Somatostatin plasma levels and biological effects following subcutaneous administration of somatostatin in man

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Abstract. The rate at which somatostatin appears in the circulation after subcutaneous bolus injection and continuous administration by pump was determined in normal subjects by serial radioimmunoassays of immunoreactive somatostatin. Following a single subcutaneous injection of 250 μg, the somatostatin peak in plasma appeared after 5 min and had only a transient effect on insulin levels. During continuous administration, somatostatin reached levels able to reduce significantly insulin and glucagon. Somatostatin plasma levels exerting biological effects were observed during the subcutaneous administration of the peptide.

The biological effects of somatostatin have been studied extensively in clinical and research fields, but its short half-life and the necessity of using intravenous administration have limited the use of this peptide. Several long-acting analogues have been studied, but are still not available for clinical purposes. Different diseases may benefit from long-term treatment with somatostatin: peptide secreting tumours, where surgical treatment is impossible (Long et al. 1979), pancreatic fistulas (Di Costanzo et al. 1982), psoriasis (Weber et al. 1981). There is evidence that somatostatin may be beneficial in the treatment of diabetes mellitus as an adjunct to insulin (Gerich 1977; Christensen et al. 1978; Dimitriadis et al. 1983; Scheen et al. 1983). However, it is impracticable to use somatostatin as a chronic therapeutic agent, because this peptide has a short biological half-life (Bethge et al. 1981) and has to be administered intravenously. In order to study these problems we have observed the somatostatin plasma levels and the biological effects following subcutaneous administration of this peptide.

Materials and Methods

Plasma levels of somatostatin, insulin, glucagon, and glucose were determined after subcutaneous administration of somatostatin in two groups of six healthy male volunteers aged 22 to 30 years, of ideal body weight, selected for the study. The first group received a single bolus subcutaneous injection of 250 μg of somatostatin (Stilamin Serono). In the second group, somatostatin was given subcutaneously into the abdominal wall through a pump in two different ways: the first by maintaining a constant infusion rate (250 μg/h) during the whole experiment; the second by increasing the infusion rate (125, 250, 500, 750 μg/h) every half hour.

Somatostatin plasma levels were determined using a modified radioimmunoassay (Ghirlanda et al. 1984) in whole plasma without extraction. An antiserum raised in rabbits (named BDC) against cyclic somatostatin (Serono) was employed. Tyr-1-somatostatin was iodinated using the chloramine-T method (Greenwood et al. 1963). The tracer was purified on a Sephadex G25 coarse column (1 × 100) which had been equilibrated with acetic acid (0.5 mol/l; pH 6.1), bovine serum albumin (0.1%), aprotinin (100 KIU/ml), and EDTA (2.8 g/l). Separation of free from antibody-bound radioactivity was made by adding dextran-coated charcoal (Norit A). According to Vinik et al. (1981), the assay
buffer and pH were modified in order to reduce the tracer degradation during incubation (72 h). The antibody affinity for standard somatostatin was not affected by these changes. The working titre used in the assay was 1:35,000; there was a bound radioactivity of 35% with negligible tracer degradation. The sensitivity of our method was about 3 ng/l in whole plasma.

Insulin, glucagon, and growth hormone were meas-
ured by radioimmunoassay (Kits Biodata Italy). The glucose plasma levels were assayed using a glucose oxidase method (Beckmann). In each group, the lyophilized somatostatin was reconstituted by adding 2 ml of aprotinin (100 000 KIU/ml) with the purpose of reducing the local somatostatin degradation as shown for insulin (Berger et al. 1980). Samples were taken before, during and after the somatostatin administration. Blood was collected in chilled tubes containing 1000 KIU/ml aprotinin and 1.2 g/l EDTA, and was immediately centrifuged. The plasma was stored at -40°C until the assay was performed. Student's t-test was employed for the statistical analysis.

**Results**

In the first group studied (Fig. 1) an immediate rise in plasma somatostatin from the mean basal level (35 ± 5 ng/l) appeared at 2 min. The maximum value was 541.8 ± 47 ng/l (P < 0.05) after 5 min, followed by a progressive descent towards the basal level in 30 min (45 ± 4 ng/l at 30 min). Insulin (basal value 11.2 ± 2.8 mU/l) showed a transient but significant reduction at 5 min (6.7 ± 1.5, P < 0.05). Glucagon, GH, and glucose were unchanged.

