SWINE PLEUROPNEUMONIA PRODUCED BY HAEMOPHILUS PLEUROPNEUMONIAE: III. AN ELECTRON MICROSCOPIC STUDY

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The aim of this investigation was to study the ultrastructure of the early lesions in experimental induced pneumonia in pigs inoculated with Haemophilus pleuropneumoniae serotype 1 as a complement of a previous work with light microscopy in order to disclose the pathogenesis of the lesions observed.

Materials and Methods: Two pigs of conventional cross breed, 52 days old were used. They were free of Bordetella bronchiseptica and Haemophilus spp by nasal sampling and the lungs appeared normal at radiographic examination. The pigs were inoculated intranasally with 2 ml of a suspension of Haemophilus pleuropneumoniae serotype 1 containing $1,5 \times 10^8$ C.F.U. Both animals were observed several times daily and carefully examined once each day when their rectal temperature was recorded and radiological studies were performed. The pigs were killed by electrocaution and bleeded, one at 24 hours and the other at 48 hours post inoculation (P.I.). Samples were taken for bacteriology and histology. For electron microscopical examination areas of the lungs tissue were cut into pieces of approximately 1 mm^3, fixed with Karnovsky, post fixed in 1% OsO4 and embeded in Araldite. Ultrathin sections were stained with uranyl acetate and lead citrate and examined in a Philips electron microscope, E.M. 201.

Results: Pathological changes found in the ultrastructural investigation comprised all components of the alveoli. At 24 hours P.I. both types of epithelial cells showed degenerative changes characterized by different degrees of intracellular oedema, vesicles formation and detachment from the basement membrane. At 48 hours P.I. almost all type II pneumocytes were lacking dense llamelar bodies. In both types of epithelial cells regresive changes were more severe and necrotic cells were observed free in the alveoli. At 24 hours P.I. all capillaries were distended with packed erythrocytes, some of them in different stages of disintegration. Blood monocytes and occasional neutrophils were also observed. A few platelets were seen associated with disintegrated erythrocytes. Fibrin was not observed. At 24 hours P.I. the capillary endothelial cells were mostly without any changes while at 48 hours P.I. the endothelium as a general feature appeared swollen with many distended vesicles.

The alveolar septa were distended by oedema, fibrin and mononuclear cells. At 24 hours P.I. the alveolar spaces were distended with fluid, cellular debris, monocytes and few neutrophilic leucocytes. At 48 hours P.I. the lumen of most alveoli was collapsed. The content appeared the same as at 24 hours P.I. Few bacteria were observed free in the lumen or inside macrophages.

Discussion and Conclusions: The degenerative changes observed in the epithelial cells were similar to those reported in pneumonia produced by Salmonella cholera suis and also to those seen in connection with hipoxia, radiation injuries and haemorrhagic shock indicating a non specific cellular response. The packed erythrocytes within the alveolar capillaries appeared to be in different stages of disintegration. This material, identified as fibrin with light microscopy, was found consisted in erythrocytes debris with electron microscope. These findings are in many respects rather similar to the hyaline or fibrinoid thrombi found in the glomerular capillaries in association with Haemophilus infections and in swine salmonellosis, an experimental model of generalized Shwartzman reaction. The marked degenerative changes seen in the endothelial cells at 48 hours P.I. could be due to hypoxemia caused by the thrombus formation or by a combination of toxic, endotoxins and hypoxic effects and explain the exudative and infiltrative changes found in the alveolar septa and in the interstitial connective tissue. In this study most of the inflammatory cells were identified as macrophages, some with phagocited osmiophilic material or bacterias. In addition some neutrophilic leucocytes were observed.

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Acknowledgements: The authors whish to thank the following persons and institutions for their kind help: Dr. C. Gomez Dumm; Dr. G. Winqvist; the Postgraduate Department of the Veterinary Faculty of La Plata, Argentina, and the Cargill Foundation,Buenos Aires, Argentina.