Glucose tolerance in subclinical pyridoxine deficiency in man[1,2]

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ABSTRACT   Oral glucose tolerance tests were carried out in 16 clinically normal subjects with pyridoxine deficiency (as adjudged by red cell transaminase activity and the 6h tryptophan load test), and in 16 nondeficient controls. The deficient subjects had fasting normoglycemia with hypoinsulinemia ($p < 0.01$), and a normal postglucose increment in insulin, but peak blood glucose and incremental glucose area were both significantly lower than in the controls ($p < 0.01$). Suppression of growth hormone during the glucose tolerance test was similar in both groups. These data are interpreted as indicating an enhanced sensitivity to the hypoglycemic action of insulin in pyridoxine-deficient individuals. Supplementation with pyridoxine in these subjects resulted in a tendency for insulin levels to rise (NS), and a significant increase in growth hormone levels. Impairment of growth hormone reserve may be the basis for the increased insulin sensitivity. Subclinical pyridoxine deficiency of the degree seen in this study is apparently associated with improved, rather than impaired, glucose tolerance.   *Am J Clin Nutr* 1983;38:440–444.

KEY WORDS   Pyridoxine deficiency, glucose tolerance tests, insulin, growth hormone

Introduction

Studies in rats have shown that pyridoxine deficiency is associated with impaired glucose tolerance (1). Similar abnormalities seen in pregnant women, and oral contraceptive users, have been attributed to concomitant pyridoxine deficiency (2–8). The relevance of these observations to the interpretation of glucose tolerance tests in subjects with nutritional deficiency of vitamin B_6 has not been determined. A previous study from this Institute showed that B_6 deficiency did not contribute to the overall glucose intolerance of diabetics (9). However, the impairment of glucose tolerance produced by B_6 deficiency is relatively modest, and may have been masked by the more severe abnormalities associated with diabetes mellitus. Since subclinical deficiencies of the B group vitamins are common in otherwise normal persons from certain sections of the Indian population, the present study was carried out to determine the effect of subclinical pyridoxine deficiency on glucose tolerance.

Materials and methods

Experimental subjects

Apparently healthy male volunteers, 23 to 32 yr old with a negative family history for diabetes, and a daily dietary intake of 2000 kcal (70 to 80% of energy as carbohydrate), were studied. All the subjects had a normal weight for height (10), a normal skinfold thickness, and were free of any clinical signs of nutrient deficiency. Oral glucose tolerance tests (OGTTs) were carried out after a 12-h overnight fast, with 100 g glucose. On the next day, 5 g l-tryptophan was administered on an empty stomach, and urine was collected over the next 6 h for xanthurenic acid (XA) estimation.

Analytic methods

Blood glucose was estimated by the Nelson-Somogyi method (11), insulin and growth hormone (GH) by double-antibody radioimmunoassays (12, 13) and XA

by colorimetry (14). Erythrocyte glutamic oxaloacetic transaminase (EGOT), and its percentage increase on incubation with an excess of pyridoxal phosphate (EGOT-PALP effect), were estimated by a colorimetric method (15), modified for erythrocytes (16).

Assessment of pyridoxine status

Subjects who had *both* an EGOT-PALP effect exceeding 40% (17), and a 6-h urinary XA excretion in the tryptophan load test of 25 mg or more (14), were considered pyridoxine deficient. The nondeficient controls were negative by both parameters.

OGTT areas

Areas under the glucose and insulin response curves were measured by planimetry, and expressed as mmol/l^{-1}/h, and mU/l^{-1}/h, respectively. Both the total and incremental area—the area enclosed above a line drawn through the fasting glucose level were measured.

Pyridoxine supplementation

Six pyridoxine-deficient subjects were supplemented with 10 mg pyridoxine hydrochloride once a day orally for 10 days. At the end of this period, fasting blood was analyzed for glucose, EGOT, insulin, and GH.

Results

Sixteen subjects were found to be pyridoxine-deficient biochemically, while 16 were taken as nondeficient controls (**Table 1**).

Blood glucose (Table 1, **Fig 1**)

Fasting blood glucose was similar in both groups. However, in the deficient group EGOT-PALP effect correlated inversely with the fasting glucose level (r = −0.53, p < 0.05), but no correlation existed in the control group (r = 0.08, NS). In other words, the more deficient the subject, the lower the fasting glucose tended to be. Total glucose area was also lower in the deficient individuals, but the difference was not significant (p < 0.10). Peak blood glucose and the incremental glucose area were significantly lower in the subjects who were deficient (p <0.01). There was no difference in the time at which the peak glucose occurred in the two groups. In 13 individuals in each group the highest value attained was at 30 min after the glucose load; in the remainder it was at 60 min.

