Embryonal Carcinomas in Syrian Hamsters after Intratesticular Inoculation of Zinc Chloride during Seasonal Testicular Growth

John Guthrie and Olive A. Guthrie

Tenovus Research Laboratory and Department of Pathology, Southampton General Hospital, Southampton, SO1 6HU, England

SUMMARY

Zinc chloride solution injected into the testes of 49 Syrian hamsters during the early spring months resulted in two embryonal carcinomas of the testis found at necropsy 10 weeks later. The zinc chloride injections were made during the period of rapid seasonal gonadal growth when spermatogonial division is activated and resulted in areas of coagulative necrosis occupying about 25% of each testis. The tumor arose adjacent to the areas of necrosis.

INTRODUCTION

The use by Michalowsky (14) of zinc chloride solution to produce partial castration in domestic cockerels had the unexpected result of inducing testicular teratomas when the intratesticular injections were made in the spring. Similar tumors have been produced by zinc sulfate (4), zinc nitrate (5), copper sulfate (6), and cadmium chloride (10). Bagg (2) induced teratomas in seasons other than spring by administering gonadotrophic hormones as well as by injecting zinc chloride into the testes (2). Zinc chloride in aqueous solution at pH 3.2 or less and in 5% concentration produces an area of necrosis within the testis, and teratomas closely resembling human testicular teratomas arise in the vicinity. In general, injection of zinc salts induces teratomas only if given in late winter or early spring (January to March), but the sensitivity of the avian gonad to changes of photoperiod is more marked in species other than domestic fowl. For this reason the Japanese quail was used for the induction of testicular teratomas during photoperiodically stimulated testicular growth out of season (12).

Although the experimental avian teratomas have served for the investigation of the effects of an antigonadotrophin (11) and might prove suitable for other studies on the control of differentiation and growth, a mammalian model could be expected to approximate the human situation more closely. Previous attempts to induce testicular teratomas in mammals have been confined to laboratory rats and mice. Injection of zinc chloride into the testes of Wistar rats has met with variable success. Several workers have produced no tumors (8, 18), but Riviere et al. (16) claimed 1 seminoma, 9 interstitial cell tumors, and 1 teratoma (this was an embryonal carcinoma). Bresler (3) reported teratoma, intraduct seminoma, and chorionepithelioma after copper sulfate inoculation and androgen stimulation in mice, but spontaneous teratomas arise in 1% of strain 129 mice (17) and in about 10% of strain 129/Sv-S7J (1).

In view of the success in inducing testicular teratomas in birds in which gonadal growth is stimulated by light change, either seasonally or artificially, it seemed useful to explore the possibility of inducing the teratomas in a mammal that might be so affected and remain so under laboratory conditions. The Syrian hamster seemed to meet this requirement (7).

As teratomas have not been reported in Syrian hamsters, it was decided to attempt their induction in this mammal by intratesticular injection of zinc chloride in the early weeks of the calendar year. At this time, in late winter the seminiferous tubules in the hamster under natural lighting contain spermatogonia in active division, with absence of mature spermatozoa.

MATERIALS AND METHODS

Animals. The Syrian hamsters used were of random-bred strains and were raised under conditions of natural light at a temperature of 20°. They were 2 months old in January, at the beginning of the experiment.

Zinc Chloride. The inoculum was prepared in a concentration of 4 g/100 ml by dissolving zinc chloride in sterile distilled water B.P. and adding sufficient 1 N hydrochloric acid to prevent precipitation as zinc hydroxide.

Procedure. Zinc chloride solution, 0.05 ml, was injected through the scrotum into the center of each testis by means of a fine No. 17 hypodermic needle. This was done under pentobarbitone anesthesia during the 1st 6 weeks of the calendar year.

The animals were fed a diet of standard rat pellets and were housed in polyethylene cages in groups of 3 and 4 and exposed to natural daylight at a room temperature of 18-20°.

