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READERS' FORUM

Increase of insulin-like immunoreactivity in submandibular salivary glands of dexamethasone-treated ageing rats

Dear Sir,

Previous reports showed that parotid and submandibular salivary glands (SSG) of various animal species, including man, contain an insulin-like immunoreactive factor (ILI) (1, 2) which can bind *in vitro* to insulin receptors and exert several biological activities of insulin, such as stimulation of amino acid uptake and lipogenesis (3). This factor is likely to be synthesized *in situ* by cells of the intercalated ducts (4), differently from most other hormones and growth factors present in the salivary glands, which are produced in the granular convoluted tubular cells (5, 6). We have previously reported that the ILI content in rat SSG is maintained throughout lifetime (7) and is increased in streptozotocin-diabetic animals (8). In the present work we investigated whether SSG-ILI was also increased in another type of experimental diabetes, induced by glucocorticoids administration to ageing rats and characterized by augmented peripheral insulin resistance (9). Male Sprague-Dawley rats of 18 months of age (620-680 g body weight) were subjected to a daily subcutaneous injection of dexamethasone (DEX) phosphate (0.125 mg/kg, dissolved in saline) for 13 days. An age-matched untreated group was used as control. Blood samples were collected from the tail vein of conscious animals during the experimental treatment for the monitoring of plasma glucose and insulin levels. Twenty-four hours after the last DEX injection, the rats were anaesthetized with sodium pentobarbital (50 mg/kg) and SSG were dissected and homogenized. The methods of extraction and evaluation of ILI have been previously described (3).

Body weights of 18-month old rats decreased by 20% after 13 days of DEX treatment and also adrenal gland weights were significantly reduced with respect to controls ($p<0.01$) (Table 1). DEX-treated animals developed hyperglycaemia and marked hyperinsulinaemia (4-5-fold increase over basal values). It should be noticed that some DEX-treated rats remained normoglycaemic although most of them became markedly hyperglycaemic.

Figure 1 illustrates that the ILI content of submandibular glands increased significantly in DEX treated animals with respect to untreated age-matched controls, and that the submandibular gland protein content was unchanged in control and DEX-treated animals respectively.

Thus, ILI present in salivary glands can be modified by a glucocorticoid treatment leading to alteration of glucose homeostasis in ageing animals. This observation is an additional example for the possibility for ILI content to be regulated, which was previously found to be the case in streptozotocin diabetics rats where SSG-ILI levels increased after 4 weeks duration of disease (8). In the latter case, the ILI increase was interpreted as a compensatory adaptation in insulin deficient diabetic syndrome which might help to preserve salivary gland function (10). Moreover DEX treatment of 18-month old rats, which was highly effective in all animals as testified by adrenal gland hypotrophy, led to a marked hyperglycaemia only in those animals in which the intervening compensatory hyperinsulinaemia was probably insufficient to restore an already precarious glucose homeostasis. Indeed, it is likely that DEX administration further aggravated a pre-existing age-dependent state of insulin resistance in target tissues, which has been reported to develop quite early during ageing (11).

It cannot be excluded that the increased ILI content

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Table 1 - *Characteristics of 18 mo old Sprague-Dawley rats treated with dexamethasone (DEX).*

Treatment	Controls	DEX
Body weight (g)		
Before treatment	696±26	731±23
After treatment	710±19	584±18**
Adrenal gland weight (mg)	23±3	12±1**
Glycaemia (mg/dl)	110±6	246±59*
Insulinaemia (ng/ml)	3.6±0.6	17.3±2.5**

Mean±SEM of 6-7 determinations.
*$p<0.05$, **$p<0.01$ *vs* controls

found in SSG of hyperinsulinaemic animals could be, at least in part, accounted for by trapped insulin through a ligand-receptor endocytotic process. However, the previously reported increase in ILI content of SSG in streptozotocin-diabetic rats with negligible circulating insulin levels (8) and the direct demonstration of the presence of insulin mRNA in the mouse salivary glands (12), support the assumption that most insulin-like material is synthesized *in situ*. On the basis of the common embryologic origin, structure and function of pancreatic and salivary tissues, it may be suggested that not only pancreatic β-cells but also ILI-producing cells in salivary glands could be stimulated by DEX treatment to produce more insulin or ILI as a compensatory adaptation. However, it should also be reminded that DEX is a well known inducer of gene expression like other steroid hormones (13) and finally, that increased ILI levels result from a direct effect of DEX treatment or rather from the hormonal induced insulin resistance remains to be determined. In summary we report here that, as in streptozotocin diabetic rats characterized by a drastic reduction of insulinaemia, ILI production in rat SSG is also enhanced in DEX-induced diabetes, characterized by peripheral insulin resistance rather than drastically impaired insulin secretion.

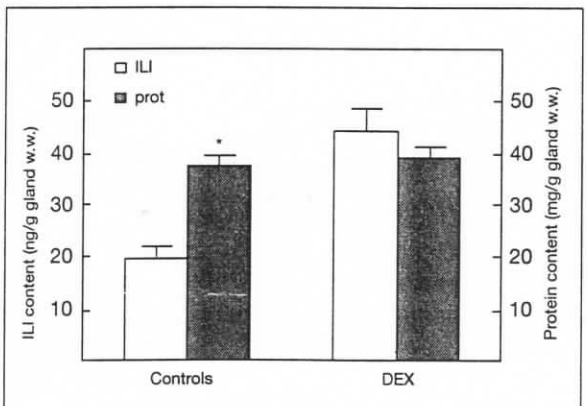

Fig. 1 - *Insulin-like immunoreactivity (ILI) and protein content in submandibular salivary glands of 18-mo old rats treated with dexamethasone (DEX). Bars represent mean ± SEM of 14 determinations, *p<0.01 vs controls.*

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