NOTE

Observation of Inclusion Bodies in Renal Epithelial Cells of Experimentally Infected Horses with African Horse-Sickness Virus

By:

A.R. Amjadi & P. Ahourai

INTRODUCTION

Since the introduction of African horse-sickness (AHS) in the middle east countries on 1958 (19,20) an extensive study has been carried out on the disease and various problem associated with its control.

During this period attempts were made to investigate the route of natural transmission of the virus (17), attenuation of the virus in mice (4), adaptation to tissue culture (1.1, 1.5). use of this new host system for vaccine production (6, 12, 13) and study of different properties of virus (14).

The aim of this contribution is to report the observation of intra-cytoplasmic inclusion bodies in renal epithelial cells of experimentally infected horses with African horse-sickness virus in order to confirm Fotherringham's finding (3) and to evaluate it in the diagnosis of the disease.

MATERIAL AND METHODS

The material was obtained from 4 horses which were experimentally infected with virulent AHS virus from the 4th passage of an Iranian strain, 10/60-MB4, (4th passage of the virulent virus in suckling mouse brain). The inoculum was contained 6.3 X 10⁶ LD_{so}/ml. of the virus.

Systematic necropsy procedures were performed immediately after death. Samples from heart, kidneys, spleen, lungs and lymphnodes were collected for virus isolation and histopathological studies.

RESULTS

Post mortem findings as well as virus isolation from internal organs confirmed that the actual cause of death was AHS in all tested animals. The pathological aspects revealed a mixed form of disease predominantly cardial type.

The gross lesion and histopathological changes serially showed the same stage as described in classical AHS by Maurer et al. (19).

The most characteristic changes were seen in the kidneys. The kidneys were moderately enlarged and covered with large amount of yellowish gelatinous edema fat. There was 1-2 cm wide band of severe congestion and hemorrhage in the cut surface of kidneys.

Microscopic section of kidneys showed non specific focal interstitial nephritis. Severe congestion, dilation of the vessels and intense interstitial hemorrhages were present in the medulla specially in the corticomedullarly junction. Besides these changes many inclusion bodies were found in the epithelial cells of the urinary tubules. These inclusion bodies were located in the cytoplasm rather than in the nucleus. They stained deeply with eosin (pink or bright red) by Haematoxilin and Eosin and dark red by Macciavello's staining methods.

The urinary tubules showed intense degeneration, cells being swollen and packed with eosinophilic inclusion bodies of various size. From one to several inclusion bodies were present in some cells. There were no visible inclusion body in other organs except kidneys.

Morphologically these inclusion bodies were round, sharply demarkated and surrounded with a distinct clear halo, standing out clearly from the rest of the cells and being readily detected under the microscope. Their size was smaller than a red corpuscle and up to the size of a single epithelial cell.

In order to make sure of the specificity of these inclusion bodies in histopathological diagnosis of AHS, the tissues from two healthy horses were also collected in the same condition and followed the same method of slide preparation as subject horses. The inclusion bodies were not found neither in the renal epithelial cells, nor in the tissues of the any other organs of these healthy horses.

CONCLUSION

In addition to virus isolation (1) several serological methods, namely neutralization test (2, 5), complement fixation test (9), hemaglutination (8), and gel diffusion precipitation test (7) have been widely used for the diagnosis of the AHS. These methods although valuable in the diagnosis of the disease, are somehow time consuming, and thus a quick but reliable method has been sought. The presence of inclusion bodies in the renal epithelial cells observed in experimentally infected cases of AHS may serve as an aid for a quick diagnosis of the disease.

SUMMARY

Eosinophilic inclusion bodies of various sizes were found in the cytoplasm of urinary tubule epithelial cells in experimentally infected cases of African horse-sickness. These inclusion bodies seem to be pathogonomic for the disease.

ACKNOWLEDGEMENT

The authors wish to thank Dr. M. Kaveh Director General and Dr. H. Mirchamsy Assistant Director of Razi Institute for their support and Drs. V. Sohrab, M. Baharsefat and A. Hazrati for their helpful advise and supervision.



Cytoplasmic inclusion bodies in renal epithelial cells.

REFERENCES

- 1— Alexander, R.A.: Studies on the neurotropic virus of AHS (1), Onderstepoort J. Vet. Sci. and Anim. Ind., 4, (1935): 291-322.
- 2— Alexander ,R.A. Studies on the neurotropic Virus of Horse sickness (3), Cnderstepoort J. Vet. and Anim. Ind.; 4, (1935): 349-378.
- 3— Fotheringham, A.: Preliminary note on the occurrence of inclusion like bodies in experimental and natural cases of AHS and the probable significance of their presence in relation to the diagnosis of the disease. J. Comp. Path. and Therap; 49, (1936) 268-273.
- 4— Hazrati, A. and Taslimi, H.: Study on AHS Virus strain isolated in Iran. Proc. XVII The World Vet. Congress; 1, (1963): 535-543.
- 5— Hazrati, A. and Czawa, Y.: Serological studies of AHS Virus with emphasis on neutralization test in tissue culture. Canad. J. Comp. Med. and Vet. Sci.; 29, (1965): 173-178.
- 6— Hazrati, A. and Ozawa, Y.: Monovalent Live Horse Sickness Vaccine. Bull. Off. Int. Epiz. 64, (1965): 59.
- 7— Huq, M.M. and Ansari, M.Y.: Gel precipitation test for the diagnosis of sout African horse sickness. Bull. Off. Int. Epiz.; 58, (1961): 691-698.
- Khorshed, B.M.: Complement fixation with horse sickness viruses. Nature, 189, (1961), 294.
- 9— McIntosh, B.M.: Complement Fixation with Horse Sickness Viruses. Onderstepoort. J. Vet. Rec. 27, (1965), 165-169.

- 10— Maurer, F.D., and McCully, R.M.: African horse-sickness with emphasis on Pathology. Am. J. Vet. Res. 24, (1963): 235-266.
- 11. Mirchamsy, H. and Taslimi, H.: Adaptation de virus de la pest equine a la culture des cellule. Acad. Sci. Paris: 255, (1962): 424.
- 12— Mirchamsy, H. and Taslimi, H.: Immunization against horse sickness with tissue culture adapted neurotropic viruses. Brit. Vet. J.; 120, (1964): 481.
- 13. Mirchamsy, H. and Taslimi, H.: attempt to vaccinate foals with living tissue culture adapted horse sickness virus; Bull. Off. Int. Epiz. 62, (1964), 911.
- 14 Mirchamsy, H. and Taslimi, H.: Visyalization of horse sickness virus by the fluorescent antibody technique. Immunology, 7, (1964): 213-216.
- 15. Ozawa, Y., Hazrati, A.: Growth of African horse-sickness virus in monkey kidney cell culture. Am. J. Vet. Res., 25, (1964), 505-511.
- 16— Ozawa, Y. Hazrati, A. and Erol, N.: African horse-sickness live virus tissue culture vaccine. Am. J. Vet. Res., 26, (1964), 154-169.
- 17— Ozawa, Y. Nakata, G.: Experimental Transmission Live Virus of African horse-sickness by means of mosquitoes. Am. J. Vet. Res., 26, (1965), 744-748.
- 18. Ozawa, Y.: Study on the replication of AHS Virus in two different cell line culture; Arch. Inst. Razi, 20, (1968), 125-128.
- 19_ Rafyi, A.: La pest equine; Arch. Inst. Razi; 13, (1961), 60-106.
- 20___ Reid, N.R.: African horse-sickness. Brit. Vet. J., 118, (1961), 137-142.