Prolonged Fasting Hypoglycemia Due to Insulin Antibodies in Patient With Non-Insulin-Dependent Diabetes Mellitus: Effect of Insulin Withdrawal on Insulin-Antibody–Binding Kinetics

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Fasting hypoglycemia, which persisted for 3 days after insulin treatment was stopped, occurred in a patient with non-insulin-dependent diabetes mellitus who had inappropriate plasma free-insulin levels (18–25 μU/ml) and extremely high antibody-bound insulin (>20,000 μU/ml) but normal counter-regulatory hormone secretion and plasma C-peptide levels. The amount of antibody-bound insulin decreased in a biphasic pattern over 13 mo of observation with an initial half-life of 35 days and a more gradual decrease with a half-life of 160 days. The number of high-affinity antibody binding sites was virtually identical to the amount of antibody-bound insulin in the patient's plasma. We conclude that the patient's fasting hyperinsulinemia and hypoglycemia were due to release of antibody-bound insulin. *Diabetes Care* 10:160–63, 1987

Most diabetic patients treated with insulin develop insulin antibodies (1). Except for the uncommon occurrence of immune insulin resistance (2), these antibodies are not generally thought to adversely influence metabolic control (3,4). However, in the syndrome of autoimmune hypoglycemia (5–9), in which insulin-binding antibodies develop without prior exposure to exogenous insulin, hypoglycemia has been a prominent feature and is presumed to occur due to the release of insulin from the insulin antibodies at inopportune times. Moreover, because insulin antibodies prolong the half-life of insulin (10,11), it has been postulated that insulin antibodies may cause delayed postprandial hypoglycemia (10,12). Harwood (13) and Albert and Popp (14) have reported cases of prolonged fasting hypoglycemia in patients with insulin-dependent diabetes occurring as a result of insulin antibodies. We report here a case of prolonged fasting hypoglycemia in a non-insulin-dependent diabetic patient due to insulin-binding antibodies and describe the serial changes of insulin-antibody–binding kinetics over 13 mo after stopping insulin treatment.

SUBJECT AND METHODS

An 84-yr-old White woman came to the emergency room of St. Mary's Hospital at midnight on July 17, 1984, because of weakness, nausea, malaise, and dizziness of ~1 wk duration. She also complained of having fallen 7 days before admission, in the middle of the night, while preparing some food because she had awakened hungry. She had been waking up drenched in sweat almost every night for >1 wk. Her plasma glucose in the emergency room was 42 mg/dl (2.3 mM), and she was admitted to the hospital for further evaluation.

Past medical history included chronic mitral and aortic valve stenosis, thought to be of rheumatic origin, atrial fibrillation, a cholecystectomy, cataract extraction, and recent onset of hypertension. She was known to have non-insulin-dependent diabetes mellitus (NIDDM) since 1959, at which time her weight was 150% of her ideal body weight (90 kg). Subsequently, she was treated solely with a hypocaloric diet until 1973, when a single daily subcutaneous injection of beef-pork insulin was begun. Although the patient's body weight had decreased from 74 kg in 1980 to 59 kg at the time of admission, her insulin dose had increased from 46 to 60 U/day during this interval.

At the time of admission, the patient was taking 0.25 mg digoxin daily, 80 mg furosemide daily, 60 U lente insulin in the morning, and 0.2 mg clonidine twice daily. On physical examination the patient was alert and oriented with increased venous jugular pressure, hepatomegaly, pitting edema, and auscultatory findings of the previously mentioned valvular heart disease. Chest X ray showed cardiomegaly, pulmonary congestion, and a small pleural effusion. Admission laboratory blood tests showed mild normocytic anemia (Hb, 11 g/dl), but the total white blood cell count, plasma electrolytes, creatinine, total protein, and urinalysis were normal.

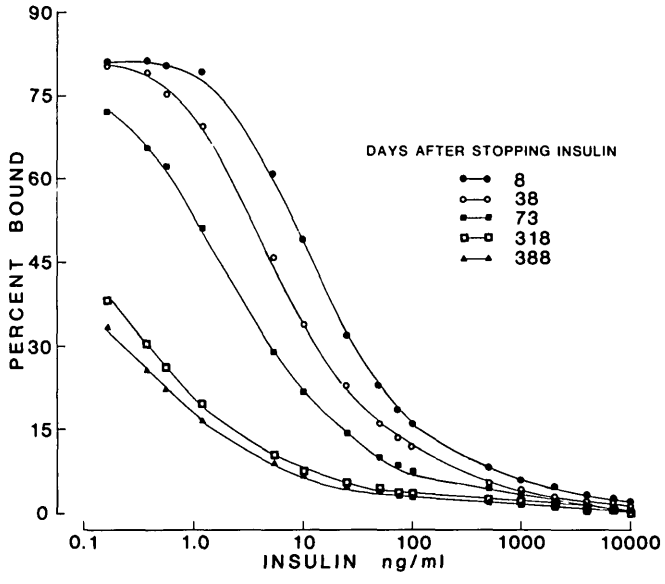

FIG. 1. Changes in insulin-antibody binding of ^{125}I–porcine insulin after stopping insulin treatment.

