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A SEROLOGICAL SURVEY FOR SWINE VESICULAR DISEASE IN DENMARK

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SØRENSEN, K. J.: *A serological survey for swine vesicular disease in Denmark.* Acta vet. scand. 1980, *21*, 324—329. — Pig sera from Danish breeding centres were examined for occurrence of antibodies to swine vesicular disease virus using the counter immunoelectrophoresis test (CIET) and the serum neutralization test (SNT). Nine hundred and ten serum samples from 28 breeding centres were tested using the SNT and the CIET in parallel, whereas in a survey of 2052 samples from 218 herds the CIET was used for initial screening. CIET positive samples were subsequently assayed by the SNT. It was concluded that of the 2962 samples tested none had significant content of specific antibody to swine vesicular disease virus, although cross reacting antibody could be demonstrated in some cases.

S V D a n t i b o d y; p i g.

Swine vesicular disease (SVD) has so far never occurred in Denmark. The absence of the disease has been confirmed by serological screenings carried out in 1973 and 1977 (*Sørensen* 1973, 1977 a). In these screenings, each comprising about 600 swine sera, significant levels of specific antibody to SVD virus were not found. However, as also known from other countries (*Golding et al.* 1976, *Adair* 1976) low levels of cross-reacting antibodies may occur.

In the present work a larger number of swine sera were tested with a view to support earlier findings and in order to obtain further knowledge of the occurrence of cross-reacting antibodies among Danish pigs.

MATERIALS AND METHODS

Sera

A total of 2962 swine sera from Danish breeding centres were tested, 910 sera representing all animals in 28 breeding centre herds and 2052 sera representing about 20 % of the adult animals in 218 other breeding centres.

As reference serum a hyperimmune serum produced in rabbits was used. Three inoculations with purified virus (*Sørensen* 1977 b) emulsified in Freund's incomplete adjuvant were given at weekly intervals. Serum was collected 1 week after the last inoculation and stored at —20°C until use.

Techniques

The serum neutralization test (SNT) and the counter immunoelectrophoresis test (CIET) have been described elsewhere (*Sørensen* 1980).

CIET positive or doubtful positive samples were retested using a modification of the technique described by *Kohn* (1970): Two wells were punched next to each other (anode wells) and a third well (cathode well) positioned equidistantly from the 2 other wells. The serum in question and a rabbit hyperimmune SVD serum were placed in the 2 anode wells and allowed to diffuse at room temperature for 90 min before antigen was placed in the cathode well, and electrophoresis was performed.

RESULTS

The 910 sera representing all breeding animals in 28 herds were examined using CIET and SNT in parallel (Table 1). In the CIET 893 of the 910 sera were found negative, whereas 17 sera were positive. When these sera were retested using the modified Kohn's technique the precipitin lines coalesced indicating the reactions as antigen-antibody reactions (Fig. 1 a, b).

In the SNT 1 of the 17 sera had a titer of 38 while the rest had titers below 20. Of the 893 CIET negative samples 2 had SNT titers of 38 and 28, respectively, while the rest had titers below 20. In the screening of 2052 sera from 218 breeding centre herds the sera were initially tested using the CIET, and samples giving positive reaction were subsequently examined in the SNT

Table 1. Results of a serological survey for SVD antibody in Denmark.

	The sera examined using the CIET and the SNT in parallel	The sera initially examined using the CIET and positive samples subsequently examined in the SNT
Number of herds	28	218
Number of samples	910	2052
CIET positive	17	60
CIET positive with SNT titer > 20	1[1]	0
CIET negative with CIET titer > 20	2[2]	0

Samples with SNT titers > 20 were retested and those still giving SNT titers > 20 in the second test are tabulated.

[1] SNT titer 38.

[2] SNT titers 28 and 38, respectively.

(Table 1). Of the 2052 samples 60 were found positive in the CIET. However, all of these had SNT titers below 20. In total 2.6 % of the sera cross-reacted in the CIET.

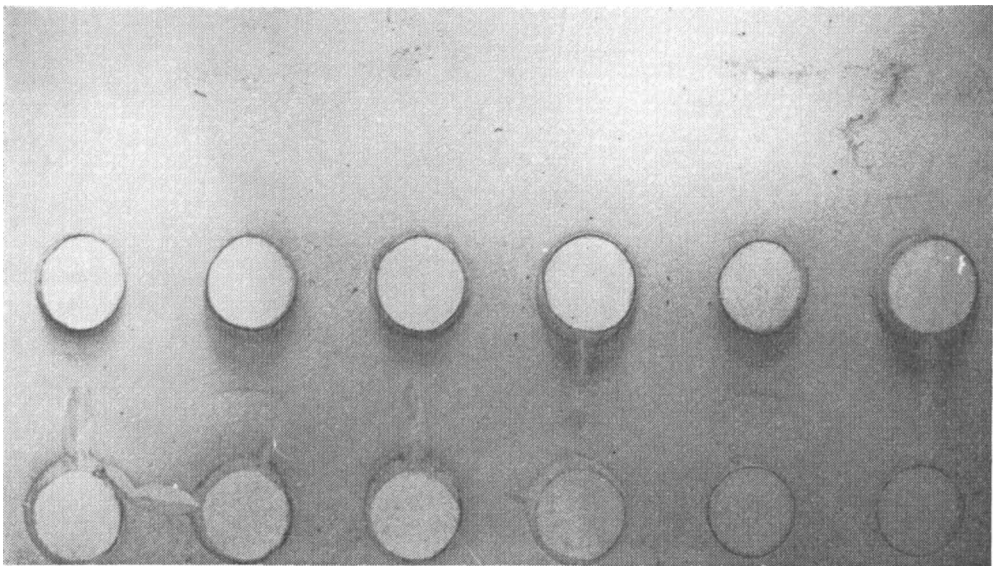

Figure 1a. Two examples of positive reaction with sera from breeding centres in the CIET.

