Increased peripheral venous somatostatin concentration and decreased glucagon response to arginine in patients with insulin dependent diabetes mellitus without residual B-cell function

Increased plasma SRIF in IDDM

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Abstract. The glucagon response to insulin hypoglycaemia is frequently reduced in patients with IDDM. In the present study arginine infusion, thought to act directly on the islet cells, was used to stimulate somatostatin (SRIF) and glucagon in IDDM without residual B-cell function.

Thirteen IDDM patients' were compared with 13 sex- and age matched normal controls following an arginine infusion. The plasma SRIF concentrations in the 'IDDM group' and normal controls increased from 24.2 ± 2.5 to 31.1 ± 3.9 pmol/l (P < 0.01) and 19.7 ± 1.7 to 23.9 ± 3.4 pmol/l respectively after 10 min (P < 0.01). The plasma glucagon concentrations increased from 27 ± 4.7 to 176 ± 23.1 pmol/l (P < 0.01) and 36 ± 5.0 to 302 ± 31.9 pmol/l (P < 0.01) respectively after 20 min.

Thus, in long standing IDDM without residual B-cell function, increased plasma SRIF concentrations are found and the glucagon response to arginine is reduced. The last observation further explains why these patients are especially vulnerable to hypoglycaemia.

The interrelationship between the three major hormones in the pancreatic islets has been extensively studied in experimental diabetes and in diabetes in man (Christensen et al. 1981; Ejiri et al. 1981; Orci et al. 1976). However, in long-term insulin dependent diabetics without residual B-cell function no studies are published on the plasma SRIF and glucagon levels. Although elevated plasma SRIF and glucagon are usually found in experimental diabetes, elevated SRIF and decreased glucagon from the pancreas has also been found (Hara et al. 1979; Schusdziarra et al. 1981). A reduced plasma glucagon response to insulin induced hypoglycaemia in IDDM of long standing has been reported by several groups. The cause of the reduced glucagon response was considered to be disturbances of glucagon release due to an intrinsic A-cell defect, probably related to the glucose sensor (Bolli et al. 1984; Gerich et al. 1973).

Earlier studies using arginine stimulation, thought to act directly on the islet cells (Fajans et al. 1971; Wasada et al. 1980), have shown no change or increased glucagon response compared to normal controls, mainly in groups of IDDM patients where the residual B-cell function was not defined (Gerich et al. 1975; Kawamori et al. 1980). We wanted to investigate the arginine response of SRIF and glucagon in a group of IDDM patients without residual B-cell function.

Materials and Methods

Thirteen IDDM patients, 18–33 years old, 7 women and 6 men, were investigated. The mean duration of the disease was 153 ± 9.3 months. They were selected because of absence of C-peptide response to glucagon (1 mg iv) and arginine. None were obese. They were treated with insulin NPH and rapid acting insulin twice daily and their mean dose of insulin was 56 ± 3.2 IU/day. Ten had insulin antibodies. Six had more than 3 microaneurisms in the ocular fundus, none had proliferative
retinopathy, I had proteinuria. None had diabetic neuropathy, judged by motoric and sensoric nerve conductance velocity (median-, posterior tibial- and surreal nerve). All were in fair metabolic control, mean HbA1c was 8.3 ± 0.4%, and all except one had HbA1c values below 10% (normal value for non-diabetics is less than 5.8%).

Thirteen normal persons 21–39 years old, 7 women and 6 men were investigated following the same procedure. All the participants gave their informed consent.

The test consisted of an iv antecubital infusion with arginine chloride (0.5 g/kg/20 min) following a 30 min recumbency period. The test was performed from 08.00 h, fasting, the diabetics did not receive their insulin. Blood samples were drawn from the opposite arm for SRIF, pancreatic glucagon, GH, C-peptide, HbA1c, insulin antibody and blood glucose measurements.

The blood samples for SRIF and glucagon measurements were collected in ice-cooled heparinized evacuated glass tubes as quoted (Skare et al. 1984). Specific RIA's were employed for hormone measurements (Folling & Norman 1972; Rutlin et al. 1977; Skare et al. 1984; von Schenck & Nilsson 1981). C-peptide was measured by a radioimmunoassay kit (Novo-Research Institute), stable HbA1c was measured using a ion-exchange chromatography method (Dahl-Jørgensen & Larsen 1982), insulin antibodies were determined according to Folling & Norman (1972) and blood glucose with a glucose dehydrogenase method. The results are expressed as mean value ± SEM.

For statistical analysis the Wilcoxon test for pair differences and the Wilcoxon rank sum test were employed. P-values equal to or less than 0.05 were considered significant.

