CALCANEAL ULTRASOUND ANALYSIS OF NIGERIAN ADULTS WITH TYPE 2 DIABETES

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This work was supported by a Minority International Research Training (MIRT) grant from the Fogarty International Center of the National Institutes of Health.

Abstract

Objective: Type 2 diabetes is a major chronic disease in northern Nigeria. Although type 2diabetes is usually associated with increased bone mineral density, we were interested in estimating the bone mineral density of patients with type 2 diabetes in a region of the world where the usual calcium intake is well below the recommended dietary recommendations.

Research Design and Methods: A total of 50 patients (25M/25F) with type 2 diabetes and 50 healthy controls (20M/30F) were recruited at the Jos University Teaching hospital in Jos, Nigeria. Information regarding age, weight, height, medication use and duration of disease were obtained. Body composition analysis to determine lean body mass and body fat was performed using bioelectrical impedance analysis. Bone quality was assessed using quantitative ultrasound of the calcaneus. Glucose control was monitored using fasting glucose concentrations.

Results: Both male and females subjects with type 2 diabetes had superior ultrasound parameters including broadband ultrasound attenuation (BUA), speed of sound (SOS) and stiffness index (SI) relative to controls. However, there were no significant differences in these parameters between the subjects with diabetes and the controls. No associations between ultrasound parameters and body mass index or body composition were found for either the diabetic subjects or controls. A non-significant trend was observed between glucose control and SI for the female diabetic subjects. A statistically

significant correlation was obtained between SI and duration of disease but only for the female diabetic subjects.

Conclusions: Calcaneal ultrasound is a relatively inexpensive means for monitoring bone quality in patients with type 2 diabetes. The more favorable bone ultrasound parameters observed for patients with type 2 diabetes may be the result of the bone-promoting effects of hyperinsulinemia.

Key words: type 2 diabetes, bone density, calcaneal ultrasound, stiffness index, hyperglycemia, Nigeria

Introduction

Bone mineral density is widely regarded as a primary predictor of bone fracture risk. While most studies of individuals with type 1 diabetes have shown a decrease in bone mineral density (1-7), there is less agreement regarding bone status in patients with type 2 diabetes. Bone mineral density has been reported to be decreased (8-11), increased (12-15) or not significantly different in patients with type 2 diabetes compared to healthy controls (1,13). Despite these discordant findings regarding bone mineral density, it is generally agreed that fracture risk is increased in both type 1 and type 2 diabetes (16-21). One factor that may contribute to high fracture rates in diabetes is the greater number of falls incurred by patients with diabetes as the result of poor vision, peripheral neuropathy, or the consequences of stroke. However, the increased risk for falls persists even after adjusting for these factors (16,22,23). Another explanation for increased fractures in patients with diabetes may be poor bone quality that is not accounted for by bone mineral density alone (24).

The rate of bone loss varies directly with the rate of bone turnover (25,26). Bone turnover has been reported to be lower in patients with type 2 diabetes compared to healthy controls or subjects with type 1 diabetes (8,27). However, even though a decrease in bone turnover may result in a higher bone mineral density, it may have an additional effect on bone that is unrelated to mineral content. For example, slow turnover of bone may permit the accumulation of microdamage in bone resulting in poorer bone quality and decreased strength, and an increased risk for fracture (28). In addition, the formation of advanced glycation end-products (AGEs) in patients with poor glucose control may decrease bone strength and increase susceptibility to fracture. In human cadaver bone, it has been shown that higher concentrations of AGEs were associated with decreased bone strength (29). Although the mechanism relating the concentration of AGEs to bone strength has yet to be determined, it has been suggested that AGEs may alter the physical properties of bone collagen (30). Angular deformation and decreased torsional strength and energy absorption were found in the bones of rats with either spontaneous or experimentally induced diabetes (31-34). These changes in

bone properties were observed in the absence of changes in bone mineral density or bone mineral content.

