NOTE

A Serological Survey on Rotavirus Infection in Dogs by Immune Adherence Hemagglutination Test

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ABSTRACT. In serological survey by immune adherence hemagglutination test, 129 of 578 dog sera (22.3%) had titers of 1:8 or higher against bovine rotavirus, Lincoln strain. Of them, the positive rate of antibody in the family dogs was significantly higher than that in the stray dogs. Furthermore, the family dogs were shown to have obviously higher incidence of antibody against Wa strain of human rotavirus than Lincoln strain.—*Key words:* IAHA, rotavirus.

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It is already known that rotaviruses are important etiological agents of diarrheal disease in human as well as in various animal species throughout the world. However, the relationship of rotavirus infection between human and other animal species remains unknown. As a clue for clarifying this point, we have been particularly interested in the rotavirus infection in dogs, since dogs have close contact with humans, above all the family dogs with children. In addition, it is reported that pups experimentally infected with human rotavirus yielded large quantities of the virus without clinical symptom [13].

Although isolation of a rotavirus from dogs has been reported by England and Poston in 1980 [1] and subsequently by other investigators [2, 5], the epidemiological status of infection in dogs and the relation of the virus to human rotavirus are not clear. Furthermore, rotavirus has not been isolated from dogs as yet in Japan. In order to elucidate circumstances of canine rotavirus infection, the authors performed a serological survey in

dogs in Japan.

This paper deals with the distribution of antibody against rotavirus among dogs by means of immune adherence hemagglutination (IAHA) test. The IAHA test has become widely available to detect many viral antigens and antibodies because of its greater sensitivity and rapidity [3, 9, 10].

Serum samples used in this study were obtained from 578 dogs in Tokyo and Gifu areas during 1961 to 1981. Of them, 432 sera were collected from family dogs referred to veterinary hospitals or Gifu University Clinic for medical treatment, vaccination or temporary custody, and remaining 146 serum samples were collected at the municipal dog pound. These serum samples were stored at -20°C until examined.

The IAHA test was carried out according to Inouye *et al.* [7]. The antigen used was prepared from culture fluid of MDBK cells infected with the Lincoln strain of bovine rotavirus by the method of Takahashi *et al.* [12]. Moreover, Lincoln strain was employed for both neutralization test (NT) and CF assay.

Human rotavirus, Wa strain, propagated in

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MA-104 cells, was also used to prepare the IAHA antigen.

The incidence of antibodies against Lincoln strain in dogs is shown in Table 1. A titer of 1:8 or higher, considered positive, was detected in 129 (22.3%) of the 578 dogs. The positive rate of IAHA antibody obtained in the present study was nearly consistent with that (5/18) of neutralizing antibody to bovine rotavirus in dogs in Japan reported by Sato *et al.* in the first place [11]. The positive rate (25.7%) of antibody in the family dogs was significantly higher than that (12.3%) in the stray dogs. ($p < 0.01$)

To establish the reliability of the IAHA test in dog serum, 113 sera were tested simultaneously by NT and IAHA assay. The comparative results indicated complete agreement in 89 sera (78.8%)—either positive in both tests or negative in both. With 22 sera, however, the NT was positive but the IAHA was negative. This finding may be mainly attributable to the IgM antibody which could not be measured by the latter test system [7]. But in most of the IAHA-positive sera in this study,

the titers were higher than NT titers. On the other hand, the percentage of positive sera ($\geq 1:8$) by CF test is 9.6. Accordingly, compared with CF, which has been most widely used for epidemiological studies on the rotavirus, the IAHA is more sensitive.

The positive rates of IAHA antibody in dogs according to years of serum collection during a period of 1961 to 1981 varied from 15.4 to 29.9%. In particular, attention should be paid to the fact that 2 of 13 dog serum samples collected in 1961, when rotavirus had not been detected from any mammals yet, had antibody. It strongly suggests that rotavirus infiltrated among dogs in Japan already in 1961.

In the prevalence of IAHA antibody against Lincoln strain in dogs, no statistically significant difference was observed in relation to breed, sex and locality. In the present study, however, the positive rate of antibody tended to increase gradually with the advance of age. Similar patterns have been observed in human beings [4]. The continuous rise of positive rate in proportion to age suggests that inter-

Table 1. IAHA antibody titer to rotavirus^{a)} in dog sera

Category	No. of tested sera	No. of sera with antibody at indicated titer ^{b)}							≥ 8	%
		<8	8	16	32	64	128	256		
Family dogs	432	321	7	19	47	22	12	4	111	25.7
Stray dogs	146	128	2	2	6	4	0	4	18	12.3
Total	578	449	9	21	53	26	12	8	129	22.3

a) Lincoln strain.

b) Reciprocal of serum dilution.