Results

The results are expressed in Table 1 and Fig. 1.

All IDDM patients had a basal serum C-peptide concentration less than 0.05 nmol/l, resistant to stimulation. There was no correlation between basal plasma SRIF and amounts of insulin antibodies. Significant differences between women and men were not found.

Comparison of the groups

The plasma SRIF concentrations were significantly higher in the IDDM patients when compared with the normal subjects (P < 0.05) at -15, -5, 0, 2, 20, 25, 30 and 45 min. The differences were thus significant in the pre- and post-stimulatory periods (P < 0.05 and 0.02, respectively, expressed as areas beneath the curves). The increment during stimulation, calculated from the respective basal levels in the two groups, was, however, not statistically different. The plasma glucagon concentrations were significantly lower in the group of diabetics at every point of time from 2 min (P < 0.002), gradually decreasing to insignificant differences at 30 min. The serum GH concentrations were highest in the group of IDDM, significant difference being detected already after 2 min (P < 0.02), maximal difference was evident after 25 min (P < 0.0006). The blood glucose values were highest in all IDDM patients at every point of time.

Table 1. IDDM patients and normal controls following arginine infusion.

<table>
<thead>
<tr>
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<th>Mean basal values</th>
<th>Earliest significant response values</th>
<th>Peak response values</th>
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<tbody>
<tr>
<td><strong>SRIF (pmol/l)</strong></td>
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<td>IDDM patients</td>
<td>24.2 ± 2.5</td>
<td>28.2 ± 2.8 (5 min, P &lt; 0.01)</td>
<td>31.1 ± 3.9 (10 min, P &lt; 0.01)</td>
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<td>Normal controls</td>
<td>17.9 ± 1.7</td>
<td>20.7 ± 2.1 (2 min, P &lt; 0.01)</td>
<td>23.9 ± 3.4 (10 min, P &lt; 0.01)</td>
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<td><strong>Glucagon (pmol/l)</strong></td>
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<td>IDDM patients</td>
<td>27 ± 4.7</td>
<td>47 ± 14.2 (1 min, P &lt; 0.01)</td>
<td>176 ± 23.1 (20 min, P &lt; 0.01)</td>
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<td>Normal controls</td>
<td>36 ± 5.0</td>
<td>160 ± 20.7 (2 min, P &lt; 0.01)</td>
<td>302 ± 31.9 (20 min, P &lt; 0.01)</td>
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<td><strong>GH (µg/l)</strong></td>
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<td>IDDM patients</td>
<td>6.5 ± 1.6</td>
<td>10.3 ± 2.5 (2 min, P &lt; 0.05)</td>
<td>19.6 ± 2.8 (30 min, P &lt; 0.01)</td>
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<td>Normal controls</td>
<td>3.3 ± 0.9</td>
<td>8.7 ± 1.6 (30 min, P &lt; 0.01)</td>
<td>8.7 ± 1.6 (30 min, P &lt; 0.01)</td>
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<td><strong>Glucose (mmol/l)</strong></td>
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<td>IDDM patients</td>
<td>9.7 ± 1.2</td>
<td>10.3 ± 1.2 (5 min, P &lt; 0.01)</td>
<td>13.6 ± 1.1 (60 min, P &lt; 0.01)</td>
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<td>Normal controls</td>
<td>3.9 ± 0.1</td>
<td>4.7 ± 0.2 (20 min, P &lt; 0.01)</td>
<td>4.7 ± 0.2 (20 min, P &lt; 0.01)</td>
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ARGinine CHLORIDE, 0.5 g/kg

Fig. 1.
The figure shows the response of plasma SRIF, glucagon, serum GH and blood glucose to arginine infusion in 13 patients with IDDM and 13 sex- and age matched normal controls. mM = mmol/l, pM = pmol/l.

Discussion

Measurements of plasma SRIF are technically demanding. Most investigators have found extraction necessary (Arimura et al. 1975; Penman et al. 1979). In our procedure this is done with ‘Vycor’ glass, and under these circumstances our assay measured mainly SRIF-14 (Skare et al. 1984).

The study shows that the unstimulated plasma SRIF is higher and the stimulated plasma glucagon levels are lower in IDDM without residual B-cell function than in comparable normal subjects. This is in agreement with studies on animals (Hara et al. 1979). Following arginine stimulation, peripheral plasma SRIF is chiefly derived from the gastrointestinal tract, the pancreatic islets possibly being the main source (Wasada et al. 1980). The increased plasma SRIF levels may be a reflection of insulin deficiency or the diabetic state in itself. Ketosis develops more rapidly in patients without B-cell function (Madsbad et al. 1982a). Since elevated free fatty acids and beta-hydroxybutyrate stimulates pancreatic SRIF release, this might be a contributing factor, even though these patients were fairly well regulated (Kawamori et al. 1980; Wasada et al. 1980).

The increased values are in the pre- and post-stimulatory state, indicating a basal hypersecretion. The increment on stimulation with arginine corresponds to that in normal subjects.

In dogs with alloxan diabetes, iv insulin infusion diminished, but did not normalize plasma SRIF (Schusdziarra et al. 1978). In our study with fairly well regulated diabetics, plasma SRIF was supranormal.
The reduced plasma glucagon response to arginine infusion in long standing IDDM found in the present study, may at first seem surprising. Most authors (Gerich et al. 1975; Kawamori et al. 1980) have found exaggerated or normal plasma glucagon response to arginine in IDDM depending on the degree of control. However, no information regarding B-cell function in these patients is available. Furthermore, they were not selected because of proved absence of B-cell function as were the IDDM patients in the present study. The hyper-response in IDDM with B-cell function is clearly due to abnormal blood glucose and insulin levels (Kawamori et al. 1980). They showed perfect normalization of excessive glucagon response to iv arginine with the 'artificial B-cell'.

The reduced response found in the present study is of considerable interest. In the animal with experimental diabetes, increased amounts of A-cells are found in the islets (Orli et al. 1976). The reduced plasma glucagon levels may be a reflection of the increased SRIF levels. However, as mentioned, the diabetic animal model usually has increased SRIF levels together with increased glucagon levels, even if elevated SRIF and decreased glucagon release from the pancreas can be found (Hara et al. 1979; Schusdziarra et al. 1981). In human IDDM of long duration, reduced plasma glucagon response to insulin induced hypoglycaemia has been reported (Gerich et al. 1973). The A-cell response was normal in subjects with recent onset of diabetes and was progressively blunted in diabetics with longer duration of the disease. In these studies it was suggested that the duration of diabetes and not the loss of B-cell function is the most important factor for the reduced glucagon response (Bolli et al. 1983). Gerich et al. (1975) demonstrated increased or normal increments of plasma glucagon on arginine stimulation in IDDM patients, depending on their degree of metabolic control. Their study was, however, carried out on heterogeneous groups of diabetics where the residual B-cell function was undetermined. Our diabetics were all fairly well regulated. Insulin itself may inhibit glucagon release (Schusdziarra et al. 1978). However, the IDDM patients in the present study did not receive their morning insulin on the day of arginine testing, so they were relatively hypoinsulinaemic compared to the normal persons who released insulin in response to arginine.

In our group of IDDM without endogenous insulin production, no correlation was shown between duration of diabetes and glucagon response. The study of Madshod et al. (1982b) showing reduced glucagon response to hypoglycaemia in patients without C-peptide response is in agreement with our results, even though we used another glucagon stimulator. A subgroup of islet cell surface antibodies, known to be more specific for the B-cell than the cytoplasmic antibodies, and thought to play a pathogenetic role in the B-cell destruction, has recently been shown to cross-react with the A-cell (Van De Winkel et al. 1982). Although islet cell antibodies were found to be of no significance in glucagon response (Gray et al. 1978), this might hypothetically contribute to a gradual decrement in the A-cell reactivity, also affecting other stimulatory mechanisms than hypoglycaemia.

The elevated serum GH concentrations in the basal period, and the rapid and exaggerated response to arginine as an indication of increased sensitivity of the GH releasing mechanism in diabetics have been confirmed several years ago (Zane Burday et al. 1968). This indicates that SRIF from the pancreas does not influence the pituitary gland significantly in IDDM.

We believe that the homogeneity in this group of patients was crucial for our results. Our findings of elevated plasma SRIF and decreased plasma glucagon in this group of IDDM may be explained by 1) elevated plasma SRIF caused by the disease, suppressing the glucagon concentration, or 2) decreased glucagon release from the A-cell, possibly because of cell damage or disturbed islet architecture combined with insulinopenia and metabolic disturbances elevating the plasma SRIF. The decreased A-cell reactivity to different stimuli and increased plasma SRIF might further increase the vulnerability of these patients to rapid lowering in blood glucose levels.

Acknowledgments

The expert technical assistance of the laboratory staff, especially Miss Vera Kriz is gratefully acknowledged. This study was supported by the Hormone Laboratory, Aker Hospital.

References


Received on January 30th, 1985.