

Short Communication

The Absence of Anti-Rabies Antibody in the Sera of Feral Raccoons (*Procyon lotor*) Captured in Hokkaido, Japan

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SUMMARY: Feral raccoons captured in Hokkaido, Japan were examined for the presence of rabies virus neutralizing antibody (VNA). Between 2000 to 2002, 741 serum samples were collected and then subjected to VNA titer determination by rapid fluorescent focus inhibition test (RFFIT). Sera showing RFFIT titers of $\geq 1:25$ have been considered as positive according to previous reports. VNA was detected in none of serum samples from the feral raccoons. The present study provides valuable background information for rabies prevention in Japan. The potential risk of raccoon-derived rabies in the wilderness has been of concern because of the increasing number of feral raccoons originally introduced from rabies-endemic countries. The continuous monitoring of sick and/or dead wild raccoons would help to prevent an epidemic spread of raccoon rabies.

Since the 19th century, wildlife rabies in the United States (U.S.) has significantly increased due to changes in human demographics, animal translocation, ecologic change, and viral adaptation (1). After an outbreak on the West Virginia-Virginia border in 1977 due to the translocation of infected raccoons (*Procyon lotor*) from the southeastern U.S., the epizootic of raccoon rabies has spread through the mid-Atlantic and northeastern states of the U.S. and into Canada (2).

The common raccoon is widely distributed throughout regions ranging from Canada to Central America; this animal has also been introduced in Russia and Western Europe, as well as in a number of islands, including Japan (3). Since the 1970s in Japan, a large number of raccoons has been imported from the U.S. as pets, and raccoons released into the wilderness have multiplied rapidly as wild raccoons. In Hokkaido, the northernmost island of Japan, the intentional release and escape of pet raccoons over the last 20 years has led to a naturalized population, which is centered principally in the southern part of the Ishikari Plain (4).

As part of a feral raccoon management program carried out by the Hokkaido Government in 1999-2002, raccoons were caught using box traps (Havahart Model 1089, Woodstream, Lititz, Pa., USA, or handmade by hunters), and the animals were euthanized according to the method of Beaver et al. (5). Juveniles (young of the year) and adults were analyzed by examining tooth eruption and root foramina closure of the canines (4). The presence of virus neutralizing antibody (VNA) in the sera was determined by rapid fluorescent focus inhibition test (RFFIT) (6). Virus neutralization was conducted using sera showing RFFIT titers higher than 1:25, since nonspecific inactivation of virus infectivity was

reported when low dilutions of raccoon sera were examined for the presence of VNA (2,7).

In western-central Hokkaido, 741 serum samples were collected from 2000 to 2002 (Fig. 1). The number of samples collected in 2000, 2001, and 2002 were 248, 213, and 280, respectively (Table 1).

The RFFIT titers of all tested samples was lower than 1:12, i.e., VNA was detected in the sera of neither juvenile nor adult raccoons captured in western-central Hokkaido from 2000 to 2002 (Table 2). On the other hand, the RFFIT titers of serum sample from feral raccoons collected at 3 weeks after the first vaccination ranged between 163 and 328. As up to 20% of the feral raccoons inhabiting rabies endemic areas were shown to have circulating antibodies to rabies virus (2,7-9), our results suggest that a natural infection of feral raccoons has not yet occurred, and that rabies was not enzootic in Hokkaido before 2002.

Coyne et al. reported that the threshold raccoon density for rabies persistence, as estimated by the lowest density of raccoons in a rabies-endemic area in the U.S., was 3.0 heads/km² (10). Maesaki et al. reported that the recent density of feral raccoons in Hokkaido was 1.0-3.0 heads/km² (11). An increase in raccoon density of up to 3.0 heads/km² in Hokkaido would indicate that an epizootic spread of raccoon rabies might be likely in cases of accidental or illegal introduction of animals infected with rabies virus into the wilderness.