Plasma digoxin level was in the therapeutic range (1.2 ng/ml). Glycosylated hemoglobin was 4.8% (normal, 4–7%).

Clinical course. The patient was given an infusion with glucose, which initially alleviated her hypoglycemia. She was placed on a low-sodium weight-maintenance diet and for the next 2 days received only 10 U lente insulin before breakfast. Because of persistent hypoglycemia before breakfast (42, 39, and 51 mg/dl; 2.3, 2.1, and 2.6 mM), insulin treatment was stopped on day 3. The patient's fasting plasma glucose the next morning (48 h after her last dose of insulin) was 59 mg/dl (3.3 mM). Over the next 6 days, her fasting plasma glucose gradually increased to 79 mg/dl (4.4 mM). Plasma free-insulin levels during episodes of hypoglycemia were 18–25 μU/ml (15).

Because of these inappropriate plasma insulin levels and apparently normal pituitary-adrenal function (plasma cortisol, 18 μg/dl AM, 10 μg/dl PM; plasma growth hormone, 7 ng/ml; plasma thyroxine, 6.4 μg/dl), the possibility of an insulinoma was considered. However, CT scan and ultrasonic examination of the abdomen were negative and plasma free C-peptide was normal (1.09 ng/ml). The patient's monocyte insulin binding (10.4%/2 × 10^7 cells) was also normal (normal range 8–12%) (16). Assay for the presence of anti-insulin–receptor antibodies was negative (17). However, when insulin-antibody binding was measured (18), the patient's plasma contained a very high titer of insulin antibodies (80% binding at 1:68 dilution and 59% binding at 1:360 dilution compared with <15% binding at 1:68 dilution usually found in insulin-treated diabetic patients) (10,11,18). At admission the patient's total plasma insulin was 23,300 μU/ml (free insulin, 18 μU/ml). Thus it seemed likely that the persistent fasting hypoglycemia with inappropriate circulating insulin levels could be ascribed to dissociation of antibody-bound insulin.

The patient was discharged 12 days after admission on only diet treatment for her diabetes with a fasting plasma glucose of 80 mg/dl (4.5 mM). Over the subsequent 13 mo, she did not have fasting plasma glucose values <72 mg/dl (4 mM).

Special investigations. To further evaluate the role of insulin-antibody–binding kinetics in the pathogenesis of this patient's hypoglycemia and to assess the effects of stopping insulin therapy on insulin-antibody binding, we determined the equilibrium binding characteristics of this patient's insulin antibodies, her total bound and free insulin, and her plasma glucose concentrations over the 13 mo after stopping insulin therapy.

Equilibrium insulin-antibody binding was determined by a modification (11) of the method of Goldman et al. (18). The capacity and affinity of the high- and low-affinity binding sites was calculated by fitting the binding data to a two-site model with a nonlinear regression computer program (19). Plasma total and free insulin were measured by radioimmunoassay with the method of Nakagawa et al. (15). Bound insulin was calculated as total minus free insulin.

RESULTS

Figure 1 shows insulin-binding curves with ^{125}I-labeled porcine insulin and unlabeled porcine insulin determined over the 13 mo after stopping insulin therapy. Comparable binding was observed with ^{125}I-labeled human insulin (Table 1). Insulin binding decreased progressively over this interval. As shown in Table 1, the patient initially had a total plasma insulin concentration of 23,300 μU/ml. Of patients with insulin-antibody–associated hypoglycemia, this was exceeded only by that (28,000 μU/ml) in the patient reported by Hirata et al. (8). Over the 13 mo after stopping insulin treatment, antibody-bound insulin decreased from >22,000 to ~1000 μU/ml. There was also a progressive decrease in the capacity and association constants of both high- and low-affinity insulin-antibody–binding sites. The initial capacities and constants were in the range previously found in patients with insulin-antibody–associated hypoglycemia (0.5–10 × 10^9 L/mol and 0.6–5 nM for the high-affinity binding sites and 0.1–10 × 10^7 L/mol and 8–1600 nM for the low-affinity binding sites, respectively; 5–9). The amount of high-affinity antibodies and their association constants decreased to a greater extent (92 and 78%, respectively) than those of the low-affinity insulin antibodies (45 and 15%, respectively).

With multiple linear regression, the changes in the amount of bound insulin were found to be significantly correlated to changes in the number of high-affinity ($r = .99$, $P < .0001$) and low-affinity ($r = .95$, $P < .02$) binding sites and the association constant of the high-affinity sites ($r = .95$, $P < .02$). The number of high-affinity binding sites was virtually identical to the amount of bound insulin over the 13 mo of observation, suggesting that this class of insulin antibodies probably was predominantly if not nearly exclusively responsible for the antibody-bound insulin.