Most studies that have investigated the bone status of patients with diabetes have used single photon or dual X-ray absorptiometry (DEXA). Although DEXA is considered the gold standard for the estimation of bone mineral density, it provides information only about bone size and mineral content. However, physical and structural properties of bone such as microarchitecture and elasticity influence bone strength independent of mineral content. *In vitro* studies of cadaver bones have demonstrated that quantitative ultrasound can provide information about bone elasticity and strength (35). In a study of diabetic patients with acute Charcot osteoarthropathy (CO), which is characterized by bone and joint destruction in the foot as a result of neuropathy, Jirkovska and coworkers (36) reported that subjects with CO had significantly lower calcanel ultrasound parameters than controls without diabetes. These authors suggested that calcaneal ultrasonometry may be useful in assessing the risk of foot fracture in CO.

Type 2 diabetes is common in Nigeria and other regions of sub-Saharan Africa and it is associated with high morbidity and mortality. These conditions are exacerbated by the lack of accessibility to methods for monitoring glucose control. In addition, this disease occurs in an environment where lifetime dietary calcium intake is well below the recommended intake of calcium (37), a circumstance which could impede attainment of peak bone mass. We therefore used calcaneal ultrasound to compare the bone quality of men and women with type 2 diabetes in northern Nigeria versus that of healthy age- and gender matched controls from the same region. Our aim was to determine if type 2 diabetes has a deleterious effect on bone status in that environment.

METHODS

Study population. This study was conducted in the city of Jos, which is located in Plateau State in northern Nigeria. Subjects were recruited from the Diabetes Clinic and the General Outpatient Clinic at the Jos University Teaching Hospital, which provides care to patients from a broad range of socioeconomic classes, with the majority of patients representing the lower economic stratum. Subjects were excluded if they were pregnant, less than 15 years of age or greater than 70 years of age, or had a history of bone disease or trauma. A total of 100 subjects were recruited into this study, including 50 adults with type 2 diabetes (25 males, 25 females) and 50 controls. The control subjects (30 males, 20 females) were matched for gender, age, height and weight to the subjects with diabetes. For the subjects with diabetes, the duration of the disease was recorded. Glucose control was determined using the fasting blood glucose concentration. The following criteria were

applied: serum glucose of ≤ 5.6 mmol/L, good; > 5.6 -6.9 mmol/L,satisfactory; 7-10 mmol/L, maginal; > 10 mmol/L, poor. The majority of the subjects were taking metformin and glibenclamide. Only three female patients and four male patients with diabetes were receiving insulin.

Written informed consent was obtained from each subject following a detailed explanation of the aims of the study and its requirements in English or Hausa, the predominant native language in Jos, by a medical personnel fluent in the language. This study was approved by the Ethics Review Committee of the Jos University Teaching Hospital, Jos, Nigeria and by the Human Research Review Committee of the University of New Mexico, Albuquerque, New Mexico, USA.

Anthropometric measurements. The weight of each subject was determined using a battery-operated scale accurate to 0.5 kg and height was measured to within 0.25 cm using a portable stadiometer. Blood pressure was measured by a physician using a nylon cuff and latex inflation system (Prestige Medical, Inc, Northridge, CA). Body composition analysis to determine fat-free mass and body fat was conducted using bioelectrical impedance analysis (RJL Systems, Inc, Detroit, MI). Reactance and resistance values, weight, height, gender, and self-reported age were used to calculate fat-free mass and body fat with the software provided by the manufacturer.

Bone ultrasound measurements. The ultrasound analysis was performed using the Achilles^{+} Solo ultrasound (Lunar Corporation, Madison, WI, USA). Each subject was seated with their right foot placed in the heel bath of the instrument. Measurements of the speed of sound transmission (SOS m/sec) and the broadband ultrasound attenuation (BUA) were made following the introduction of water containing surfactant into the heel bath. BUA is defined as the slope of the regression line derived from the ratio of the signal amplitude of the calcaneus to that of water (reference) at each frequency of ultrasound (dB/MHZ). The SOS is a measurement of the time it takes for the soundwave to pass through the heel compared to the time required for the signal to pass through the water bath alone. The SI is calculated as (0.67 X BUA) + (0.28 XSOS) – 420. Reproducibility was monitored by measuring the same control subject each day the study subjects were measured. The calibration of the instrument was checked weekly using a phantom heel provided with the instrument.

Statistical analysis . Descriptive statistics and group comparisons were performed using the Number Crunching Statistical Software program (NCSS 2000, Kaysville, UT). Comparisons of anthropometric and ultrasound parameters between the patients and controls were made using the two-sample *t* test. Relations between various anthropometric characteristics and ultrasound parameters

were tested using regression analysis. A p-value of 0.05 was considered statistically significant.

