Bacteroides caccae sp. nov., Bacteroides merdae sp. nov., and Bacteroides stercoris sp. nov. Isolated from Human Feces

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Three new saccharolytic Bacteroides species that have DNAs with guanine-plus-cytosine contents of 40 to 46 mol%, produce major amounts of succinate, and were isolated principally from human feces are described: Bacteroides caccae, with ATCC 43185 as the type strain; B. merdae, with ATCC 43184 as the type strain; and B. stercoris, with ATCC 43183 as the type strain. These groups previously have been referred to as the “3452A,” “T4-1,” and “subsp. a” groups, respectively.

In 1978, Johnson (3) and Johnson and Ault (4) described, but did not name, three DNA homology groups of saccharolytic anaerobic gram-negative rods that grow well in 20% bile and form succinate as a major product: groups “3452A,” “T4-1,” and “subsp. a.” Except for a few strains of group “3452A” from clinical specimens, all strains studied were isolated from fecal samples. Because the strains are nonmotile, anaerobic, gram-negative rods, produce major amounts of succinate from glucose, have DNA with a guanine-plus-cytosine (G + C) content of 40 to 46 mol% (3), and cluster with Bacteroides fragilis and related species by RNA homology studies (5), they are members of the genus Bacteroides (2). For them we propose the names Bacteroides caccae, Bacteroides merdae, and Bacteroides stercoris.

MATERIALS AND METHODS

Strains. Strains were from the Virginia Polytechnic Institute and State University Anaerobe Laboratory collection and, with the exception of OC-9, are the same strains of Bacteroides homology groups (“3452A,” “T4-1,” and “subsp. a”) studied by Johnson (3) and Johnson and Ault (4). Strain OC-9 could not be recovered from storage and was omitted from this study. The type strain (3452A) and 18 other strains of Bacteroides caccae were isolated from human feces; one strain (PrVot 2302) was isolated from hog cecum contents. The type strain (B5-21) and 17 other strains of B. stercoris were isolated from human feces.

Methods of characterization. Cultures for tests for fermentation of sugars, hydrolysis of esculin, digestion of gelatin, milk, and meat, reduction of nitrate and resazurin, growth in 20% bile and 6.5% NaCl, and production of catalase, urease, hydrogen, and fermentation acids were grown in prereduced anaerobically sterilized media, and tests were performed as described in the Virginia Polytechnic Institute and State University Anaerobe Laboratory Manual (1). Neutral red reduction was tested in peptone-yeast extract-fructose medium containing 3 mg of neutral red per 100 ml of medium (the neutral red was added from a suspension of 100 ml of neutral red in 100 ml of 60% aqueous absolute alcohol); disappearance of the red color was interpreted as a positive reaction. Strains also were characterized with the RapID-ANA (Innovative Diagnostics Systems, Inc., Atlanta, Ga.) panels following the directions of the manufacturer. Antibiotic susceptibility results were determined by the methods of Wilkins and Thiel (6). Any reaction that was atypical of results obtained with other strains in the group was repeated.

RESULTS AND DISCUSSION

Bacteroides caccae (cac'ca, pronounced kak'ke) Gr. n. kakke feces; NL gen. n. caccae of feces, referring to source of isolate. Previously referred to as “3452A” DNA homology group (3, 4). Cells of the type strain from peptone-yeast extract-glucose broth cultures are 1.4 to 1.6 by 2.5 to 12 μm and occur singly or in pairs. Cells may appear vacuolated or beaded in strains from broth cultures in media with a fermentable carbohydrate.

Surface colonies on supplemented brain heart infusion blood agar plates (1) incubated for 48 h are 0.5 to 1 mm in diameter, circular, entire, convex, gray, translucent, shiny, and smooth. Rabbit blood may be slightly hemolyzed.

Glucose broth cultures are turbid with a smooth sediment and have a final pH of 5.0 to 5.2. Strains grow equally well at 30 and 37°C but less well at 25 and 45°C. The type strain reduces neutral red and does not produce hydrogen sulfide.

From peptone-yeast extract-glucose broth cultures, major amounts of succinate and acetate, often with trace amounts of propionate and isovalerate, are detected; only a trace of hydrogen is detected in headspace gas. Pyruvate is converted to acetate. Lactate and threonine are not utilized.

Other characteristics of the species are given in Table 1. Characteristics by which B. caccae can be differentiated from phenotypically similar Bacteroides species are given in Table 2.

Type strain: ATCC 43185 (VPI 3452A). The G + C content of the DNA is 40 mol% for the type strain and 40 to 42 mol% for the 20 other strains examined (3).

Bacteroides merdae (mer'dae) L. gen. n. merdae of feces, referring to source of isolate. Previously referred to as “T4-1” DNA homology group (3, 4).

Cells of the type strain from peptone-yeast extract broth cultures are 1.6 by 3.1 to 12 μm and occur singly or in pairs or short chains.

Surface colonies on supplemented brain heart infusion blood agar plates (1) are 0.5 to 1.0 mm in diameter, circular to slightly irregular, entire, convex, white, shiny, and smooth. Rabbit blood is slightly hemolyzed.

