

71. *C. sticklandii*  
hh. Isovaleric not produced.
21. *C. cochlearium*  
gg. Butyric acid not produced.  
h. Formic acid produced.
54. *C. purinilyticum*  
hh. Formic acid not produced.
4. *C. acidurici*  
ff. Nonmotile.
70. *C. sporosphaeroides*

### List of species of the genus *Clostridium*

1. *Clostridium absonum* Nakamura, Shimamura, Hayase and Nishida 1973, 426.<sup>44</sup>

ab'so.num. L. adj. *absonus* inharmonious, not corresponding with; intended to mean "deviating from *C. perfringens*."

This description, unless otherwise indicated, is based on study of the type strain, two other strains having a high degree of DNA homology with the type (Nakamura et al., 1973), and other phenotypically similar strains, as well as on the description by Nakamura et al. (1973) and Hayase et al. (1974).

Cells in PY broth cultures are reported to be nonmotile (Nakamura et al., 1973), but we have found that cells of vigorously growing young cultures of some strains are motile with a single subpolar flagellum. This observation also was made by Stanley M. Harmon (personal communication). Cells in PYG broth cultures are Gram-positive and  $0.9\text{--}1.7 \times 1.7\text{--}11.8 \mu\text{m}$ .

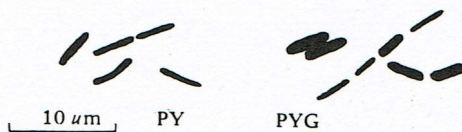


Figure 13.5. *Clostridium absonum*.

Spores are oval, subterminal, and do not swell the cell. Sporulation occurs most readily on egg-yolk agar incubated for 2 days. Spores are seldom seen in broth cultures although the organisms from such cultures survive heating at 80°C for 10 min.

Cell walls contain *meso*-DAP (Weiss et al., 1981).

Surface colonies on blood agar are 1–3 mm, circular to scalloped, slightly opaque, convex, grayish, shiny, smooth, and  $\beta$ -hemolytic.

Glucose broth cultures are turbid with a smooth to mucoid sediment and have a pH of 4.5–4.8 after incubation for 5 days.

Growth is equally abundant at 30, 37 and 45°C, and somewhat less at 25°C. Growth is inhibited by 6.5% NaCl or 20% bile.

Abundant gas is detected in PYG agar deep cultures.

Gelatin is slowly digested; there is no digestion of milk or chopped meat.

Some strains of the species produce hydroxysteroid dehydrogenases that are active in bile acid metabolism (Macdonald et al., 1981, 1983c; Macdonald and Roach, 1981; Sutherland and Macdonald, 1982; Macdonald and Hutchison, 1982).

Products in PYG broth cultures are butyric and acetic acids; lactic and formic acids may be present; ethanol and butanol may be detected. Abundant H<sub>2</sub> is produced. Pyruvate is converted to butyrate and acetate; lactate is not converted to propionate; threonine is not utilized. Ammonia is produced. Neutral red and resazurin are reduced.

All strains tested are susceptible to penicillin G, tetracycline, and chloramphenicol; the type strain is resistant to erythromycin, and four strains including the type are resistant to clindamycin.

Culture supernatants injected intraperitoneally are not toxic to mice; toxin is weakly produced by all strains (Nakamura et al., 1973). Intravenous injection of culture filtrates is lethal to mice (Nakamura et al., 1973; Hayase et al., 1974).

Other characteristics of the species are given in Table 13.12.

Isolated primarily from soil. Similar strains have been isolated from bear feces; one strain is from a case of gas gangrene in a human wound contaminated with soil (Nakamura et al., 1979).

The mol% G + C of the DNA of the type strain has not been reported.

Type strain: ATCC 27555 (DSM 599).

Further comments. *C. absonum* is most easily differentiated from *C. baratii* by its hydrolysis of gelatin, and from *C. perfringens* by not producing H<sub>2</sub>S and by its relative lack of toxicity for mice. The lecithinase of *C. absonum* produced on half-antitoxin Nagler agar cannot be neutralized completely by *C. perfringens* type A antitoxin as can the lecithinases of *C. baratii* and *C. perfringens* (Nakamura et al., 1973).

2. *Clostridium aceticum* (ex Wieringa 1940) Gottschalk and Braun 1981, 476.<sup>VP</sup>

a.ce'ti.cum. L. n. *acetum* vinegar; L. adj. suff. *-icus* belonging to; N.L. neut. adj. *aceticum* related to acetic acid, which it produces.

This description is based on the descriptions by Adamse (1980), Braun et al., (1981), and Gottschalk and Braun (1981).

