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Increase in gastric secretion induced by 2-deoxy-D-glucose is impaired in capsaicin pretreated rats

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Gastric acid secretion was determined following intravenous administration of 2-deoxy-D-glucose (2-DG; 60 mg kg^{-1}) or electrical stimulation of the vagus nerve in urethane-anaesthetized rats pretreated when newborn with either capsaicin or the vehicle. The secretory response to 2-DG was substantially reduced in the capsaicin pretreated rats, while that induced by electrical vagal stimulation (1 mA, 1 ms, 3 Hz) was unaffected.

These results suggest that capsaicin-sensitive fibres are involved in the afferent branch of the reflex response activated by 2-DG to stimulate gastric acid secretion.

Introduction There is morphological and functional evidence indicating that the stomach receives an afferent innervation sensitive to the neurotoxin capsaicin and the physiological role of these nerves has recently received considerable attention (see Maggi & Meli, 1988, for review).

Sensory denervation by capsaicin did not influence carbachol- (Alfoldi *et al.*, 1986) or bethanechol- (Raybould & Taché, 1988) induced increase in gastric acid secretion (GAS) while controversial results have been reported for histamine- (Alfoldi *et al.*, 1986; Raybould & Taché, 1988) and pentagastrin-induced (Alfoldi *et al.*, 1986; Dugani & Glavin, 1986; Raybould & Taché, 1988) increase in GAS. When GAS was stimulated by gastric distension, an indirect vago-vagal reflex was activated and sensory denervation induced by capsaicin inhibited the response (Raybould & Taché, 1988).

In view of these findings, it appeared worthwhile to determine the influence of systemic capsaicin sensory denervation on the increase in GAS induced by a central stimulant 2-deoxy-D-glucose (2-DG), that is considered to act through vagal stimulation (Hirschowitz & Sachs, 1965; Colin-Jones & Himsworth, 1970). The stimulation of GAS by electrical stimulation of the vagus nerve was also studied to determine whether the vagal efferent branch was sensitive to capsaicin.

Methods Male and female albino rats, Sprague-Dawley Nossan strain, were housed at constant room temperature (21 ± 1°C), relative humidity (60%) and with 12 h light-dark cycle (light on 06 h 00 min). The animals were deprived of food for 20 h before the experiments but allowed free access to tap water.

Capsaicin was administered subcutaneously (50 mg kg^{-1}), under ether anaesthesia, on the second day of life. This dose was reported to cause a permanent degeneration of unmyelinated afferent neurones (Maggi & Meli, 1988) and a marked depletion of calcitonin gene-related peptide-like immunoreactivity in the stomach (Evangelista & Renzi, unpublished data; see also Sternini *et al.*, 1987). Control animals received equal volumes of vehicle (10% ethanol, 10% Tween 80 and 80% saline, v/v/v). The animals were used two months later.

Acid secretion was determined according to Ghosh & Schild (1958) in urethane (1.5 g kg^{-1} s.c.)-anaesthetized rats, maintained at 36–37°C by means of a heating lamp. The animals were tracheotomized and polyethylene tubes inserted into the stomach lumen through the œsophagus and the duodenum. The stomachs were flushed with 50 ml of warm saline to remove any solid material and thereafter perfused at a rate of 0.8–0.9 ml min^{-1} by means of a peristaltic pump (De Saga, Heidelberg, F.R.G.). A period of 45 min was allowed for equilibration after all surgical procedures had been completed. After collection of two samples, acid secretion was stimulated by intravenous (via the jugular vein) administration of 2-deoxy-D-glucose (Sigma; 60 mg kg^{-1}), dissolved in saline in a volume of 1 ml kg^{-1} or by electrical stimulation of the vagus nerve. In this latter case, the animals were artificially ventilated by means of a ventilator (Basile, Varese, Italy) for small rodents (60 strokes min^{-1}, 0.8 ml 100 g^{-1} body wt). The vagi were cut bilaterally at cervical level and the peripheral end of the left vagus was placed on a bipolar hook-shaped platinum electrode and stimulated by means of an electronic stimulator (WPI, New Haven, CT, U.S.A.) with square wave pulses of 1 ms duration, at 3 Hz and 1 mA.