RESULTS

Subjects. A summary of the characteristics of the patients with type 2 diabetes and their corresponding controls is given in Table 1. Although the male subjects with diabetes were slightly older than the control subjects (53.3 vs. 48.9 years, respectively) there was no significant difference in age between the two groups. There also was no significant difference between the male subjects with diabetes and male controls for any other of the anthropometric characteristics (weight, height, BMI, body fat, per cent body fat, fat-free mass and per cent fat-free mass). The median time since diagnosis for the male subjects was 6 years and ranged from 1 to 24 years.

As was seen with the male subjects, there were no significant differences in any of the anthropometric characteristics between the female subjects with diabetes and the controls (Table 1). The median time since diagnosis for the female subjects was 5 years and ranged from 1 month to 28 years.

Ultrasound parameters.

A summary of the ultrasound analyses for the diabetic subjects and controls is shown in Table 2. Although the male subjects with diabetes had higher mean BUA and SOS values than the male controls, the differences were not statistically significant. The mean SI, which is calculated from the BUA and SOS data, was also higher for the male subjects with diabetes, but was not significantly different from that of the controls.

The female subjects had comparable BUA values but slightly higher SOS and SI values than the female controls. As was true for the male subjects, there were no statistically significant differences in the ultrasound parameters between the patients with diabetes and the female controls. The SI values we report herein for male and female patients and controls are similar to those we found previously for healthy Nigerian adults (38)

When the relation between the ultrasound parameters (BUA, SOS and SI) and anthropometric characteristics for both male and female subjects were examined using regression analysis, no significant correlations were found between any of the parameters and weight, height, or BMI. The only trend we observed was a negative relation between age and SI in the female controls ($r = -0.43$, $p = 0.059$). A significant positive relation between duration of disease and SI was obtained for the female subjects with diabetes (Fig. 1, $r = 0.45$, $p = 0.02$) but not for male patients with diabetes. In addition, a non-significant trend was observed for the relation between SI and glucose control only for the female diabetics (Fig. 2).

Figure 1.

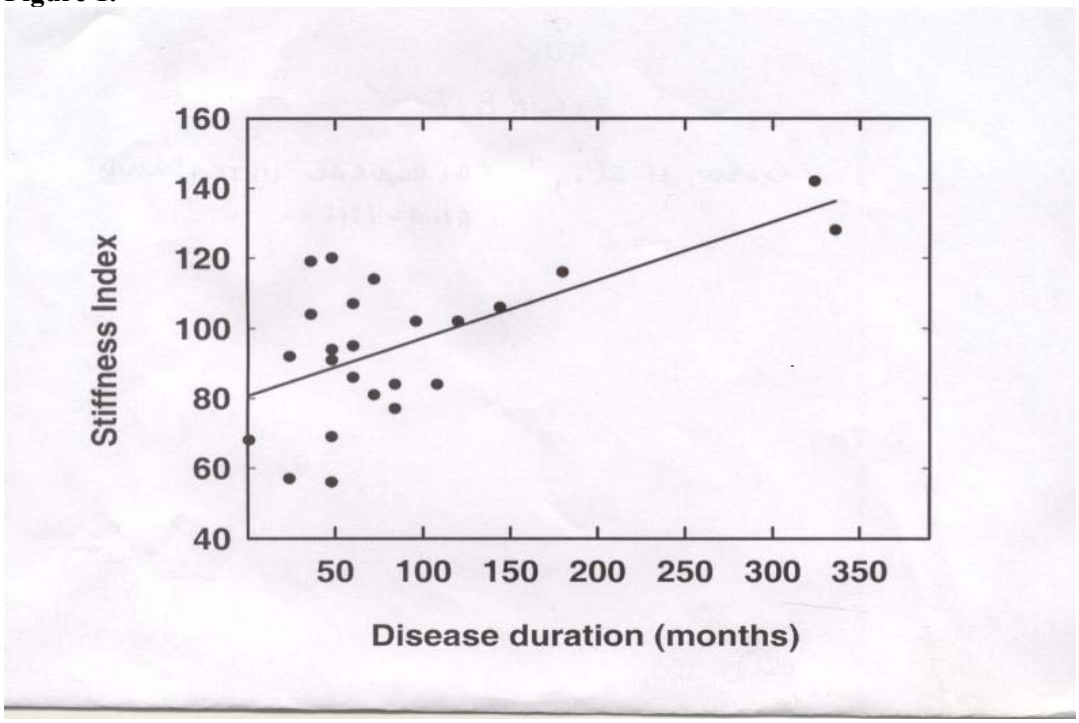

Fig. 1. The relation between stiffness index and duration of disease in female patients with type 2 diabetes (r = 0.45, p = 0.02).

