JOURNAL OF CLINICAL MICROBIOLOGY, July 1995, p. 1950–1953 Vol. 33, No. 7
0095-1137/95/$04.00+0

Brain Abscess Due to *Bacillus macerans* following a Penetrating Periorbital Injury

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Received 21 November 1994/Returned for modification 28 December 1994/Accepted 17 April 1995

We report a case of a brain abscess due to *Bacillus macerans* and *Clostridium* sp. following a penetrating periorbital injury by a wooden branch. Intracranial penetration by and retention of a foreign body were not suspected initially, and neurological symptoms developed only 2.5 months later. Previously reported cases of brain abscesses due to *Bacillus* species are reviewed.

Periorbital puncture wounds by wooden objects are particularly dangerous because of the risk of unsuspected intracranial penetration and retention of a foreign body. Months after an apparently trivial injury, serious infectious complications, such as a brain abscess, can occur (4, 5, 18). Non-anthrax *Bacillus* species, which are often dismissed as contaminants when recovered from clinical specimens, are now increasingly recognized as opportunistic pathogens (8, 9, 11, 22, 23). However, there have been very few reports of brain abscesses due to these bacteria. This report describes a case of intracranial infection due to *Bacillus macerans* following a penetrating periorbital injury.

A previously healthy 52-year-old man fell from a scaffold onto a *Thuja* hedge. He did not lose consciousness and suffered only a right orbital hematoma. A skull X-ray revealed no fracture. Three weeks later, he developed an abscess on the right eyelid which was treated by drainage and a 3-week course of oral amoxicillin-clavulanate (2 g/day). Two and one-half months after the initial trauma, the patient began to experience severe frontal headaches and giddiness and became increasingly disoriented. A cranial computerized tomography scan showed a right frontal lesion with dilated ventricles and a major mass effect. The patient was transferred to a neurosurgical unit. At the time of admission, he was conscious but lethargic and complained of severe headaches. On the following day, his condition began to deteriorate, with seizures, confusion, and facial paralysis. Magnetic resonance imaging showed a right frontal lesion with surrounding edema and ventricular dilatation due to obstruction of the interventricular foramen. Surgical exploration after frontal craniotomy revealed a major inflammatory granuloma, containing a limited amount of pus, which originated from a fracture line in the orbital roof and progressed into the right frontal lobe. Small fragments of bark were found between the dura mater and the bone. Specimens of abscess pus, the foreign body surrounded by the granuloma, and ventricular fluid were taken to the microbiology laboratory. The patient was started on intravenous amoxicillin (12 g/day) and metronidazole (1.5 g/day). On the day following the intervention, his condition deteriorated, with an extension response to stimuli of the upper limbs and nonreactive pupils. Transcranial Doppler analysis revealed intracranial hypertension, and a computerized tomography scan showed a major cerebral edema with attenuation of the frontal lobes and the presence of blood and gas. A second surgical intervention was undertaken to set up an external ventricular shunt. The patient died during the postoperative course.

