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Pasteurella multocida Infection of the Calf in a Patient who had Moxa Cautery Treatment for Degenerative Arthritis

Yunsop Chong,* Hee Joo Lee,* Samuel Y. Lee,* Jun Seop Jahng and Kyu Hyun Yang****

Departments of Clinical Pathology and Orthopedic Surgery,** Yonsei University College of Medicine, Seoul, Korea*

Pasteurella multocida infection in man is rare. This organism was isolated from a calf infection of a 48-year-old woman with degenerative arthritis who had the history of treatments with prednisolone, acupuncture and moxa cauterization. She did not have any animal contact. It was considered probable that the orgasnism invaded through the cauterization ulcers. The organism was difficult to identify, because of its superficial resemblance to other organisms. Oxi/Ferm and N/F systems failed to identify the isolate. The organism was susceptible to many antimicrobial agents tested except to amikacin and clindamycin.

Key Words: P. multocida infection.

Pasteurella multocida is an aerobic gram-negative coccobacillary or rod shaped organism pathogenic to animals as well as to man (Weaver & Hollis, 1980). *P. multocida* infection in human is rare and includes three types, i. e., localized, pulmonary and systemic infections (Deboer & Dumler, 1963; Holloway *et al.*, 1969; Hubbert and Rosen, 1970a; Hubbert & Rosen 1970b; Nelsen & Hammer, 1981).

P. multocida exists as a commensal in the upper respiratory tract of various animals (Saphir & Carter, 1976; Billie *et al.*, 1978). Therefore most of the human infections are caused by animal contacts, either by a bite or some other way. But there are occasional cases in which no known history of animal contact can be found (Hubbert & Rosen, 1970b).

Recently we isolated a strain of *P. multocida* from the aspiration of a calf infection in a 48-year-old woman with degenerative arthritis of the knee who received various treatments including prednisolone, acupuncture and moxa cauterization. It was considered probable that the organism invaded the tissue through the cauterization ulcer. To our knowledge, there has been no report of such infection in Korea. Bacteriological findings and clinical observations are presented.

CASE REPORT

A 48-year-old woman (unit no. 1208919) was

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admitted to Yonsei Medical Center through the emergency room on the 28th of August 1981 with the complaints of painful swelling and limitation of motion of the left knee. During the last three years she received various treatments for her degenerative arthritis including aspirations of joint fluid and injections of prednisolone. Two months ago she received treatments by acupuncture and moxa cauterization and a severe inflammation developed in her left knee.

Physical examination on admission revealed a mentally alert obese woman. Marked swelling, local heat and tenderness of the left knee and leg were noted. Numerous cauterization scars and three ulcerations of 2 cm in size were noted on the left knee. Cephalexin and ampicillin were given for 4 days and an arthrotomy was performed on the 5th day. About 100 ml of yellowish green pus was removed from the joint space. Cartilages of the distal femur and proximal tibia were denuded. Pus drained from the ends of femur and tibia when they were curretted. These observations were consistent with acute pyogenic arthritis and osteomyelitis.

Fever persisted after the operation. Because of the swelling, local heat and pain of the left calf, incision and drainage were done on the 13th hospital day and about 800 ml of brownish pus was drained.

Laboratory findings included a WBC count of 16,400/μl on the day of admission. Increased WBC count of 22,800/μ1 was noted on the day of knee operation, which returned to an almost normal count of 10,000/μl three days after surgery. Bacterial cultures yielded various organisms. From the specimen of ulcer taken on the 4th hospital day, a heavy grwoth of *Staphylococcus aureus* and *Streptococcus pyogenes was obtained.* The specimen taken from inside of the knee joint on the 5th day yielded a moderate growth of *Pseudomonas aeruginosa* and a *Pseudomonas* species. The aspirated fluid of the calf taken on the 13th hospital day yielded a heavy growth of *P. multocida.* Following her operation, she was on dibekacin and cephaloridine, and no more *P. multocida* was isolated. But, later it was found that her wound was infected by *Enterobacter* this time.