Figure 2

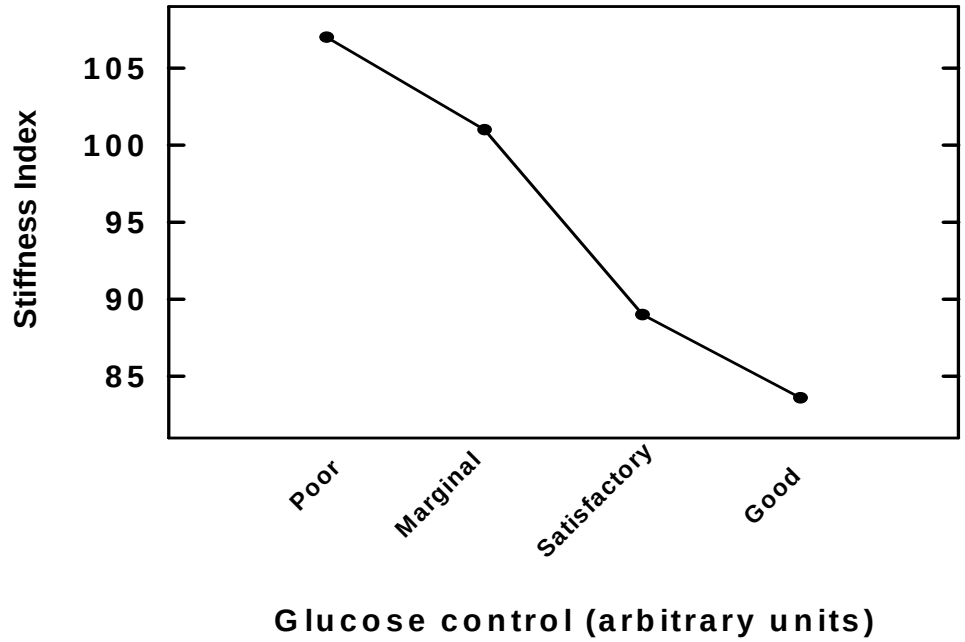

Fig. 2. The relation between glucose control and stiffness index in female patients with type 2 diabetes. See Methods section for classification with respect to glucose control.

Table 1. Summary of the anthroprometric characteristics of the subjects with diabetes and the healthy controls

	Males		Female
Diabetic subjects (n = 25) Parameters (Mean ± SD)	Controls (n = 30) (Mean ± SD)	Diabetic subjects (n = 25) (Mean ± SD)	Controls (n = 20) (Mean ± SD)
Age (years) 53.3 ± 8.5	48.9 ± 12.0	48.1 ± 11.0	49.2 ± 15.0
Weight (kg) 72.3 ± 15.0	72.2 ± 12 .0	69.6 ± 13.0	69.8 ± 13.0
Height (m) 1.70 ± 0.06	1.68 ± 0.1	1.58 ± 0.06	1.58 ± 0.07
BMI (kg/m^2) 24.9 ± 4.4	25.5 ± 3.5	27.8 ± 4.4	28.1 ± 5.2
Phase Angle 6.31 ± 0.80	6.60 ± 1.00	6.35 ± 0.90	6.05 ± 0.80
FFM (kg) 59.0 ± 7.9	58.6 ± 7.4	41.3 ± 4.7	41.1 ± 4.3
FFM percent 82.8 ± 5.9	81.8 ± 4.8	60.5 ± 6.2	60.4 ± 8.7
Fat (kg) 13.2 ± 7.1	13.6 ± 5.7	28.0 ± 10.0	28.5 ± 11
Fat percent 17.2 ± 5.9	18.2 ± 4.8	39.5 ± 6.2	39.6 ± 8.7

Abbreviations: FFM, fat-free mass; PA, phase angle

There was no statistically significant difference in any parameter between the diabetic subjects and their respective gender controls.

