animals. Under Nembutal anaesthesia (40 mg/kg b.wt) the right vena saphena magna was exposed at the knee. A freshly prepared sterile solution of 6-OH-dopamine (Hässle, Sweden) dissolved in 0.9% saline containing 0.1 mg/ml ascorbic acid, was slowly injected into the vein. One of the animals received 36 mg/kg b.wt and survived for 24 h. The other 2 animals both received 50 mg/kg b.wt and survived for 24 and 48 h respectively after injection. At sacrifice the animals were perfused with 5% glutaraldehyde under Nembutal anaesthesia as described elsewhere[9]. The mandibular incisors were removed and decalcified for 6 days in a cold solution of 4% EDTA dissolved in a 300 mOsm cacodylate buffer (pH 7.3). The solution was exchanged daily. The decalcified teeth were osmicated, dehydrated in acetone and embedded in Vestopal W. Thin transverse sections, covering the entire root canal, were cut from the apical portion of the roots of 3 incisors from each animal. The thin sections were collected on one hole copper grids coated by carbon-stabilized formvar. After staining with uranyl acetate and lead citrate, the sections were examined in a Philips EM 301 electron microscope.

Results and discussion. In 2 cats sacrified 24 h after injection of 6-OH-dopamine, very subtle alterations were found in the incisor pulps. These were confined to a few (2-4) unmyelinated axons in which degenerative features similar to those seen after surgical sympathectomy[10] were observed. These axons were swollen, presented a loss of microtubules and contained electron-dense inclusions. In the cat surviving for 48 h, marked alterations were found in several unmyelinated fibres. Here, various degenerative features, such as axonal swelling, loss of axonal organelles and an intra-axonal content of a fragmented material, were present (figure 1). Large electron-dense bodies were commonly seen within the axoplasm of the affected fibres. Schwann cells devoid of intact axons were found encluded in a folded basement membrane with several empty pockets. The Schwann cells themselves presented largely a normal picture (figure 2). In addition, protrusions of Schwann cell cytoplasm into unmyelinated axons occurred (figure 3). Degenerating unmyelinated axons were seen together with intact fibres in the same Schwann cell. In no case could any alterations in the myelinated fibres be found. Likewise the non-nervous pulpal tissue had a normal appearance. Examination of pulps of normal permanent incisors in several adult cats failed to reveal unmyelinated fibres with the degenerative features mentioned above.

It seems clear from the present preliminary study that 6-OH-dopamine can be used as a tool for producing a selective degeneration of sympathetic nerve fibres in the root canal of feline mandibular incisors. Axonal degeneration has been achieved at some distance from the site of 6-OH-dopamine uptake at the terminal and preterminal segments. A dose level of 50 mg/kg b.wt. i.v. and a survival time of about 48 h appears to be suitable for this type of study.

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The timing of cyclophosphamide therapy in tumor-bearing rats affects the resistance to tumor challenge in survivors

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Summary. Cyclophosphamide given to rats 2 or 5 days after an injection of Yoshida ascites sarcoma cured approximately the same proportion of animals, but the resistance to a subsequent tumor challenge was found only in rats treated with the drug 5 days after tumor injection.

Cyclophosphamide effectively inhibits the growth of a variety of rodent neoplasms[1] and is widely used in the therapy of human tumors. However, cyclophosphamide is also strongly immunosuppressive[2], and therefore may interfere with the host immune reaction to the tumor. Inasmuch as the drug-induced immunosuppression is more profound in the proliferative phase of the immune response[2,3], the damage to the immune system caused by the drug applied for the purpose of eradication of the tumor may depend upon the stage of the immune response of the host to tumor antigens. The present paper reports on an experiment where the time period between tumor and cyclophosphamide injection significantly influenced the resistance of the treated animals to a subsequent tumor challenge 4 months later.

Materials and methods. 5-month-old female and male rats of WVM strain (derived from Wistar stock) weighing 200-250 g were used as recipients of rat Yoshida ascites sarcoma (YAS) which has been maintained in our laboratory for more than 10 years by weekly passages in WVM rats. Cyclophosphamide (Bosnalijek, Sarajevo) was given i.v. in a dose of 120 mg/kg. Dead animals were checked for the presence of the tumor in the abdominal cavity and other tissues. The 4-month survival rates were compared using the χ-square test with Yates correction.

