Effect of Insulin on Motilin Release in Man

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Funakoshi, A., Schteingart, D.E. and Vinik, A.I. Effect of Insulin on Motilin Release in Man. Tohoku J. exp. Med., 1987, 152 (3), 247-251 —— The effect of insulin on motilin release was investigated by use of the euglycemic glucose clamp technique. By use of this technique plasma glucose concentration was maintained constant at 80–90 mg/100 ml, and plasma insulin immuno-reactivity (IRI) was increased from 15±6 μU/ml to 171±22 μU/ml in 10 min, and remained at this level for 2 hr. Plasma motilin like immunoreactivity (MLI) concentration decreased within 10 min from 199±36 pg/ml to 120±28 pg/ml and remained low during the course of study. A significant negative correlation between MLI and IRI concentrations (r = −0.72, p <0.01) was observed. The present results indicate that the suppressive effect of insulin on motilin release is a direct action of insulin and is not mediated by glucose.

Motilin, a 22 amino-acid peptide hormone has been known as one of the gut hormones which affect the motility of the stomach and intestine (Brown et al. 1971; Lee et al. 1974; Jennewein et al. 1975). It has been shown that intraduodenal administration of glucose and amino-acids solution, which increase insulin release, inhibit the release of endogenous motilin (Christofides et al. 1979). We have shown that following insulin administration plasma motilin-like immunoreactivity (MLI) concentration fell sharply in healthy subjects and diabetic patients with and without autonomic neuropathy (Funakoshi et al. 1982). The fall of plasma MLI concentration was not correlated with changes in plasma glucose and it may have been due to the direct action of insulin on the inhibition of MLI release.

The present study was designed to examine the effect of insulin on MLI release during maintaining blood glucose at a constant level utilizing the glucose clamp technique. The present study clearly demonstrated that it is the increase
in insulin concentration that causes a fall in plasma MLI concentration.

**MATERIALS AND METHODS**

Six healthy subjects were studied. Four women and two men whose ages ranged from 22 to 32 years and body weight from 100% to 114% (mean = 105%) of ideal, as determined from Metropolitan Life Insurance Company Tables, volunteered for the study. No subjects had a family history of diabetes mellitus or lipid abnormality and none were on medication. All subjects had a minimum of three normal sleeping nights prior to study and were on their usual meals plans. An informed consent was obtained from each subject before study. The study was approved by the Committee for the protection of human rights of the University of Michigan, Ann Arbor, MI, USA.

**Glucose/insulin clamp technique**

The euglycemic glucose clamp technique developed by Andres and colleagues (Andres et al. 1966; Sherwin et al. 1974; DeFronzo et al. 1979) was carried out as follows: An antecubital vein was cannulated in an antegrade manner to administer infusates. A priming dose of insulin (2.0 mU/kg/min) was administered over 10 min to raise the serum insulin to the desired level. The dose was administered in a logarithmically falling manner until 10 min when the continuous infusion was begun. The 1.0 mU/kg/min of insulin infusion rate was maintained constant by infusion of porcine monocomponent insulin. Blood glucose was maintained between 80-90 mg/100 ml by monitoring glucose concentrations at 5 min intervals using a Beckman glucose analyzer 2 (Fullerton, CA, USA) and by adjusting the infusion rate of a 20% glucose solution using previously described algorithms based upon servo-feedback control (Andres et al. 1966; Sherwin et al. 1974).

**Laboratory method**

In all studies, blood samples were kept on ice until the plasma was separated by centrifugation and stored at −20°C. The plasma MLI concentration was measured by a specific RIA using a COOH-terminal specific antiserum MO3. Natural porcine motilin was used as the standard, and as the tracer in the assay. Both were kindly donated by Dr. J.C. Brown, University of British Columbia, Vancouver, Canada. The sensitivity of the assay was 2 pg/tube. The intraassay coefficient variation was 5% at motilin concentrations found in normal plasma. Details of this assay have been published (Funakoshi et al. 1982). Plasma insulin was measured by a double antibody RIA (Hayashi et al. 1974). The least detection limit was 0.15 μU/tube, and the intraassay coefficient variation was 3.2%.

**Statistical method**

All results were expressed as mean values ± S.E. of the mean. The significance of difference was tested by the Student's t-test for paired comparison, with p values of <0.05 regarded as significant.

**RESULTS**

The results of the effect of hyperinsulinemia during a euglycemic clamp are depicted in Fig. 1. The blood glucose concentration was maintained at 90-95 mg/100 ml during the infusions of insulin. The circulating insulin concentrations rose from a fasting concentration of 15 μU/ml to 132-157 μU/ml. During the euglycemic hyperinsulinemic clamp there was clear evidence of marked depression of MLI level from 199±36 pg/ml to 120±28 pg/ml at 10 min, and a steady state plasma MLI concentration of 85-135 pg/ml was obtained. A
significant negative correlation between MLI and insulin ($r = -0.72$, $p < 0.05$) was observed during the euglycemic state between each pair of subject.

**DISCUSSION**

Cyclic changes of MLI in plasma during the interdigestive period coincide with phasic changes of interdigestive myoelectric activity (IMC) of the proximal small bowel (Chey et al. 1978; Peeters et al. 1980; You et al. 1980). Potentially, this may result in erroneously high basal levels. This problem was addressed by obtaining at least three to five blood samples at 10 min intervals during the basal state and averaging these values. We showed previously the absence of a relationship among the periodic fluctuations of plasma concentrations of MLI, insulin and glucose in the fasting state, and the significant inverse correlation between the rise in plasma insulin concentration after a meal and a fall of plasma MLI concentration (Funakoshi et al. 1985). The absence of a relationship between the periodic fluctuations of plasma concentrations of MLI, insulin and glucose in the fasting state suggests that episodic or varying secretory rates may be occurring indepen-
dentely in endocrine pancreas and gut.

The mechanism responsible for the decrease of MLI concentration following a meal is not known. It has been suggested that a gastrointestinal peptide which has a hormonal role should be released after a meal. Motilin is released by ingestion of fat (Christofides et al. 1979). It is inhibited, however, by ingestion of glucose and protein (Christofides et al. 1979). This indicates that motilin release is indeed under the control of food stimuli but that the control mechanism is complex. After a balanced meal, for example, competitive food components may tend to both stimulate and suppress motilin release, resulting in a rather small net change.

It is possible that release of gut peptides such as somatostatin (Patel et al. 1981), pancreatic polypeptide (Adrian et al. 1980), and secretin (Jenssen et al. 1986) after a meal inhibit motilin release. However, we have previously shown that administration of insulin cause the inhibition of MLI release (Funakoshi et al. 1982). The present study reveals a strong inverse correlation between MLI and insulin during euglycemic clamp. Therefore, taking together our previous studies (Funakoshi et al. 1982, 1985) and the present study suggests that hyperinsulinemia per se or some factor or factors associated with the hyperinsulinemic state inhibits the release of motilin.

Since motilin is an important determinant of the interdigestive motor complex in the small gut (Itoh et al. 1976) and intestinal dysfunction is not uncommon in patients with diabetic autonomic neuropathy (Vinik and Glowniak 1982; Achem-Karam et al. 1985) who have high plasma MLI levels (Funakoshi et al. 1982), it seems that insulin may have significant effects on bowel motility reflected in changes in motilin concentration.

References