Cells in broth cultures in an atmosphere of 67% H<sub>2</sub> and 33% CO<sub>2</sub> as described by Braun et al. (1981), are Gram-negative rods, motile and peritrichous, and  $0.3\text{--}1.0 \times 4.0\text{--}8.0 \mu\text{m}$ . With fructose as a substrate, cells may be up to 40  $\mu\text{m}$  in length.



Figure 13.6. *Clostridium aceticum*.

Spores are round, terminal and swell the cell. Sporulation occurs most readily on fructose agar medium incubated for 2 days.

Cell walls contain *meso*-DAP and are composed of at least two layers (Braun et al., 1981).

Colonies do not form readily. After prolonged incubation in roll tubes in an agar medium containing mud extract and a CO<sub>2</sub>-H<sub>2</sub> (1:2) atmosphere (Adamse, 1980), "a barely visible, light-brown tuft of cellular material" can be seen.

The optimum temperature for growth is 30°C. Growth occurs between 25°C and 37°C, but is poor at 45°C. Optimum pH for autotrophic growth is 8.3; growth occurs between pH 7.5 and 9.5. Strains of this species grow chemolithotrophically in an atmosphere of CO<sub>2</sub> and H<sub>2</sub>, converting these substrates to acetate. They also utilize the organic

Table 13.12.

Characteristics of species in the genus *Clostridium*. Acid from glucose, gelatin hydrolyzed.<sup>a</sup>

Characteristics	1. <i>C. absonum</i>	7. <i>C. auranti-butyricum</i>	11. <i>C. bifer-mentans</i>	12. <i>C. botulinum</i>			14. <i>C. cadav-eris</i>	18. <i>C. chauvoei</i>	24. <i>C. difficile</i>	27. <i>C. fel-sineum</i>	31. <i>C. haemo-lyticum</i>	40. <i>C. litus-eburense</i>
				Types C, D	Types B, E, F (saccharo-lytic)	Types A, B, F (proteo-lytic)						
Products from PYG <sup>b</sup>	BAL (f2,4)	LBA	AF(iv icpibbs2)	BPA(vls)	BA(1)	ABiVib (icvp2,3,4)	BA2,4 (fpIs)	ABF(1s)4	BAicivib (fv12,4)	AB4 (1sf)	APB(s)	BAiV pfb (2,3,i4)
Motility	±	+	+	±	+	±	±	d	±	d	+	+
H <sub>2</sub> produced	4	4	4	4	4	4	4	4	4	4	4	-
Indole produced	-	-	+	±	-	-	+	-	-	-	+	-
Lecithinase produced	+	-	+	±	-	-	-	-	-	-	+	+
Lipase produced	-	+	-	+	+	+	-	-	-	-	-	-
Esculin hydrolyzed	+	+	±	-	-	+	-	+	+	+	-	-
Starch hydrolyzed	±	+	-	-	±	-	-	-	-	±	-	-
Nitrate reduced	+	+	-	-	-	-	-	±	-	-	-	-
Acid produced from <sup>c</sup>												
Amygdalin	-w	-	-	-	-w	-	-	-	-	-	-	-
Arabinose	-	w	-	-	-	-	-	-	-	+w	-	-
Cellobiose	+	+	-	-	-	-	-	-	+w	+w	-	-
Fructose	+	+	d	w-	+w	-w	d	-w	+	+	d	+
Galactose	+	w	-	-w	±	-	-	w+	-	+	-w	-
Glycogen	-	w	-	-	-w	-	-	-	-	-	-	-
Inositol	-	-	-	±	-w	-	-	-	-	-	d	-
Inulin	-	-	-	-	-w	-	-	-	-	d	-	-
Lactose	+	+	-	-	-	-	-	+w	-	+w	-	-
Maltose	+	+	w-	d	+w	-w	-	+w	-	±	-w	+
Mannitol	-	-	-	-	-	-	-	-	±	-	-	-
Mannose	+	w	-w	d	+w	-	-w	+w	±	+	d	+w
Melezitose	-	-	-	-	d	-	-	-	d	-	-w	-
Melibiose	±	w	-	-w	-	-	-	-	-	-	w-	-
Raffinose	-	+	-	-	-	-	-	-	-	±	-w	-
Rhamnose	-	-	-	-	-	-	-	-	-	+w	-w	-
Ribose	+w	-	-	d	d	-	-	d	-	-	d	-w
Salicin	+w	w	-	-	-	-	-	-	-w	+	-	-
Sorbitol	-	-	-w	-	±	-w	-	-	-w	-	-	-
Starch	-w	w	-	-	d	-	-	-	-	±	-	-
Sucrose	+	+	-	-	+w	-	-	+w	-	+	-	+w
Trehalose	w+	-	-	-	w+	-	-	-	-w	-	-w	-
Xylose	-	-	-	-	-	-	-	-	-w	+	-	-
Milk reaction	c	c	d	dc	c-	d	cd	c	-	c	cd	cd
Meat digested	-	-	+	±	-	+	+	-	-	-	±	+

