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Massage-like stroking influences plasma levels of gastrointestinal hormones, including insulin, and increases weight gain in male rats

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Abstract

The aim of the present study was to investigate the effects of repeated massage-like stroking on plasma levels of some gastrointestinal hormones, insulin included, glucose and weight gain. For this purpose, male rats were exposed to stroking on the ventral side of the abdomen for 3 or 14 times. The treatments were given every second day. Control rats were picked up at the same time but received no stroking. Body weight was measured regularly. Rats were decapitated 10 min after the last treatment. Hormone levels were radioimmunoassayed and glucose was measured by spectrophotometry.

In rats exposed to 3 sessions of massage-like stroking plasma levels of insulin ($p<0.05$) and somatostatin ($p<0.01$) were significantly decreased 10 min after the last treatment. After 14 treatments of massage-like stroking, decreased plasma levels of insulin ($p<0.01$) and gastrin ($p<0.01$) as well as increased glucose levels ($p<0.01$) were observed 10 min after the last treatment. In addition, weight gain was significantly increased (ANOVA $p<0.0001$) in rats exposed to 14 treatments.

In conclusion, repeated massage-like stroking decreased plasma levels of gastrin, insulin and somatostatin, increased plasma levels of glucose and promoted weight gain. The effects were influenced by the number of treatments.

Keywords: CCK; Gastrin; Glucose; Insulin; Oxytocin; Somatostatin; Weight gain; Rat; Massage-like stroking

1. Introduction

Gastrointestinal hormones are regulated by the presence of food in the gastrointestinal tract in combination with the activity in the autonomic nervous system, the vagal nerve in particular. The vagal nerve exerts both stimulatory and inhibitory effects on the release of the gastrointestinal hormones and is also of importance for the regulation of insulin (Frohman et al., 1967; Uvnäs-Moberg, 1983; Siaud et al., 1990).

Plasma levels of insulin, gastrin, cholecystokinin (CCK) and somatostatin have been shown to change in response to non-noxious sensory stimulation. Suckling in rats, sows and humans, low intensity electrical stimulation of the sciatic nerve and brushing in anaesthetized cats and rats, respectively, induce such effects, as does also warm temperature applied to the ventral side of the abdomen, vibration and acupuncture-like electrical stimulation (Uvnäs-Moberg et al., 1986, 1987; Widström et al., 1988, 1990; Linden et al., 1990; Uvnäs-Moberg, 1991; Algers et al., 1991; Eriksson et al., 1994).

Non-noxious sensory stimulation also influences a broad range of other autonomic and endocrine functions in an anti-stress direction. It e.g. reduces blood pressure and spontaneous locomotor activity, changes the secretion of corticotrophin-releasing hormone (CRH) and increases nociceptive thresholds in rats. (Kaufman et al., 1977; Hotta et al., 1992; Kurosawa et al., 1995; Uvnäs-Moberg et al., 1996; Lund et al., 1999). The elevation of the nociceptive thresholds becomes sustained after repeated treatments and increases in size, by each treatment (Lund et al., 2002).

The increase in nociceptive thresholds caused by massage-like stroking is abolished by an antagonist to the peptide oxytocin (OT) (Ågren et al., 1995). Since various types of non-noxious stimulation, as well as the technique of massage-like stroking used in the present study, also

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releases OT into plasma and cerebrospinal fluid (CSF) (Stock and Uvnäs-Moberg, 1988; Uvnäs-Moberg et al., 1993; Lund et al., 2002), OT may be one important mediator of some of the effects caused by non-noxious sensory stimulation, the effects on vagally controlled hormones included. Repeated exposure to OT causes long-term effects by influencing the activity in other receptor populations e.g. α_2-adrenoreceptors and opioidergic function. (Diaz-Cabiale et al., 2000; Petersson et al., 1996, 1998).

Connective tissue massage in human volunteers produces pain relief. Moreover, it increases the plasma concentration of β-endorphin, which increases pain threshold suggesting that β-endorphin or other endogenous opioids may also be involved in the regulation of some of the above-mentioned endocrine and anti-stress like effects (Kaada and Torsteinbo, 1989).

