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MALIGNANT LYMPHOMAS IN SHEEP

A thesis presented in partial fulfilment
of the requirements for the degree of
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Alastair Campbell Johnstone

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ABSTRACT

Malignant lymphoma is the general term applied to any neoplastic disorder of lymphoid tissue, including Hodgkin's disease and reticulum cell sarcoma. This group of neoplasms are among the most commonly occurring spontaneous neoplasms of sheep in New Zealand, being exceeded in frequency only by carcinomas of the small intestine and primary neoplasms of the liver. It has not been established whether malignant lymphoma is of sporadic or enzootic occurrence in this country, but from limited epidemiological observations in which on two occasions the disease was seen in two animals from the same property, it is possible that the latter distribution occurs. This and most other series indicate that mature sheep are most frequently affected but the disease does occur in younger sheep and lambs.

A study has been made of the pathology of ovine malignant lymphoma, based on 22 cases, most of which were collected from slaughter-houses and histological material from a further 18 cases filed previously at this laboratory. In nearly all cases nodular or diffuse lesions, consisting of accumulations of invasive neoplastic cells of lymphoid origin, were distributed widely throughout the body. Gross evidence of tumour in the lymph nodes was present in all except three cases of the disease. This involvement was usually multiple, with many of the carcass and visceral lymph nodes containing tumours. The spleen contained neoplastic lesions in 73 per cent. of the cases examined. Of the non-lymphoid organs affected by malignant lymphoma, the liver, kidney, bone marrow, heart, small intestine and abomasum predominated. Of the 40 cases examined, 37 were classified as lymphosarcomas and three as reticulum cell sarcomas. The degree of cellular differentiation in the lymphosarcomas varied from primitive lymphoblastoid to well differentiated lymphocytic cell types, with the less

differentiated forms being more common. Some nuclear and cytoplasmic abnormalities which have previously been described as being non-specific in other types of neoplasms were seen in specimens from 10 cases which were examined with an electron microscope.

Support for the hypothesis that "malignant lymphoma of sheep is transmissible" was sought by attempts to experimentally transmit this disease to lambs using intraperitoneal injections of cell-free tumour extracts during gestation or within 12 hr of birth. To date none of these sheep, which are only three years old, have developed overt neoplasia but 20 of them have developed elevated numbers of circulating lymphocytes. This has persisted for periods of two years or more and there is evidence to indicate that this should be interpreted as a preclinical phase of malignant lymphoma.

To investigate an hypothesis that malignant lymphoma of sheep is due to infection by an oncornavirus, electron microscopic examinations were made on specimens from fresh tumour as well as on preparations from tissue cultures which had been inoculated with various ovine lymphoid tumour homogenates. These studies were inconclusive and it was not until cultures of phytohaemagglutinin-stimulated lymphocytes derived from the sheep with experimentally transmitted lymphocytosis were examined that virus-like particles, consistent with the morphology of "type-A" oncornaviruses could be demonstrated. They were present in membrane-limited vacuoles and cytoplasm of these cells and less frequently extracellularly in the five experimentally inoculated sheep examined but were not present in any of the lymphocytes from an equal number of control sheep.

Lymphocyte cultures from 15 of the experimentally inoculated sheep showed sensitization to antigens in ovine malignant lymphoma homogenates when tested by a macrophage migration inhibition test. This test also

demonstrated the presence of common tumour specific antigens in five of the six tumour extracts used either for sheep inoculations or in testing for macrophage inhibitory factor production.

In an attempt to accelerate the development of overt neoplasia in three of the experimentally inoculated animals showing a persistent lymphocytosis, they were given a combined course of the immunosuppressive agents azathioprine and horse anti-sheep lymphocyte globulin. This resulted in a profound fall in circulating lymphocytes and while the cell mediated immunity was suppressed, as judged by the survival of skin allografts, macrophage migration inhibition in response to tumour antigens was not significantly altered in two of the three animals under treatment.

A close relationship was demonstrated in the experimentally inoculated sheep between the occurrence of lymphocytosis, the development of tumour specific cell mediated immunity and the presence of virus-like particles in phytohaemagglutinin-stimulated lymphocytes. This evidence combined with the demonstration of common apparently tumour specific antigens in malignant lymphoma extracts used in these studies provides strong support for the hypothesis "that this disease of sheep is transmissible and is most probably due to an infection with an oncogenic virus". The exploitation of the sheep model described is of potential value for the comparative study of preneoplastic events in the general field of cancer research.