substrates fructose, ribose, glutamate, fumarate, malate, pyruvate, serine, formate, ethylene glycol, and ethanol, but in the presence of organic substrates CO<sub>2</sub> and H<sub>2</sub> are not converted to acetate (Braun and Gottschalk, 1981). Dulcitol, adonitol, citrate, succinate, glycine, threonine, lactate, methanol, isopropanol and glycerol are not utilized (Braun et al., 1981). Atmospheric N<sub>2</sub> is fixed (Rosenblum and Wilson, 1949).

H<sub>2</sub> is produced only in the stationary growth phase and inhibits growth in fructose medium at pH 8.5 if the bicarbonate concentration is very low (Braun and Gottschalk, 1981).

Gluconate is fermented to pyruvate and glyceraldehyde-3-phosphate by a modified Entner-Doudoroff pathway (Andreesen and Gottschalk, 1969).

Other characteristics of the species are given in Table 13.13.

Isolated from soil, lake sediment, and sewage sludge.

The mol% G + C of the DNA is 33 (T<sub>m</sub>) (Braun et al., 1981).

Type strain: DSM 1496 (ATCC 35044).

Further comments. The species is most easily differentiated from *C.*

*formiceticum*, which it most closely resembles phenotypically, by its ability to form acetate from CO<sub>2</sub> and H<sub>2</sub>, and to utilize formate, serine or ethylene glycol, but not methanol as substrates. Also see Further Comments under *C. thermaceticum*.

### 3. *Clostridium acetobutylicum* McCoy, Fred, Peterson and Hastings 1926, 483.<sup>AL</sup>

a.ce.to.bu.ty'li.cum. English n. *acetone*; N.L. adj. *butylicum* butylic; N.L. neut. adj. *acetobutylicum* referring to production of acetone and butyl alcohol.

This description is based on the description by Smith and Hobbs (1974), Holdeman et al. (1977), and on study of the type and eight other strains.

Cells in PYG broth cultures are straight rods, motile and peritrichous, 0.5–0.9 × 1.6–6.4 μm. Granulose (a starch-like polymer) is often present (O'Brien and Morris, 1971). Gram-positive, becoming Gram-negative in older cultures.

Table 13.12.—continued

Characteristics	44. <i>C. novyi</i> types			45. <i>C. oceanicum</i>	50. <i>C. perfringens</i>	53. <i>C. puniceum</i>	56. <i>C. putrificum</i>	60. <i>C. roseum</i>	62. <i>C. sardinense</i>	65. <i>C. septicum</i>	66. <i>C. sordellii</i>	69. <i>C. sporogenes</i>	81. <i>C. thermosulfurigenes</i>
	A	B	C	ALb (fics pibiv2,3,4)	ABL (pfs)	AB4(lf)	ABibiv2 (fpicvls)	BA <sub>s</sub> 4	BAL(Fp)	BA(Fpl2)	A(FiCp ibivl)	ABivib2 (picvls4)	2AL
Motility	±	±	+	±	-	+	+	∓	+	±	±	±	+
H <sub>2</sub> produced	4	d	1	4	4	4	4	4	4	4	4	4	4
Indole produced	-	±	+	-	-	-	-	-	-	-	+	-	-
Lecithinase produced	+	+	-	∓	+	-	-	-	+	-	+	-	NT
Lipase produced	+	-	-	-	-	-	-	-	-	-	-	+	NT
Esculin hydrolyzed	-	-	-	+	d	+	d	+	+	+	∓	+	NT
Starch hydrolyzed	-	-	-	d	±	+	-	-	d	-	-	-	+
Nitrate reduced	-	-	-	-	±	-	-	-	±	d	-	-	-
Acid produced from													
Amygdalin	-	-	-	-	-w	-w	-	-	-	-	-	-	+
Arabinose	-	-	-	-	-	-w	-	+	-	-	-	-	+
Cellobiose	-	-	-	d	∓	d	-	+	+w	+w	-	-	+
Fructose	-w	d	-	w+	+	w+	-w	+	+	+	d	-w	NT
Galactose	d	-	-	d	+w	+	-	+	+	+w	-	-	+
Glycogen	-	-	-	-	d	-	-	-	-	-	-	-	NT
Inositol	±	±	w	-	±	-	-	-	-	-	-	-	+
Inulin	-	-	-	-	-w	NT	-	-	-	-	-	-	NT
Lactose	-	-	-	-	+	∓	-	+	+w	+	-	-	-
Maltose	d	d	-	+w	+	+w	-w	-	+w	+	w+	-w	+
Mannitol	-	-	-	-	-	-	-	-	-	-	-	-	+
Mannose	-	w+	w	+w	+	d	-	+	+	+	-w	-	+
Melezitose	-	-	-	-	-	-	-	-	-	-	-	-	-
Melibiose	-	-	-	-	-w	-w	-	-	-	-	-	-	+
Raffinose	-	-	-	-	d	-w	-	-	-	-	-	-	-
Rhamnose	-	-	-	-	-	-	-	+	-	-	-	-	+
Ribose	d	-w	-	-	d	-w	-	-	d	d	-w	-	-
Salicin	∓	-	-	-w	-	±	-	+	+w	d	-	-	+
Sorbitol	-	-	-	-	∓	-	-	-	-	-	-	-	-
Starch	-	-	-	-	d	d	-	+	-w	-	-	-	+
Sucrose	-	-	-	-	+	+	-	+	+w	-	-	-	+
Trehalose	-	-	-	-w	d	∓	-	-	d	+w	-	-	+
Xylose	-	-	-	-	-	d	-	+	-	-	-	-	+
Milk reaction	c	d	-	d-	dc	c	d	cd	c	cd	d	d	NT
Meat digested	-	+	+	+	±	-	±	-	-	-	+	+	NT