As shown in Fig. 2, the amount of bound insulin and the number of high-affinity binding sites decreased in a biphasic

TABLE 1
Changes in insulin-antibody binding, plasma insulin, and plasma glucose after stopping insulin treatment

Days after stopping insulin	Bo (%)*	High-affinity binding sites*		Low-affinity binding sites*		Plasma insulin (μU/ml)			Plasma glucose (mg/dl)
		No. (nM)	Association constant (10^9 L/mol)	No. (nM)	Association constant (10^6 L/mol)	Total	Bound†	Free	
0						23,300	23,282 (155)	18	47
8	81 (83)	113 (141)	1.04 (1.08)	2136 (2440)	2.92 (3.11)	17,600	17,575 (114)	25	100
38	80 (79)	73 (98)	0.99 (1.02)	1635 (1823)	2.87 (2.97)	10,800	10,792 (72)	8	77
73	72 (70)	34 (48)	0.89 (0.86)	1424 (1391)	2.88 (2.89)	4784	4764 (31)	20	78
318‡	38 (37)	14 (17)	0.33 (0.33)	657 (761)	2.74 (2.69)	1597	1480 (11)	17	141
388‡	34 (36)	9 (8)	0.23 (0.26)	1171 (1131)	2.66 (2.40)	1205	1188 (8)	18	117

Bo, percent bound at tracer ^{125}I-insulin.
*Values in parentheses obtained with ^{125}I–human insulin with or without unlabeled human insulin.
†Values in parentheses are in nM.
‡3- to 4-h postprandial sample.

pattern; initially, there was a rapid decrease ($t_{1/2}$ ~35 days), and later a more gradual decrease ($t_{1/2}$ ~160 days). The latter is comparable to the rate of decrease of insulin-antibody binding found by Ionescu-Tirgoviste et al. (20) after stopping insulin treatment in patients with NIDDM, whereas the former is close to the half-life of immunoglobulin G (21). It is possible that this biphasic pattern could be due to the patient's endogenous insulin acting as a minor stimulus for continued antibody production. Certainly the high levels of bound insulin found in the patient's plasma >1 yr after stopping insulin therapy must represent endogenously secreted insulin that had become bound to her insulin antibodies.

DISCUSSION

The question of why hypoglycemia at the degree of severity observed in our patient is not more frequently observed in patients with NIDDM who have been treated with insulin arises for several reasons. First, the titer of insulin antibodies found in our patient and the patient's total plasma insulin were extremely high. Such values are very rare, being found only in cases of immune insulin resistance (2) and autoimmune hypoglycemia (5–9). Second, patients with NIDDM have various degrees of tissue insulin resistance (16). Third, in contrast to patients with IDDM (12), counterregulatory mechanisms are relatively intact in NIDDM patients (22). Fourth, patients with NIDDM still have endogenous insulin secretion and can thus, to a certain extent, buffer the effect of insulin released from insulin antibodies by reducing endogenous secretion of insulin (22).

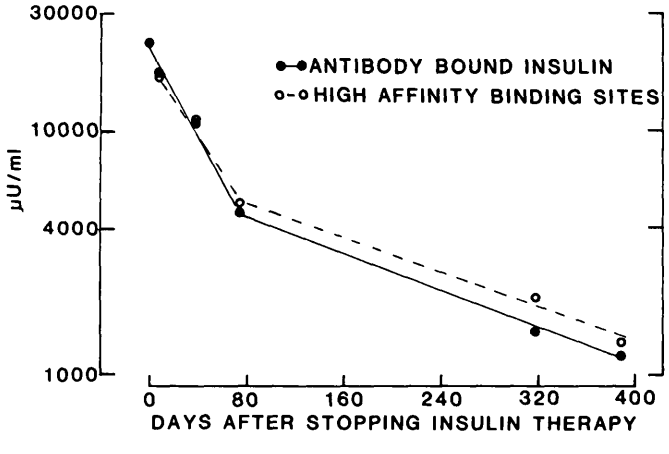

FIG. 2. Changes in amount of insulin bound to insulin antibodies and number of high-affinity insulin binding sites after stopping insulin treatment.

In summary, at the time of the hypoglycemia, the patient had a very high total plasma insulin associated with free-insulin levels that were inappropriate for the level of glycemia. After excluding other known causes of hypoglycemia, it follows that release of insulin bound to insulin antibodies was responsible. As far as we could find in the literature, this is the first report of a prolonged (postabsorptive) hypoglycemia in an insulin-treated patient with NIDDM shown to be due to insulin antibodies. The importance of the high-affinity insulin-antibody binding sites as the reservoir of bound insulin is suggested by the finding that the bound-insulin level was virtually identical with the capacity of the high-affinity insulin binding sites present in the patient's plasma. This is consistent with the recent finding of a positive correlation between apparent distribution space of insulin and the capacity of the high-affinity insulin antibodies in insulin-treated diabetic patients (11). Our study provides evidence that insulin released from these antibodies can give rise to an inappropriate plasma free-insulin level that induces hypoglycemia.

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