The Expanding Spectrum of Animal Cancer Models

Bruce E. LeRoy, DVM, PhD, Diplomate ACVP (Clinical Pathology)
Department of Pathology and Comparative Oncology Group, University of Georgia,
Athens, GA (bleroy@vet.uga.edu)

Introduction

For many years, animals with cancer have been used as model systems for studying a wide variety of neoplasms that also occur in human beings. Animals with spontaneously-developing cancers that are prevalent in both humans and animals, such as lymphoma, bladder cancer, and melanoma, are important biological resources that serve to facilitate investigations regarding the pathogenesis and response to treatment of these important diseases. It is in this context that the role of the veterinary pathologist as a comparative biomedical researcher/clinician may be fully realized. From a historical perspective, the study of cancers in animals as model systems for human disease was heavily biased towards observations of naturally-occurring cancers in rodents. However, in recent years there has been a great increase in genetically-modified rodents that manifest a variety of neoplastic and pre-neoplastic conditions, as well as an increasing focus on companion animals with cancer as pre-clinical models to evaluate new chemotherapy agents. These changes have served to radically change the landscape of this field. Finally, as veterinary pathologists, we must constantly strive to balance our intellectual hunger for understanding these diseases with compassion for pet owners of pets with cancer.

Animal Models of Cancer—A Historical Perspective

Animal models used for studying cancer include:

- Neoplasms induced by xenobiotics
- Naturally-occurring neoplasms
- Transplantation of a primary neoplasm into an immunodeficient rodent (xenografts)
- Genetically-modified rodents

One of the earliest and most important contributions of an animal-based experiment to cancer research was the realization by Yamagiwa and Ichikawa that the application of coal tar to the ears of rabbits would result in malignant skin cancers\(^1\). This experiment lent strong support to the hypotheses of Pott and others that coal products were carcinogenic to humans, and was a crucial step in the development of the multistage model of carcinogenesis. At the time, model systems useful for identifying the contributions of individual hydrocarbons that humans working in the coal processing and petroleum industries were exposed to were not available. In 1930, Kennaway and others used this same model to demonstrate that specific polycyclic aromatic hydrocarbons such as dibenzanthracene would induce skin cancer\(^2\). The rabbit skin
carcinogenesis assay, while not a naturally-occurring cancer, provided an efficient animal-based system allowing the identification of compounds potentially carcinogenic to humans.

In 1910, Peyton Rous successfully transplanted pieces of a spontaneous sarcoma from a barred Plymouth Rock hen to chickens of the same breed. Subsequently, he found that inoculation of cell-free filtrates from whole-tissue extracts of this sarcoma resulted in the development of the same kind of tumor found on the original hen\(^3\). Later work showed that these “filterable agents” (as viruses were called) could also induce tumors in mammals. The molecular basis for this transforming activity was discovered in 1976, when Peter Duesberg and co-workers identified the nucleotide sequence responsible for viral transformation of cells as the \(v\)-src gene, one of the first proto-oncogenes\(^4\). This directly led to the realization that the protein product of this viral gene was a tyrosine kinase, and provided the key first step towards understanding the foundational role that phosphoproteins play in intracellular signal transduction. Thus, this animal model of cancer can be directly traced to the large number of phosphorylation-based signaling pathways that are dysregulated in virtually all cancers. For this work, Peyton Rous received the Nobel Prize in Physiology or Medicine in 1966.

The role of the pituitary and testis to cause alterations in the size of the accessory sex organs such as the prostate was noted in domestic animals by several investigators, with the first report being published in 1786\(^5\). However, Charles Huggins, an MD surgeon that had been charged with creating a urology service at the new University of Chicago Medical School, was the first to apply this principle as a medical therapy to men with prostatic diseases. While gaining experience abroad at the Lister Institute in London, Huggins studied the acid and alkaline phosphatases. Returning to Chicago, he developed a system to isolate prostatic secretions in normal dogs and dogs with prostatic hyperplasia. This animal model allowed him to monitor the changes in serum and seminal fluid phosphatase activity and prostatic cell morphology following castration, androgen replacement, and estrogen treatment\(^6\). He found that castration and estrogen administration resulted in:

- Shrinkage of the prostate gland
- A marked decrease in prostatic secretions
- A reduction in serum acid phosphatase levels
- Profound prostatic epithelial cell atrophy

Based on these results, Huggins began to treat men with prostatic hyperplasia or prostatic carcinoma with castration and estrogen treatment. The success of this treatment modality is with us today, as androgen-deprivation therapy is used in virtually every man with prostate cancer. Interestingly, for this work Huggins was awarded the Nobel Prize in Physiology or Medicine in 1966, along with another scientist he greatly admired, Peyton Rous.

