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Vancomycin-resistant *Enterococcus* sp.

Infection with vancomycin-resistant *Enterococcus* is associated with increased morbidity and mortality. The majority of VRE are associated with the species *Enterococcus faecium* (77%) and *E. faecalis* (9%), with the remaining 14% of isolates representing species less frequently implicated in serious infections, including *E. gallinarum, E. casseliflavus, E. avium*, and *E. raffinosus.*
Resistance to glycopeptides in *Enterococcus* spp. is mediated by the vancomycin resistance (Van) operon. This operon may be carried on the chromosome or on a plasmid. Nine variants have been identified in the ligase gene: *vanA, vanB, vanC, vanD, vanE, vanG, vanL, vanM, and vanN*. The most frequent are *vanA* and *vanB*.

E. faecium and *E. faecalis* with acquired vancomycin resistance (VRE) are multidrug resistant bacteria which can be responsible for health care-associated infections. Detection of this resistance trait is particularly important for the prevention and epidemiological surveillance of these infections.
The *vanA* gene procures a high-level resistance to vancomycin and teicoplanin, while isolates with the *vanB* gene are still susceptible to teicoplanin in vitro. Pathogenicity of VRE is low except among immunocompromised patients, but they are highly transmissible within the hospital setting (especially *E. faecium*) which is the reason to implement infection control measures in order to limit their spread.

Clinical sample

From isolated colonies:

- Identification tests:
  - MALDI-TOF
  - or
    - at the genus *Enterococcus* level: Gram + cocci, catalase -, PYR test +
    - at the species level: Vitek (GP ID card) or Phoenix (PMIC/ID Panels) for example
- Susceptibility/detection tests:
  - Disk diffusion test of Vancomycin and teicoplanin(EUCAST v. 8.0, 2017)

	S ≥	R <
Vancomycin (5 µg)	12	12
Teicoplanin (30 µg)	16	16

According to EUCAST guidelines: If the zone edge is fuzzy, colonies grow within the zone or if you are uncertain, then perform confirmatory testing with PCR or report resistant (see pictures below) even if the zone diameter is ≥ 12 mm. Isolates must not be reported susceptible before 24 h incubation.

  - MIC with Etest® of vancomycin and teicoplanin (EUCAST v. 8.0):

	S	R
Vancomycin	≤4	>4
Teicoplanin	≤2	>2

Some strains of VRE confer a low MIC values. If suspected, the bacterial suspension can be increased to McFarland 2.0 and the incubation time extended to 48 h. The detection of the *van* genes should be done to confirm the resistance.
  - If available in the lab, molecular diagnostic can be performed on colonies (Xpert *vanA/vanB or vanR* PCR BD max kits)

Screening sample (colonization)

- Rectal swab or stool specimens.
- Inoculate the specimen into an enrichment broth (8-9 ml BHI with a 30 µg disc of vancomycin, final concentration of 3-4 µg/ml).
- After O/N incubation, soak a cotton swab into the broth and inoculate a chromogenic agar specific for VRE (VRE-select [Biorad], VRE Brilliance [Oxoid], chromID VRE [bioMérieux], etc.).
- Incubate the chromogenic plate during the time required by the manufacture. Of note, with the use of the enrichment broth, it is usually unnecessarily to prolong this incubation to 48h.
- On characteristic colonies: identification and susceptibility testing similar to clinical sample or according to manufacture recommendation.

N.B. Direct inoculation on chromogenic agar is also possible. The enrichment broth increases the sensitivity of detection of 10 to 15%. All chromogenic agar from different manufactures have similar performances. The incubation time and supplementary tests might be different from one agar to another (Suwantarat et al., 2014, J Clin Microbiol, Comparison of five chromogenic media for recovery of VRE from fecal samples).

During an outbreak, or to screen for contact patients of an index case, detection of VRE may be performed using a rapid PCR test directly on the rectal swab should be considered, as the turnaround time is reduce to few hours instead of two days. The negative predictive value is usually very high; however, the positive predictive value for a *vanB* result is low because of the presence of this gene in anaerobic commensal bacteria from human faeces (e.g. *Clostridium* sp.; Ballard et al. AAC, 2005, Jan;49(1):77-81).