Xenopsin stimulates exocrine pancreatic secretion in the dog[1]

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Summary. The octapeptide xenopsin, previously isolated from amphibian skin, stimulates exocrine pancreatic secretion of bicarbonate and protein in conscious dogs and increases the volume of secretion. This effect is shown in the dose range of 1.25 up to 160 pmoles $kg^{-1}min^{-1}$. The high potency of this peptide is suggestive of a possible physiological role of xenopsin in mammals.

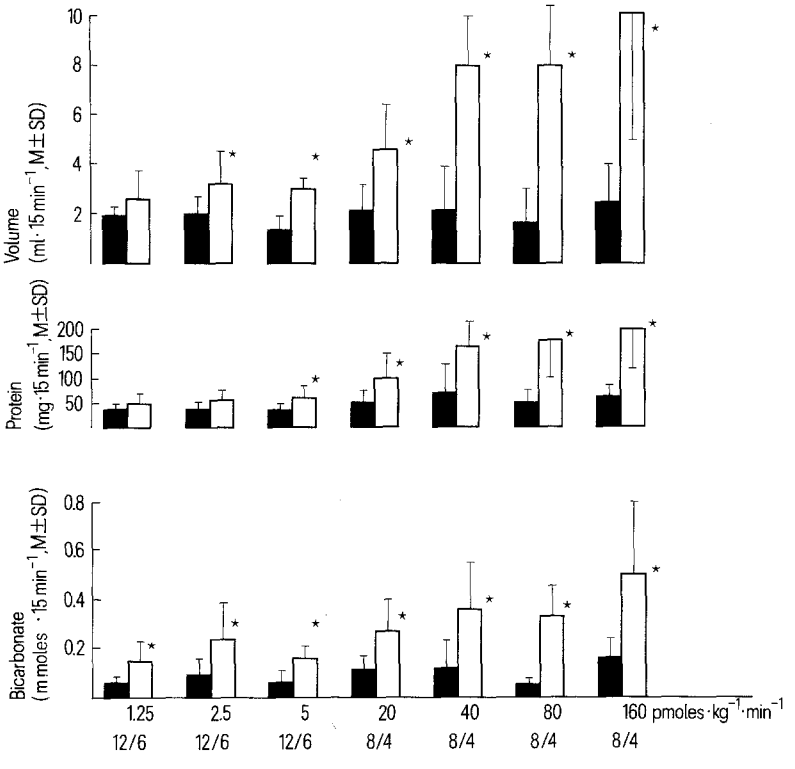
Numerous peptides isolated from the skin of amphibians such as caerulein[2], ranatensin[3], bombesin[4], dermorphin, sauvagine[5] and others have been found to be biologically active also in mammals. Xenopsin, an octapeptide isolated from the skin of xenopus laevis[6], has been shown to induce contraction of rat stomach[6,7] duodenum, and ileal muscular strips[7], to induce vascular permeability[7], to raise blood glucose level and the plasma levels of insulin, glucagon, gastrin[8,9] pancreatic polypeptide (PP) and substance P[10]. Recently xenopsin has been reported to stimulate blood flow in the canine pancreas[9,10]. We therefore tested the effect of xenopsin on exocrine pancreatic secretion in dogs.

Methods. 6 mongrel dogs were provided with pancreatic fistulae of the modified Herrera type. After an interval of at least 2 weeks 20, 40, 80, 160 and in a 2nd series 1.25, 2.5, and 5 pmoles $kg^{-1}min^{-1}$ synthetic xenopsin[6] was infused i.v. into the femoral vein for 30 min, dissolved in 30 ml of normal saline solution. Normal saline solution was infused for the following 30 min. This interval served as the basal period for the next dose of xenopsin. The different doses of each series were administered in randomized order. The experiments were always performed in the afternoon. Exocrine pancreatic secretion was collected in 15-min intervals. Protein concentration was determined by the biuret method, bicarbonate concentration by adding HCl and back-titration with NaOH after boiling. Student's t-test for paired samples was used for statistical analysis.

Results. The result is shown in the figure. All doses caused a significant rise of bicarbonate output, the smallest dose of xenopsin stimulating volume secretion significantly was 2.5 pmoles $kg^{-1}min^{-1}$, the smallest dose stimulating protein output was 5 pmoles $kg^{-1}min^{-1}$. The rise of bicarbonate and protein output was mainly the consequence of an increased volume secretion, as protein and bicarbonate concentration was not affected significantly.

In control studies, 0.08 pmoles $kg^{-1}min^{-1}$ secretin stimulated volume secretion by 1.2 ± 0.3 ml 15 min^{-1} and bicarbonate output by 0.14 ± 0.08 mmoles 15 min^{-1}; 0.5 pmoles kg^{-1} min^{-1} CCK 33 stimulated protein output by 49 ± 27 mg 15 min^{-1} above basal. 40 g kg^{-1} commercial dog food stimulated volume secretion to 11 ml · 15 min^{-1}, protein output to 250 mg · min^{-1}, and bicarbonate to 1.1 mmoles · 15min^{-1}.

