Augmentation of chemically induced pancreatic and bronchial cancers by epidermal growth factor

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SUMMARY The incidence of Syrian golden hamsters with pancreatic cancer induced by subcutaneous injections of *N*-nitroso-*bis*(2-oxopropyl)amine for 19 weeks (each 10 mg/kg) increased from 44% to 75% ($p = 0.016$) when epidermal growth factor was also administered from week 5 through week 8 (5 μ g every three days for 10 injections). Epidermal growth factor increased pancreatic weight and body weight. The incidence of animals with bronchial cancer doubled. Epidermal growth factor could be a cocarcinogen as a result of its mitogenic activity.

Proliferation of new cell populations is required for carcinogenesis and promotes it.^{1,2} Because the polypeptide hormone epidermal growth factor (EGF) is ordinarily mitogenic, EGF augments neoplasia.³ In mice, EGF shortens the latent period of development of methylcholanthrene-induced cutaneous cancer⁴ and increases by three-fold the number of anal squamous cancers after administration of the colonic carcinogen 1,2-dimethylhydrazine (DMH).⁵ Reduction of the salivary-enteric complement of EGF in male mice by removal of the submandibular salivary glands, with their rich store of EGF and other growth factors, is associated with a one third reduction in the number of animals with colonic cancers after 20 weeks of treatment with DMH.⁶

The experiments to be described examined the effect of EGF on pancreatic cancer induced in hamsters by *N*-nitroso-*bis*(2-oxopropyl)amine (BOP).⁷ Hamster liver converts BOP to *N*-nitroso(2-hydroxypropyl)(2-oxopropyl)amine (HPOP), which is apparently the proximate carcinogen.⁸ BOP-induced pancreatic cancers, arising mainly in association with ductal goblet cells,⁹ are similar to human pancreatic cancers in terms of their histology, distribution, and multiplicity; production of diabetes and thromboses; and presence of immunologic and enzymic markers.^{10,11} Unlike human carcinomas, they arise in pancreatic acini as well as in ducts.

Methods

ANIMALS

Female Syrian golden hamsters were divided among four treatment groups. The ratio of survivors to the initial numbers of animals is indicated in parentheses: EGF + BOP (32/45), BOP alone (25/25), EGF alone (10/10), saline solution (9/10). Animals receiving BOP were given subcutaneous injections for 19 weeks (10 mg/kg). Epidermal growth factor was administered every three days for a course of 10 injections (each 5 μ g) (Figure). Pancreatic cancers were diagnosed by standard criteria.¹²

Results

The Figure shows that hamsters receiving EGF alone were heavier by 24–36% than all other groups at the end of the experiment. The heaviest pancreases per gram of body weight were found in animals given EGF alone and in those given EGF + BOP. The increments were 44% greater than values in the saline-injected control group and 22% greater than values in the BOP group. Details are reported in reference⁷.

Increased cellularity and an increased number of cancers were observed in the EGF-BOP group. The Table shows the attack rates.

Discussion

A cocarcinogenic effect of EGF in these experiments

Figure 7 Body weight of EGF-treated hamsters during 19 weeks of BOP treatment. Reproduced by permission of Cancer Research, ref. 7.

nearly doubles the incidence of pancreatic and bronchial cancers induced by BOP. The conditions did not permit discriminating between an effect of EGF on initiation or on promotion of neoplasia nor on stimulation of RNA synthesis or DNA synthesis.¹³

Neither is it possible in considering the pancreatic weights of hamsters treated with EGF to separate the contributions of increased cellularity, of a generalized effect on visceral mass, or of the mass of the cancers themselves. In view of the mitogenic action of EGF on myriad cells and organs,³⁷ including pancreas and cultured pancreatic cancer cells, in the present experiments, a direct stimulatory effect on pancreatic cell proliferation seems likely. The trophic effect of cholecystokinin on the hamster pancreas can increase

by six-fold the number of animals with BOP-induced carcinomas.¹⁴ But absence of a cocarcinogenic effect of caerulein (an analogue of cholecystokinin) in hamsters treated with the carcinogen *N*-nitrosobis (2-hydroxypropyl)amine is not easy to rationalise with these results.¹⁵ Moreover, human cancers are more frequent as body weight increases,¹⁶ and azoxymethane-induced colonic carcinogenesis in rats is inhibited by severe weight loss.^{17,18} The cocarcinogenic effect of EGF observed on bronchial carcinoma in our hamsters is not within the scope of this report.

Do the results of these experiments have counterparts in natural carcinogenesis? In mice, the pancreas is the richest source of mRNA for prepro-EGF, except for the kidney and the unique circumstances of the lactating breast and the submandibular glands.¹⁹ Exposure of human KB carcinoma cells to EGF increases the levels of their own EGF-receptor mRNA and protein at a post-transcriptional level.²⁰ Epidermal growth factor binds to receptors on cultured human pancreatic cells²¹ and can stimulate phosphorylation of membrane proteins.²² Although the presence of an endogenous mitogen is far from proving the existence of autocrine regulation,²³ especially inasmuch as any EGF secreted by the pancreas could be used strictly for exocrine purposes, the possibility exists. The EGF receptor is the cellular homologue of the avian erythroblastosis virus *erbB* proto-oncogene,^{24,25} and the genes for pancreatic neuropeptide Y and *erbB* overlap on human chromosome 7.²⁶ Perhaps endogenous EGF enhances expression of the surface receptors on the cells to which EGF binds,²⁷ leading to greater susceptibility to chemical carcinogenesis.^{28,29}

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Table Incidence of cancers after BOP treatment

Treatment	Animals (no)	Pancreatic cancers, local or invasive (% of animals)	Pancreatic cancers, invasive (% of those animals having cancer)	Bronchial cancers (% of animals)
EGF	10	0	0	0
BOP	25	44	73	28
EGF/BOP	32	75*	63†	56‡
Controls	9	0	0	0
Total	76	46	66	33

Comparison with BOP group:

* $\chi^2 = 5.69$, $p = 0.016$; † $\chi^2 = 1.29$, $p = 0.26$; ‡ $\chi^2 = 4.55$, $p = 0.03$.

Local cancers were confined to the pancreas. Invasive cancers infiltrated peripancreatic fat, connective tissue, or lymph nodes.

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