In the present study we examined the effects of massage-like stroking, given 3 or 14 times, on plasma levels of gastrin, CCK, insulin, somatostatin and glucose in rats. In addition, weight gain was measured continuously during the entire experimental period.

2. Materials and methods

2.1. Animals

Forty-four male Sprague–Dawley rats (290 g at arrival, Scanbur-BK AB, Sollentuna, Sweden) were used. The animals arrived 7 days before the start of the experiments allowing them to adapt to their new environment. They were maintained under constant controlled conditions of light–dark cycle (12 : 12 h, lights on 06.00), temperature 20 ± 2 °C, and relative humidity (55–60%). Food (R36: Ewos, Södertälje, Sweden) and tap water was freely available in the home cage. The animals were housed five or six per cage (Macrolon IV). All experiments were performed in the daytime 10 a.m.–3 p.m. The experiments were approved by the Ethical Committee for Animal Experiments in Stockholm.

2.2. Massage-like stroking

Massage-like stroking was performed according to the procedure described by Kanetake, 1982 and modified by Kurosawa et al., 1995. During the massage-like stroking the rat was held across the scapula and neck region and were exposed to manual stroking for 5 min of the ventral (~10 cm^2) side of the abdomen with a speed of ~20 cm s^{-1} and a frequency of 0.67 Hz (i.e. 1 stroke every 1.5 s or 40 strokes per minute), and with an estimated pressure of 100 mm H_20. In comparison, the most sensitive mechanoreceptors have a threshold lower than 4 μm for a single stepwise indentation (Sato et al., 1997). Adominal massage-like stroking is more efficient than stroking of the lateral side of the abdomen with the same frequency, or than stroking on the ventral side with other frequencies or other time-spans (Kurosawa et al., 1995; Uvnäs-Moberg et al., 1996).

The person performing the experiments had trained the procedure of massage-like stroking on other rats at a regular basis in order to standardize speed and pressure in the present experiments.

2.3. Experimental design

The animals were assigned randomly to treatment, i.e. massage-like stroking or to the control group. Rats in the treatment group were given massage-like stroking 3 ($n = 6$) or 14 ($n = 8$) times. Treatments were given every second day. Control rats ($n = 6$ and $n = 8$) were picked up at the same time as the animals receiving massage-like stroking, but were only held. The rats were decapitated 10 min after the last treatment.

2.4. Body weight

All rats were weighed prior to the massage-like stroking during the whole experimental period. Weight measurements (Mettler PE200) were accurate to a tenth of a gram.

2.5. Collection of blood

10 min after the last treatment rats were decapitated one by one in a separate room to avoid unspecific stress responses (Brodin et al., 1994). Trunk blood was collected in ice-chilled tubes containing 10 IU ml^{-1} of heparin (Lövens Läkemedel, Malmö, Sweden) and 500 IU ml^{-1} of Trasylol® (Bayer, Germany). Blood samples were centrifuged and thereafter plasma was separated, frozen and stored at −20 °C until analysis.

2.6. Radioimmunoassays (RIA)

Gastrin and insulin were determined by RIA as previously described (Petersson et al., 1999). Limit of detection for gastrin was 6.2 pmol l^{-1} and for insulin 0.05 ng ml^{-1}. Somatostatin and CCK were determined by RIA after SEP-PAK® C_{18} extraction (Waters Corporation, Milford, Mass., USA), as previously described (Petersson et al., 1999). Limit of detection for somatostatin was 1.2 pmol l^{-1} and for CCK 3.5 pmol l^{-1}.

2.7. Spectrophotometry

Glucose was measured with GOD-PAP spectrophotometric method (Cat. No. 14365, Diagnostica Merck, Darmstadt, Germany).

2.8. Statistical analysis

The results are presented as means ± SD. *P*-values of 0.05 or less were regarded as statistically significant. All data were analysed with the computer software program Statistica® version 6.0. The homogeneity of variances in the data was tested with Levene's test (Table 1). If the test