Hormone levels

Fasting plasma insulin was 2½ times higher in the control group compared to the deficient group (p < 0.01). The insulin response to the glucose load was, however, the

TABLE 1
Glucose tolerance, insulin, and GH
in relation to pyridoxine status
(all values are mean ± SEM)

		Nondeficient (n = 16)	Deficient (n = 16)
Basal EGOT activity (mmol/l/h)		85.8 ± 3.68	72.1 ± 6.4*
EGOT-PALP effect (% stimulation)		32.3 ± 1.4	65.3 ± 3.6†
6-h Urinary XA during tryptophan load test (mg)		8.0 ± 2.1	37.5 ± 5.8†
Blood glucose (mmol/l)	Fasting	4.7 ± 0.15	4.5 ± 0.19
	Peak	8.0 ± 0.20	7.0 ± 0.30‡
OGTT glucose area (mmol/l^{-1}/h)	Total	14.9 ± 1.0	13.5 ± 0.7
	Incremental	4.6 ± 0.7	2.8 ± 0.4‡
Plasma insulin (mU/l)	Fasting	18.8 ± 3.2	7.4 ± 2.2‡
	Peak	79.5 ± 9.2	67.7 ± 7.3
OGTT insulin area (mU/l^{-1}/h)	Total	127.4 ± 17.3	97.2 ± 8.5
	Incremental	72.8 ± 7.3	81.4 ± 8.9
Integrated glucose increment per mU increment in insulin	(/umol/mU)	64 ± 5	31 ± 4‡
Plasma growth hormone (μg/l)	Fasting	2.5 ± 0.6	2.1 ± 0.5
	Nadir	Undetectable	Undetectable

* p < 0.05.
† p < 0.001.
‡ p < 0.01, by *t* test.

same in the two groups. Growth hormone levels, both fasting and during the GTT, were similar in the control and the deficient subjects.

Glucose-insulin relationships

The integrated change in blood glucose during GTT associated with the overall increment in insulin, was assessed by the ratio incremental glucose area/incremental insulin area, and expressed as μmol of glucose increment per mU of insulin. In the pyridoxine-deficient group the rise in glucose for every mU of insulin secreted, was less than half that in nondeficient subjects. Since the

insulin secretory response was the same in the two groups, this mathematical relationship may be interpreted as an enhanced sensitivity to the hypoglycemic action of endogenously secreted insulin.

Effect of supplementation (Table 2, Fig 2)

In four of the six deficient individuals who were supplemented with pyridoxine, fasting plasma insulin levels showed an increase; in one patient there was only a small increase, while in the 6th patient the insulin level after supplementation was lower. The mean difference before and after treatment was not significant (2.2 ± 1.3 mU, p < 0.10), probably because of the small numbers. Fasting blood glucose was not affected by supplementation. Plasma GH increased significantly after supplementation (p < 0.01), the
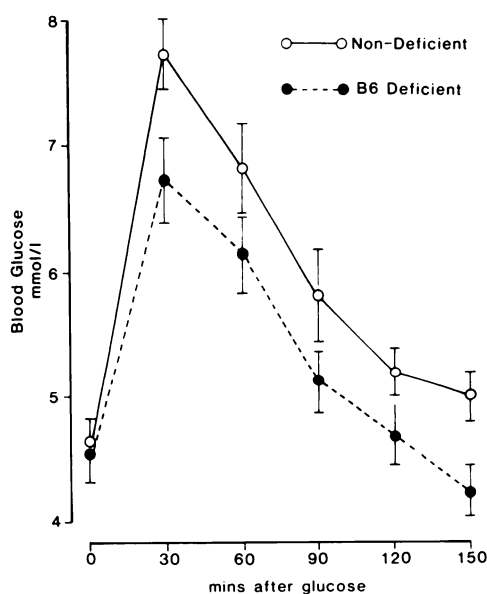

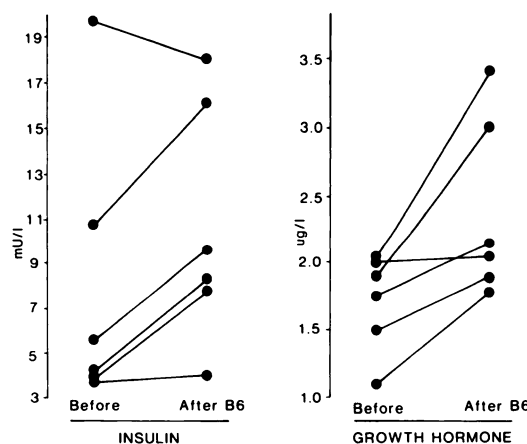FIG 1. Glucose tolerance curves in B₆-deficient subjects and nondeficient controls (mean ± SEM).

