The airborne excretion by pigs of swine vesicular disease virus

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SUMMARY

The air of loose-boxes holding pigs affected with swine vesicular disease was sampled for virus. In the multistage impinger virus to a titre of $10^{2.6}$ TCID 50 was associated with particles greater than 6 μ m., $10^{1.6}$ with particles 3–6 μ m. and $10^{1.4}$ or less with particles less than 3 μ m. In the noses of workers in contact with the pigs for periods not less than 5 min., virus to a titre of $10^{2.4}$ TCID 50 was found. Virus was recovered from the air for 2–3 days during the disease and maximum titre in pigs infected by injection or by contact occurred on the second to third day after generalization of the lesions. The amounts of virus were about 160-fold less than those recovered from pigs affected with foot-and-mouth disease, and the quantity and time of excretion suggest that the source of swine vesicular disease virus in the aerosol may be from the lesions and skin rather than from the respiratory tract.

INTRODUCTION

Outbreaks of swine vesicular disease due to a virus have been described in Italy (Nardelli *et al.* 1968), Hong Kong (Mowat, Darbyshire & Huntley, 1972) and Great Britain (Dawe, Forman & Smale, 1973). At the time of the first outbreaks in Great Britain in December 1972 it was important to determine the method of spread of the disease and investigations were carried out to measure the amounts of swine vesicular disease virus present in the air of loose-boxes containing infected pigs. This paper records the results of two experiments, where pigs were infected by inoculation or by contact.

MATERIALS AND METHODS

Animals

Large white pigs, weight 30-40 kg., were housed in loose-boxes.

Virus

The strain of swine vesicular disease used came from a field case and was used either as the original field material (vesicular fluid and extract of vesicular epithelium – Dawe *et al.* 1973) or after one passage in pigs.

Infection of animals

In one experiment eight pigs housed in a loose-box were inoculated on the bulbs of the heel of both fore feet with virus fluid diluted 1/10 in phosphate buffered

Days	No. of animals affected*			Virus recovery		
after infection		Generalised	Sites affected†	Multistage impinger	Collector	Examiner
1	No pigs affected			PBo	No recovery	
2	6/8	3/8	12/40	$1 \cdot 8 \ddagger \leqslant 1 \cdot 2 \leqslant 1 \cdot 2 \leqslant 1 \cdot 2$	₹ 1.2	2·4§
3	7/8	7/8	22/40	$\ll 1.2 (a)$ $\ll 1.2 (b)$ $\ll 1.2 (c)$	< 1.2	<i>≷</i> 1·2
4	7/8	7/8	29/40	$\stackrel{1 \cdot 6}{<} 1 \cdot 2 \\1 \cdot 4$	<i>≷</i> 1·2	$\leqslant 1.2$
5	8/8	7/8	32/40	$\begin{array}{c} 2 \cdot 6 \\ 1 \cdot 6 \\ \leqslant 1 \cdot 2 \end{array}$	1.8	1·4 2·4
6	8/8	8/8	33/40	$\begin{array}{c} 1 \cdot 8 \\ \leqslant 1 \cdot 2 \\ \leqslant 1 \cdot 2 \end{array}$	1.6	2·0 1·6
7	8/8	8/8	34/40	$\leqslant 1 \cdot 2$ $\leqslant 1 \cdot 2$ $\leqslant 1 \cdot 2$	1.6	1·8 2·0

Table 1. Extent of lesions and recovery of virus from pigs inoculated intradermally with swine vesicular disease virus

* Numerator: number of animals affected or number of sites affected. Denominator: number of animals inoculated or number of sites available.

(a), (b), (c): top, middle and bottom stages of multistage impinger (> 6μ m., 3- 6μ m., < 3μ m.).

† Sites: 4 feet and/or legs. Snout, tongue and/or lips.

‡ Total virus (log TCID 50) recovered over 30 min. from multistage impinger.

§ Total virus (log TICD 50) recovered from nasal swab.

saline. In the other experiment four pigs were placed in contact with two pigs showing lesions of swine vesicular disease. Both groups of animals were observed daily and the extent of lesions recorded.

Air sampling

The air inlet and outlet in the box were blocked and the walls sprayed with water to maintain a high relative humidity. The air was sampled for 30 min. with a multistage impinger (May, 1966). In addition nasal swabs were taken from people operating the multistage impinger (collectors) and from those examining the animals (examiners) as previously described (Sellers, Donaldson & Herniman, 1970). The large volume sampler was also used initially, but it was not possible to disinfect it adequately after use owing to the resistance of swine vesicular disease virus to the cleaning process that could be used.

Days after exposure	No. of animals affected*	Sites affected†	Virus recovery, multistage impinger
1-3		No pigs affected	
4	3/4	5/20	$\leqslant 1.2 (a) \ \leqslant 1.2 (b) \ \leqslant 1.2 (c)$
5	4/4	15/20	1·4‡ 1·25 ≷ 1·2
6	4/4	16/20	2·4 1·25 ≪ 1·2
7	4/4	18/20	$ \leqslant 1 \cdot 2 \\ \leqslant 1 \cdot 2 \\ \leqslant 1 \cdot 2 $

Table 2. Extent of lesions and recovery of virus from pigs exposedto pigs suffering from swine vesicular disease

* Numerator: number of animals showing lesions or number of sites affected. Denominator: number of animals exposed or number of sites available.

† Sites: 4 feet and/or legs. Snout, tongue and/or lips.

(a), (b), (c): top, middle and bottom stages of multistage impinger (> 6 μ m., 3-6 μ m., < 3 μ m.).