<sup>a</sup> Symbols: +, reaction positive for 90–100% of strains (pH of sugars below 5.5); -, reaction negative for 90–100% of strains; ±, 61–89% of strains positive; ∓, 11–39% of strains positive; d, 40–60% of strains positive; w, weak reaction (pH of sugars 5.5–5.9); numbers (hydrogen) represent abundant (4) to negative on a “-” to “4+” scale; c (milk), curd; d (milk), digestion; NT, not tested. Where two reactions are listed, the first is the more usual and occurs in 60–90% of strains.

<sup>b</sup> Products (listed in the order of amounts usually detected): a, acetic acid; b, butyric acid; c, caproic acid; l, lactic acid; f, formic acid; p, propionic acid; s, succinic acid; v, valeric acid; ib, isobutyric acid; ic, isocaproic acid; iv, isovaleric acid; 2, ethanol; 3, propanol; 4, butanol; i4, isobutanol. Capital letters indicate at least 1 meq/100 ml of culture; small letters indicate less than 1 meq/100 ml. Products in parentheses are not detected uniformly.

<sup>c</sup> Ammonia produced from hydrolysis of peptone may mask acid production in this group.



Figure 13.7. *Clostridium acetobutylicum*.

Spores are oval and subterminal, slightly swelling the cell.

Cell walls contain *meso*-DAP, and glucose, rhamnose, galactose and mannose (Cummins and Johnson, 1971). The wall is triple-layered (Cho and Doy, 1973).

Surface colonies on blood agar plates are 1–5 mm, flat to raised, granular, translucent to semiopaque with irregular margins and occasionally with a mosaic internal structure.

Cultures in PYG broth are turbid with a smooth sediment, and have a pH of 4.5–5.0 after incubation for 5 days.

The optimum temperature for growth is 37°C. A fermentable carbohydrate, biotin, and *p*-aminobenzoic acid are required. No growth in the presence of 6.5% NaCl or 20% bile.

Acetyl methyl carbinol is produced. Neutral red is reduced.

Abundant gas is produced in glucose agar deep cultures.

H<sub>2</sub>S is produced by one of nine strains tested.

Fixes atmospheric N<sub>2</sub> (Rosenblum and Wilson, 1949).

Strains produce an inducible carboxymethyl cellulase and cellobiase (Allcock and Woods, 1981). NADH and NADPH-ferredoxin and rubredoxin oxidoreductases also are present (Petitdemange et al., 1977, 1981). Superoxide dismutase (Hewitt and Morris, 1975) and deoxyribonuclease (Johnson and Francis, 1975) are produced.

Table 13.13.

