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EXPERIMENTS ON AN EARLY PROTECTION AGAINST FOOT-AND-MOUTH DISEASE VIRUS

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Summary. - The influence of the peptide diacetylsplenopentin (SP5) on an early protection of guinea pigs against foot-and-mouth disease (FMD) was investigated. 80 % protection was achieved if SP5 was applied in a dose of 2 mg one day before challenge with FMD virus (FMDV) type O_1 Lausanne strain. In comparison with this a conventional commercial adsorbate vaccine protected guinea pigs about 7 days after vaccination. An earlier protection can be obtained in general by vaccination with a higher content of the immunogen. A tenfold increase of the 146 S particle dose in a conventional oil adjuvanted FMDV vaccine protected pigs about 2 days earlier as observed after inoculating a „normal" vaccine (about 11 days).

Key words: foot-and-mouth disease virus; synthetic peptide; early protection; guinea pig; swine

Since 1992 the member states of the European Community have ceased vaccinating their cattle population against FMDV. Therefore it is of special interest to dispose of a vaccine for ring vaccination in the case of an outbreak giving protection against this important virus disease as soon as possible. For this purpose concerning an inactivated vaccine there are three possibilities.

(1) *Use of a suitable adjuvant.* It has already been proved that aluminiumhydroxide is less suitable. So we did not succeed in achieving a high and early protection from types O_1 and A_5 in swine even when using a vaccine with strikingly increased antigen content. Using a suitable oil adjuvant protection is given 7-14 days after vaccination even with 1/10 of antigen content per immunizing dose (Nöckler *et al.*, 1990 *a, b*). According to Dudnikov (1991) universal vaccines against types O, A, C and Asia 1 respectively, containing oil adjuvants too, protect cattle, pigs and sheep as soon as 1-3 days after vaccination against homologous virus very efficiently.

(2) *Increase of antigen content.* By increasing antigen content not only a higher antibody titer in general but also an earlier protection can be achieved (see results). However, e. g. using a formol-adsorbate vaccine (146 S particle) with twofold antigen content an increase of titers of neutralizing

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anibodies by only 0.2 log units was detected (Pay and Hingley, 1987; Liebermann *et al.*, 1988). Thus the cost-benefit relationship and so the prime cost are of special interest both for the producer and for the used.

(3) *Use of special immune modulators.* As soon as in 1989 Forner *et al.* (1989) reported on a normalizing effect of splenine sequences, mainly of splenopentin (SP5), i. e., its acyl derivatives on the immune system. They stimulate the differentiation of T- and B-cells presumably in a direct manner. Amongst others they seem to be suitable for treatment of AIDS and other viral infections.

At first we studied the effect of SP5 (Ac-R-K(Ac)-E-V-Y) in guinea pigs by means of FMDV challenge. We used approximately 7 months-old guinea pigs (500–550 g) from our institute breeding. They were inoculated subcutaneously with a dose of 2 mg SP5 in phosphate buffered saline pH 7.4. One to ten days later they were challenged with more than 500 ID_{50} of guinea pig adapted O_1 Lausanne virus and observed for 10 days. Simultaneously a conventional formol adsorbate vaccine containing aluminiumhydroxide and saponine was tested. The results are summarized in Table 1. Here, the protection against generalization means that only primary vesicles were allowed to occur at inoculation site, i. e., on the rear left leg. From the table it becomes obvious that the highest rate of protection was already achieved 1–2 days after application of SP5. 7 days later, however, the protection rate ammounted only to about 20 % whereas in the case of standard vaccine it rised to 80 %. Only short time protection by SP5 occured, because this small peptide was probably quickly degraded.

These results provoke to apply SP5 and standard vaccine simultaneously. Such experiments have to be performed later on. It is neccessary to investigate the effect of different combinations of SP5 and oil adjuvanted vaccine on the beginning of protection.

As mentioned above, the increase of antigen content in a conventional vaccine might be an other way for an earlier protection. Fig. 1 shows the influence of an

Table 1. Protection effect of diacetylsplenopentin (SP 5) against FMDV challenge

	The day of virus challenge after inoculation of SP5 or vaccine	Protection against generalization protected / infected	%
SP5*	1	14/17	82
	3	6/11	55
	7	1/ 5	20
Standard vaccine (146 S antigen)**	1	10/21	48
	3	13/22	48
	7	4/ 5	80
Negative control		2/17	12

* Dose: 1 × 2 mg
** Dose: 1 × 1.5 μg

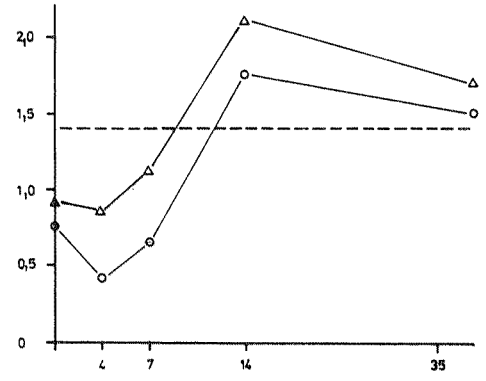

Fig. 1
Development of neutralizing antibodies in pigs after immunization with FMDV type O_1, inactivated by binary ethyleneimine
Adjuvant: oil-adjuvant.
Ordinate: titer in plaque neutralization test, measured as virus titre reduction in logarithmic units.
Abscissa: days post vaccination.

◁- -◁ : 20 μg of 146 S antigen per animal
O. . . . .O : 2 μg of 146 S antigen per animal
(------ : minimal value of a protective O_1 vaccine tested against virus type O_1

increased antigen dose on the time of onset of protection. Here, only the neutralization test was used because of correlation between antibody titer and protection of pigs (Thalmann and Nöckler, 1987). In the case of a tenfold increase of the 146 S antigen amount in an oil adjuvanted vaccine in pigs protection occurs only approximately 2 days earlier. The results of Dudnikov *et al.* (1991) and our results with SP5 illustrate, that there is obviously more than one possibility to obtain an early protection against FMD.

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