Results and discussion. Rats were injected i.p. with 10^6 YAS cells/animal and were divided into 3 groups of 21 animals each. The 1st group did not receive any further treatment. Rats in the 2nd group were given cyclophosphamide 2 days after tumor injection, and rats in the 3rd group were injected with the drug 5 days after YAS injection. Within 2 weeks after injection, YAS killed all rats that were not given cyclophosphamide. 14 rats survived in the group that had been treated with cyclophosphamide 2 days after YAS injection, and 10 survived in the group that had been given the drug 5 days after tumor inoculation. The difference in

the survival rate between the 2 latter groups was not statistically significant. 4 months after YAS injection, all surviving rats were given a 2nd i.p. injection of 10^6 YAS cells. All rats that had been treated with cyclophosphamide 2 days after the 1st YAS injection died, while 7 of 10 rats that had been given cyclophosphamide 5 days after the 1st YAS injection rejected tumor challenge and survived ($p < 0.001$). These 7 rats were resistant to further YAS challenges (data not shown).

Therefore, although the application of cyclophosphamide 2 or 5 days after YAS injection appeared to be equally effective against the tumor, resistance to a subsequent tumor challenge was demonstrable only in rats treated with the drug 5 days after YAS injection. The difference was probably due to the greater susceptibility of the immune system to cyclophosphamide during the early phase of the immune response to the tumor than later when the proliferation of the lymphoid cells subsided[2,3]. The data indicate that the timing of even a successful tumor chemotherapy may greatly influence the future resistance to the tumor in the cured animals.

Survival of rats treated with cyclophosphamide[a] after injection of Yoshida ascites sarcoma (YAS) cells[b], and resistance of the survived animals to the 2nd YAS inoculum

Cyclophosphamide given after the 1st YAS injection	No. of surviving/No. of treated rats After the 1st YAS injection[c]	After the 2nd YAS injection
No cyclophosphamide	0/21	-
2 days	14/21	0/10
5 days	10/21	7/10

[a] 120 mg/kg i.v.; [b] 10^6 cells/recipient i.p.; [c] Tumor-free survivors 4 months after tumor injection; [d] Challenged 4 months after the 1st YAS injection; tumor-free survivors 2 months after the challenge.

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Radioprotective effect of a protein free parathyroid extract

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Summary. The radioprotective effect of a bovine protein-free parathyroid extract was studied in rats, being administered orally after irradiation. A significant increase in survival was found after treatment compared to controls. It is assumed that the extract contains a new, probably as yet unknown bioactive agent responsible for the radioprotection.

Rixon et al[1,2] have shown that the parathyroid extract (200 USP units parathormone, Eli Lilly and Co.) significantly prolonged the survival rate of whole body X-ray irradiated rats. They assumed that the radioprotective effect of the parathyroid extract was mainly due to the calcium mobilizing action of the parathormone, since the rise of calcium concentration in the mammalian tissues is known to reduce their radiosensitivity[3,4].

Present work is concerned with the protective efficacy of a protein-free and consequently calcium-inactive parathyroid extract (PF-PTE against 600 R, or 850 R, or 1000 R doses of X-ray irradiation. Preparation of PF-PTE was described previsouly[5]. The lyophylized PF-PTE was dissolved in physiological saline and administered perorally in 2 ml volume by a gastric tube to male CFE rats weighing 120–150 g. The whole body irradiations were delivered by a 'Super Liliput 200' X-ray apparatus 180 kV, 4 mA, 0,5 mm Cu filter, 50 cm from the target at a dose-rate of 7,8 R/min. Different groups of animals (n = 40 in each group) were irradiated with 600 R, or 850 R, or 1000 R. One other group was kept under the same conditions but without X-ray exposure. 20 of each group of the irradiated animals were treated with PF-PTE (0.1 mg/100 g of b.wt) firstly 3 h after the irradiation and on 3 successive days thereafter. The irradiated, but non-treated animals received physiological saline only. Survival in each group was observed and expressed as the number of rats still living 30 days after exposure (percent survival). The dose reduction factor (DRF) was also caclulated according van Bekkum[6]. All the nonirradiated, non-treated animals survived the 30th day. In the non-treated 600 R irradiated and the 600 R irradiated PF-PTE treated groups, all the animals were still alive on the 30th day.

The PF-PTE proved to be effective in increasing the 30 day survival of 850 R whole body irradiated animals (figure). The dose-reduction factor (DRF) at 850 R whole body irradiation is 1.35. The PF-PTE treatment was not effective