In the second group, the first experiment (Fig. 2), performed with a constant infusion rate (250 µg/h), gave a constant somatostatin plasma level of 208 ± 28 ng/l. This level was obtained in 15 min and lasted until the end of the infusion, with a fast return to the basal level.

In the subjects treated with different dosages (Fig. 3), the somatostatin plasma levels rose from the basal level proportionally to the increasing infusion rate. The highest value observed (750 ± 45 ng/l) corresponded to the infusion of 750 µg/h. The basal level of both insulin (12.2 mU/l) and glucagon (25 ± 4.8 ng/l) was significantly reduced by a somatostatin infusion rate of 125 µg/h (7.1 ± 2, P < 0.05; 17 ± 3, P < 0.05, respectively). The maximum inhibition was obtained by an infusion rate of 250 µg/h of somatostatin (4.1 ± 1.9 mU/l, P < 0.01). On the other hand, glucagon showed a progressive reduction with increased infusion rates. The lowest glucagon level (3 ± 1.3 ng/l, P < 0.01) was observed during the highest infusion rate (750 µg/h). Growth hormone and glucose were unchanged.

**Discussion**

Synthetic somatostatin-14 has been administered to humans in a large number of clinical studies.
Plasma levels of somatostatin, insulin, glucagon, growth hormone, and glucose after subcutaneous somatostatin infusion by pump at different rates. The black circles indicate statistically significant changes from the basal levels. The asterisks show statistically significant changes from the preceding value.

(Gerich 1977; Christensen et al. 1978; Dimitriadis et al. 1983; Scheen et al. 1983; Long et al. 1979; Di Constanzo et al. 1982; Weber et al. 1981). However, all these studies have employed a venous route for infusion with different dosage schedules (250–500 µg/h), and most of them ignored the somatostatin plasma levels attained during the infusion. However, Skamene & Patel
(1984) showed that intravenous infusion rates as low as 25 µg/h give a somatostatin plasma level of 149 ± 3 ng/l and can exert an inhibitory effect on the islet hormones.

Our results show that somatostatin is absorbed after subcutaneous injection, but the bolus is cleared rapidly, and the serum level decreases in a short time. The continuous subcutaneous infusion on the contrary, promotes, a rapid increase in somatostatin plasma level reaching a dose-dependent concentration plateau within 15 min after start of the infusion. The somatostatin plasma levels obtained by a subcutaneous infusion of 125 µg/h (105 ± 25 ng/l) are comparable with the levels attained by the dose of 25 µg/h iv, the minimum dose previously shown to be effective in humans. Our experiments also confirm that this level of somatostatin exerts an inhibitory effect on plasma insulin and glucagon. At a given somatostatin infusion, the plasma levels increase rapidly and reach concentrations that last until the end of the infusion. The somatostatin plasma levels correlated with the speed of infusion and the highest values (700 ± 45 ng/l) appeared during the fastest infusion rate (750 µg/h). On discontinuing the infusion, plasma somatostatin decreased rapidly, and insulin and glucagon returned to basal value without rebound phenomena.

Numerous studies have demonstrated an inhibitory effect of somatostatin on basal and stimulated release of GH, insulin, and glucagon. In this study we correlated somatostatin plasma levels to the degree of inhibition of pituitary and islet hormone secretion in order to evaluate the sensitivity of these hormones. Insulin secretion was more sensitive than glucagon to the inhibitory effects of somatostatin (Fig. 3), the somatostatin levels that produced maximum insulin and glucagon inhibition being 208 ± 28 and 700 ± 45 ng/l, respectively. Beta cells, therefore, have the higher sensitivity to somatostatin inhibition, followed by alpha cells. The relatively constant values of GH are related to the basal condition of the study, whereas published data have shown an impaired GH secretion during somatostatin iv infusion after insulin hypoglycaemia (Adamson et al. 1982), or arginine test (Skamene & Patel 1984).

There is a direct relationship between somatostatin plasma levels and the biological effect of the peptide. Many studies performed in animals and man have shown that a meal may exert a stimulatory effect on the somatostatin plasma levels. Different authors, including our group (Uccioli et al. 1986), have demonstrated that after a test meal, somatostatin can reach values of 100 ng/l or more. We have here shown that these values can suppress the insulin and glucagon secretion, and it is thus possible to conclude that fluctuations in plasma somatostatin, occurring physiologically, may influence somatostatin sensitive cells by means of an endocrine mechanism. In conclusion, in those clinical conditions where somatostatin may be used as a therapeutic tool, continuous subcutaneous infusion by pump can induce biologically active plasma levels.

References


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