Several large retrospective studies examining the incidence and tumor types of naturally-occurring cancers in domestic animals were undertaken in the 20\(^{th}\) century by
investigators such as Feldman, Jackson, and Mulligan. Specifically, a role for naturally-occurring tumors as models for investigating human cancers has been proposed for non-Hodgkin’s lymphoma, mammary carcinoma, prostatic carcinoma, melanoma, osteosarcoma, and urinary bladder [transitional cell] carcinoma.

Xenografted tissue has been used since the late 1960’s as a model system for investigating a variety of characteristics of many neoplasms. The first successful cancer transplantation occurred when pieces of a colonic adenocarcinoma from a man were inoculated and successfully propagated in nude mice. Since then, xenografts of virtually all human (and some animal) cancers have been developed—a recent PubMed search using “xenograft” and “cancer” returned over 11,000 articles! These models have been used to evaluate numerous properties of cancer cells, such as cell proliferation & death, local invasion, angiogenesis, and the development of distant metastases of the implanted cancer cells. Xenografts are useful for predicting the response to treatment for a variety of chemotherapeutics and neoplasms, and have played a vital role as assay systems for in vivo preclinical development of anticancer drugs. While there certainly are limitations of model systems utilizing immunodeficient mice, xenografts have made and continue to provide many valuable contributions to cancer biomedical research and treatments.

Evolution and Refinement—Models of Prostate cancer and Metastasis

Currently, genetically-modified mouse models of prostate cancer include transgene and gene-inactivation based systems. Depending upon the model, the prostate glands may exhibit prostatic intraepithelial neoplasia (PIN), adenocarcinoma, or both. Metastasis is not common in these mice, unless the transgene contains simian virus 40 large-T and/or small-t tumor antigens. The use of gene expression studies has identified several genes implicated in human prostate cancer (Nkx3.1, Pim-1, and prostate stem-cell antigen) that are differentially expressed in some of these models.

Animal based model systems have been extremely useful for understanding the steps that cells from a primary neoplasm must take to establish productive metastases at distant sites in the body. One of the earliest studies describes metastasis development following the injection of rat and mouse neoplasms intra-venously, subcutaneously, and into the bone marrow and joint spaces. Currently, only a few naturally-occurring animal cancers result in bone metastases, but dogs with prostate cancer often develop bone metastases, commonly with a marked amount of new bone formation. Most transgenic mice models have a low incidence of metastasis, but cell lines derived from these tumors can be selected in vivo for increased incidence of bone metastasis. A unique model system involving severe combined immunodeficient (SCID) mice implanted with human fetal bone fragments demonstrated the ability of prostate cancer cells to specifically home to bone and other organs.
Animal Models of Cancer: The UGA Experience

Work at our institution involves:

- Dogs with prostate cancer as models for the human disease
- Use of the inhibitor of apoptosis protein survivin as a prognostic factor for canine lymphoma
- A xenograft model of feline vaccine-associated sarcomas
- Molecular abnormalities in ocular neoplasms of domestic animals
- Transcription factor studies in squamous cell carcinoma of domestic animals

Future Directions for Animal Cancer Models

Validation of models is and will continue to be a monumental challenge for veterinary pathologists. Currently, validation is defined as the process of delineating the attributes (characteristics) of an experimental system that accurately match the attributes (characteristics) of the human disease. Validation involves a synthesis of data sets collected at the molecular, physiological, and morphological levels, and it is in this capacity that pathologists will continue to function as the “final arbiter” of the relevancy of the model system. In addition to morphologic assessment of model characteristics, data from molecular studies such as comparative functional genomics will likely aid pathologists in providing accurate “validity” assessments of models.

Future directions for many of the xenograft models include enhanced quantification of tumor burden using bioluminescent imaging (BLI) of luciferase/GFP transfected cells. Additionally, novel approaches utilizing animal models to evaluate the role of the stroma in the development of cancer and the progression of metastasis are also being developed.

Finally, one of the most exciting aspects of animal models of cancer involves the National Cancer Institute’s Center for Cancer Research and its recent development of a Comparative Oncology Program (CCR-COP). Under the guidance of Dr. Chand Khanna, a veterinary oncologist, the CCR-COP is developing a partnership between the NCI and academic institutions and the pharmaceutical industry to utilize pet animals with cancer (specifically those with prostate cancer, non-Hodgkin’s lymphoma, or osteosarcoma) to participate in preclinical trials.
References:


