A significant increase in antibody titre against the *A/equi*-1 virus has been found in two horses by CF test and in seven by HAI test. Such results have been obtained only in animals which showed an increase of eightfold or greater in antibody titre against *A/equi*-2. This is more suggestive of a booster response to a common antigenic fraction in the *A/equi*-2 and in the *A/equi*-1 particles than of a recent or concomitant infection with the latter agent.

Our own unpublished investigations on horse sera collected in Switzerland in 1963 and 1964 show that at this time no animal had any antibody against influenza *A/equi*-2. It is therefore not surprising that the virus spread quickly and produced serious symptoms when introduced in such a receptive population.

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[1] Sovinova, O., Tumova, B., Pouska, F., and Nemec, J., *Acta Virol. (Prague)*, **2**, 52 (1958).
[2] Waddell, G. H., Teigland, M. B., and Sigel, M. M., *J. Amer. Vet. Med. Assoc.*, **143**, 587 (1963).
[3] Beveridge, W. I. B., Mahaffley, L. W., and Rose, M. A., *Vet. Rec.*, **77**, 57 (1965).
[4] Report of World Health Org. informal meeting on "Co-ordinated Study of Animal Influenzas" (Geneva, 1964).
[5] Lief, F. S., *J. Immunol.*, **92**, 286 (1964).
[6] Henle, W., Lief, F. S., and Fabiyi, A., *Lancet*, i, 818 (1958).
[7] Berlin, B. S., McQueen, J. L., Minuse, E., and Davenport, F. M., *Virology*, **21**, 665 (1963).
[8] Lief, F. S., and Henle, W., *Bull. World Health Org.*, **20**, 411 (1959).

Oncogenicity of Bovine Adenovirus Type 3 in Hamsters

THE prototype strain (*WBR* 1) of bovine adenovirus type 3 (ref. 1) is pathogenic for colostrum-deprived calves[2], and serological evidence indicates that, in addition to bovine adenovirus types 1 and 2, this serotype is involved in natural outbreaks of respiratory disease in cattle[3]. It has now been shown that the *WBR* 1 strain can induce the formation of tumours in hamsters, a capacity shared by certain other adenoviruses including human types 3, 7, 12, 18 and 31 (refs. 4–8), the chick embryo lethal orphan (CELO) virus[9] and a number of simian adenoviruses[10].

Nine litters of Syrian hamsters were inoculated by various routes on the day of birth with different doses of virus (strain *WBR* 1) grown in primary bovine kidney (BK) cultures and, in a number of these animals, tumours later developed at the site of inoculation. These tumours were either of a relatively firm consistency, resembling tumours induced by other adenoviruses or, as the virus dose was increased, were of a cystic type which appeared more rapidly (see Table 1). The cysts were largely filled with blood, although they also contained nodules of firm tumour tissue. Neither type of primary tumour was progressive; the firm tumours did not appear to grow after reaching a size of approximately 2–3 cm but remained static, whereas the cystic types regressed within 10 days leaving small subcutaneous nodules. Primary tumours of the firm type were transplanted in weanling and adult hamsters; in all these animals growth of the transplant tumours was progressive. Hamsters inoculated with uninfected BK cells remained healthy and free from tumours.

Table 1. INDUCTION OF PRIMARY TUMOURS IN HAMSTERS WITH BOVINE ADENOVIRUS TYPE 3 (STRAIN *WBR* 1)

Dose (log_{10})	Route	No. with tumours/ No. in litter inoculated*	Type of tumour	Latent period (days)
3·9	s/c	1/4	Firm	67
4·6	s/c	4/9	Firm	60–62
4·9	s/c	3/5	Firm (1)	53
			Cystic (2)	26
5·0	s/c	3/4	Cystic	24–26
5·2	s/c	1/6	Cystic	26
5·2	s/c	4/7	Cystic	24–27
4·0	i/c	2/3	Firm	34
4·3	i/t	2/3	Firm	47
4·6	i/p	2/4	Firm	55
		Total 22/45		

s/c, Subcutaneous. i/t, Intrathoracic. i/c, Intracerebral. i/p, Intraperitoneal.
* Survivors 7 days after inoculation.