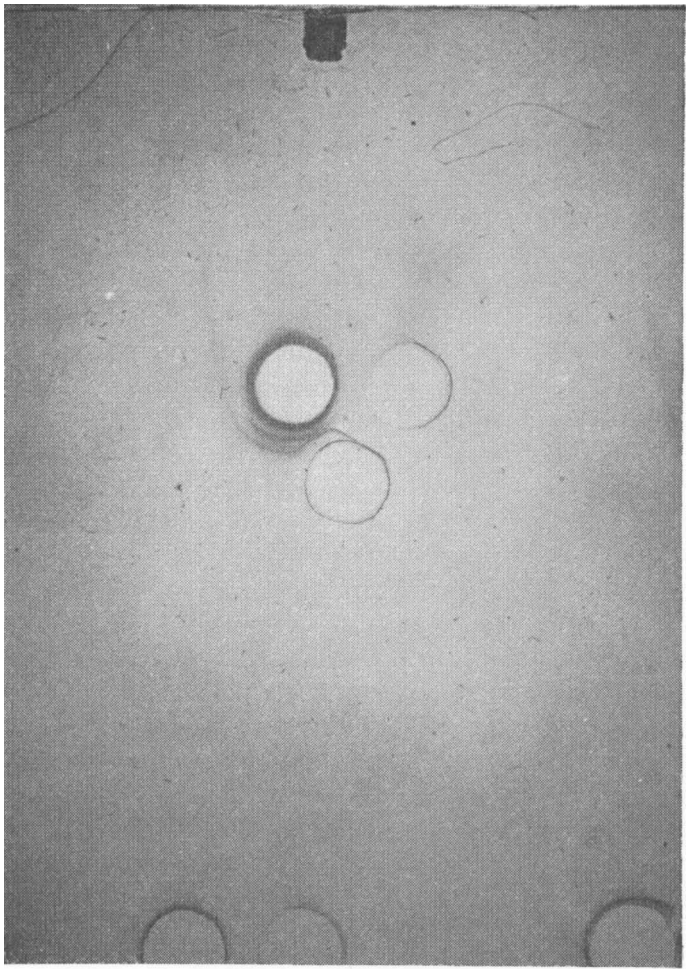

Figure 1 b. The precipitin line of a test sample (left) coalescing with the precipitin line of the rabbit hyper-immune anti SVDV serum (right) in the modified Kohn's technique.

DISCUSSION

Swine vesicular disease virus is an enterovirus and has been proposed to form a separate serogroup within the porcine enteroviruses (*Dunne* 1975). Cross-neutralization at low levels between serogroups of porcine enteroviruses is well known (*Dunne*). Likewise it is also possible to detect cross-reacting antibodies at

low levels against SVD virus in sera from pigs, which have never experienced infection with the virus (*Adair* 1976, *Golding et al.* 1976). Further the virus is neutralized by antiserum to coxsackie B5 viruses (*Graves* 1973).

The occurrence of cross-reacting antibody may complicate the interpretation of serological findings. *Watson & Hedger* (1974), *Golding et al.* and *Hendrie et al.* (1977, 1978) considered "final dilution" titers of 45 or greater in the SNT with supporting evidence from immunodiffusion tests as positive.

In the present work comprising a total of 2962 serum samples a positive CIET reaction was obtained with a total of 77 sera. Although these reactions were confirmed to be antigen-antibody reactions in the modified Kohn's test, only 1 had an SNT titer of 38 ("final dilution", titer 76), while the rest had titers below 20. Two samples which were negative in the CIET had SNT titers of 38 and 28, respectively. The 3 animals with SNT titers higher than 20 originated from 2 herds with 51 and 24 animals, respectively, all of which were tested. The testing of all animals in the herds gave a good opportunity to evaluate the specificity of the few positive reactions of low titers. If SVD had occurred, several animals in the herds would probably have been affected, and several serum samples should have reacted undisputably positive, with higher titers than those obtained. This was not found to be the case. The 3 SNT reactions were therefore interpreted as non-specific cross-reactions. Thus it was concluded that of a total of 2962 samples tested none had a significant content of specific antibody to SVDV, although cross-reacting antibody could be demonstrated in some cases.

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SAMMENDRAG

Serologiske undersøgelser for smitsom blæreudslæt (SVD) i Danmark. Svinesera fra danske avlscentre er undersøgt for forekomst af antistof mod SVD virus ved counter immunoelektroforesetest (CIET) og serum neutralisationstest (SNT). I den følgende undersøgelse af 2052 prøver anvendtes CIET i den indledende undersøgelse. Prøver, der reagerede ved CIET, blev derefter undersøgt ved SNT. Det kunne konkluderes, at i ialt 2962 undersøgte prøver kunne der ikke påvises specifikt antistof mod SVD virus, skønt krydsreagerende antistof kunne påvises i nogle tilfælde.

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