

***Mycobacterium bovis* Infection in a Herd of Japanese Shika Deer (*Cervus nippon*)**

Ryu ITOH, Yukio KAGABU, and Fumio ITOH

Akita Chuo Livestock Hygiene Service Center, 1-2 Shimoyabase, Yabase, Akita-shi 010, Japan

(Received 21 June 1991/Accepted 28 April 1992)

J. Vet. Med. Sci. 54(4): 803-804, 1992

KEY WORDS: deer, Japanese Shika deer, *Mycobacterium bovis*, tuberculosis.

It has been reported in recent years that about 50 dairy cattle are sacrificed as tuberculin reactors every year in Japan, and no typical tuberculous lesions have been observed in those animals both macroscopically and histologically. Although the outbreaks of tuberculosis in beef cattle have not been reported since 1963, there have been a few sporadic outbreaks in animals kept in zoological gardens [2, 4, 5, 7, 10].

We isolated *Mycobacterium bovis* (*M. bovis*) from a dead Japanese Shika deer reared in a zoological garden, and then carried out a survey on the dissemination of *M. bovis* infection in the herd. This is the first report on tuberculosis in deer due to *M. bovis* in Japan.

Seventeen Shika deer were kept in a 1,500 m² paddock and visitors were able to observe them from the outside of a fence. Two or 3 head of this group developed respiratory disorders like cough and gape in the autumn of 1984. One of them aged 18 months exhibiting severe symptoms died after anesthesia, and the animal was brought to our laboratory for detailed pathological examination.

Gross lesions were observed mainly in the lungs and pulmonary lymph nodes. The lungs were swollen with scattered grayish white lesions of 1-10 mm in diameter throughout the lobes (Fig. 1). Similar lesions were also observed in the pulmonary lymph nodes. The lungs, pulmonary lymph nodes, liver, spleen, kidneys and heart were fixed in 10% buffered formalin, and paraffin sections were stained routinely with haematoxyline eosin and Zeihl-Neelsen stains. Histopathological examinations revealed granulomatous lesions in the lungs and pulmonary lymph nodes, which were characterized by the presence of caseous necrotic foci or calcified foci surrounded by epithelioid granulomatous tissue, containing Langhans' giant cells and epithelioid cells (Fig. 2). Acid-fast bacilli stained with the Ziehl-Neelsen were observed in these cells. Based on these pathological findings, this case was diagnosed as tuberculosis. No remarkable lesions were noted in the other organs.

The lungs, liver, spleen, kidneys and heart which had been pretreated with 4% sodium hydroxide were inoculated onto 1% Ogawa's egg medium (Eiken Chemical Co., Ltd., Tokyo) and Dorset's egg medium (Nissui Pharmaceutical Co., Ltd., Tokyo) and then incubated at 37°C for 1 month. Acid-fast organisms isolated from the lung were found to grow only on Dorset's egg medium. This isolate was identified as *M. bovis* because of niacin negative, nitrate reduction negative, catalase (68°C, 20 min.) negative, Tween 80 hydrolysis negative, urease

production positive and arylsulfatase negative.

The survey on the dissemination of tuberculosis in this herd was carried out by subjecting the animals to the old tuberculin test (National Institute of Animal Health, Japan). This survey was performed 3 times a month at intervals. Animals with a positive or suspected reaction were sacrificed, and eventually all the deer were sacrificed. Table 1 shows the results of the tuberculin tests, the appearance of the gross lesions, histological lesions, and the isolation of mycobacteria. *M. bovis* was isolated from 6 deer showing a positive or suspected reaction to the tuberculin test except deer No. 4 with advanced calcifications of the lung. Microscopic examination revealed the presence of tuberculous lesions in the retropharyngeal lymph-nodes, even in the animals with the suspected reaction. Typical gross lesions were observed in 5 out of 7 with a positive or suspected tuberculin reaction except deer Nos. 1 and 7. It was considered that the tuberculous lesions appeared in the retropharyngeal lymph nodes as a primary focus of the infection. We may therefore reasonably conclude that the tuberculin test is an effective



Fig. 1. The lungs showing marked swelling and greyish white nodular lesions.



Fig. 2. Typical granulomatous lesions of tuberculosis in the lungs. Zeihl-Neelsen stains. $\times 72$.

Table 1. Results of tuberculin test, isolation of *Mycobacterium bovis* and appearance of tuberculous lesion

Tuberculin test	No. of deer	Gross lesion site	Histological changes	Isolation
Positives	1	—	RPLy	+
	2	Lu, Tra, RPLy	Lu, Tra, RPLy	+
	3	Lu, Tra, RPLy	Lu, Tra, RPLy	+
	4	Lu, RPLy	Lu, RPLy	—
Suspects	5	RPLy	RPLy	+
	6	RPLy	RPLy	+
	7	—	RPLy	+
Negative	8~16	—	—	—

RPLy: Retropharyngeal lymph node. Lu: Lung. Tra: Trachea.

method for the antemortem diagnosis of tuberculosis in deer.

In Europe and the U.S.A., a number of investigators have reported the outbreaks of tuberculosis in deer [1, 3, 6, 7]. In Japan, however, attention has been paid mainly to tuberculosis of cattle and swine. The results of this survey suggests, a possible transmission of tuberculosis from captive animals to man. Keeping deer as a domestic animal is becoming popular, and the number of stock farms where deer are reared has recently been increasing in Japan. This report which is the first case of isolation of *M. bovis* from tuberculosis in deer, issues a warning to zoological gardens and stock farms from the viewpoint of livestock hygiene. Strict surveillance should therefore be mounted on tuberculosis in animals kept zoological gardens and stock farms of deer.

REFERENCES

- Boever, W. J. 1978. pp. 779-815. In: Zoo and Wild Animal Medicine (Fowler, M. E. et al. eds.), W. B. Saunders Company, Philadelphia, London, Toronto.
- Hashikawa, H., Tamamura, F., Watanabe, T., Nakamura, A., Kashima, H., and Chiba, T. 1986. In: The 34th Meeting of Veterinarians and Zootechnicians of Japanese Association of Zoological Gardens and Aquarium (in Japanese).
- Hime, J. M., Keymer, I. F., Boughton, E., and Birn, K. J. 1971. *Vet. Rec.* 88: 616-617.
- Imaizumi, K., Endo, M., Asakawa, T., Takahashi, H., Ogawa, H., and Chiba, T. 1959. *J. Jpn. Vet. Med. Assoc.* 21: 13-14 (in Japanese).
- Itoh, R., Komatsu, M., Kakino, J., and Itoh, K. 1985. *J. Jpn. Poult. Dis.* 21: 75-80 (in Japanese).
- Jones, D. M. and Manton, V. J. A. 1976. *Vet. Rec.* 98: 525-526.
- Kobara, J. 1958. *J. Jpn. Assoc. Zoo. Aqua.* 2: 77-83 (in Japanese).
- Matthews, P. R. J., McDiarmid, A., and Collins, P. 1981. *Br. Vet. J.* 137: 60-66.
- Orr, M. B., Hunter, A. R., Brand, T., and Owen, D. 1978. *Vet. Rec.* 102: 484-485.
- Yugi, H., Hatakeyama, H., and Nemoto, H. 1963. *J. Jpn. Vet. Med. Assoc.* 16: 383-384 (in Japanese).