Acid output was determined at 15 min intervals by titration of the perfusate with 0.005 N NaOH to pH 7 using a digital pH meter (Radiometer, Copenhagen, Denmark).

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Statistical analysis was performed by means of Student's t test for unpaired data.

Results Mean basal acid output of controls or that from capsaicin pretreated rats did not differ significantly (Figure 1a, b). Intravenous 2-DG (60 mg kg^{-1}) produced a significant increase in GAS as compared to pre-injection values which reached maximal values between 75–105 min after administration (Figure 1a).

In capsaicin pretreated rats, 2-DG elicited only a slight increase in GAS, which was not significantly different from pre-injection values. The increase in GAS induced by 2-DG was significantly different from that in control animals at all time periods (Figure 1a).

As shown in Figure 1b, the increase in GAS induced by electrical vagal stimulation (1 mA, 1 ms, 3 Hz) reached its maximal values 30 min after the beginning of the stimulation. The increase in GAS produced by vagal stimulation in capsaicin-pretreated rats was not significantly different from that observed in vehicle-treated animals at any time tested (figure 1b).

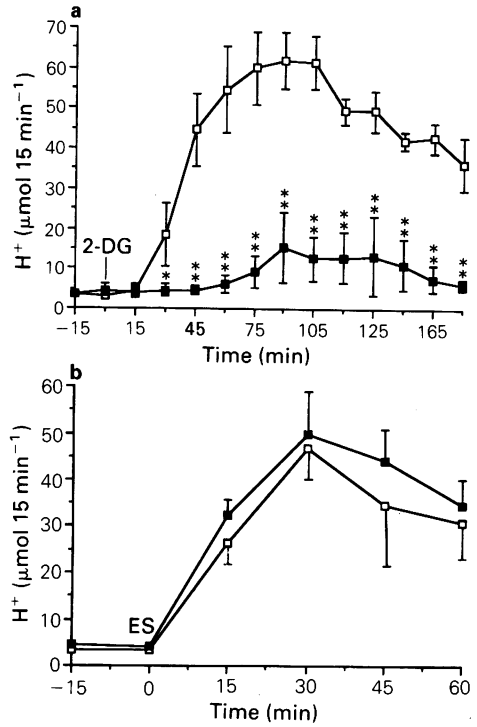

Figure 1 Effect of 2-deoxy-D-glucose (2-DG, 60 mg kg^{-1} i.v., a) or electrical stimulation (ES) of the vagus nerve (b) on gastric acid secretion (mean of at least 5 animals, s.e. indicated by vertical bars) in rats pretreated with capsaicin (■) or the vehicle (□). Statistically significant differences from the control groups are shown as * $P < 0.05$; ** $P < 0.01$.

Discussion The influence of the central nervous system in regulating GAS is well known. Electrical or pharmacological stimulation of selective brain areas, in particular the hypothalamus can induce changes in GAS mediated through the vagus nerve (see Taché, 1987, for review). Our results show that capsaicin pretreatment abolished 2-DG-induced increase in GAS but not increase in GAS induced by direct vagal stimulation. The increase in GAS elicited by 2-DG administration has been reported to be due to activation of chemoreceptors in the lateral hypothalamic area (Colin-Jones & Himsworth, 1970) which initiate and sustain a vagally-mediated response (Hirschowitz & Sachs, 1965). Neurotoxic damage at hypothalamic level has been described after sensory denervation with capsaicin (Ritter & Dinh, 1988), although a specific effect on the lateral hypothalamic area has not been reported. In conclusion, the present findings indicate that capsaicin-sensitive nerves play a crucial role in the reflex increase in GAS produced by chemically induced glucopenia brought about by 2-DG. Further studies are needed to assess whether the site of action of capsaicin in preventing the GAS response to 2-DG is central or might also have a peripheral component. In either case, the present data indicate that the efferent vagal control of GAS is unaffected by capsaicin pretreatment.

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