Table 2. Detection of IAHA antibody to human and bovine rotaviruses in dogs

Antigen	Category	No. of tested sera	No. of positive (%) ^{a)}
Human (Wa)	Family dogs	193	73(37.8)
	Stray dogs	89	13(14.6)
Bovine (Lincoln)	Family dogs	193	54(28.0)
	Stray dogs	89	13(14.6)

a) $\geq 1:8$

mittent rotavirus infections may occur among dog population.

Table 2 summarizes the results of IAHA test with Lincoln and Wa strains of rotavirus in family and stray dogs. In the family dogs, which might be closely in contact with human being, the sero-positive rates against both antigens were significantly higher than those in the stray dogs. Moreover, it was shown that the family dogs had obviously higher incidence of antibody against Wa strain of human rotavirus than Lincoln strain.

Kapikian *et al* [8] reported by IAHA assay that some animal rotaviruses are closely related with human rotavirus subgroup 1.

In addition, it has been recently shown that canine rotavirus has antigenic relationship with certain another animal rotavirus including one of human rotavirus serotypes [6]. These findings including the present results seem to support that rotavirus may be capable of causing zoonotic disease. Certain evidence which rotavirus takes on an important part as a zoonotic disease-causing agent, however, has not been obtained so far.

Further investigations are needed to demonstrate the relationship of rotavirus infection between human and animals.

REFERENCES

1. England, J. J., and Poston, B. S. 1980. *Am. J. Vet. Res.* 41: 782-783.
2. Fulton, R. W., Johnson, C. A., Pearson, N. J., and Woode, G. N. 1981. *Am. J. Vet. Res.* 42: 841-843.
3. Gershon, A. A., Kalter, Z. G., and Steinberg, S. 1976. *Proc. Soc. Exp. Biol. Med.* 151: 762-765.
4. Ghose, L. H., Schnagl, R. D., and Holmes, I. H. 1978. *J. Clin. Microbiol.* 8: 268-276.
5. Hoshino, Y., Wyatt, R. G., Scott, F. W., and Appel, M. J. 1982. *Arch. Virol.* 72: 113-125.
6. Hoshino, Y., Wyatt, R. G., Greenberg, H. B., Kalica, A. R., Flores, J., and Kapikian, A. Z. 1983. *Infect. Immun.* 41: 169-173.
7. Inouye, S., Matsuno, S., and Kono, R. 1981. *J. Clin. Microbiol.* 14: 241-246.
8. Kapikian, A. Z., Cline, W. L., Greenberg, H. B., Wyatt, R. G., Kalica, A. R., Banks, C. E., James, H. D. Jr., Flores, J., and Chanock, R. M. 1981. *Infect. Immun.* 33: 415-425.
9. Lennette, E. T., and Lennette, D. A. 1978. *J. Clin. Microbiol.* 7: 282-285.
10. Matsuno, S., and Nagayoshi, S. 1978. *J. Clin. Microbiol.* 7: 310-311.
11. Sato, K., Inaba, Y., Shinozaki, T., and Matsumoto, M. 1981. *Vet. Microbiol.* 6: 259-261.
12. Takahashi, E., Inaba, Y., Kurogi, H., Sato, K., Satoda, K., and Omori, T. 1977. *Natl. Inst. Anim. Health, Q. (Jpn.)* 17: 33-38.
13. Tzipori, S., and Makin, T. 1978. *Vet. Microbiol.* 3: 55-63.

要 約

犬におけるロタウイルスの抗体調査 (短報): 杉山 誠・源 宣之・金城俊夫・橋本 晃¹⁾ (岐阜大学農学部獣医公衆衛生学教室, ¹⁾附属家畜病院)——免疫粘着赤血球凝集反応を用いて、犬におけるロタウイルスに対する抗体調査を行った。578頭中129頭 (22.3%) が1 : 8以上の抗体価を示した。そのうち、飼育犬の抗体陽性率は放浪犬より明らかに高く、しかも飼育犬では牛ロタウイルスよりヒトロタウイルスに対する抗体陽性率が明らかに高かった。