

ISOLATION AND ELECTRON MICROSCOPICAL EXAMINATION
 OF A PORCINE ROTAVIRUS-LIKE VIRUS
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Rotavirus has been demonstrated in feces from many species with diarrhoea. Until recently all isolates of rotavirus have been antigenically related through the group antigen. In 1980 Saif et al, and Bridger and in 1981 McNulthy et al found rotaviruses in pigs and chickens, but the antigenic relation to other rotaviruses could not be demonstrated.

In a routine survey for rotavirus from piglets with diarrhoea, in which the virus was detected both by EM and ELISA, rotavirus could be demonstrated in two feces samples by EM only. The two feces samples originated from piglets born by different gilts from the same herd. Rotavirus could not be detected in three other samples from the same herd neither by EM nor ELISA. The anamnestic information indicated that neonatal diarrhoea in piglets born by gilts had been a problem in the herd for about one month. The virus isolate from the two feces samples has now been passed three times in colostrum deprived piglets kept in isolation. The inoculation material was prepared by grinding a 20% feces suspension in medium. After low speed centrifugation the supernatant was treated with penicillin and streptomycin for three hours at room temperature. The piglets were inoculated intranasally, and they all developed diarrhoea 24 to 96 hours post infection. The piglets were killed at different intervals in this period. Feces samples from the field material and from the experimentally infected piglets were examined by EM and ELISA for content of rotavirus according to Askaa and Bloch 1981, but virus could only be found by EM. The partially purified feces samples were examined after negative staining with uranyl acetate. The virions appeared indistinguishable from other rotaviruses. However, in contrast to other rotavirus isolates the majority of the virions was complete - with both inner and outer capsid - and aggregated in groups, that could not be disintegrated by ultrasonic treatment. The same observations were made in the feces samples from the field materials.

A hyperimmune serum produced in rabbit by immunization with partially purified feces samples containing rotavirus-like virus was tested by indirect fluorescent antibody technique on cell cultures infected with bovine and porcine rotavirus without finding specific fluorescence. With this serum it was possible to demonstrate virus antigen on cryostat sections of the small intestine from piglets experimentally infected with the rotavirus-like virus. Especially enterocytes of the middle part of the villi and a few at the top were affected.

In one μ m thick toluidine blue stained sections of duodenum the cranial and caudal part of jejunum and ileum groups of pale cells could be found on the lower half part of the villi. By electron microscopical examination such groups of cells could be seen to form syncytia. The cells were swollen, and the cytoplasmic membrane between the enterocytes had often disappeared completely. However, interdigitating membrane fragments with des-

mosomes could be seen between the nucleic and tight junction complexes at the brush border were often present. The brush border of the affected cells was shortened and sometimes missing, and the difference between such cells and not affected cells was obvious. In the cytoplasm of affected cells inclusions of viroplasm containing virus could be seen. The viroplasm could be partly surrounded by the membrane of a dilated cisternae of the endoplasmatic reticulum, and virus particles could be seen to bud through the membrane and in this way obtain the outer capsid layer. In the cisternae incomplete and a few complete virus particles were found. Virus from the second piglet passage was adapted to secondary swine kidney cell cultures by treating the inoculation material with 100 μ g trypsin/ml for one hour at 37^oh and by adding 2 μ g trypsin/ml and 0,5% bovine serum albumin to the maintenance medium. The isolate has until now been passed three times in this manner, and a marked cytopathogenic effect has been observed on the cell cultures after 48 hours. The last isolate has been examined by EM, and the majority of the detected virions was found to be complete.

A rotavirus-like virus was isolated and grown on cell cultures. The existence of syncytia formation of the enterocytes at the lower part of the villi, the great relative abundance of complete virions in feces samples and in the cell culture harvest together with the lack of antigenic relation to other rotaviruses, all indicates a difference of the isolated virus from porcine rotavirus.

Selected references: Askaa, J. & Bloch, B. Acta Vet. Scand. 1981, 22, 32. Bridger, J. et al. Vet. Rec. 1980, 107, 532. McNulthy, M.S. et al. J. Gen. Virol. 1981, 55, 405. Saif, L.J. et al. J. Clin. Microbiol. 1980, 12, 105.