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## STUDIES ON EQUINE INFECTIOUS ANEMIA VIRUS IN TISSUE CULTURE\*

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Only recently, has there been the likelihood that tissue culture systems may support the propagation of equine infectious anemia (EIA) virus.

To determine the susceptibility of tissue culture lines of horse cells to EIA virus, four lines of cells were derived from embryonic horse kidney (EHK-3 line), muscle (EHM-5 line), spleen (EHS-5 line) and lung (EHLu-6 line). However, not all these cell lines were susceptible to EIA virus.

While the author was undertaking the above experiments, KOBAYASHI reported the successful propagation of the EIA virus in cultures of horse leucocytes. Two strains of the EIA virus provided from KOBAYASHI were inoculated into horse kidney cells. No evidence of propagation of the virus was observed in the cells but unknown cytopathic (CP) agents were isolated.

In another experiment, EIA virus was inoculated into cultures of horse leucocytes, and the culture fluid from it was passed successively nine times through cultures of horse leucocytes. Then two ml of the resulting culture fluid was injected intravenously into a horse, which showed clinical symptoms characteristic of EIA. The findings suggested that the propagation of EIA virus is possible in cultures of horse leucocytes. However, CP effect accompanied by the propagation of EIA virus could not be clearly defined because of unknown CP-agents in the horse leucocytes.

Eight strains of unknown CP-agents were isolated from ten cultures of leucocytes all obtained from different horses. The characteristics of the CP-agents are as follows: (i) formation of type A intranuclear inclusion and syncytium in horse kidney cells; (ii) filtration through Chamberland L-3, but not Seitz EK pad or millipore filter (pore size 100 m $\mu$ ); (iii) sedimentation by centrifuging (27,000 g, 60 minutes); (iv) inactivation by heating (52°C, 30 minutes) or ether treatment (4°C, overnight); (v) non-propagation in HeLa cells or chick embryos; (vi) serological difference from equine abortion virus or Japanese B encephalitis virus; (vii) non-agglutination of chicken, chick and guinea pig red blood cells.

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\* The original report of this work will appear in this journal in the near future.