Induction of Tumours of the Stomach in Rats and Mice by *N*-nitroso-*N*-alkylurethanes

The induction of squamous carcinoma of the stomach and of the œsophagus in the rat by *N*-nitroso-*N*-methylurethane (NMU)[1] and by some other nitrosamines[2] has been reported. Squamous tumours of the stomach have been known to develop following treatment with various other carcinogens; but very few instances have been reported of the induction of tumours of the glandular part of the stomach in rodents. Such tumours have usually been produced by direct intramural injection of carcinogenic polycyclic aromatic hydrocarbons[3]. One adenocarcinoma resulted after administration of 4-nitroquinoline-*N*-oxide through a stomach fistula to 16 rats which had in addition percutaneous applications of 20-methylcholanthrene[4]. Recently, Stewart *et al.*[5] induced adenocarcinomata of the glandular stomach in four out of 47 rats, given 2,7-*bis*(acetamido)fluorene in diet.

It is therefore of interest that in the course of further experiments using NMU under conditions similar to those described previously[1] a rat, which survived 2 years after the first of two oral doses (2·5 mg per dose, at 1-month interval), was found to have a large papillary growth on the glandular part, projecting into the lumen of the stomach (Fig. 1). The squamous part was very small, and the duodenum greatly distended. Microscopically, this lesion had characteristics similar to those described as adenocarcinoma by Baba *et al.*[4]; but no definite evidence of invasiveness was seen in the section. Other animals of the same series had squamous carcinomata, sometimes accompanied by foci of hyperplasia in the glandular part of the stomach. Various degrees of hyperplasia of the glandular part of the stomach have been noted already among rats in the previous series[1].

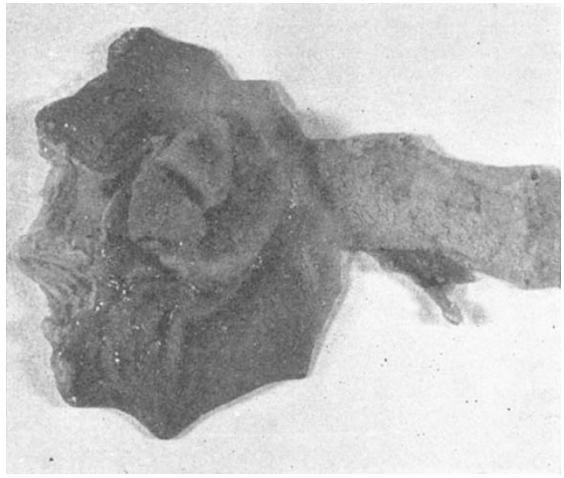

Fig. 1. Stomach and duodenum of a female rat which was given two doses of *N*-nitroso-*N*-methylurethane by stomach tube (2·5 mg per dose at 1-month interval). Killed 2 years after the first dose. Note the small size of the stomach, especially of its squamous part, the distended duodenum and the large papillary growth in the middle of the glandular part (× *c.* 1·3)

In view of the striking carcinogenic properties of NMU it was of interest to test its homologue, *N*-nitroso-*N*-ethylurethane (NEU). Among seven male white mice which survived 11–13 months after the first of four oral doses of NEU (1–2 mg per dose in aqueous ethanol, given in the course of nine months), three developed squamous carcinomata, one had in addition a sarcoma and one mouse had a papillary growth, with features suggestive of neoplasia, on the glandular part of the stomach, but again no evidence of invasion was seen.

Thus NEU induces in mice stomach tumours and lesions similar to those seen in rats given NMU. The type of tumour and the site of its appearance may depend on the site of the deposition of the *N*-nitroso-*N*-alkylurethane from the stomach tube. This can be introduced deep into the glandular part, or only as far as the squamous part of the stomach or the œsophagus. Thus, even a slight variation in the conditions of administration may determine the type and the site of tumours induced by nitroso-alkylurethanes. Like NMU[6], NEU undergoes decomposition with evolution of nitrogen in contact with —SH containing compounds or tissues, at neutral *p*H.

The induction of proliferative lesions of the glandular part of the stomach of mice and rats by these *N*-nitroso-*N*-alkylurethanes is of particular interest, and suggests that in time unequivocally malignant tumours might develop among the surviving animals.

I thank Dr. P. N. Magee for the microscopic evaluation of the lesions.

Note added in proof. Since this communication was submitted an undoubtedly malignant adenocarcinoma of the glandular stomach, with local spread, was found in a rat treated with NMU.

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Pinealoma: a Variety of Argentaffinoma?

Serotonin (5-hydroxytryptamine) contained in the gastrointestinal tract is intimately associated with the Kultschitzky type of cells[1]. Carcinoid tumours arise from such cells so their serotonin content is very important[2]; apparently, the majority, or maybe all, the symptoms of the so-called carcinoid syndrome must be attributed to the presence of serotonin at the level of the tumour[3].

Histochemically, the serotonin contained in Kultschitzky cells and in carcinoid tumours may be demonstrated as intracytoplasmic granules intensely argentaffin and faintly positive or negative to chromaffin reaction[4].

The parenchymal cells of pinealoma (Fig. 1) behave as Kultschitzky cells and carcinoid cells; thus, they reduce silver ammoniacal solutions and do not show affinity for chromium salts. For that reason it is possible that they also contain serotonin. This substance is very abundant in the nervous tissue of the brain, especially at the level of the basal nuclei, close to the pineal body. Here, serotonin could be secreted, stored or eliminated by the brain[5].

We do not know of any reference relating serotonin to either pineal body or pinealomata, nor any which refers to a clinical syndrome comparable with the carcinoid tumour in pinealoma. Since this syndrome was not discussed until 1956, it is possible that it has not been noticed in patients with pinealoma either; on the other hand, only in malignant carcinoids having hepatic metastases has this syndrome been demonstrated, and no cases of metastatic pinealoma are so far known.

Pinealoma is a rare tumour. From Frazier-Grant's collection we were only able to utilize one, and from Vincent's collection we obtained five more. Since all of them had been fixed in 10 per cent formalin for a long time we could not undertake biological determinations of either serotonin or catecholamines. On the other hand, the content of serotonin and of norepinephrin is relatively low in the normal pineal body, but such a consideration