The specimens were cultured onto the following media at 37°C for 48 h: blood agar (incubated aerobically and anaerobically), chocolate agar (incubated in 5% CO_2), and Sabouraud agar. Thioglycolate broth was also inoculated with the ventricular fluid and incubated for 5 days. Direct Gram staining of the abscess pus revealed numerous altered polymorphonuclear leukocytes and rare gram-variable rods, but no bacteria were seen in the granuloma and the ventricular fluid. Aerobic cultures of the pus and the granuloma on blood agar and chocolate agar yielded a moderate amount of a nonhemolytic, spore-forming, gram-variable rod in 48 h. The same organism was recovered in 72 h from the thioglycolate broth inoculated with ventricular fluid. The spores were ellipsoidal and subterminal and swelled the bacilli. The isolate was motile, catalase positive, and facultatively anaerobic. It utilized glucose, xylose, arabinose, mannitol, and galactose but not inositol and sorbitol. A *Bacillus* species was suspected. It was identified as *B. macerans* with API 20E and API 50CHB strips (Biomérieux) and a few supplementary tests (16, 24). The main growth and biochemical characteristics of the isolate are shown in Table 1. Antibiotic susceptibility was tested by the disk diffusion method on Mueller-Hinton agar in accordance with the guidelines of the French Antibiogram Committee (1). The results were as follows: susceptibility to amoxicillin, ticarcillin, piperacillin, cefalotin, cefoxitin, cefotaxime, gentamicin, tobramycin, netilmicin, amikacin, minocycline, pefloxacin, and trimethoprim-sulfamethoxazole and resistance to colistin. Meanwhile, anaerobic cultures of the granuloma grew a few colonies of a gram-positive, spore-forming, catalase-negative, strictly anaerobic rod producing beta-hemolytic colonies on blood agar. The rapid ID 32A and API 20A systems (Biomérieux) identified the organism as *Clostridium butyricum* or *C. beijerinckii*. However, gas-liquid chromatography analysis of fermentation products showed that the isolate produced a lot of acetic acid and very little butyric acid. This result is inconsistent with *C. butyricum* or *C. beijerinckii*, which has a much higher butyric acid/acetic acid ratio (17). *C. indolis* was suspected on the basis of gas-liquid chromatography analysis but eventually excluded because of three inconsistent characteristics (indole negative and rhamnose and xylose positive). The strain could therefore not be identified to the species level. This diagnosis was confirmed by a reference center (Institut Pasteur, Paris, France). Susceptibility testing by the disk diffusion method on Wilkins-Chalgren agar showed that the isolate was susceptible to penicillin G, amoxicillin, ticarcillin, piper-

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TABLE 1. Main characteristics of the *B. macerans* isolate in comparison with other *Bacillus* species[a]

Test and characteristic	Present isolate	*B. cereus*	*B. subtilis*	*B. licheniformis*	*B. circulans*	*B. polymyxa*
API 20E						
ONPG[b]	+	−	+	+	+	+
ADH[c]	−	±	−	+	−	−
Citrate	−	+	+	+	−	−
Urease	−	±	−	±	−	−
Indole	−	−	−	−	−	−
Voges-Proskauer reaction	−	+	+	+	±	+
Gelatin	−	+	+	+	±	+
Nitrate	+	±	+	+	−	+
API 50 CHB						
D-Arabinose	+	−	−	−	±	−
L-Arabinose	+	−	+	+	+	+
Ribose	+	+	+	+	+	+
D-Xylose	+	−	+	+	+	+
β-Methylxyloside	+	−	−	−	+	+
Galactose	+	−	±	+	+	+
D-Glucose	+	+	+	+	+	+
D-Mannose	+	−	+	+	+	+
Rhamnose	+	−	−	±	±	±
Inositol	−	−	+	±	±	−
Mannitol	+	−	+	+	+	+
Sorbitol	−	−	+	+	±	−
Maltose	+	+	+	+	+	+
Lactose	+	−	±	+	+	+
Melezitose	+	−	−	−	±	±
D-Raffinose	+	−	+	±	+	+
D-Arabitol	+	−	−	−	−	−
Other						
Anaerobic growth	+	+	−	+	±	+
Growth at 50°C	+	−	+	+	+	−
Growth at pH 5.7	+	+	+	+	±	+
Growth in 7% NaCl	−	+	+	+	±	−
Gas from glucose	+	−	−	−	−	+
Casein hydrolysis	−	+	+	±	±	+

[a] Some of the data shown here are from references 16 and 24. +, ≥85% of strains positive; ±, 16 to 84% of strains positive; −, ≤15% of strains positive.
[b] ONPG, *o*-nitrophenyl-β-D-galactopyranoside.
[c] ADH, aldehyde dehydrogenase.

acillin, imipenem, cefoxitin, chloramphenicol, minocycline, and metronidazole and resistant to clindamycin.

Cases of delayed intracranial infections following periorbital injuries have often been reported (4, 5, 10, 18). The penetrating object is usually a wooden stick, a branch, or a pencil. Wood is especially dangerous since it is porous, often grossly contaminated, easily fragmented, and difficult to detect radiologically. Because the head is unconsciously thrown backward at the instant of injury, the traversing object is most often directed toward the superior orbital plate, which is very thin and allows easy access to the frontal lobes. Organisms present on the wood, or skin bacteria contaminating it during the impact, may be thus transported intracranially. Most of the time, the initial wound appears to be superficial, does not damage the ocular globe, and is considered to be trivial. Intracranial penetration by and retention of a foreign body are not suspected, since immediate neurological symptoms are rare and ordinary X-rays often appear normal. Serious infectious complications can occur days to years after the initial wounding. Brain abscess, which is the most frequent complication and the main cause of death, occurred in half of the 42 cases of intracranial injury following periorbital wounds reviewed by Miller et al. (18) and 15 of the 21 cases of cerebral injury by pencils reviewed by Bursick and Selker (5). The pathogen most frequently involved was *Staphylococcus aureus*. The case we have described differs from those reported in the literature by its very unusual bacterial etiology.