Since 1958 no rabies case has been reported in Japan, except for an imported case of a man bitten by a dog in Nepal in 1970. The rabies-free status in Japan has been extensively maintained by quarantining dogs imported from abroad and the annual vaccination of pet dogs. Cats, raccoons, skunks, and foxes have also been added to the quarantine list since 2000. However, the potential risk of raccoon rabies in Japan appears to be increasing due to the increase in the number of feral raccoons.

The strict prohibition of the introduction of raccoons into the wilderness, as well as the control of the raccoon population in general might be necessary to reduce the potential

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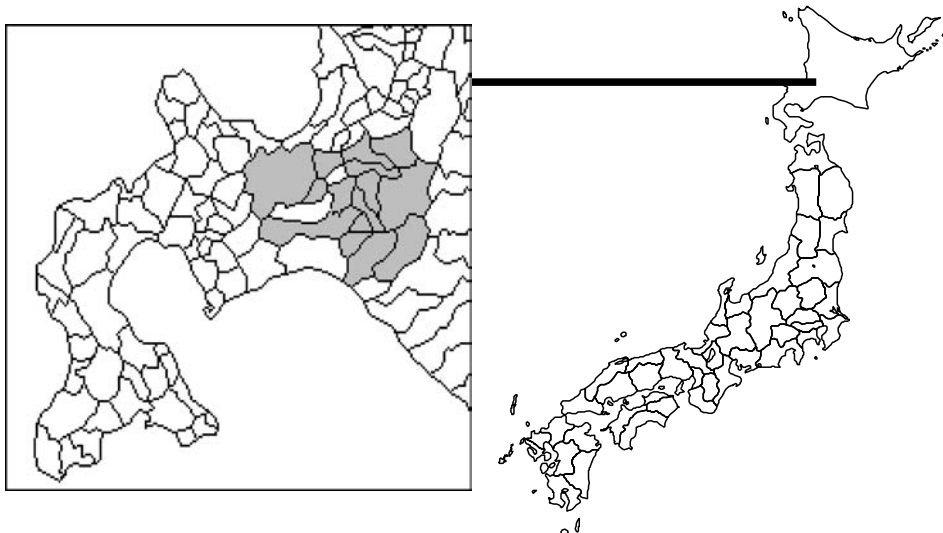


Fig. 1. Countries in which feral raccoons were trapped in Hokkaido from 2000 to 2002 (dark region). The study site included rural areas of Sapporo, Ebetu, Kitahiroshima, Iwamizawa, Kurisawa, Naganuma, Chitose, Mikasa, Yuubari, Kuriyama, Yuni, Oiwake, Hayakita, Atsuki, and Hobetsu. The raccoons were caught by using box traps (Havahart Model 1089, Woodstream, Lititz, Pa., USA) or traps handmade by hunters.

Table 1. The number of raccoons captured in west-central Hokkaido from 2000 to 2002

Year	Juvenile (%) ¹⁾	Adult (%)	Total (%)
2000	88 (35.5)	160 (64.5)	248 (100)
2001	92 (43.2)	121 (56.8)	213 (100)
2002	107 (38.2)	173 (61.8)	280 (100)

¹⁾: Juvenile were age of <1 year.

Table 2. VNA in sera of raccoons captured in west-central Hokkaido from 2000 to 2002

VNA	Year		
	2000	2001	2002
Positive ¹⁾	0 ²⁾	0	0
Negative	248	213	280

¹⁾: Positive: the RFFIT titer of ≥ 25 was considered significant.

²⁾: The number of raccoons.

risk of an outbreak of raccoon rabies in Japan.

Understanding the seroepidemiology of rabies among feral raccoons might offer valuable background data for future rabies prevention programs in Japan. As the anti-rabies antibody is not demonstrated in the sera of rabid animals until they show clinical symptoms, the continuous monitoring of sick or dead wild raccoons with a convenient rabies diagnostic test would also help to detect the possible invasion of raccoon rabies into Japan.

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