FIG 2. Fasting plasma insulin and growth hormone before and after pyridoxine supplementation in six B₆-deficient subjects.

TABLE 2
Effect of pyridoxine supplementation
in six B₆-deficient subjects
(all values are mean ± SEM)

	Before	After	Change
EGOT-PALP effect (% stimulation)	57.6 ± 3.9	32.7 ± 2.5	−25.0 ± 3.4*
Fasting blood glucose (mmol/l)	4.6 ± 0.33	4.3 ± 0.09	−0.31 ± 0.38
Fasting plasma insulin (mU/l)	8.0 ± 2.6	10.2 ± 2.0	+2.2 ± 1.3
Fasting growth hormone (μg/l)	1.7 ± 0.2	2.4 ± 0.2	+0.7 ± 0.2†

* p < 0.001.
† p < 0.01 by paired t test.

entire excursion staying within the normal range of 0 to 5 µg/l.

Discussion

In rats, pyridoxine deprivation results in a fall in insulin levels both in the plasma and in the pancreas (18). This reduction has been attributed to the suppression of insulin synthesis in the pancreas of a pyridoxine-depleted animal (19). The resultant impairment of glucose tolerance is accompanied by an increased sensitivity to the hypoglycemic action of insulin (18, 20–22).

The pyridoxine-deficient subjects studied here show many of the features that have been observed previously after experimental deprivation of pyridoxine in animals. Fasting plasma levels of insulin were significantly lower, and although the fasting hypoglycemia seen in rats (18, 20, 21) was not observed, individuals with a greater functional deficit (ie, a higher EGOT-PALP effect) tended to have lower fasting blood glucose levels. During GTT a similar increment in insulin was associated with a lower peak blood glucose and a smaller incremental glucose area in the deficient subjects than in the nondeficient controls. The lower postglucose increments are unlikely to have been due to malabsorption, since these subjects had no evidence of malnutrition. Moreover, all the subjects came from similar socioeconomic classes, which in earlier studies were shown to consume a diet providing at least 2000 kcal/day (23). Hence malnutrition severe enough to cause malabsorption, and lead to diminished glucose absorption from the gut, is extremely unlikely to have been present. These findings of reduced glucose increments in the face of a normal insulin secretory response can be interpreted as providing indirect evidence of an enhanced insulin sensitivity in the pyridoxine-deficient subjects.

The pathophysiological basis for the increased insulin sensitivity is not entirely clear, but a coexisting deficiency of one of the counter-regulatory hormones is one possible explanation. Such a deficiency, eg, of GH or cortisol, is known to be associated with increased sensitivity to insulin and increased insulin binding at the tissue receptor level (24). In experimental pyridoxine deprivation, the pituitary shows marked depletion of GH (25) although the plasma levels are unaltered (26). Moreover, the pituitary is depleted of pyridoxine well in advance of the pancreas (25). Thus impairment of GH reserve might precede the suppression of insulin synthesis. This would lead to enhanced insulin sensitivity without glucose intolerance, as seen in the subjects studied here. The lower fasting insulin levels and their tendency to rise on pyridoxine supplementation, together with the significant increase in GH levels on repletion, provide supportive evidence for these arguments. The unimpaired glucose tolerance and the normal postglucose increment in insulin suggest that insulin synthesis and reserve were not affected by the degree of pyridoxine deficiency seen here.

Subclinical pyridoxine deficiency was thus associated with an improved rather than impaired glucose tolerance. More severe degrees of deficiency might well lead to a suppression of insulin synthesis, and result in impaired glucose tolerance, as in the experimental animal. However, deficiency of this severity would rarely, if ever, be encountered alone in human subjects. It would therefore be difficult to assess to what extent vitamin B_6 deficiency per se contributes to the glucose intolerance seen in severe undernutrition. ▣

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