1. Four hundred and forty three enterococcal strains were isolated from 10,765 clinical samples (stool and urine) from patients admitted to PGIMER, Chandigarh.

2. The break up of identification of enterococcal isolates was as follows: *E. faecalis* (256), *E. faecium* (160), *E. gallinarum* (18) and *E. casseliflavus* (9).

3. From stool samples, 250 enterococcal strains were isolated. Maximum number of strains belonged to *E. faecalis* (129) followed by *E. faecium* (94), *E. gallinarum* (18) and *E. casseliflavus* (9).

4. From urine samples, 193 enterococcal strains were isolated. Maximum number of strains belonged to *E. faecalis* (127) followed by *E. faecium* (66). No other strain of enterococci was isolated from urine samples.

5. All the 443 enterococcal isolates were checked for VRE by E-test and agar dilution method. Thirty six (8.14%) isolates turned out to be VRE: 28 from stool samples and 8 from urine samples.

6. The breakup of 36 VRE was as follows: 18 *E. gallinarum* (all from stool samples), 9 *E. casseliflavus* (all from stool samples), 7 *E. faecium* (6 urine sample & 1 stool sample) and 2 *E. faecalis* (all from urine samples).

7. The level of vancomycin resistance amongst 36 VRE was as follows:
   - MIC >512 µg/ml: 3 (all *E. faecium*; all urine isolates).
   - MIC 6-16 µg/ml: 4 (all *E. faecium*; 3 urine isolates and 1 stool isolate).
   - MIC 8 µg/ml: 2 (all *E. faecalis*, all urine isolates).
   - MIC 8-16 µg/ml: 18 (all *E. gallinarum*; all stool isolates).
   - MIC 6-16 µg/ml: 9 (all *E. casseliflavus*; all stool isolates).
   - MIC ≤ 4 µg/ml: 153 *E. faecium* isolates (60 urine isolates and 93 stool isolate).
   - MIC ≤ 4 µg/ml: 254 *E. faecalis* isolates (125 urine isolates and 129 stool isolate).

8. Vancomycin resistance in 36 VRE was found to be associated with four van genotypes i.e. *vanA*, *vanB*, *vanC1* and *vanC2*. 

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vanA gene was present in *E. faecium* resistant to >512 \( \mu \text{g/ml} \) of vancomycin.

- vanB gene was present in *E. faecalis* and *E. faecium* resistant to 6-16 \( \mu \text{g/ml} \) of vancomycin.
- vanC1 gene was present in *E. gallinarum*.
- vanC2 gene was present in *E. casseliflavus*.

9. All VRE having vanA genotype were also resistant to teicoplanin (>32\( \mu \text{g/ml} \)). VRE having vanB, vanC1 and vanC2 genotypes were found to be sensitive to teicoplanin.

10. van genotypes were in agreement with phenotypes for all VRE isolates.

11. The nucleotide sequence of PCR products corresponding to vanA, vanB, vanC1 and vanC2 were submitted to GenBank and appear with accession numbers AY754011, AY786179, AY786180 and AY751080, respectively.

12. The results of BLAST analysis carried out for DNA sequence alignment of van genes present in various microbes were as follows:

- vanA gene showed 99% homology with vanA sequence present in *E. faecium* transposon Tn1546 (M97297), *E. faecium* plasmid pUW786 (AF516335) and *E. faecium* plasmid pIP816 (X56895).
- vanB gene showed 99% identity with strain MLG856-2 D-alanine:D-lactate ligase (vanB) gene (AY655711) and 97% identity with *E. faecalis* vancomycin resistance protein (vanB) gene (EFU72704) and 96% identity with *E. faecalis* vanB gene (AY665551).
- vanC1 gene showed one significant match of 99% identity with the *E. gallinarum* vanC vancomycin-resistance gene cluster (AF162694), as well as *E. gallinarum* strain N04-0414 vancomycin resistance gene cluster (DQ022190).
- vanC2 gene showed 97% identity with *E. casseliflavus* vanC2 gene (AB070704), (AB070698) and 96% identity with *E. casseliflavus* vanC2 gene (AB070703).

13. From stool samples 28 VRE were isolated belonging to 17 males and 11 females whose age ranged from 7 months to 65 years with an average of 23.9 years and median of 26.5 years. *E. gallinarum* (vanC1), *E. casseliflavus* (vanC1) and *E. faecium* (vanB) were isolated from 18, 9 and 1
patient, respectively, and isolated from different medical wards. Out of them, 10 were children whose age ranged from 7 months to 10 years. 5 of them had diarrhea, 2 had malnutrition. 1 child each had typhoid, intussusception, acute colitis, and acute dysentery. One child was HIV positive. Of 17 adults, 6 were admitted in adult gastroenterology unit, 6 underwent renal transplantation, and 3 were admitted in emergency ward. One patient who had undergone renal transplantation was transferred to male medical ward (MMW).

14. From urine samples 8 VRE were isolated belonging to 5 males and 3 females, Age ranged from 3 months to 73 years. 7 patients developed nosocomial UTI, out of which 4 patients developed sepsis and died. Urinary catheterization (present in all), followed by renal failure (5/8, 62.5%), surgery on urinary tract (3/8, 37.5%), treatment with third generation cephalosporins (3/8, 37.5%) and vancomycin (2/8, 25%) were some of important risk factors.

15. Gastroenterology unit adult gastroenterology (AGE) and paediatric gastroenterology (PGE) and renal transplant unit were the most affected units for all patients in both stool and urine with VRE.

16. Prolonged hospital stay and vancomycin in stool sample were most important risk factors with significant difference between patients and control group. Prolonged hospital stay, renal failure, dialysis, third generation cephalosporin and aminoglycoside in urine sample were most risk factors between patients and control group with significant difference.