Characteristics of species in the genus *Clostridium*. Gelatin not hydrolyzed, no acid from glucose.<sup>a,b</sup>

Characteristics	2. <i>C. acetivum</i>	4. <i>C. acidurici</i>	5. <i>C. aminovalericum</i>	21. <i>C. cochlearium</i>	28. <i>C. formic-aceticum</i>	37. <i>C. kluyveri</i>	38. <i>C. leptum</i>	41. <i>C. male-nominatum</i>	51. <i>C. polysaccharolyticum</i>	52. <i>C. propionicum</i>	54. <i>C. purinilyticum</i>	57. <i>C. querciculum</i>	70. <i>C. sporosphaeroides</i>	71. <i>C. sticklandii</i>
Products from PYG <sup>c</sup>	A	Af	Af	Bap(flS4)	A(Fs)	CBa	A(2)	BA(fpls)	fabp2	PiVibb as(1)	FA	APb3	ABp	Aivbpib
Motility	+	+	+	±	+	+	-	±	+	+	+	+	-	+
H <sub>2</sub> produced	-	-	4	4	-	2	4	4	3	4	-	4	4	1
Indole produced	NT	-	-	±	-	-	-	+	-	-	-	-	-	-
Esculin hydrolyzed	-	-	+	-	-	-	±	-	+	-	-	-	-	-
Starch hydrolyzed	NT	-	+	-	-	-	-	-	+	-	-	-	-	-
Nitrate reduced	NT	-	-	-	-	-	-	±	-	-	-	-	-	-
Acid produced from														
Amygdalin	-	-	-	-	-	-	-w	-	-	-	-	-	-	-
Cellobiose	NT	-	-	-	-	-	-	-	w	-	-	-	-	-
Fructose	+	-	-	-	w	-	-w	±	-	-	-	+	-	-
Glycogen	NT	-	-	-	-	-	-w	-	w	-	-	-	-	-
Lactose	-	-	-	-	-	-	±	-	-	-	-	-	-	-
Maltose	-	-	-	-	-	-	+	±	-	-	-	-	-	-
Ribose	+	-	-	-	-	-	w-	-	-	-	-	-	-	-
Starch	-	-	-	-	-	-	-	-	w	-	-	-	-	-
Sucrose	-	-	-	-	-	-	±	-	-	-	-	-	-	-
Trehalose	NT	-	-	-	-	-	w-	-	-	-	-	-	-	-
Xylose	-	-	-	-	-	-	w-	-	-	-	-	-	-	-

<sup>a</sup> Strains in this group do not produce lecithinase or lipase; acid is not produced from arabinose, galactose, inositol, inulin, mannitol, mannose, melezitose, melibiose, raffinose, rhamnose, salicin or sorbitol; there is no reaction in milk; meat is not digested.

<sup>b</sup> Symbols: +, reaction positive for 90–100% of strains (pH of sugars below 5.5); -, reaction negative for 90–100% of strains; ±, 50–89% of strains positive; ±, 11–50% of strains positive; w, weak reaction (pH of sugars 5.5–5.9); numbers (hydrogen) represent abundant (4) to negative on a “-” to “4+” scale; NT, not tested. Where two reactions are listed, the first is the more usual and occurs in 60–90% of strains.

<sup>c</sup> Products (listed in order of amounts usually detected): a, acetic acid; b, butyric acid; f, formic acid; c, caproic acid; l, lactic acid; p, propionic acid, s, succinic acid; iv, isovaleric acid; ib, isobutyric acid; 2, ethanol; 3, propanol; 4, butanol. Capital letters indicate at least 1 meq/100 ml of culture; small letters indicate less than 1 meq/100 ml. Products in parentheses are not detected uniformly.

Fermentation products include acetic, butyric and lactic acids, butanol, acetone, CO<sub>2</sub> and large amounts of H<sub>2</sub>. Small amounts of succinic acid may be formed. Ethanol was detected with high pressure liquid chromatography (Ehrlich et al., 1981). During exponential growth, products are acetate and butyrate. Production of butanol and acetone is highest after 18 h when the organisms are in their stationary growth phase and is associated with morphological changes in the cells (Jones et al., 1982).

Pyruvate is converted to acetate, butyrate and butanol. Neither lactate nor threonine is utilized.

The type strain produces the amino acids lysine, arginine, aspartic acid, threonine, serine, glutamic acid, alanine, valine, isoleucine, leucine and tyrosine in broth (Matteuzzi et al., 1978).

For information on stimulation of solvent production by *C. acetobutylicum*, see reports of Bahl et al. (1982a, b), Andersch et al. (1982), Monot et al. (1982), Jones et al. (1982), George et al. (1983), and Lin and Blaschek (1983).

Strains are susceptible to chloramphenicol, clindamycin, erythromycin, penicillin G, and tetracycline.

Culture supernatants are nontoxic to mice.

Other characteristics of the species are listed in Table 13.14.

Isolated from soil. Also reported from lake sediment, well water, and clam gut (Ehrlich et al., 1981), from bovine feces (Princewill and Agba, 1982), canine feces (Balish et al., 1977), and human feces (Drasar et al., 1976; Finegold et al., 1983).

The mol% G + C of the DNA is 28–29 (*T<sub>m</sub>*) (Cummins and Johnson, 1971).

Type strain: ATCC 824 (NCIB 8052, DSM 792).