‡ Total virus (log TCID 50) in stage of multistage impinger.

Virus assay

The virus present in the stages of the multistage impinger and in the nasal swabs (suspended in a volume of 10 ml.) was assayed in tissue-culture tubes of IB-RS-2 cells (de Castro, 1964). The results were expressed as tissue culture ID 50 per sample.

RESULTS

In the experiment where pigs were infected by inoculation, primary lesions were not observed until the second day, at which time some had ruptured and in three animals generalization had occurred. Further lesions appeared over the next 5 days involving the coronary band, the interdigital spaces, supernumerary digits, snout, lips and the skin over the hocks. Two peaks of virus recovery were found – on the second day at the time of development of primary lesions and on the fifth day 2-3 days after development of secondary lesions (Table 1). On average there was more virus in the nose of the examiners than in the nose of the collectors, although after the fifth day the difference was not great.

In the other experiment, when pigs were exposed to contact infection, the lesions first appeared on the fourth day, at one site on the feet in two animals, and at three sites in the third. On subsequent days lesions were found at sites on all feet and also on the lips and tongue. Maximum virus recovery was on the sixth day, 2 days after lesions were first observed (Table 2).

The greatest infectivity was associated with particles in the top stage (> $6 \mu m$.), a mean of 83% compared with a mean of 11% and 6% in the middle and bottom stages respectively.

DISCUSSION

The virus of swine vesicular disease was found to be present in the air surrounding affected pigs. The amounts of virus recovered were less than those found in loose-boxes containing pigs affected with foot-and-mouth disease by a factor of 160-fold or more (Sellers & Herniman, 1972) and virus was detectable for 2-3 days compared to 5 days for foot-and-mouth disease (Sellers & Parker, 1969). In swine vesicular disease the maximum amount was recovered 2-3 days after generalization had occurred; in contrast, in pigs affected with foot-and-mouth disease the maximum amount of virus was recovered at the time of generalization of the disease. These results suggest that the source of airborne virus may be different in the two diseases. In foot-and-mouth disease (Sellers, Herniman & Donaldson, 1971) it appears that the upper respiratory tract may be the source and in more recent work infection of the lung of pigs may be involved (Terpstra, 1972). Present investigations (Dawe et al. 1973; Burrows & Mann, personal communication, 1973) indicate that the virus in high titre is present in the vesicles and in the skin at the time of disease, and it might be that the main source of virus in swine vesicular disease is virus shed into the surroundings as the result of rupture of the lesions. That the source of airborne virus may be different between the two diseases is also reinforced by the finding that the partition of infectivity associated with particle size is different (foot-and-mouth disease - 63 %, 27 %, 10 % - Sellers & Herniman, 1972).

With the finding that smaller amounts of swine vesicular disease virus were excreted by infected pigs over a shorter period, spread by the airborne route would not be expected to the same extent as in foot-and-mouth disease. In investigations in the field during the present epidemic, most of the spread was attributed to movement of pigs or feeding of infected swill. Only 2 of the 103 outbreaks could be attributed to local spread. The large size of the particle associated with infectivity would require strong and turbulent winds to lift the infected particles and maintain them in the airborne state. In addition, pigs are not as efficient samplers of virus as are cattle exposed to foot-and-mouth disease virus and recent experiments have shown that a large dose is required to infect the pig by the nasal or oral routes (Burrows & Mann, personal communication, 1973).

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REFERENCES

MOWAT, G. N., DARBYSHIRE, J. H. & HUNTLEY, J. F. (1972). Differentiation of a vesicular disease of pigs in Hong Kong from foot-and-mouth disease. Veterinary Record 90, 618-21.

DAWE, P. S., FORMAN, A. J. & SMALE, C. J. (1973). A preliminary investigation of the swine vesicular disease epidemic in Britain. *Nature, London* 241, 540-2.

DE CASTRO, M. P. (1964). Behaviour of the foot-and-mouth disease virus in cell cultures: susceptibility of the IB-RS-2 cell line. Archivos do Instituto biológico, São Paulo 31, 63-78. MAY, K. R. (1966). Multistage liquid impinger. Bacteriological Reviews 30, 559-70.

- NARDELLI, L., LODETTI, E., GUALANDI, G. L., BURROWS, R., GOODRIDGE, D., BROWN, F. & CARTWRIGHT, B. (1968). A foot-and-mouth disease syndrome in pigs caused by an enterovirus. *Nature, London* **219**, 1275–6.
- SELLERS, R. F. & PARKER, J. (1969). Airborne excretion of foot-and-mouth disease virus. Journal of Hygiene 67, 671-7.
- SELLERS, R. F., DONALDSON, A. I. & HERNIMAN, K. A. J. (1970). Inhalation, persistence and dispersal of foot-and-mouth disease virus by man. *Journal of Hygiene* 68, 565-73.
- SELLERS, R. F., HERNIMAN, K. A. J. & DONALDSON, A. I. (1971). The effects of killing or removal of animals affected with foot-and-mouth disease on the amounts of airborne virus present in loose-boxes. *British Veterinary Journal* 127, 358-65.
- SELLERS, R. F. & HERNIMAN, K. A. J. (1972). The effects of spraying on the amounts of airborne foot-and-mouth disease virus present in loose-boxes. *Journal of Hygiene* 70, 551-6.
- TERPSTRA, C. (1972). Pathogenesis of foot-and-mouth disease in experimentally infected pigs. Bulletin. Office International des Épizooties 77, 859-74.