

Isolation of *Arcobacter skirrowii* from a Patient with Chronic Diarrhea

The genus *Arcobacter* currently includes four species (7). Two species, *Arcobacter cryaerophilus* and *Arcobacter butzleri*, have been associated with human disease. They were mainly isolated from patients with diarrhea and bacteremia (2, 3, 6). *Arcobacter nitrofigilis* has been associated with roots of a salt marsh plant, *Spartina alterniflora*. The fourth species, *Arcobacter skirrowii*, has been recovered from lambs with diarrhea; from aborted porcine, ovine, and bovine fetuses; and from the preputium of bulls. Until now, this species has not been isolated from clinical specimens from humans. We report here a case of diarrhea in an elderly patient from whom *A. skirrowii* was isolated.

A 73-year-old man with a prosthetic aortic heart valve was admitted to the hospital after two months of persisting diarrhea. The diarrhea had not responded to dietary measures and treatment with nifuroxazide. The patient lost weight, was anorexic, and became asthenic. He was treated for progressive coughing with oral levofloxacin (500 mg/day) for 4 days just before hospitalization. At day 6 of hospitalization, he was given intravenous ampicillin (2 g) and netilmicin (150 mg) as endocarditis prophylaxis 30 min before a colonoscopy was performed and 500 mg of amoxicillin 6 h after intervention. This procedure showed diverticulosis but no malignancies. He was hospitalized for 14 days. During this stay, his symptoms resolved gradually without any specific treatment. At day 10, the diarrhea had disappeared.

Stool specimens were cultured for conventional enteric pathogens and *Campylobacter* species at days 1, 2, 10, and 12 of hospitalization. In addition to a selective *Campylobacter* medium incubated at 42°C in a microaerophilic atmosphere (containing approximately 80% N₂, 5% O₂, 10% CO₂, and 6% H₂), a filtration technique was used. A sterile cellulose acetate membrane filter with a 0.65-μm pore size was placed on a Columbia agar plate with 5% defibrinated horse blood and was inoculated with 5 drops of fecal suspension in nutrient broth. After 20 min, the filter was removed and the plate was incubated at 37°C in a microaerophilic atmosphere (with 6% H₂).

The conventional cultures for enteric pathogens and *Campylobacter* were negative. After 4 days of incubation, the Colum-

bia blood agar plate inoculated with the first stool sample showed oxidase-positive and strongly-catalase-positive grayish colonies of gram-negative curved rods. The strain was able to grow at 15°C, which is a distinctive feature that differentiates *Arcobacter* species from *Campylobacter* species and other *Campylobacter*-like organisms. The results of the phenotypic tests of the isolate were compatible with those of *A. skirrowii* (Table 1). Differentiating among *Arcobacter* species, however, is difficult when using classical tests and may give erroneous results. A multiplex PCR assay with five primers targeting the 16S and 23S rRNA genes, developed for the simultaneous identification of *A. butzleri*, *A. cryaerophilus*, and *A. skirrowii*, confirmed the organism as *A. skirrowii* (1). Polyacrylamide gel electrophoresis of the whole-cell proteins was performed. The obtained profile was compared with a database for the identification of *Campylobacter* species and related organisms by means of a numerical analysis of whole-cell protein profiles (7). The identification of the isolate as *A. skirrowii* was confirmed once more.

We believe this to be the first isolation of *A. skirrowii* from a human stool sample. There was no association with farm animals or pets, and food anamnesis was not relevant. The source of the *A. skirrowii* remains obscure. It is not clear whether this strain was etiologically associated with the patient's diarrhea. Because of the fastidious growth of *Arcobacter* species, it is not easy to evaluate their possible role in human disease. However, 20 years of experience with the filtration technique gives us reason to believe that even if *A. skirrowii* is pathogenic, its role in human disease is limited.

REFERENCES

1. Houf, K., A. Tutenel, L. De Zutter, J. Van Hoof, and P. Vandamme. 2000. Development of a multiplex PCR assay for the simultaneous detection and identification of *Arcobacter butzleri*, *Arcobacter cryaerophilus* and *Arcobacter skirrowii*. FEMS Microbiol. Lett. 193:89–94.
2. Hsueh, P. R., L. J. Teng, P. C. Yang, S. K. Wang, S. C. Chang, S. W. Ho, W. C. Hsieh, and K. T. Luh. 1997. Bacteremia caused by *Arcobacter cryaerophilus* 1B. J. Clin. Microbiol. 35:489–491.
3. Lerner, J., V. Brumberger, and V. Preac-Mursic. 1994. Severe diarrhea associated with *Arcobacter butzleri*. Eur. J. Clin. Microbiol. Infect. Dis. 13:660–662.

TABLE 1. Phenotypic properties of *Arcobacter* species and the patient strain^{a,b}

Characteristic	<i>A. butzleri</i>	<i>A. cryaerophilus</i>	<i>A. nitrofigilis</i>	<i>A. skirrowii</i>	Patient strain
Catalase activity	V	V	+	+	+
Nitrate reduction	+	+	+	+	+
Urease	–	–	+	–	–
H ₂ S (TSI)	–	–	–	–	–
Hippurate hydrolysis	–	–	–	–	–
Indoxylacetate hydrolysis	+	+	+	+	+
Growth at 15°C	+	+	+	+	+
Growth at 25°C	+	+	+	+	+
Growth at 42°C	V	–	–	V	W
Growth in 1% glycine	–	V	–	– (V ^c)	–
Growth on MacConkey agar	V	V	–	–	–
Susceptibility to nalidixic acid	V	V	S	S	R
Susceptibility to cephalothin	R	R	S	R (S ^c)	S

^a According to Nachamkin (5).

^b +, positive reaction; –, negative reaction; TSI, triple sugar iron; W, weak reaction; V, variable reaction; S, susceptible; R, resistant.

^c According to Nachamkin (4).

4. **Nachamkin, I.** 1999. *Campylobacter* and *Arcobacter*, p. 717. In P. R. Murray, E. J. Baron, M. A. Phaller, F. C. Tenover, and R. H. Tenover (ed.), Manual of clinical microbiology, 7th ed. ASM Press, Washington, D.C.
5. **Nachamkin, I.** 2003. *Campylobacter* and *Arcobacter*, p. 907. In P. R. Murray, E. J. Baron, J. H. Jorgensen, M. A. Phaller, and R. H. Tenover (ed.), Manual of clinical microbiology, 8th ed. ASM Press, Washington, D.C.
6. **Vandamme, P., P. Pugina, G. Benzi, R. Van Etterijck, L. Vlaes, K. Kersters, J.-P. Butzler, H. Lior, and S. Lauwers.** 1992. Outbreak of recurrent abdominal cramps associated with *Arcobacter butzleri* in an Italian school. J. Clin. Microbiol. **30**:2335–2337.
7. **Vandamme, P., M. Vancanneyt, B. Pot, L. Mels, B. Hoste, D. Dewettinck, L. Vlaes, C. Van Den Borre, R. Higgins, J. Hommez, K. Kersters, J.-P. Butzler, and H. Goossens.** 1992. Polyphasic taxonomic study of the emended genus *Arcobacter* with *Arcobacter butzleri* comb. nov. and *Arcobacter skirrowii* sp. nov., an aerotolerant bacterium isolated from veterinary specimens. Int. J. Syst. Bacteriol. **42**:344–356.

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