Further comments. This species is most easily differentiated from *C. beijerinckii* by its absolute requirement for a fermentable carbohydrate

for growth, and by requirement for biotin and *p*-aminobenzoic acid; *C. beijerinckii* requires other vitamins and amino acids as well. *C. acetobutylicum* differs from *C. aurantibutyricum* and *C. felsineum* by its failure to digest gelatin.

4. *Clostridium acidurici* (corr.) (Liebert, 1909) Barker 1938, 323.<sup>AL</sup>  
a.ci.du'ri.ci. N.L. n. *acidum uricum* uric acid; N.L.gen.n. *acidurici* of uric acid.

Based on the description by Smith and Hobbs (1974) and study of the type strain.

Cells in PY-0.3% uric acid broth are Gram-variable to Gram-negative, motile and peritrichous, 0.5–0.7 × 2.5–4.0 μm, occurring singly.



Figure 13.8. *Clostridium acidurici*.

Spores are oval, terminal and subterminal, and swell the cell. Sporulation occurs most reliably on chopped meat-uric acid agar slants incubated at 30°C for 1 week in an atmosphere of N<sub>2</sub>.

Cell walls contain meso-DAP (Weiss et al., 1981).

Surface colonies on uric acid agar are spreading, rhizoid, transparent, colorless and flat, clearing the agar.

No growth in gelatin or milk, or on egg-yolk or blood agar.

Table 13.14.

Characteristics of species in the genus *Clostridium*. Acid from glucose, gelatin not hydrolyzed, meat not digested.<sup>a</sup>

Characteristics	3. <i>C. acetobutylicum</i>	6. <i>C. arcticum</i>	8. <i>C. baratii</i>	9. <i>C. barkeri</i>	10. <i>C. beijerinckii</i>	13. <i>C. butyricum</i>	15. <i>C. carnis</i>	16. <i>C. celatum</i>	17. <i>C. cellobioparum</i>	19. <i>C. clostridioforme</i>	20. <i>C. coccoides</i>	22. <i>C. cocleatum</i>	23. <i>C. colinum</i>
Products from PYG <sup>b</sup>	BAl4(s)	PA(b)	BAL (fps)	BLpa	BA (Fpls,2,4)	BAF (ls2,4)	BALf(s)	AFb2(s)	Alf2	A(Fls2)	AS	AF(Ls)	FAP(l)
Motility	±	+	-	-	+	±	±	-	+	∓	-	-	+
H <sub>2</sub> produced	4	tr	4	4	4	4	4	4	4	4	4	4	4
Indole produced	-	+	-	-	-	-	-	-	-	∓	-	-	-
Lecithinase produced	-	NT	+	-	-	-	-	-	-	-	-	-	-
Lipase produced	-	NT	-	-	-	-	-	-	-	-	-	-	-
Esculin hydrolyzed	+	+	+	+	+	+	+	+	+	+	+	+	+
Starch hydrolyzed	+	-	d	-	±	+	-	-	-	-	-	-	-
Nitrate reduced	-	-	d	-	-	-	-	d	-	±	-	-	-
Acid produced from													
Amygdalin	-	-	-	-	∓	±	w	+	-	-w	+	±	w-
Arabinose	d	-	-	-	±	±	-	-	+	d	+	-	-
Cellobiose	+	w	+	-	+	+	w+	+	+	±	+	+	d
Fructose	+	+	+	+	+	+	d	+	+	+	+	+	+
Galactose	+	NT	+w	-	+	+	d	+	+	w+	+	+	w
Glycogen	d	-	∓	-	d	+	-	-	-	-	+	-	-
Inulin	-	NT	-	-	d	∓	-	-	-	-	-	+w	d
Lactose	d	-	+w	-	+	+	d	+	w	±	+	+	-
Maltose	+	±	+w	-	+	+	w+	+	+	+w	+	∓	+
Mannitol	∓	w	-	+	d	∓	-	-	-	-	+	-	d
Mannose	+	+	+	-	+	+	+w	+	+	+w	+	+	+
Melezitose	-	-	-	-	∓	∓	-	-	-	∓	+	-	-
Melibiose	-	-	-w	-	d	+	-	-	+	d	+	-	+w
Raffinose	-	-	-	-	±	+	-	-	-	±	+	∓	+
Rhamnose	-	-	-	-	∓	-	-	-	-	±	+	-	-
Ribose	-	-	-w	-	∓	+	-	d	+	d	+	-	w-
Salicin	+	w	+w	-	∓	+	w	+	w	±	+	d	w+
Sorbitol	-	-	-	+	∓	-	-	-	-	-	+	-	-
Starch	+	-	d	-	±	+	-w	-	-	-w	-	-	-w
Sucrose	+	-	+	-	+	+	+w	+	-	+	+	+	+
Trehalose	∓	-	-	-	±	+	-	+	-	±	+	-	+
Xylose	±	+	-	-	+	+	-	-	+	+	+	-	-
Milk reaction	c	a	c	-	c	c	-	c	-	c	c	c	-