Members of the genus *Bacillus* are gram-positive, aerobic, spore-forming rods that are ubiquitous in the environment (6). Whereas *B. anthracis* is a well-known pathogen, non-anthrax *Bacillus* species have often been dismissed as culture contaminants when isolated from clinical material. However, their role as opportunistic pathogens has been increasingly recognized over the past few decades (3, 8, 9, 11, 22, 23). The most frequently involved organisms are *B. cereus* and, to a lesser extent, *B. subtilis* and *B. licheniformis*, whereas other species have been very rarely incriminated. In a recent review of the medical importance of *Bacillus* species (15), Logan found only one case of human infection due to *B. macerans*. It was a wound infection following removal of a malignant melanoma (11). This species was described by Schardinger in 1905 (21). Its spores are rarely found in soil, but it multiplies in plant materials (6). It has been isolated from cases of cattle abortion in Scotland (15).

Approximately 30 cases of *Bacillus* central nervous system infections, consisting mainly of meningitis and ventricular shunt infections, have been reported (25). Most of them were due to *B. cereus* and associated with predisposing factors, such as surgical procedures or head trauma. However, case reports of *Bacillus* brain abscesses are exceptional. Our review of the

TABLE 2. Case reports of brain abscesses due to *Bacillus* species since 1960

Case (reference)	Age (yr)	Predisposing factors	Location(s)	Route of infection	Species	Associated pathogen(s)	Treatment	Outcome
1 (11)	63	Leukemia, chemotherapy	Cingulate gyrus	Hematogenous spread	*B. cereus*[a]	None	Antibiotics	Death
2 (11)	3	Leukemia, chemotherapy	Multiple	Hematogenous spread	*B. subtilis*[a]	None	Antibiotics	Death
3 (20)	56	Leukemia, chemotherapy	Multiple	Hematogenous spread	*B. subtilis*[a]	None	Antibiotics	Death
4 (12)	3	Leukemia, chemotherapy	Multiple	Hematogenous spread	*B. cereus*	None	Antibiotics	Memory deficit, hemiparesis
5 (13)	41	None	Frontal	Penetrating orbital injury	*B. licheniformis*	*Fusobacterium* sp., *Bacteroides asaccharolyticus*	Aspiration, antibiotics	Recovery
This study	52	None	Frontal	Penetrating orbital injury	*B. macerans*	*Clostridium* sp.	Excision, antibiotics	Death

[a] Species isolated from blood and sputum cultures. Gram-stained histopathologic sections of the brain revealed bacilli, but they were not cultured.

literature yielded only five cases since 1960 (Table 2). In cases 1 to 3 (11, 20), brain abscess was a postmortem finding in immunocompromised patients with disseminated infections. Case 4 (12) also occurred in a neutropenic patient, and the cerebral location was probably a metastatic focus subsequent to hematogenous spread of bacteria. In contrast, the case reported by Jones et al. (13) is quite similar to ours: *B. licheniformis* was isolated from a frontal brain abscess following a penetrating periorbital injury by a dried plant stem. Small fragments of wood were found in the abscess cavity, and anaerobic organisms were associated with the *Bacillus* species. The patient was successfully treated by drainage and antibiotics.

