification techniques, rare enterococcal species are more frequently submitted to the NRC for confirmation. Linezolid resistance and tigecycline resistance is still low.