MATERIALS AND METHODS

Specimens of pus aspirated from calf was inoculated onto blood agar and MacConkey agar plates and into thioglycollate medium. Blood agar was incubated in a CO_2 incubator. Cultural and biochemical characteristics of the isolate were tested in the conventional way. Acid production from carbohydrates was tested using cystine tryptic agar (Difco) with 1% carbohydrates and the final results were read after 72 hours incubation. API 20E (Analytab Products, Plainview, N.Y.), Oxi/Ferm (Roche Diagnostics, Nutley, N.J.) and N/F (Flow Laboratories, Roslyn, N.Y.) systems were also used to compare their identifications. Antimicrobial susceptibility was tested by the disk diffusion method(NCCLS, 1979).

RESULTS

On the gram-stained direct smear of calf pus, numerous pus cells and a moderate number of gram-negative bacteria were found. On the blood agar plate after 24 hours incubation a large number of nonhemolytic small greyish colonies of 0.5 to 1 mm in diameter were found. The colonies reached 1 to 2 mm in diameter after 48 hours incubation. The organism did not grow on MacConkey plate. The growth in thioglycollate medium was finely granular in appearance. The stained smear showed a gram-negative organism somewhat resembling

Table 1. Cultural and biochemical characteristics of *Pasteurella multocida* isolate

Characteristic	Result*
Hemolysis	−
Oxidase	+
Catalase	+
Motility	−
Indole	+
Methyl red	−
Voges-Proskauer	−
Citrate	−
Phenylalanine deaminase	−
Urease	−
Nitrate reduction	+
Gelatinase	−
Lysine decarboxylase	−
Ornithine decarboxylase	+
Arginine dihydrolase	−
Esculine hydrolysis	−
Deoxyribonuclease	−
β-D-galactosidase	−
MacConkey growth	−
42°C growth	+
TSI slant acid	+
butt acid	+
H_2S	−
Acid from	
Adonitol	−
Arabinose	−
Dulcitol	−
Fructose	+
Glucose	+
Inositol	−
Lactose	−
Maltose	−
Mannitol	+
Raffinose	−
Rhamnose	−
Salicin	−
Sorbitol	+
Sucrose	+
Trehalose	−
Xylose	−

* +, positive or growth; −, negative or no growth.

Table 2. Antimicrobial susceptibility of the *Pasteurella multocida* isolate

Antimicrobial agent	Susceptibility
Amikacin	R*
Ampicillin	S
Cefamandole	I
Cephalothin	S
Chloramphenicol	S
Clindamycin	R
Colistin	S
Dibekacin**	S
Erythromycin	S
Gentamicin	I
Kanamycin	I
Penicillin G	S
Tetracycline	S
Tobramycin	S
Co-trimoxazole	S

* S, susceptible; R, resistant; I, intermediate.

** Showa disk (Japan) method was used.

Neisseria, *Acinetobacter* or *Moraxella*. The isolate was nonmotile, oxidase positive, and grew at 25, 35 and 42°C. Other biochemical and cultural characteristics are shown in table 1. The isolate was susceptible to many antimicrobial agents including penicillin G, but was resistant to clindamycin and amikacin (Table 2).

DISCUSSION

P. multocida infection of man is known to be rare. According to Hubbert and Rosen (1970a; 1970b) there were only 194 cases of reported human infections upto 1965. The same authors reported 316 infections from 1965 to 1968. Although it is true that various types of human infection have been increasingly reported in recent years (Nelson & Hammer, 1981), the infection of *P. multocida* is still considered a rare one. At the Yonsei Medical Center, where over 30,000 specimens are processed anually,