When isolating a *Bacillus* species from a clinical specimen, microbiologists need to distinguish between a real pathogen and a contaminant. A *Bacillus* isolate is generally considered clinically significant when it is recovered from at least two blood cultures drawn on different occasions or one blood culture with a microbiologically documented source of infection or isolated in pure culture from a surgical specimen obtained during an open surgical procedure with clinical evidence of infection at that site (3, 22). The present isolate was recovered from three distinct surgical specimens, and although it was not purely cultured from one of them it was considered to be responsible for the infectious process. Clinically significant isolates should be identified to the species level. Moreover, we think that species identification should be systematically performed for isolates from patients at high risk of *Bacillus* infection, i.e., granulocytopenic patients with intravascular catheters and intravenous drug abusers (3, 8, 20, 22, 23). Species identification is difficult because of the great number of species, some of them ill defined, and the considerable variation between strains within each species. It has been greatly facilitated by the availability of miniaturized test strips (API 50CHB and API 20E) which, in conjunction with morphological observations, can identify up to 38 *Bacillus* species and subspecies (16). Our isolate, on the basis of its subterminal, ellipsoidal spore swelling the bacilli and its variable Gram staining, was assigned to morphological group 2 (24). The most frequently isolated species, *B. cereus*, *B. subtilis*, and *B. licheniformis*, belong to group 1, characterized by gram-positive, nonswollen bacilli. Differentiation from the closely related species *B. circulans* and *B. polymyxa* was achieved by the following tests (Table 1): utilization of D-arabinose and D-arabitol, growth at 50°C, and production of gas from carbohydrates. Non-anthrax *Bacillus* species are generally resistant to most β-lactam agents (3, 22, 25). However, there may be some variation among the different species (7). Our isolate was susceptible to all of the β-lactams tested. We found no previous reports of the susceptibility of this particular species to antibiotics.

Clostridia are anaerobic, spore-forming organisms that are ubiquitous in the environment. They can be found in brain abscesses following craniotomy (2), retention of foreign bodies (19), and lawn dart injuries (14). In the present case, the *Clostridium* isolate associated with the *Bacillus* strain could not be identified to the species level, since it was closely related to *C. beijerinckii* according to its sugar fermentation pattern and to *C. indolis* according to gas-liquid chromatographic analysis of its fermentation products.

Intracranial penetration should be considered in all periorbital puncture wounds. Patients should undergo careful radiological examination, especially computerized tomography scanning, to detect retained foreign bodies and, as a minute fragment could go undetected, follow-up monitoring with interval computerized tomography for early diagnosis of a brain abscess (10). Antibiotic therapy is recommended, even in the absence of signs of intracranial involvement (4), and should be directed not only against *S. aureus* but also against ubiquitous organisms such as *Bacillus* and *Clostridium* species. Microbiologists should be aware of the increasing medical importance of *Bacillus* species and be able to routinely identify them.

We thank J. P. Carlier and M. J. Butel for identification of the *Clostridium* strain and C. Gillet for secretarial assistance.

REFERENCES

1. **Acar, J., E. Bergogne-Bérézin, J. M. Brogard, Y. Chabbert, R. Cluzel, P. Courvalin, H. Dabernat, H. Drugeon, L. Dubreuil, J. Duval, J. P. Flandrois, J. Fleurette, F. Goldstein, M. Meyran, C. Morel, A. Philippon, J. Sirot, C. J. Soussy, A. Thabaut, and M. Veron.** 1993. Communiqué 1993 du Comité de l'Antibiogramme de la Société Française de Microbiologie. Pathol. Biol. **41:**741–748.
2. **Ariza, J., A. Casanova, P. Fernandez Viladrich, J. Linarez, R. Pallares, G. Rufi, R. Verdaguer, and F. Gudiol.** 1986. Etiological agent and primary source of infection in 42 cases of focal intracranial suppuration. J. Clin. Microbiol. **24:**899–902.
3. **Banerjee, C., C. I. Bustamante, R. Wharton, E. Talley, and J. C. Wade.** 1988. *Bacillus* infections in patients with cancer. Arch. Intern. Med. **148:**1769–1774.
4. **Bard, L. A., and W. H. Jarrett.** 1964. Intracranial complications of penetrating orbital injuries. Arch. Ophthalmol. **71:**332–343.
5. **Bursick, D. M., and R. G. Selker.** 1981. Intracranial pencil injuries. Surg. Neurol. **16:**427–431.
6. **Claus, D., and R. C. W. Berkeley.** 1986. Genus *Bacillus* Cohn 1872, p. 1105–1139. *In* P. H. A. Sneath, N. S. Mair, M. E. Sharpe, and J. G. Holt (ed.), Bergey's manual of systematic bacteriology. The Williams & Wilkins Co., Baltimore.
7. **Coonrod, J. D., P. J. Leadley, and T. C. Eickhoff.** 1971. Antibiotic suscepti-

