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EC MICROBIOLOGY
Research Article

Draft Genome Sequence of *Enterococcus Faecium* Strain 24-10 Isolated from the Patient with Wound Infection

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Received: February 27, 2017; **Published:** March 30, 2017

Abstract

We present a pyrosequencing-based draft-genome sequence of pathogenic Enterococcus faecium strain 24-10. The strain was isolated from the patient with the wound infection. The genome length is 2,980,950 bp and it presumably carries four plasmids, pathogenicity island, a number virulence genes and genes of vancomycin resistance.

Keywords: *Enterococcus faecium Strain 24-10; Draft Genome Sequence; Wound Infection*

Introduction

Enterococci are widely known as the common inhabitants of humans and animals [1,2]. *Enterococcus faecium* strains are known to be one of the first inhabitants of newborns gastrointestinal tract [3]. Although in the last few decades *E. faecium* strains emerged as a cause of various infections all over the world [4-6]. Cases of endocarditis, bacteremia, central nervous system infections, respiratory tract, urinary tract and other infections were registered to be caused by *E. faecium* [7]. Vancomycin resistant enterococci (VRE) are the most virulent among enterococcal species. Moreover some *E. faecium* strains are resistant to more than one antibiotic [8]. Enterococcal genomes are known to have mosaic structure which could be the result of multiple horizontal genes transfer [9]. We report of a new pathogenic *E. faecium* 24-10 strain isolated in Saint-Petersburg (Russia) which carries in its genome a number of virulence factors including genes of vancomycin resistance. Understanding the spread of VRE in different parts of the world will help to realize the scope of the problem. Search on all known and putative enterococcal pathogenicity genes will help to understand this new emerged pathogen and will help to prevent its dangerous spread.

Methods

Enterococcus faecium 24-10 was isolated from the patient with wound infection in 26th Municipal Hospital (Saint-Petersburg, Russia). The genomic DNA was extracted from the bacterial culture by DNeasy Blood and Tissue Kit (Qiagen, USA). The genome was sequenced de novo using 454 pyrosequencing on the GS FLX platform (Roche 454 Life Sciences). Barcoded DNA libraries were prepared using GS DNA Library Preparation Kit in combination with GS Multiplex Identifiers Kits. The GS emPCR kit II was used for emulsion-based clonal amplification of the MID-adapted library. GS LR70 Sequencing Kit in combination with GS FLX PicoTiterPlate Kit (70x75) was used to obtain sequences. All the procedures were carried out according to manufacturer's instructions (Roche). Reads were assembled *in silico* using Newbler assembler, Genome annotation was performed using the standard operation procedures from the Rapid Annotation using Subsystems Technology (RAST) server [10]. Center of genomic epidemiology services and NCBI BLAST were used to determine antibiotic resistance genes (ResFinder 2.1), virulence factors (PathogenFinder 1.1, VirulenceFinder 1.5) and plasmids (PlasmidFinder 1.3) [11-13].

Results

Enterococcus faecium 24-10 genome size is 2,980,950 bp with a mean of GC content 37,6%. The draft genome sequence consists of 191 contigs of different sizes (from 201 bp to 156127 bp). A total of 3188 coding sequences (CDS) and 65 structural RNA's were predicted.

Four plasmids belonging to the following rep-families: repUS15, rep2, rep11 and rep17 were predicted by Plasmid Finder.

E. faecium 24-10 was known as VRE and we confirmed it by finding genes related to vancomycin resistance *vanA* and *vanB*. Moreover it turned out that the strain's genome carries other antibiotic resistance genes: *ant(6)-Ia* and *aph(3')-III* giving it aminoglycoside resistance; *erm(B)* giving it macrolide resistance; *lnu(B)* giving it lincosamide resistance and *msr(C)* giving it streptogramin B resistance as well as aminoglycoside and lincosamide resistance.

Analysis of the genome of *E. faecium* 24-10 revealed the presence of the following pathogenicity genes: two genes encoding pili – *pilA* and *pilB*, gene of collagen-binding protein *acm*, genes encoding surface adherence factors *esp* and *efaA*, gene encoding hemolysin *hylE*. The whole pathogenicity island associated with *esp* gene was found as well.

The strain *E. faecium* 24-10 belongs to ST-78 cluster.

Nucleotide sequence accession numbers. This Whole Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession MVEB00000000. The version described in this paper is the version MVEB00000000.

Conclusion

In recent years vancomycin resistant enterococci (VRE) have become one of the leading cause of hospital infections. Spreading of VRE all over the world indicates that this new emerged pathogen is a very important target for the medical science society. The strain *Enterococcus faecium* 24-10 isolated from the patient with the severe wound infection proved to have genetic determinants of resistance to four groups of antibiotics – glycopeptides, aminoglycosides, lincosamides and streptogramins. The last three antibiotic groups are usually used for the treatment of VR enterococcal and VR staphylococcal infections. We detected a number of genes associated with pathogenicity in *E. faecium* 24-10 genome – adherence factors, collagen-binding protein and hemolysin. The study also revealed that *Enterococcus faecium* 24-10 presumably carries four plasmids which role and structure need further investigation. The data obtained from the analysis of the *Enterococcus faecium* 24-10 whole-genome sequence correlates with strain's isolation source.

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Volume 7 Issue 2 March 2017