A case of nontraumatic gas gangrene in a common marmoset (Callithrix jacchus)

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ABSTRACT. The common marmoset is widely used in neuroscience and regenerative medicine research. However, information concerning common marmoset disorders, particularly infectious diseases, is scarce. Here, we report a case involving a female common marmoset that died suddenly due to gas gangrene. The animal presented with gaseous abdominal distention at postmortem, and Clostridium perfringens type A was isolated from several tissues. Vacuoles, a Gram-positive bacteremia and intravascular hemolysis were observed microscopically in the muscles, liver and lungs. On the basis of these findings, we diagnosed nontraumatic gas gangrene caused by Clostridium perfringens type A in this common marmoset.

KEY WORDS: Clostridium perfringens, common marmoset, gas gangrene


The common marmoset (Callithrix jacchus) is a small primate species that is widely used in neuroscience and regenerative medicine research. However, information concerning common marmoset disorders, particularly infectious diseases, is scarce. Here, we report a case involving a common marmoset that developed Clostridium perfringens infection and unexpectedly succumbed to gas gangrene.

At our institute, common marmosets are obtained from CLEA Japan, Inc. (Tokyo, Japan) or bred in our facility. The animals are housed in a single, pairs or families in stainless cages (82 × 61 × 158 cm), fed a commercial diet supplemented with honey and vitamins A, C, D3 and E and provided ad libitum access to tap water. The animal room is maintained at a temperature of 25–27°C, with a relative humidity of 40–60% and a 12-hr light–dark cycle.

A 4-year-old female common marmoset, weighing 365 g, died suddenly. It had not been used in any experiment for 6 months and showed no apparent external injuries. It had recovered from chronic watery diarrhea and associated weight loss 3 to 4 months before death and exhibited only facial erythema the day before death. At the time of discovery, the animal was believed to be dead for 6–12 hr. It showed uniform abdominal bloating with marked flatulence and unclear fluctuation (Fig. 1). The skin showed decreased turgor, with lightening of the color to light bronze (Fig. 1). The fever was 37°C, and the body weight was 365 g.

At necropsy, severe straining of the abdominal wall due to gas buildup throughout the intestine was observed (Fig. 1), accompanied by a foul smell resembling that of rotten eggs and viscous, blood-tinged ascitic fluid. The skeletal muscles were grayish-yellow and watery. Generalized hemorrhage was observed in the subcutaneous tissues. The liver and spleen were dark red in color, and enlarged and scattered dark red spots were present on the lungs.

Stamp-smear samples of the spleen, liver, lungs and blood and tracheal swabs were inoculated onto 5% horse blood agar plates for aerobic and anaerobic bacteriological culture and observed after incubation at 37°C for 48 hr. The stamp-smear samples were stained with Giemsa and Gram stains. Following a catalase test and Gram staining, individual colonies were identified using biochemical tests and a polymerase chain reaction (PCR) assay [20]. All bacterial colonies were anaerobically and purely isolated and were gram-positive, catalase-negative, lecithinase-positive, large, nonmotile, grayish to grayish-yellow and rough with irregular edges. The isolates were identified as C. perfringens using biochemical tests and a PCR assay with species-specific primer pairs. Genotypes of C. perfringens isolates were determined by PCR to amplify the genes encoding enterotoxin and alpha, beta, epsilon, iota and beta2 toxins [6]. All isolates showed only the alpha toxin gene and were accordingly classified as C. perfringens type A. Giemsa and Gram-stained stamp-smear samples of the spleen, liver, lungs and peripheral blood demonstrated numerous boxcar-shaped, gram-positive organisms and few leukocytes (Fig. 2).

Samples of the quadriceps femoris muscle, liver, pancreas, intestine and kidney were fixed in 20% neutral buffered formalin and embedded in paraffin. Sections of all samples...
Fig. 1. a) Postmortem findings in the abdominal and femoral regions. The abdominal skin is edematous and discolored with necrotic bullae. b) Necropsy findings in the abdominal cavity. Note the diffuse flatulence in the small and large intestines.

Fig. 2. Photomicrograph of a peripheral blood smear sample with Gram stain. Several boxcar-shaped, Gram-positive bacilli can be seen. Bar: 5 µm.

Fig. 3. Representative histopathological findings of gas gangrene in the skeletal muscle stained with HE (a, c) and Gram (b, d) stains. The lesions contain vacuoles and edema (a), Gram-positive bacilli accumulation (b) and myonecrosis (c) with invasion of the interstitial tissue by Gram-positive bacilli (d). Bars: 50 µm.

Fig. 4. Histopathological findings in the cecum stained with HE (a) and Gram (b) stains. Several Gram-positive bacilli are distributed in clusters and have invaded the dilated vessels in the lamina propria and submucosa. Bars: 50 µm.
C. perfringens causes gas gangrene. It has been reported that C. perfringens is most likely to invade the hepatic–cystic system [7], with the following mechanism suggested by experts. When the mucosal reticuloendothelial system is damaged, C. perfringens easily invades the hepatic portal vein and causes bacteremia. In the present case, it is speculated that C. perfringens entered the bloodstream via the damaged mucosal barrier in the large intestine, considering the absence of trauma, and multiplied rapidly in the vessels and submucosa, which provide an anaerobic environment. Then, it invaded the muscles and finally caused myonecrosis.

Several studies have reported C. perfringens infection as a cause of gastric dilatation syndrome in nonhuman primates [3, 5, 16], including one report involving a common marmoset [16]. However, the condition of the present case differs from the pathological condition of acute gastric dilatation syndrome. With regard to nonhuman primates, only one case of C. perfringens infection in a rhesus macaque (Macaca mulatta) has been reported to date [12], although the primate presented with necrotizing clostridial infection caused by skin wounds [12]. Therefore, the present case is the first, to the best of our knowledge, to report nontraumatic gas gangrene caused by C. perfringens type A in nonhuman primates. This should also attract the attention of humans engaged in breeding nonhuman primates with regard to appropriate sanitary managements.

REFERENCES

ASM press, Washington, DC.