bility of *Bacillus* species. J. Infect. Dis. **123:**102–105.
8. **Cotton, D. J., V. J. Gill, D. J. Marshall, J. Gress, M. Thaler, and P. A. Pizzo.** 1987. Clinical features and therapeutic interventions in 17 cases of *Bacillus* bacteremia in an immunosuppressed patient population. J. Clin. Microbiol. **25:**672–674.
9. **Farrar, W. E.** 1963. Serious infections due to "non-pathogenic" organisms of the genus Bacillus. Am. J. Med. **34:**134–141.
10. **Hansen, J. E., S. K. Gudeman, R. C. Holgate, and R. A. Saunders.** 1988. Penetrating intracranial wood wounds: clinical limitations of computerized tomography. J. Neurosurg. **68:**752–756.
11. **Ihde, D. C., and D. Armstrong.** 1973. Clinical spectrum of infection due to *Bacillus* species. Am. J. Med. **55:**839–845.
12. **Jenson, H. B., S. R. Levy, C. Duncan, and S. Mcintosh.** 1989. Treatment of multiple brain abscesses caused by *Bacillus cereus*. Pediatr. Infect. Dis. J. **8:**795–798.
13. **Jones, B. L., M. F. Hanson, and N. A. Logan.** 1992. Isolation of *Bacillus licheniformis* from a brain abscess following a penetrating orbital injury. J. Infect. **24:**103–104.
14. **Lew, J. F., B. L. Wiedermann, J. Sneed, J. Campos, and D. McCullough.** 1990. Aerotolerant *Clostridium tertium* brain abscess following a lawn dart injury. J. Clin. Microbiol. **28:**2127–2129.
15. **Logan, N. A.** 1988. *Bacillus* species of medical and veterinary importance. J. Med. Microbiol. **25:**157–165.
16. **Logan, N. A., and R. C. W. Berkeley.** 1984. Identification of *Bacillus* strains using the API system. J. Gen. Microbiol. **130:**1871–1882.
17. **Magot, M., J. P. Carlier, and M. R. Popoff.** 1983. Identification of *Clostridium butyricum* and *Clostridium beijerinckii* by gas-liquid chromatography and sugar fermentation: correlation with DNA homologies and electrophoretic patterns. J. Gen. Microbiol. **129:**2837–2845.
18. **Miller, C. F., J. S. Brodkey, and B. J. Colombi.** 1977. The danger of intracranial wood. Surg. Neurol. **7:**95–103.
19. **Pencek, T. L., and K. J. Burchiel.** 1986. Delayed brain abscess related to a retained foreign body with culture of *Clostridium bifermentans*. Case report. J. Neurosurg. **64:**813–815.
20. **Pennington, J. E., N. D. Gibbons, J. E. Strobeck, G. L. Simpson, and R. L. Myerowitz.** 1976. *Bacillus* species infection in patients with hematologic neoplasia. JAMA **235:**1473–1474.
21. **Schardinger, F.** 1905. *Bacillus macerans*, ein Aceton bildender Rottebacillus. Zentralbl. Bakteriol. Parasitenkd. Infectionskr. Hyg. Abt. II **14:**772–781.
22. **Sliman, R., S. Rehm, and D. M. Schlaes.** 1987. Serious infections caused by *Bacillus* species. Medicine **66:**218–223.
23. **Tuazon, C. U., H. W. Murray, C. Levy, M. N. Solny, J. A. Curtin, and J. N. Sheagren.** 1979. Serious infections from *Bacillus* species. JAMA **241:**1137–1140.
24. **Turnbull, P. C. B., and J. M. Kramer.** 1991. *Bacillus*, p. 296–303. *In* A. Balows, W. J. Hausler, Jr., K. L. Herrmann, H. D. Isenberg, and H. J. Shadomy (ed.), Manual of clinical microbiology, 5th ed. American Society for Microbiology, Washington, D.C.
25. **Weisse, M. E., J. W. Bass, R. V. Jarrett, and J. M. Vincent.** 1991. Nonanthrax *Bacillus* infections of the central nervous system. Pediatr. Infect. Dis. J. **10:**243–246.