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Acid from:

- Amygdalin: 50.0%
- L-Arabinose: 100.0%
- Celllobiose: 74.0%
- Dextrin: 42.0%
- Esculin: 90.0%
- Glycogen: 16.0%
- Gum arabic: 0.0%
- Inulin: 100.0%
- L-Arabinobiose: 100.0%
- Rhamnose: 63.0%
- Xylose: 5.0%
- Hydrolysis of esculin: Yes
- Digestion of gelatin: Yes

Production of:

- Indole: 5.0%
- Catalase: 0.0%
- Hydrogen: Weakly
- Phosphatase: 25.0%
- β-Galactosidase: 100.0%
- α-Glucosidase: 89.0%
- β-Glucosidase: 100.0%
- α-Galactosidase: 100.0%
- α-Fucosidase: 95.0%
- Tetrazolium reduction: 58.0%

Aminopeptidase for:

- Glycine: 100.0%
- Proline: 5.0%
- Phenylalanine: 74.0%
- Arginine: 100.0%
- Serine: 74.0%
- Pyroglutamidase: 0.0%

Susceptibility to:

- Chloramphenicol: 100.0%
- Clindamycin: 100.0%
- Penicillin G: 5.0%
- Tetracycline: 47.0%

Glucose broth cultures are turbid with a smooth sediment and have a final pH of 5.3. The optimum temperature for growth is 37°C. The type strain produces hydrogen sulfide after incubation for 5 days.

From peptone-yeast extract-glucose broth cultures, major amounts of succinate and acetate, often with trace amounts of propionate, isobutyrate, or isovalerate, are detected; no hydrogen, or only a trace amount, is detected in headspace gas. Pyruvate is converted to acetate and propionate. Lactate, threonine, and gluconate are not utilized.

Other characteristics of the species are given in Table 1. Characteristics by which B. merdae can be differentiated from phenotypically similar Bacteroides species are given in Table 2.
**Bacteroides stercoris** (ster'co.ris) L.N. stercus feces; L. gen. n. stercus of feces, referring to source of isolate. Previously referred to as the "subsp. a" DNA homology group (3, 4).

Cells of the type strain from peptone-yeast extract-glucose broth cultures are 1.6 by 2.4 to 12.6 μm and occur singly and in pairs. Vacuolated cells sometimes are seen in cultures in broths that contain a fermentable carbohydrate.

Surface colonies on supplemented brain heart infusion blood agar (1) with rabbit blood are 0.5 to 1 mm in diameter, circular, entire, convex, transparent to translucent, shiny, smooth, and β-hemolytic.

Glucose broth cultures are turbid with a smooth to stringy sediment and have a final pH of 5.0 to 5.4. The optimum temperature for growth is 37°C. Resazurin is reduced; neutral red is not reduced.

From peptone-yeast extract-glucose broth, major amounts of succinate and acetate, often with moderate amounts of formate and propionate, and usually trace amounts of isobutyrate and isovalerate, are produced; no hydrogen is detected in headspace gas. Pyruvate is converted to acetate. Neither lactate, threonine, nor gluconate is used.

**TABLE** from phenotypically similar *B. merdae*.

Neither lactate, threonine, nor gluconate is used.

Detected in headspace gas. Pyruvate is converted to acetate.

**TABLE 2.** Characteristics that help to differentiate *B. caccae* and *B. merdae* from other saccharolytic *Bacteroides* species that grow well in 20% bile and that do not produce indole*.

<table>
<thead>
<tr>
<th>Species</th>
<th>Trehalose</th>
<th>Melezitose</th>
<th>Arabinose</th>
<th>Catalase</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>B. caccae</em></td>
<td>A</td>
<td>A</td>
<td>A</td>
<td>–</td>
</tr>
<tr>
<td><em>B. merdae</em></td>
<td>A</td>
<td>A</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td><em>B. distasonis</em></td>
<td>A</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td><em>B. vulgatus</em></td>
<td>–</td>
<td>–</td>
<td>A</td>
<td>v</td>
</tr>
<tr>
<td><em>B. fragilis</em></td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
</tbody>
</table>

* A, pH below 5.5 for 90 to 100% of strains. +, Positive reaction for 90 to 100% of strains. −, pH 5.5 or above for 90 to 100% of strains or negative reaction. v, Reaction variable among strains.

**TABLE 3.** Characteristics that help to differentiate *B. stercoris* from other indole-positive saccharolytic *Bacteroides* species that grow well in 20% bile

<table>
<thead>
<tr>
<th>Species</th>
<th>Fermentation of:</th>
<th>Melezitose</th>
<th>Cellobiose</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>B. stercoris</em></td>
<td>–</td>
<td>–</td>
<td>A</td>
</tr>
<tr>
<td><em>B. uniformis</em></td>
<td>–</td>
<td>–</td>
<td>A</td>
</tr>
<tr>
<td><em>B. thetaiaotaomicron</em></td>
<td>A</td>
<td>A</td>
<td>A</td>
</tr>
<tr>
<td><em>B. ovatus</em></td>
<td>A</td>
<td>A</td>
<td>A</td>
</tr>
</tbody>
</table>

* A, pH below 5.5 for 90 to 100% of strains; –, pH 5.5 or above for 90 to 100% of strains.

* Reactions from data of Johnson and Ault (4).

of a change in the composition of the substrate available today, as compared with the substrate that we were using 10 years ago.

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**LITERATURE CITED**