They were killed by ether in the last week of April and the first week of May, and necropsies were performed. The testes were fixed in 10% neutral formalin and sliced into 0.3-cm thick blocks. The pituitary glands were dissected out and fixed either in formalin or osmium tetroxide for later studies.

Material for light microscopy was processed for paraffin sections, cut at 5 μm, and stained by Harris's hematoxylin and eosin.

Received May 20, 1974; accepted June 7, 1974.
RESULTS

In 6 cases, no testis could be found but, in 3 of these 6 cases, fibrous-walled cavities in the vicinity of the epididymis probably represented the remains of testes that had been totally destroyed. In all the others, 43 in number, there were areas of coagulative necrosis similar to those found in the previous experiments on domestic fowl and quail (9, 12). These were surrounded by a zone of pigmented and foamy macrophages and a variable amount of surviving testis. In most cases, only a rim of seminiferous tubules surrounded the lesion, and here spermatogenesis was present.

In 2 hamsters killed 10 weeks after inoculation, the left testis showed in each a small focus of embryonal carcinoma adjacent to the area of necrosis produced by the injection of zinc chloride (Fig. 1). Both tumors were just under 0.3 cm in diameter and were essentially similar in structure. The solid cellular growth consisted of large cells with relatively large nuclei, prominent nucleoli, and moderate numbers of mitotic figures (Fig. 2). Spermatogenesis was present in these testes, but in the immediate vicinity of the tumors, the tubules were lined by spermatogonia and Sertoli cells. In both cases, the growth infiltrated among the seminiferous tubules at its periphery. The structure corresponded closely to embryonal carcinoma in man or malignant teratoma anaplastic of the British Testicular Tumour Panel (15).

DISCUSSION

Spontaneous tumors in hamsters are rare, although 1 strain (B10 4.24) has a high incidence of adenoma of the adrenal cortex, and another (B10 15.16) is supersensitive to the action of the polycyclic hydrocarbons (13). As no spontaneous testicular tumors have been reported in hamsters, the discovery in this experiment of 2 embryonal carcinomas only 10 weeks after the injection of zinc would seem to be clearly related to the experimental procedure. No metastases were found, but the tumors must be very early in development. It is of interest that, although teratomas in humans show a similar ratio from right to left, as do testicular tumors as a whole (5:4), the more malignant growths, malignant anaplastic teratomas and malignant trophoblastic teratomas favor the left side (15). In the zinc-induced teratomas in fowl, significantly more left- than right-sided tumors are of the dwarf embryonal variety (9).

The results of this experiment would seem to justify further experiments with injection of the metallic salt in the different phases of spermatogenic development under increasing light periods. It would also be relevant to allow further growth and possible differentiation of a number of tumors to take place before assessing the significance of type or relationship to either testis.

REFERENCES

Fig. 1. Experimentally induced embryonal carcinoma in testis of hamster HR23/73. Note part of necrotic area produced by zinc chloride at the top and the tumor on the right. Seminiferous tubules in the bottom part of the picture show no spermatogenesis and centrally show invasion by the tumor. H & E X 88.

Fig. 2. Experimentally induced embryonal carcinoma in testis of hamster HR 23/73. Upper portion, the edge of the tumor seen is composed of a solid cellular growth apparently syncytial in places. The nuclei are large, somewhat convoluted, and have 1 or 2 large nucleoli. Lower portion, seminiferous tubules show absence of spermatogenesis; center portion, 1 tubule contains a multinucleated giant cell. H & E, X 220.
Embryonal Carcinomas in Syrian Hamsters after Intratesticular Inoculation of Zinc Chloride during Seasonal Testicular Growth

John Guthrie and Olive A. Guthrie

Cancer Res 1974;34:2612-2614.

Updated version  Access the most recent version of this article at:
http://cancerres.aacrjournals.org/content/34/10/2612

E-mail alerts  Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions  To request permission to re-use all or part of this article, contact the AACR Publications Department at permissions@aacr.org.