Broth cultures supplemented with 0.3% uric acid have a smooth sediment with no turbidity and a pH of 7.4-7.7 after incubation under N<sub>2</sub> for 6 days. Most rapid growth occurs in media at an initial pH of 7.6-8.1; there is poor growth below pH 6.5 or above 9.0. Growth occurs between 19°C and 37°C (Barker and Beck, 1942). Uric acid, xanthine, guanine or hypoxanthine is required as a carbon and energy source (Barker and Beck, 1941). Selenite and tungstate stimulate xanthine dehydrogenase and formate dehydrogenase activity (Wagner and Andreesen, 1979).

Products in PY-urate broth are acetate, NH<sub>3</sub>, and CO<sub>2</sub>. No H<sub>2</sub> is produced. No carbohydrates are fermented.

The type strain is resistant to erythromycin, penicillin and tetracycline. It is moderately sensitive to chloramphenicol and clindamycin.

Culture supernatants of the type strain are nontoxic to mice.

Other characteristics of the species are given in Table 13.13.

Strains are widely distributed in soil. They have been isolated also from chicken droppings (Barker, 1978), and from wild birds (Barker and Beck, 1942).

The mol% of the G + C is 28 (T<sub>m</sub>) (Dürre et al., 1981).

Type strain: ATCC 7906 (DSM 604).

Further comments. For reviews concerning the metabolic degradation of purines by this organism see Vogels and Van der Drift (1976) and Yoch and Carithers (1979). See also Champion and Rabinowitz (1977), Wagner and Andreesen (1977), Waber and Wood (1979), and Dürre and Andreesen (1983).

*C. cylindrosporium* was isolated at the same time and from the same sample as *C. acidurici*. *C. cylindrosporium* has been validated as a species (Andreesen et al., 1985, 207) and has been studied widely in conjunction with studies on *C. acidurici* (Barker and Beck, 1941, 1942; Champion and Rabinowitz, 1977; Wagner and Andreesen, 1977, 1979; Tanner et al., 1982). Metabolically the organisms are very similar; both ferment uric acid, neither ferments any carbohydrate. The mol% G + C of *C. cylindrosporium*, however, is 32 by T<sub>m</sub> (Tonomura et al., 1965), and a partial catalog of the 16S-rRNA of *C. cylindrosporium* indicates that it is not closely related to *C. acidurici* (Tanner et al., 1982). Originally these organisms were differentiated on the basis of cell morphology and the size, shape and position of their spores (Barker and Beck, 1942). Champion and Rabinowitz (1977) have found that *C. cylindrosporium* forms formate from uric acid while formate is not produced by *C. acidurici*. Wagner and Andreesen (1977, 1979) have found that the two organisms have different metal ion requirements for formation of formate dehydrogenases and different requirements for hypoxanthine metabolism.

The mol% of the G + C is 28 (T<sub>m</sub>) (Dürre et al., 1981).

*C. cylindrosporium* type strain: ATCC 7905 (DSM 605).

See Further Comments under *C. purinolyticum* for differentiation of these three species.

