ANIMAL MODEL OF HUMAN DISEASE

Hypercalcemia of Malignancy

Animal Model: VX-2 Carcinoma of Rabbits

Contributed by: D. M. Young, DVM, PhD, Comparative Pathology Section, and J. M. Ward, DVM, PhD, Tumor Pathology Section, National Cancer Institute, Bethesda, MD 20014; and David J. Prieur, DVM, PhD, Department of Veterinary Microbiology and Pathology, Washington State University, Pullman, WA 99164.

Biologic Features

Hypercalcemia is a frequent and serious complication of many types of neoplastic disease.[1] Although the majority of cases of hypercalcemia of malignancy are associated with an occurrence of osteolytic skeletal metastasis, a significant number of hypercalcemic patients have no demonstrable neoplastic involvement of bone. Therefore, it has been postulated that osteotropic humoral factor(s) secreted by neoplastic cells are of etiologic importance in the development of the hypercalcemia.[2] The pathogenesis of hypercalcemia resulting from the interaction (humoral and/or cellular) of neoplasia and bone is poorly understood.

Animal Models

Four distinct neoplastic diseases of laboratory animals have been suggested as potential models of hypercalcemia of malignancy: Leydig cell tumor of rats,[3] fibrosarcoma of mice,[4] Walker carcinosarcoma 256 of rats,[5] and VX-2 carcinoma of rabbits.[6] Although the rodent model neoplasms have an associated hypercalcemia, their morphologic characteristics and biologic features are not homologous with the human condition. Studies in our laboratory indicated that the VX-2 carcinoma is

Publication sponsored by the Registry of Comparative Pathology of the Armed Forces Institute of Pathology and supported by Public Health Service Grant RR 00301 from the Division of Research Resources, US Department of Health, Education and Welfare, under the auspices of Universities Associated for Research and Education in Pathology, Inc. Dr. Prieur is the recipient of Research Career Development Award K04 RR 00003 from the National Institutes of Health. Address reprint requests to Dr. D. M. Young, Department of Veterinary Science, Veterinary Research Laboratory, Montana State University, Bozeman, MT 59717.

0002-9440/78/0199-0619/$01.00

an appropriate model for studying the mechanisms of osseous-mediated hypercalcemia of malignancy in the absence of skeletal metastases.[7]

Biologic Features of the Rabbit Model

The VX-2 carcinoma is the result of a malignant transformation of the viral-induced Shope papilloma.[8] The biologic behavior of the readily transplantable VX-2 carcinoma has been well characterized.[9] Subsequent to implantation in muscle, the carcinoma grows rapidly with local infiltration and metastases to regional lymph nodes and lungs. Histologically the VX-2 carcinoma is characterized by high cellularity, scant stroma, anaplasia, and prominent pleomorphism (Figure 1). A progressive fulminating hypercalcemia develops from 2 to 4 weeks after tumor implantation. Dissection of the primary injection site after 3 weeks reveals a necrotic neoplastic mass with cystic central areas. Peripheral margins are well vascularized and contain a rim of viable-appearing neoplastic tissue. Metastatic involvement of the sublumbar lymph nodes may be so extensive that major vessels and lymphatics become occluded. Pulmonary metastases are diffuse, consisting of numerous, gray to white, well-circumscribed, small (1 to 2 mm) nodules distributed evenly throughout the lungs. Death occurs predominantly at 4 to 8 weeks, with evidence of pulmonary metastases, hypercalcemia, nephrocalcinosis, and renal failure. Extensive examination at necropsy has failed to reveal any evidence of distant metastasis to bone. Local invasion may erode regional limb bones and cause pathologic fractures. Varying degrees of increased osteoclastic activity and bone resorption are evident histologically at different skeletal sites.[10] Although evidence of enhanced bone resorption has been reported to occur at locations distant from the transplantation site, it is greatest in the bones of the limb harboring the neoplastic growth.

Comparison With Human Disease

The VX-2 carcinoma model is of epithelial origin, with many histologic and ultrastructural features similar to human tumor types associated with hypercalcemia. The VX-2 neoplasm is highly malignant but does not metastasize to bone. The hypercalcemia which develops is insidious and progressive and results in nephrocalcinosis, which has been reported in humans. In addition, the development of the hypercalcemia has been associated with production of prostaglandins (and metabolites thereof) by neoplastic cells in both the VX-2 carcinoma and in human carcinomas associated with hypercalcemia. Similar histologic osseous lesions indicative of enhanced bone resorption have been reported in rabbits and humans.

