EFFECT OF STOMACH AND GUT JUICES ON INFECTIVITY OF LOW AND HIGH
PASSAGED STRAINS OF T.G.E. CORONAVIRUS: PROPERTIES OF A VIRUS
MUTANT RESISTANT TO INACTIVATION BY STOMACH JUICE OBTAINED BY
CYCLES OF SURVIVOR SELECTION IN TISSUE CULTURE

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The coronavirus causing transmissible gastroenteritis (T.G.E.) of swine is considered a major etiologic agent of the neonatal enteritis. Sows orally immunized with virus strains modified by serial passages in cell culture confer little or no protection to their suckling pigs (lactogenic immunity), in contrast to the protection seen in piglets milking sows given field virus strains. Improvement of immunisation methods of pregnant sows by oral route using live virus vaccine is the main objective of the present work. Induction of an efficient lactogenic immunity (passive protection of young piglets by milk) is strongly dependant of local antigenic stimulation of the maternal gut before farrowing. Stability of virus vaccine infectivity in digestive fluids (during the transit between mouth and gut epithelium) is a critical parameter for oral immunization of adult swine. Viruses, pig cell lines (RP.TG and S.T.) plaque assay are previously described (1). Virus is diluted 10-fold in i) stomach juice (18 samples or ii) gut juice (22 samples) collected from adult pigs or iii) in McIlvaine buffer (0.1 M, pH 2 and 3). The mixture is placed in stirring water bath at 37°C. T.G.E. virus is highly fragile in digestive fluids. No significant differences are observed between low passaged and high passaged strains. The amount of virucidal activity of digestive fluids is variable according the physiologic status of the digestive tract. These results suggest that virus infectivity of the vaccine dose given orally could be dramatically reduced during transit in stomach and gut lumen. This feature can partly explain the disappointing results of numerous attempts to immunize pregnant sows by oral route with live virus vaccine against T.G.E.

The "152-SG" virus mutant was selected in tissue culture after 152 cycles of survivor selection in stomach juice. This mutant is characterized by a high resistance to acidity (pH = 2.0) and by small plaques (0.5 mm) in a swine testis cell line (S.T.). In contrast, normal plaques (1.5 - 2.0 mm) are observed in RP.TG cell line. Other T.G.E. virus strains (L.P. and H.P. strains) show normal plaques both in S.T. and in RP.TG cells. Two litters of 4 days old S.P.F. piglets were inoculated by oral route with 4×10^6 and 4×10^7 P.F.U. of the "152-SG" mutant of T.G.E. virus. Mortality was nil and mild diarrhea was observed between 4th and 7th day post inoculation only in the litter given 4×10^7 P.F.U. An immune response (neutralizing antibodies) was also observed in serum and in secretions. These results suggest that the "152-SG" mutant is attenuated for the newborn S.P.F. piglet. Testing of immunogenic activity in the pregnant sow is in progress.

Table I

Stability of T.G.E. coronavirus infectivity in low pH buffer
(10 minutes/37°C in Mc.Ilvaine Buffer)

Virus strain	pH = 7.0	pH = 3.0	pH = 2.0
152 SG	4×10^6 *	2×10^6	1×10^6
Purdue 115	3×10^7	4×10^7	1×10^4

* P.F.U./0.3 ml

REFERENCES

1. H. LAUDE, J. GELFI and J.M. AYNAUD. In vitro properties of low and high passaged strains of transmissible gastroenteritis coronavirus of swine. Am. J. Vet. Res., 42:447 (1981).