5. *Clostridium aminovalericum* Hardman and Stadtman 1960, 552.<sup>AL</sup>

Table 13.14—continued

Characteristics	25. <i>C. durum</i>	26. <i>C. fallax</i>	30. <i>C. glycolicum</i>	34. <i>C. indolis</i>	35. <i>C. innocuum</i>	43. <i>C. nexile</i>	46. <i>C. oroticum</i>	47. <i>C. papyrosolvens</i>	48. <i>C. paraputrificum</i>	49. <i>C. pasteurianum</i>	58. <i>C. ramosum</i>	59. <i>C. rectum</i>
Products from PYG <sup>b</sup>	laf2	ABL(s)	AiViB2,3, i4i5(pfls)	AF2	BLa(fs)	AF2(ls)	AF2(ls)	AL2	BAL(sf)	ABf(ls)	FAI(s2)	Bapv
Motility	+	±	±	+	-	-	-	+	±	±	-	-
H <sub>2</sub> produced	4	4	4	4	4	4	4	4	4	4	d	4
Indole produced	-	-	-	+	-	-	-	-	-	-	-	-
Lecithinase produced	-	-	-	-	-	-	-	-	-	-	-	-
Lipase produced	-	-	-	-	-	-	-	-	-	-	-	-
Esculin hydrolyzed	+	+	±	+	+	+	+	+	+	-	+	+
Starch hydrolyzed	-	-	-	±	-	-	-	-	+	-	-	-
Nitrate reduced	-	±	-	±	-	-	±	-	±	-	-	-
Acid produced from												
Amygdalin	-	-	-	-	-	-w	-	-	d	-	+	-
Arabinose	-	-	-	-w	-	-	+	+	-	w	-	-
Cellobiose	-	-w	-	+w	+	-	-	+	+	-	+	-
Fructose	+	+	+w	+w	+	w+	+	±	+	+	+	-
Galactose	+	w+	-	w+	+	+w	+	+	+	w-	+	w
Glycogen	-	-	-	-	-	-	-	-	±	-	-	-
Inulin	-	-	-	-	+	d	+w	-	-	-	-	-
Lactose	-	w-	-	w+	-	+w	+	-	+	-w	+	w
Maltose	+	+	d	w+	-	-w	+	-	+	+	+	-
Mannitol	-	-	-	-w	+	-	±	-	-	+	±	-
Mannose	+	+w	-	d	+	-w	-	-	+	+	+	-
Melezitose	w	-	-	-	-	-	±	-	-	+	-	-
Melibiose	-	-	-	-w	-	w-	-	-	-	+	±	-
Raffinose	+	-	-	+w	-w	d	+	-	-	+	+	-
Rhamnose	-	-	-	±	-	-	+	-	-	-	d	-
Ribose	-	w+	-	-w	±	-	+	+	w-	-	d	-
Salicin	-	-	-	w-	+	±	+	-	+	-	+	w
Sorbitol	-	-	±	-	-	-	-w	-	-	+	-	-
Starch	-	w+	-w	-	-	-w	-	-	+	-w	±	-
Sucrose	+	-	-	d	+	+w	+	-	+	+	+	w
Trehalose	+	-	-	w-	+	-w	±	-	±	+	+	-
Xylose	-	-	±	d	-w	w+	+	+	-	w	-w	-
Milk reaction	-	c	-	c	-	-c	c	-	c	-	c	c

a.mi'no.va.ler'i.cum. L. adj. suff. *-icus* related to; N.L. neut. adj. *aminovalericum* referring to ability to ferment aminovaleric acid strongly.

Description based on those of Smith and Hobbs (1974) and Holde-man et al. (1977), and on study of the type strain (received from three different sources).

Cells in PYG broth cultures are straight rods, motile and peritrichous, 0.3–0.5 × 1.5–5.2 μm, occurring singly and in pairs. Cells stain Gram-positive but rapidly become Gram-negative as cultures reach maximum stationary phase.

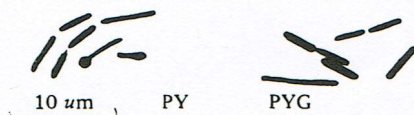


Figure 13.9. *Clostridium aminovalericum*.

Spores are small, spherical and terminal, swelling the cell. The sporulation occurs most reliably on chopped meat agar slants incubated at 30°C for 5 days.

Cell walls contain *meso*-DAP (Weiss et al., 1981).

Surface colonies on blood agar plates are 0.5–1 mm, circular, entire,

flat to convex, translucent to opaque, granular, gray, dull, smooth and weakly hemolytic.

Cultures in PYG broth are turbid with a smooth sediment and have a pH of 5.9–6.2 after incubation for 1 week.

Optimum temperature for growth is 37°C; grows at 25°C and 30°C; poor growth at 45°C. Growth is inhibited by 6.5% NaCl and by 20% bile.

Hippurate is hydrolyzed by the type strain. Neutral red and resazurin are reduced.

Abundant gas is produced in PYG agar deep cultures.

Deoxyribonuclease is present (Smith 1975a).

Products in PYG broth at a pH of 6.1 include major amounts of acetic acid and abundant H<sub>2</sub>. From aminovalerate as the sole energy source, at a pH of 7.4–7.7, acetate, ammonia, propionate and valerate are produced (Hardman and Stadtman, 1960). Phenylacetic acid has been detected in the one strain tested (Mayrand and Bourgeau, 1982).

Culture supernatants of the type strain are nontoxic to mice.

The type strain is sensitive to chloramphenicol, erythromycin, penicillin G and tetracycline. Resistance to clindamycin is variable.

Other characteristics of the species are given in Table 13.13.

Isolated from sewage sludge and from rumen contents of bloating calves (Jayne-Williams, 1979); also isolated from urine specimens from pregnant women with bacteriuria (Meijer-Severs et al., 1979), from hamster feces (Bartlett et al., 1978) and from human feces (Drasar et al., 1976; Finegold et al., 1983).