Discussion. The hithero known effects of xenopsin in mammals were induced by i.v. injections of 1.2–12 mmoles kg^{-1}[7-9] and i.v. infusions of 40 pmoles $kg^{-1}min^{-1}$ (Zinner et al.[10]). Our study shows, that in the dog a dose 32 times less, i.e. 1.25 pmoles $kg^{-1}min^{-1}$ xenopsin stimulates exocrine pancreatic secretion. As this potency of xenopsin approaches that of cholecystokinin (CCK 33)[11] and as xenopsin has no structural resemblance to CCK or secretin, the exocrine pancreas may have receptors not only for the 2 classic hormones but also for the amphibian peptide xenopsin and structurally related substances such as neuroten-



Effect of different doses of xenopsin on the pancreatic secretion of volume, bicarbonate, and protein in conscious dogs. In the lowest line observations/dogs are noted. Black columns indicate basal, open columns indicate stimulated secretion. Volume secretion was stimulated significantly by 2.5 ($p < 0.05$). 5, 20, 40, 80, and 160 (in all these doses with a $p < 0.001$) pmoles $kg^{-1}min^{-1}$ xenopsin; protein output was significantly elevated after 2.5 ($p < 0.01$) 5, 20, 40, 80, and 160 (all with a $p < 0.001$) pmoles kg^{-1} min^{-1} xenopsin; bicarbonate output was stimulated significantly by 1.25 ($p < 0.05$), 2.5 ($p < 0.05$), 5 ($p < 0.01$), 20, 40, 80, and 160 (all $p < 0.001$) pmoles kg^{-1} min^{-1} xenopsin.

sin[12]. Of course, an indirect way of action via intestinal hormones or nerves must also be considered. At any rate, this observation will stimulate a search for the presence of xenopsin in mammalian tissue to elucidate the possible physiological role of xenopsin in mammals.

1 This work was supported by the Deutsche Forschungsgemeinschaft Fe 127/4.
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Thymic control of the polyploidization of hepatocytes during aging

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Summary. Relative DNA-contents of hepatocyte nuclei have been determined in young, old, and thymus-grafted old mice. There is an age-dependent increase in the occurrence of tetraploid nuclei, which is in agreement with previous data regarding the nuclear volume of hepatocytes. The neonatal thymus grafted into old recipients decreased the percentage of tetraploid cells to a statistically significant extent.

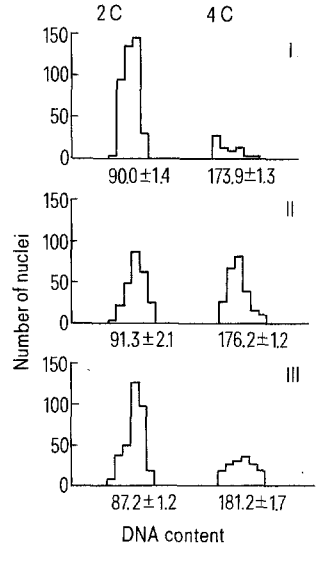
The increase in hepatic cell volume with advancing age is a phenomenon known since the beginning of this century[1]. An age-dependent increase in the mean cellular and nuclear volume was also observed in hepatocytes of haired heterozygous Balb/c-nu mice from our own colony[2]. Furthermore, neonatal thymus grafted into old recipients proved to be able to reverse the age-dependent increase of the nuclear volume to a significant extent[2]. It has been suggested that the increase of the nuclear volume may be the result of the polyploidization of hepatocyte nuclei[3-6]. Experimental evidence has proved the existence of an increased nuclear ploidity level in old animals[7]. The phenomenon of age-dependent polyploidization has been attributed to a hypothesized inability of G_2 phase nuclei to enter the M phase[7]. Therefore, both the age-dependent increase and the thymus-dependent recovery of the mean nuclear volume of hepatocytes[2] may be related to some parallel modifications of the DNA-content of the cell nuclei. The present paper describes an experimental approach to verify this hypothesis.

DNA contents of hepatocyte nuclei were measured by a microfluorimetric method in smears from young (2 months old) and old (24 months old) as well as old, thymus-grafted (24 months old) Balb/c-nu mice. Implantation of the neonatal thymus was performed at an age of 23 months. Smears of the liver were prepared on glass slides by pression, fixed in formaldehyde and stained by the classical Feulgen method. Microfluorimetric measurements of the relative DNA contents were performed according to Fujita[8] by using an OPTON MPM 01 automatic cytofluorimeter connected to a PDP 36/11 computer. Each group consisted of 3 animals, and altogether more than 450 hepatocyte nuclei were measured from each group. The data are presented in a single pool per group in the form of histogram (fig.). Percentual distributions of diploid and tetraploid nuclei are given as averages of the individual values of each group (table). The old animals show a significant increase in the occurrence of tetraploid nuclei as compared to young mice, which is in agreement with the

Percentual distribution of diploid and tetraploid hepatocyte nuclei in young, old, and thymus-grafted old Balb/c-nu mice

Group of animals	2C nuclei (%)	4C nuclei (%)	Significance p<	
Young	83.58±3.42	16.42±3.42	0.01	
Old	49.61±6.28	50.39±6.28	0.02	0.05
Old+thymus	69.06±6.15	30.94±6.15		

Values are means±SD for 3 animals per group. Significance values were calculated by using Student's t-test.



Age- and thymus-dependence of the ploidy levels of hepatocytes in young (I), old (II) and thymus-grafted old (III) Balb/c-nu mice. 2C = diploid cells; 4C = tetraploid cells.