

## Kurze Mitteilungen / Short Communications

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### **Detection by Immunofluorescence of Transmissible Gastroenteritis (TGE) Viral Antigen in Pigs, Using Cat Anti Feline Infectious Peritonitis (FIP) Virus Conjugate**

By

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*With 2 figures*

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#### **Introduction**

An antigenic relationship between feline infectious peritonitis (FIP) and transmissible gastroenteritis (TGE) viruses, two members of the Coronaviridae family (1, 3, 8, 10, 11), has been reported recently by different workers (4, 9, 13). WITTE et al. (13) demonstrated TGE viral antigen in epithelial cells of the small intestine from an experimentally infected pig, using FITC-conjugated  $\gamma$ -globulin from a leopard which had died of FIP. Antibody titers to TGE in pigs hyperimmunized with TGE virus are remarkably low (7, 12), resulting in low working dilutions of FITC-conjugated immunoglobulins for diagnostic purposes; in contrast, antibody titers to FIP virus in sera and peritoneal fluids from FIP field cases are usually high (4, 6). In this paper the practical use of anti-FIP virus conjugate for the diagnosis of TGE in pigs is reported.

#### **Material and Methods**

Preparation of FITC conjugated globulins was performed as described previously (12). An anti-FIP virus conjugate was prepared from the peritoneal fluid of a cat which had suffered from a natural FIP infection as confirmed by post mortem examination. The anti-FIP virus antibody titer of this preparation, as determined by indirect immunofluorescence test (4) was 2560. The conjugate was absorbed with feline kidney monolayer cells, using standard procedures (2). An anti-TGE virus conjugate was prepared from the serum of a specified pathogen-free pig immunized with TGE virus. The neutralizing antibody titer of this serum when tested against 100 TCID<sub>50</sub> of TGE virus

was 256 (12). Cryostat sections were made from porcine intestine specimens, sampled from TGE suspected farms via the Provincial Animal Health Services in the Netherlands. The IFT was carried out as described previously (12).

To demonstrate the specificity of anti-FIP antibodies for the viral antigen an indirect IFT was performed on TGE viral antigen positive sections of the small intestine from a pig, using six sera and five peritoneal fluids from cats which had antibodies to FIP virus and five negative sera from an SPF breeding colony, respectively (5). These sera and peritoneal fluids were used in a 1 : 10 dilution; a FITC conjugated rabbit anti-cat IgG preparation served as second antibody.

Spleen sections from three cats with clinical FIP symptoms showing bright fluorescence with the anti-FIP virus conjugate, were used to determine whether fluorescence with an anti-TGE virus conjugate could be observed, in order to confirm the one-way antigenic relationship between TGE and FIP viruses, reported by WITTE *et al.* (13).

### Results

Sequential sections through intestine specimens from 218 pigs, collected from 120 TGE suspected farms were examined using anti-FIP and anti-TGE virus conjugates in parallel; 61 of the specimens examined (39 farms) contained TGE viral antigen as demonstrated with the anti-TGE virus conjugate. The same sections also showed fluorescence with the anti-FIP virus conjugate,

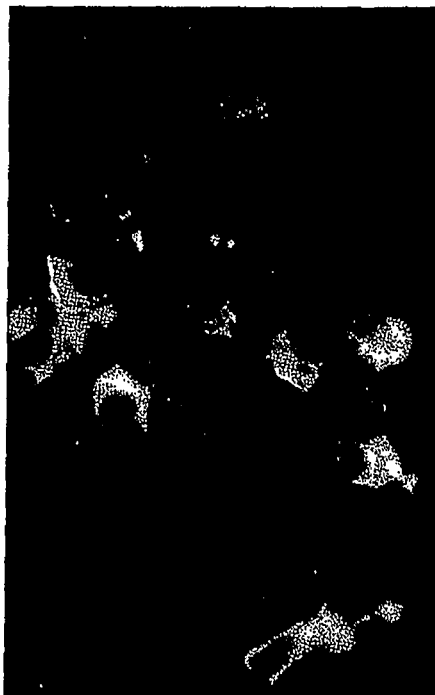


Fig. 1. Section through a porcine jejunum sample positive for TGE viral antigen; fluorescence is demonstrated using an anti-FIP virus conjugate

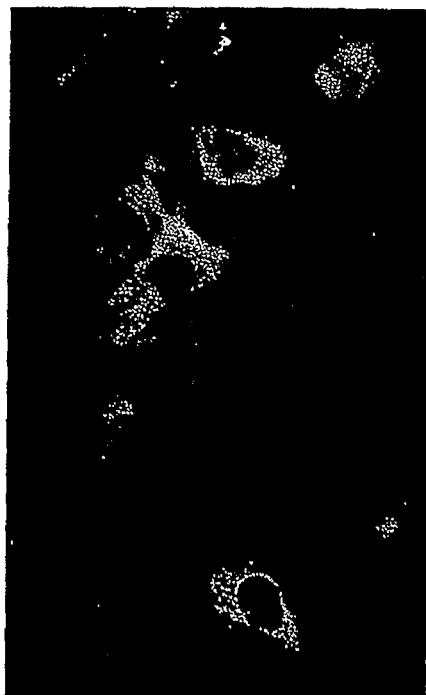


Fig. 2. Subsequent section from sample shown in Fig. 1 stained with the homologous anti-TGE virus conjugate

whereas the preparations negative in the homologous reaction were also negative in the heterologous test. Intensity and localization of the fluorescence with both conjugates in subsequent sections of the same preparations were comparable (Figs. 1 and 2). The working dilution for the anti-FIP virus conjugate (1:20) proved to be fourfold higher than the dilution for the anti-TGE virus conjugate (1:5).

Confirmation of the specificity of the anti-FIP fluorescence for TGE viral antigen was obtained by indirect IFT. The six cat sera and five peritoneal fluids with anti-FIP antibodies also gave positive results on TGE virus-positive sections; SPF feline sera consistently gave negative results also in the heterologous test. Sections from FIP positive cat spleens were examined using the anti-TGE virus conjugate with negative results.

#### Discussion

Identical results were obtained with an anti-TGE and an anti-FIP virus conjugate on sections of small intestines of a large number of pigs suspected of a TGE infection. Conjugates prepared from feline peritoneal fluids of clinical FIP cases consequently may replace anti-TGE virus conjugate for diagnosis of the porcine disease, although the influence of possible strain differences of FIP virus on the specificity of the antibodies in the feline peritoneal fluid cannot be excluded so far. In order to avoid non-specific reactions, only SPF pigs should be used to raise anti-TGE sera, which is a time-consuming and expensive procedure. Since antibodies to porcine viruses are unlikely to occur in cats, as shown by others (9), peritoneal fluids from FIP infected cats constitute good starting material for the preparation of conjugates for serological TGE diagnosis. FIP ascitic fluids can be obtained in large quantities from field cases and their homologous antibody titers tend to be very high (4, 6). Furthermore, our results confirm the antigenic relationship between TGE and FIP viruses (4, 9, 13); absence of fluorescence in FIP virus-positive spleen sections using an anti-TGE virus conjugate is taken as additional evidence for the one-way antigenic relationship reported by WITTE et al. (13).

#### Summary

Based on an antigenic relationship between TGE and FIP viruses, the use of FITC labelled anti-FIP virus preparations for the diagnosis of TGE in pigs was studied. Complete correlation between the results obtained with FITC conjugates directed against both viruses was demonstrated. The advantage of using an anti-FIP virus conjugate for the diagnosis of TGE is discussed. A one-way antigenic relationship between TGE and FIP virus was confirmed.

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#### Zusammenfassung

Verwendung eines Anti-Katzenperitonitis-(FIP)-Virus-Konjugates zur Immunfluoreszenzdiagnose der übertragbaren Gastroenteritis (TGE) der Schweine

Aufgrund der antigenen Verwandtschaft zwischen TGE- und FIP-Virus wurde die Verwendung eines FITC markierten Anti-FIP-Virus Konjugates

zur TGE-Virus Diagnose untersucht. Mit FITC-Konjugaten gegen beide Viren konnten übereinstimmende Ergebnisse erzielt werden. Diskutiert wird der Vorzug der Verwendung eines Anti-FIP-Virus Konjugates in der TGE-Virus Diagnostik. Die "one-way"-Antigenverwandtschaft zwischen TGE- und FIP-Virus wird mit diesen Ergebnissen bestätigt.

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