

PROPAGATION OF BOVINE ROTAVIRUS BY CATS AND DOGS

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Résumé

PROPAGATION DU ROTAVIRUS BOVIN PAR LE CHIEN ET LE CHAT. — Des chats gnotobiotés ont été infectés expérimentalement, par voie orale, à l'aide de rotavirus bovin. Ils ont excrété du rotavirus dans leurs matières fécales pendant au moins quinze jours après infection. La séroconversion observée après une semaine chez tous les animaux n'a suffi ni à interrompre l'excrétion virale, ni à prévenir la réexcrétion suite à une seconde inoculation avec une souche identique ou différente du rotavirus bovin.

Des chats et des chiots sentinelles placés pendant un certain temps dans la même cage que les animaux infectés ont également excrété du virus, mais n'ont pas fait de séroconversion.

Aucun des animaux n'a souffert de diarrhée au cours de l'expérience.

Le chat est donc capable d'assurer la multiplication du rotavirus bovin et ce virus est transmissible du chat au chat et du chat au chien. Le chat peut dès lors, comme le chien, intervenir dans la dissémination du rotavirus bovin.

Rotavirus infection is a very common event in different animal species, including calf and man, and is often associated with diarrhoea (Pastoret and Schoenaers, 1977; Flewett and Woode, 1978; Mc Nulty, 1978; Scherrer and Cohen, 1978).

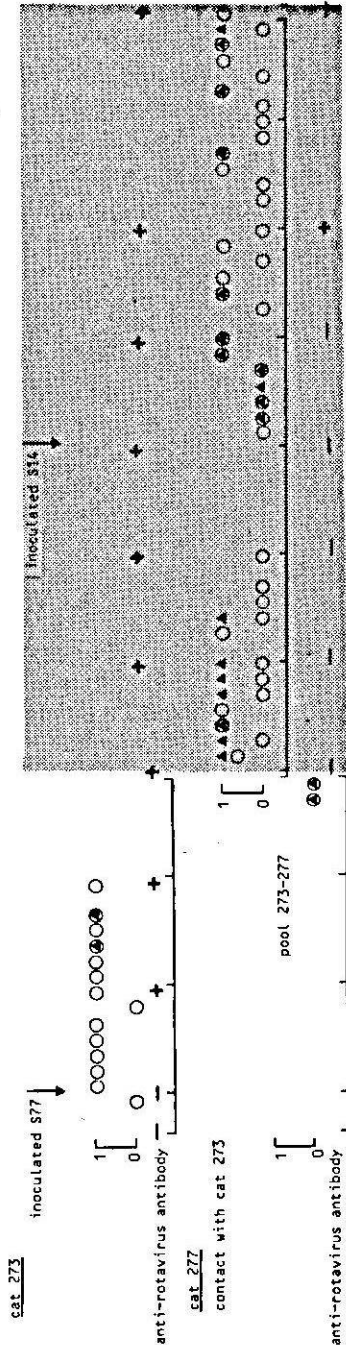
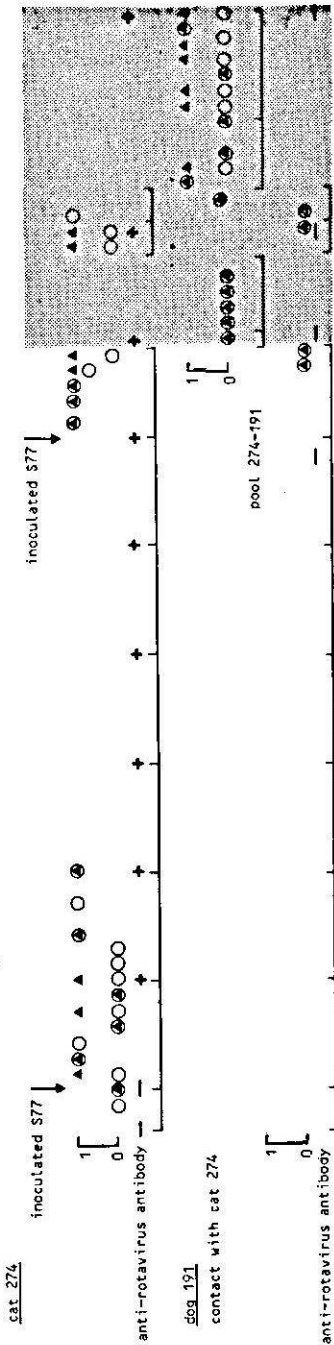
Anti-rotavirus antibodies have been demonstrated in cat sera in Ireland (Mc Nulty *et al.*, 1978), Scotland (Snodgrass *et al.*, 1979), Belgium (Dagenais *et al.*, 1980b) and USA (Hoshino *et al.*, 1981).

