Genome sequences of 11 human vaginal Actinobacteria strains

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Bacterial vaginosis (BV) is a vaginal dysbiosis associated with serious health complications (1–6). It is characterized by the absence of Lactobacillus species in the vagina and overgrowth of a polymicrobial community often containing members of the phylum Actinobacteria, including Gardnerella vaginalis, Atopobium spp., and others. In fact, G. vaginalis was recently shown to elicit several features of BV in a mouse vaginal infection model (7, 8). Bifidobacteria are also commonly isolated from the vagina, although members of this genus are rarely found in pathological contexts. Here, we isolated 11 vaginal bacteria from the phylum Actinobacteria. Vaginal swabs were collected from nonpregnant and pregnant women according to Washington University institutional review board (IRB)-approved protocols (201108155 and 20110382). Organisms isolated from vaginal swabs were cultured anaerobically, and identification was performed by 16S rRNA gene sequencing. Genomic DNA was obtained using the Wizard genomic DNA purification kit (Promega). Methodological details on isolation and clinical information will be described elsewhere.

Genomes were assembled de novo using the One Button Velvet assembly pipeline (version 1.1.06) (9) with hash sizes of 31, 33, and 35 after downsizing the sample input data to 100× coverage. An internal core gene screen on the assembly tested for completeness of the genome. After assembly, the minimum length for contigs was set to 200 bp, and an internal core gene screen was performed as defined by the Human Microbiome Project (HMP) (10). Then, adapters were removed, and low-quality regions were trimmed. Finally, a screen for contamination was performed. The process of gene annotation included generating both ab initio and evidence-based (BLAST) predictions. Functional predictions of coding sequences were made using GeneMark and Glimmer3 (11, 12). Loci were then defined by clustering predictions with the same reading frame. We evaluated predictions using the nonredundant (NR) and Pfam databases and resolved overlaps between adjacent coding genes. Intergenic regions not spanned by GeneMark and Glimmer3 were subject to a BLAST search against NCBI’s NR database and predictions generated based on protein alignments. tRNA genes were determined using tRNAscan-SE (13) and noncoding RNA genes by RNAmmer (14) and Rfam (15). Metabolic pathways and subcellular localization were predicted using KEGG and PSORTb, respectively (16, 17), and functional domains were evaluated using InterProScan (18).

**Accession number(s).** These whole-genome shotgun projects have been deposited in GenBank under the accession numbers listed in Table 1. We have also made the strains available to the genome. After assembly, the minimum length for contigs was set to 200 bp, and an internal core gene screen was performed as defined by the Human Microbiome Project (HMP) (10). Then, adapters were removed, and low-quality regions were trimmed. Finally, a screen for contamination was performed. The process of gene annotation included generating both ab initio and evidence-based (BLAST) predictions. Functional predictions of coding sequences were made using GeneMark and Glimmer3 (11, 12). Loci were then defined by clustering predictions with the same reading frame. We evaluated predictions using the nonredundant (NR) and Pfam databases and resolved overlaps between adjacent coding genes. Intergenic regions not spanned by GeneMark and Glimmer3 were subject to a BLAST search against NCBI’s NR database and predictions generated based on protein alignments. tRNA genes were determined using tRNAscan-SE (13) and noncoding RNA genes by RNAmmer (14) and Rfam (15). Metabolic pathways and subcellular localization were predicted using KEGG and PSORTb, respectively (16, 17), and functional domains were evaluated using InterProScan (18).

**Table 1** Identifiers and nucleotide sequences for sequenced strains of vaginal Actinobacteria

<table>
<thead>
<tr>
<th>Genus/species</th>
<th>Strain</th>
<th>BEI catalog no.</th>
<th>Nucleotide accession no.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Actinomyces neui</td>
<td>MIR8396A</td>
<td>HMS-1266</td>
<td>LRPJ00000000</td>
</tr>
<tr>
<td>Alloscardovia omnicolens</td>
<td>CMW7705A</td>
<td>HMS-1282</td>
<td>LRPK00000000</td>
</tr>
<tr>
<td>Atopobium vaginae</td>
<td>CMW7778A</td>
<td>HMS-1300</td>
<td>LSOA00000000</td>
</tr>
<tr>
<td>Bifidobacterium bifidum</td>
<td>MIR8628B</td>
<td>HMS-1264</td>
<td>LRPQ00000000</td>
</tr>
<tr>
<td>Bifidobacterium breve</td>
<td>GED8481</td>
<td>HMS-1261</td>
<td>LRPQ00000000</td>
</tr>
<tr>
<td>Bifidobacterium longum</td>
<td>CMW7750</td>
<td>HMS-1299</td>
<td>LSRB00000000</td>
</tr>
<tr>
<td>Corynebacterium sp.</td>
<td>CMW7794</td>
<td>HMS-1295</td>
<td>LSRZ00000000</td>
</tr>
<tr>
<td>Gardnerella vaginalis</td>
<td>GED7275B</td>
<td>HMS-1272</td>
<td>LSRD00000000</td>
</tr>
<tr>
<td>Gardnerella vaginalis</td>
<td>GED7760B</td>
<td>HMS-1284</td>
<td>LSRQ00000000</td>
</tr>
<tr>
<td>Gardnerella vaginalis</td>
<td>CMW7778B</td>
<td>HMS-1298</td>
<td>LSRC00000000</td>
</tr>
<tr>
<td>Propionibacterium avidum</td>
<td>MIR7694</td>
<td>HMS-1291</td>
<td>LRVD00000000</td>
</tr>
</tbody>
</table>
research community by depositing them with the Biodefense and Emerging Infections (BEI) Research Resource Repository (see BEI numbers in Table 1).

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REFERENCES