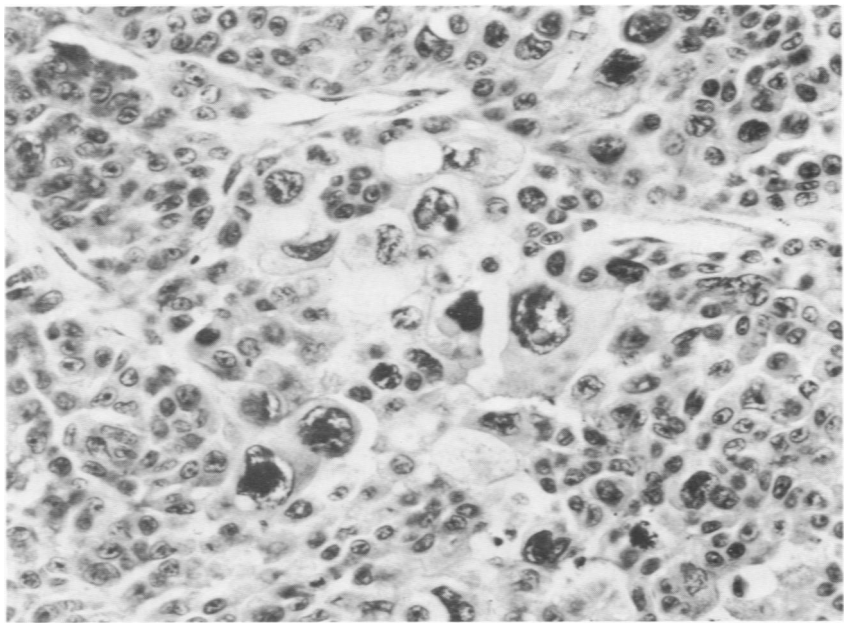

Figure 1—Section of VX-2 carcinoma 21 days after implantation in quadriceps muscles. Note high cellularity, nuclear hyperchromatism, and pleomorphism. (H&E, ×330)

Potential Usefulness of the Model

Because the VX-2 carcinoma has many morphologic, biochemical, and biologic features in common with the human carcinomas associated with hypercalcemia, this model should be useful in the elucidation of the mechanisms of the hypercalcemia of malignancy. The natural host for the neoplasm is an ideal size for the collection of blood, urine, and tissue for experimental studies. The VX-2 model provides an opportunity for investigation of a variety of substances which may interact with bone, eg, osteoclast activating factor, tumor angiogenesis factor, and prostaglandins.[11] The VX-2 carcinoma grows readily in athymic nude mice.[12] Therefore, studies could be performed in the same species, comparing the biologic behavior of suspected human carcinomas with the VX-2 carcinoma. In addition, the VX-2 carcinoma has been shown to be sensitive to therapeutic modalities and, therefore, offers an opportunity for comparative therapeutic studies.[13,14]

Availability

The VX-2 carcinoma is readily transplantable from rabbit to rabbit. In addition to serial transplantation of the neoplasm, we have found that

storage in the frozen state (−190 C) for 5 years does not affect its viability or biologic behavior. Several laboratories are studying the VX-2 carcinoma and are maintaining a supply of the tumor for future investigations.

Addendum: After the submission of this manuscript, Wolfe et al reported a different spectrum of biologic behavior of the VX-2 carcinoma.[15] The original supplier for our study and for Wolfe's study was Arthur D. Little, Inc.; however, recent investigations have revealed that Arthur D. Little, Inc., has two sublines.[16] We received VX-2 1F, whereas Dr. Wolfe's source was derived from VX-2/1. Our subline (VX-21F) is more malignant and produces a greater hypercalcemia than does the Wolfe subline (VX-2/1).

References

1. Chopra D, Clerkin EP: Hypercalcemia and malignant disease. Med Clin North Am 59:441–447, 1975
2. Myers WPL: Hypercalcemia associated with malignant diseases. Endocrine and Nonendocrine Hormone-Producing Tumors. Chicago, Year Book Medical Publishers, Inc., 1971
3. Rice BF, Roth LM, Cole FE, MacPhee AA, Davis K, Ponthier RL, Sternberg WH: Hypercalcemia and neoplasia: Biologic, biochemical, and ultrastructural studies of a hypercalcemia-producing Leydig cell tumor of the rat. Lab Invest 33:428–439, 1975
4. Tashjian AH Jr, Voelkel EF, Levine L, Goldhaber P: Evidence that the bone resorption-stimulating factor produced by mouse fibrosarcoma cells is prostaglandin E_2. J Exp Med 136:1329–1343, 1972
5. Hilgrad P, Schmitt W, Minne H, Ziegler R: Acute hypercalcemia due to Walker carcinosarcoma 256 in the rat. Horm Metab Res 2:255–256, 1970
6. Wilson JR, Merrick H, Woodward ER: Hypercalcemia simulating hyperparathyroidism induced by XV-2 carcinoma of rabbit. Ann Surg 154:485–490, 1961
7. Young DM, Fioravanti JL, Prieur DJ, Ward JM: Hypercalcemic VX-2 carcinoma in rabbits: A clinicopathologic study. Lab Invest 35:30–46, 1976
8. Kidd JG, Rous P: A transplantable rabbit carcinoma originating in a virus-induced papilloma and containing the virus in masked or altered form. J Exp Med 71:813–838, 1940
9. Voelkel EF, Tashjian AH Jr, Franklin R, Wasserman E, Levine L: Hypercalcemia and tumor-prostaglandins, the VX_2 carcinoma model in the rabbit. Metabolism 24:973–986, 1975
10. Hough A Jr, Seyberth H, Oates J, Hartmann W: Changes in bone and bone marrow of rabbits bearing the VX-2 carcinoma: A comparison of local and distant effects. Am J Pathol 87:537–552, 1977
11. Young DM, Ward JM, Prieur DJ: Neoplasia and skeletal homeostasis. An animal model. Calcif Tissue Res 22 (Suppl):505–507, 1978
12. Young DM, Ward JM, Prieur DJ: Unpublished observations
13. Young DM, Fioravanti JL: Adriamycin: An animal model for detection of oncolytic and cardiotoxic activity. Proc Am Assoc Cancer Res 16:73, 1975 (Abstr)
14. Dickson JA, Shah SA, Waggott D, Whalley WB: Tumor eradication in the rabbit by radiofrequency heating. Cancer Res 37:2162–2169, 1977
15. Wolfe HJ, Bitman WR, Voelkel EF, Griffiths HJ, Tashjian AH: Systemic effects of the VX_2 carcinoma on the osseous skeleton. Lab Invest 38:208–215, 1978
16. Wodinsky I: Personal communication, 1978