Rotavirus particles have been occasionally observed in stools of diarrhoeic kittens (Snodgrass *et al.*, 1979; Chrystie *et al.*, 1979), and a specific feline rotavirus, antigenically distinct from other known rotaviruses, has been recently isolated and characterized (Hoshino *et al.*, 1981), but seems apathogenic for cats.

As rotavirus from one species may often infect members of other species, cats are probably also able to multiply rotavirus from non-feline origin. Pigs (Woode and Bridger, 1975) and dogs

▲ Virus isolation; ○ counter immunoelectroosmophoresis examination : positive, 1; negative, 0
 anti-rotavirus antibodies in sera : positive, +; negative, -

▨ contact between two animals
 ↓ oral inoculation (10^8 pfu/2 ml of rotavirus S14 or S77)
 † accidental death



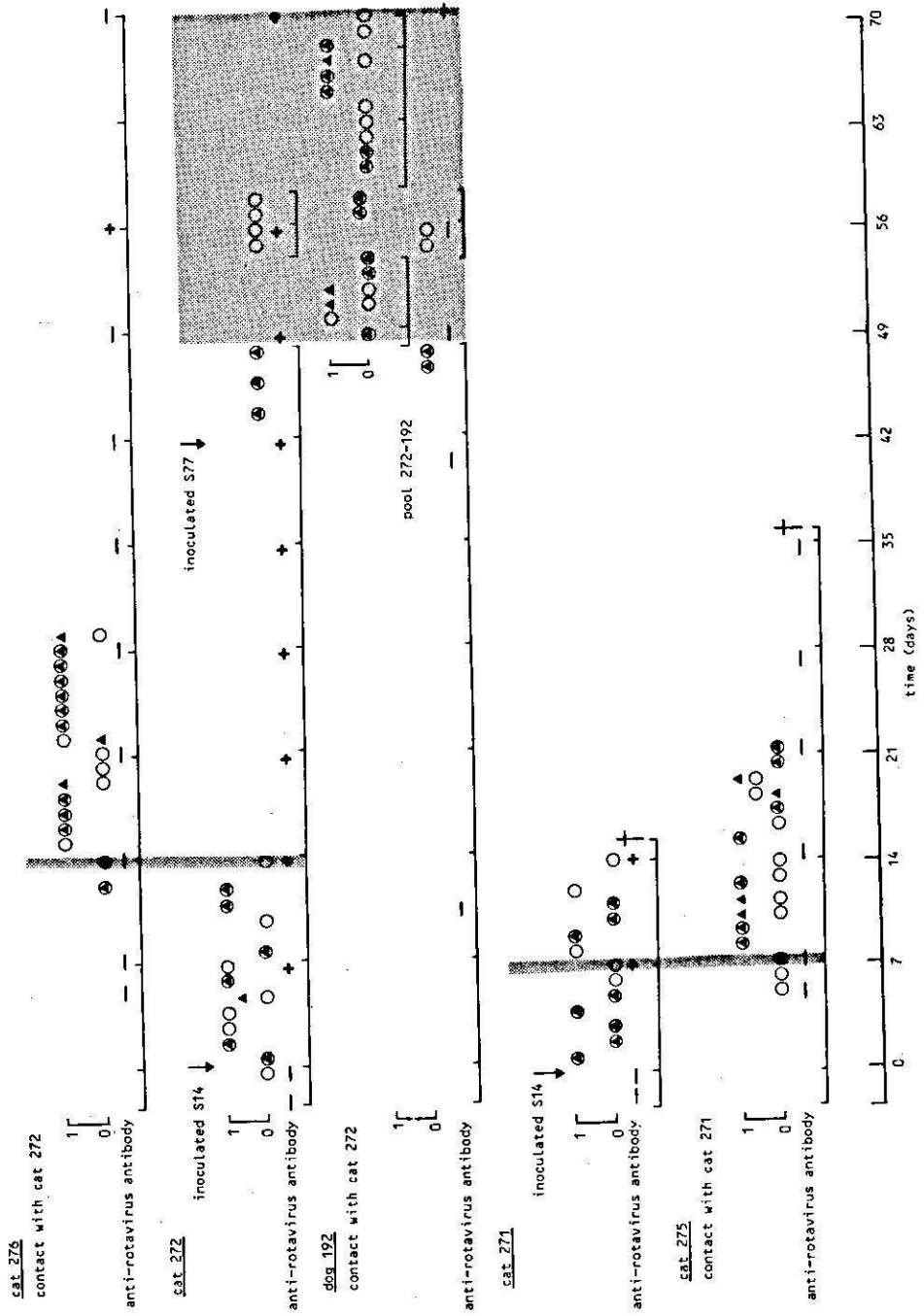


Fig. 1.— Experimental procedure and evolution of oral excretion by cats and dogs.

(Dagenais *et al.*, 1981b; Schwers *et al.*, 1983; Hoyois *et al.*, 1982) have already been shown to propagate bovine rotavirus and it was therefore interesting to know if the same was true for cats, and if cat to cat and cat to dog transmission of bovine rotavirus occurred.

Materials and Methods

Viruses and cell cultures

Rotavirus strains S14 and S77 were isolated from stools of diarrhoeic calves in our laboratory (Dagenais *et al.*, 1981a), following the method of Babiuk *et al.* (1977).

Rhesus monkey kidney cells MA 104, kindly provided by Prof. Bohl (Ohio, USA), were grown in Earle's minimum essential medium (MEM) supplemented with non-essential amino-acids, 0.85 µg/ml sodium bicarbonate, 10 % foetal bovine serum (FBS), 120 units/ml penicillin, 10 µg/ml streptomycin and 0.8 µg/ml natamycin (Pimarufin \oplus).

The same medium was used for virus production, but devoid of FBS and containing 2.5 µg/ml trypsin (Difco).

Virus titres were measured by plaque assay, using the methods described by Matsuno *et al.* (1977) and Dagenais *et al.* (1981a).

Experimental procedure

Seven SPF cats and two SPF beagle dogs, devoid of anti-rotavirus antibodies, were used in this experiment. The animals were kept in individual cages, in the same room of a controlled animal house.

Four cats were inoculated orally on day 0 and three of them on day 42, with 2 ml of a suspension containing 5×10^7 PFU (plaque forming units) /ml of rotavirus S14 or S77.

Two contact cats were placed for 24 hours in the same cage as one of the inoculated animals, respectively on days 7 and 14, and a third one from day 21 to day 70.

Two dogs were kept in the same cage as two reinoculated cats from day 49 to the end of the experiment.

The experimental procedure is described in figure 1.

Faeces were collected daily during 14 days after each inoculation or contact, or as long as the animals stayed in contact. Serum samples were taken weekly from day 0 to day 70.

Counter-immuno-electro-osmophoresis

The presence of rotavirus in stools was tested by counterimmuno-electro-osmophoresis (CIEOP), following the method described by Middleton *et al.* (1976) and currently used in our laboratory (Dagenais *et al.*, 1980a, b, c).

The sera were tested for the presence of anti-rotavirus antibodies using the same technique. Reference anti-rotavirus antiserum was prepared by hyperimmunisation of rabbits with purified bovine rotavirus (S14), as previously described (Schwers *et al.*, 1982).

Rotavirus antigen was produced and purified as previously described (Lansival *et al.*, 1981; Schwers *et al.*, 1983), excepted that it was concentrated by ultracentrifugation (100 000 g, 90 min) instead of ultrafiltration on PEG 6000.

Virus isolation

Virus isolation was performed as previously described (Babiuk *et al.*, 1977; Dagenais *et al.*, 1980d; Schwers *et al.*, 1983). The specificity of the cytopathogenic effect was controlled by CIEOP.

Results

The four inoculated animals excreted bovine rotavirus, as shown by CIEOP and virus isolation. Seroconversion was observed on day 7 for all of them. Viral excretion began one or two days after inoculation and persisted for two weeks, despite the presence of anti-rotavirus antibodies in their sera.

Contact cats became infected one or two days after contact; however, these animals did not seroconvert (a late and very transient production of anti-rotavirus antibodies was observed on day 56).

A second inoculation was followed by detectable viral excretion in two out of the three remaining animals.

Dogs kept in the same cage as the reinoculated cats became infected and excreted virus, showing that all the cats were contagious. No seroconversion was observed in those dogs. None of the animals in this experiment developed diarrhoea. The viral excretion pattern for all the animals is shown with more details in figure 1.

Cat 271 accidentally died on day 14 and cat 275 on day 36.

Positive isolates were always confirmed by CIEOP.

Discussion

All the inoculated animals excreted rotavirus. Cats are therefore able to multiply and to disseminate bovine rotavirus, as already shown for dogs (Dagenais *et al.*, 1981b).

Viral isolation on MA 104 cells seems to be a more sensitive method than CIEOP for the detection of rotavirus particles in cats stools (fig. 1).

Specific antibodies were detected in the sera of all the animals seven days after inoculation, contrary to what was observed for dogs, in which seroconversion is late and inconstant after infection with bovine rotavirus (Dagenais *et al.*, 1981b; Schwerts *et al.*, 1983).

The immune response is not sufficient to stop viral excretion, as infectious virus was present in faeces for at least one week after seroconversion.

A primary viral excretion followed by seroconversion is not sufficient to prevent further excretion of the same (274) or another (272, 273) viral isolate given later. Cat 272 did not excrete rotavirus at detectable amounts, but excreted enough to infect his contact dog (192).

A second infection was also followed by viral excretion in dogs (Schwerts *et al.*, 1983) and calves (Schwerts *et al.*, unpublished data).

Control cats kept in the same cage as the infected ones for 24 h or longer became infected and excreted virus for one to two weeks. The infection of a control cat (277) three weeks after inoculation of its contact may be due either to the fact that the inoculated animals excreted infectious rotavirus over a period of at least 21 days, or to the resistance of rotavirus in the environment.

None of these animals produced specific antibodies, notwithstanding prolonged contact

and viral excretion (277). A very transient peak of circulating antibodies was however-observed on day 56 in the two surviving contact animals. The dose of virus to which the animals were exposed might have been too weak to provoke a viral multiplication sufficient to induce a persistent antibody response, since the sensitivity of the CIEOP technique is rather low.

Dogs in contact with infected cats excreted rotavirus, but did not seroconvert. Seroconversion has already been shown to be inconstant after infection of dogs with bovine rotavirus (Dagenais *et al.*, 1981b; Schwerts *et al.*, 1983).

It is interesting to note that one dog (192) became infected in the absence of detectable virus in the faeces of its contact cat (272), showing that very low amounts of virus are sufficient to determine infection and viral multiplication and excretion in dogs.

Bovine rotavirus is thus transmissible between cats, and between cat and dog.

The infection is asymptomatic in both species. In natural conditions, on a farm, dogs may excrete bovine rotavirus (Hoyois *et al.*, 1982). The same is probably true for cats, and these two species may therefore play a significant role in the maintenance of this viral infection.

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Summary

SPF cats were experimentally infected orally with bovine rotavirus. All of them excreted virus over a period of at least two weeks after inoculation. Seroconversion was observed after one week for all the animals, but it did not stop viral excretion or prevent further excretion of the same or another rotavirus strain given later.

Cats or dogs kept in the same cage as inoculated animals became infected and excreted virus, but seroconversion was not observed in these contact animals. None of the animals developed diarrhoea during the experiment.

Cats are thus able to multiply bovine rotavirus, and transmission of this virus occurs between cats or between dogs and cats. Therefore, cats, like dogs, may play a role in the epizootiology of rotavirus infection in calves.

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