this is our first isolation of *P. multocida.*

The most frequent type of infection due to this organism in man is known to be respiratory, either primary or secondary (Holloway *et al.*, 1969; Hubbert & Rosen, 1970; Schmidt *et al*, 1970; Itoh *et al.*, 1980; Nelson & Hammer, 1981). Infections of extremities are also frequent and are usually caused by animal contacts or bites (DeBoer & Dumler, 1963; Barth *et al.*, 1968; Bell *et al.*, 1969; Hawkins & Colorado, 1969; Hubbert & Rosen, 1970a; Griffin & Barber, 1975; Maurer *et al.*, 1975; Arvan & Goldberg, 1977; Spagnuolo, 1978; Gomez-reino *et al.*, 1980; Lucas & Bartlett, 1981). This organism has also caused infections of the central nervous system as well as septicemia (Itoh *et al.*, 1980; Stern *et al.*, 1981). Lately even nosocomial infections were reported (Itoh *et al.*, 1980). Most of the patients were reported to have various underlying conditions such as rheumatoid arthritis and steroid treatment. Seventy-five per cent of the patients unrelated to animal bite were older than 40 (Hubbert & Rosen, 1970b). Barth *et al.*, (1968) isolated this organism from the calf abscess of a 53-year-old woman who was on steroid therapy for 15 years because of her rheumatoid arthritis.

Our patient was a 48-year-old woman suffering from degenerative arthritis for three years and receieved prednisolone therapy. Two months before admission she received acupuncture and cauterization treatment which resulted in skin ulcers of the knee. It is most likely that *P. multocida* and other organisms invaded through these ulcers.

P. multocida is isolated from healthy animals. In some studies it was reported that 67% of the cats, 54% of the dogs and 14% of wild rats were carriers (Hubbert & Rosen, 1970a). Recent studies of dogs showed 6% (Billie *et al.*, 1978) to 12% (Saphir & Carter, 1976) were carriers. Infections of *P. multocida* usually follow animal bite or scratch. However, our patient did not have a history of animal bite or close contact, although she had a household dog.

Oberhofer (1981) divided *P. multocida* of human origin into 11 biotypes, i.e., A to K. Biotypes A and B were frequent isolates from cat associated infections. Dog associated infections did not show correlations with biotypes. Our isolate belonged to his biotype I.

P. multocida is a difficult organism to identify and can be mistaken for *Haemophilus, Neisseria* or *Acinetobacter* (Barth *et al.*, 1968; Lucas & Bartlett, 1981). There was no similarity of our isolate to *Haemophilus,* because of the larger colonies and positive oxidase. However we at first thought it might be *Neisseria* or *Moraxella* because of its characteristics such as no growth on MacConkey agar, the small size of colonies after 24 hours incubation on blood agar, positive oxidase, the odor similar to that of *Neisseria* and gram-negative cocci-like appearance. To rule out the possibility of being one of the glucose nonfermenting gram-negative rods, TSI and Motility indole ornithine medium were inoculated with the isolate. In these media the isolate gave positive results for glucose fermentation and indole production indicating that it was neither *Neisseria* nor a glucose nonfermenter.

After testing various cultural and biochemical characteristics it was possible to identify the organism as *P. multocida.* Even with the use of API 20E, or Oxi/Ferm system, known to be good methods for the correct identification of oxidase positive, fermentative organism such as *Aeromonas* and *Vibrio,* the identification of *P. multocida* were reported to be difficult. In one study (Collins *et al.*, 1981) it was reported that only 64% and 76% of *P. multocida* were correctly identified with API 20E and Oxi/Ferm respectively. In another study (Oberhofer, 1981) the reliabilities were reported to be 68% and 81%

each. It was our experience that API 20E correctly identified our isolate only when the inoculum was heavy enough, while Oxi/Ferm failed to identify it correctly. N/F system also failed to identify the isolate.

An interesting characteristic of this gram-negative organism is the susceptibility to many antimicrobals including penicillin G. Despite its *in vitro* susceptibility, this organism seems to survive in tissue even after a reasonable period of antimicrobial treatment. Barth *et al.*, (1968) reported recurrent infections of *P. multocida* in a rheumatoid arthritis patient when cephalothin administration for 25 days was stopped. Our isolate was resistant to amikacin and clindamycin. To other antimicrobials it was susceptible or intermediate. Our patient was on dibekacin for 9 days and on cephaloridin for 4 days before the isolation, but we were able to isolate the organism.

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