On histological examination[11] of fixed tissues, the firm tumours were seen to comprise sheets of polygonal cells with numerous mitotic figures interspersed with multinucleate giant cells, some with ten or more nuclei. The general appearance suggested an undifferentiated sarcoma, but origin from epithelial tissue could not be excluded. In the cysts, the wall contained foci of tumour cells and the histological appearance of the nodules resembled that of the firmer tumours.

Monolayer cultures of primary or transplant tumours were comprised of cells resembling the types seen in sections. Such cells grown *in vitro* were capable of inducing progressive tumours when inoculated into adult hamsters. Attempts to demonstrate infective virus in tumours or their cultured cells have been unsuccessful to date.

Sera collected at various intervals after the appearance of tumours were used in complement fixation tests with 10 per cent extracts of tumour tissues or with tissue culture medium containing infective homologous virus. Complement-fixing antibody to viral antigen could be demonstrated in all hamsters, often in high titre, but could not be shown in significant amounts in those with transplant tumours. These findings may account for the stasis or regression of the primary tumours and for the progression of the transplant tumours. Complement-fixing antibody to tumour antigen was detected in low titre in one hamster with a primary tumour but not in any of the other hamsters. Complement fixation tests for the presence of polyoma or *SV*40 antibodies in the sera, or for antigens in the tumours, yielded negative results[12]. Neutralizing antibodies to bovine adenovirus type 3 were demonstrated in the sera of a number of the hamsters bearing primary tumours.

Bovine adenovirus types 1 and 2 were not found to be oncogenic for hamsters within an observational period of 90 days[7], whereas the present experiments indicate that bovine adenovirus type 3 is the first non-primate adenovirus, other than the avian CELO virus, which can induce tumours in hamsters.

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[1] Darbyshire, J. H., Dawson, P. S., Lamont, P. H., Ostler, D. C., and Pereira, H. G., *J. Comp. Path.*, **75**, 327 (1965).
[2] Darbyshire, J. H., Jennings, A. R., Dawson, P. S., Lamont, R. H., and Omar, A. R., *Res. Vet. Sci.*, **9**, 81 (1966).
[3] Darbyshire, J. H., Jennings, A. R., Omar, A. R., Dawson, P. S., and Lamont, P. H., *Nature*, **208**, 307 (1965).
[4] Huebner, R. J., Casey, M. J., Chanock, R. M., and Schell, K., *Proc. U.S. Nat. Acad. Sci.*, **54**, 381 (1965).
[5] Girardi, A. J., Hilleman, M. B., and Zwickey, R. E., *Proc. Soc. Exp. Biol. and Med.*, **115**, 1141 (1964).
[6] Trentin, J. J., Yabe, Y., and Gaylor, G., *Science*, **137**, 835 (1962).
[7] Huebner, R. J., Rowe, W. P., and Lane, W. T., *Proc. U.S. Nat. Acad. Sci.*, **48**, 2051 (1962).
[8] Pereira, M. S., Pereira, H. G., and Clarke, S. K. R., *Lancet*, i, 21 (1965).
[9] Sarma, P. S., Huebner, R. J., and Lane, W. T., *Science*, **149**, 1108 (1965).
[10] Hull, R. N., Johnson, I. S., Culbertson, C. G., Reiner, C. B., and Wright, H. F., *Science*, **150**, 1046 (1965).
[11] Chesterman, F. C. (personal communication).
[12] Pereira, H. G. (personal communication).

Fractionation of Partially Degraded Tobacco Mosaic Virus Ribonucleic Acid

IN the step-wise analysis of nucleotide sequence in tobacco mosaic virus ribonucleic acid (TMV–RNA)[1], occasional random splitting of internucleotide bonds results in the production of partially degraded molecules. These