Table 2. Summary of the ultrasound parameters for the subjects with diabetes and the healthy controls

Males		Females	
Diabetic subjects (n = 25) Parameters (Mean ± SD)	Controls (n = 30) (Mean ± SD)	Diabetic subjects (n = 25) (Mean ± SD)	Controls (n = 20) (Mean ± SD)
BUA (dB/MHz) 128 ± 16	119 ± 16	122 ± 21	122 ± 11
SOS (m/sec) 1565 ± 49	1550 ± 46	1554 ± 47	1545 ± 31
SI 102 ± 23	93 ± 22	97 ± 24	92 ± 16

Abbreviations: BMI, body mass index; BUA, broadband ultrasound attenuation; SOS, speed of sound; SI, stiffness index.

There was no statistically significant difference in any ultrasound parameter between the diabetic subjects and their respective gender controls.

Discussion

Using calcaneal ultrasound to assess the effect of diabetes on bone quality, we found that the ultrasound properties of the calcaneous of both the men and women with type 2 diabetes in Nigeria compared favorably to that of their healthy counterparts. This conclusion is borne out by the fact that there were no statistically significant differences between the controls and diabetic subjects with respect to the three ultrasound parameters, SI, SOS and BUA.

Our findings using calcaneal ultrasound to assess the bone quality agree with those of previous studies that used DEXA or single-photon absorptiometry. In the Rotterdam Study, van Daele and coworkers found that adult patients with type 2 diabetes had normal to increased bone mineral density (12). A study of the bone mineral density of Japanese patients with type 2 diabetes (39) also found that patients with good glycemic control were protected against bone loss. A similar study by Christensen and Svendsen in Denmark (40) concluded that postmenopausal women with type 2 diabetes had a higher BMD than non-diabetic controls and seemed to have been protected from bone loss by their diabetic condition .

There are several factors that could potentially affect bone status in patients with diabetes. These include: duration of disease, obesity, hyperinsulinemia, hypercalciuria

(associated with glycosuria), impaired renal function, increased levels of advanced glycation end-products, microvascular complications, altered concentrations of anabolic hormones and elevated cytokines. These factors may have opposing effects on bone density.

Because a high body mass has been shown to be positively correlated with bone mineral density, the increased bone density observed in type 2 diabetes has been attributed to the increased weight or obesity commonly seen in patients with type 2 diabetes (16,41). In our own study of patients with type 2 diabetes the patients and controls were closely matched with regard to age, weight and height (Table 1), and the mean BMI values were identical between the two groups. This indicates that some other factor besides weight must have been contributing to accrual of bone mass in these Nigerian diabetic subjects.

Type 2 diabetes is characterized by a period of insulin resistance and hyperinsulinemia that precedes the onset of the disease. Although glucose transport may be adversely affected, the other anabolic effects of insulin may still be operable. Insulin may act directly on its own receptor in the bone or by binding to the receptor for insulin-like growth factor (IGF1). Insulin-like growth factor-1 plays an important role in bone remodeling. Its overall anabolic effect on bone is the result of suppression of bone collagen degradation, enhancement of bone matrix deposition, and recruitment of osteoblastic cells (42,43). In a study of type diabetic men and women in the U.S., Barrett-Connor and Kretz-Silverstein (44) found that among women at least, fasting insulin was significantly and positively associated with bone density of the radius and spine and was independent of age, BMI, waist-hip ratio, or other factors associated with bone density. The authors concluded that hyperinsulinemia may be responsible in part for the observed association of both diabetes and obesity with BMD in women. Dennison and coworkers (12) also concluded that the greater bone mineral density found in patients with type 2 diabetes is the result of hyperinsulinemia. We did not have measurements of the subjects' insulin levels in the present study. However, because all of the diabetic patients in the present study were taking glucose-lowering drugs (metformin or glibenclamide) and only a few were receiving insulin, we assume that the majority of subjects with diabetes had some residual endogenous insulin.

Since HbA_{1c} measurements were not widely available in Nigeria at the time our study was conducted, glucose control in patients with diabetes was assessed using fasting glucose concentrations. When we examined the relation between SI and glucose control based on fasting glucose concentration, a negative trend was observed between the SI value and the degree of glucose control for the female diabetics (Fig. 1). This finding is contrary to what we anticipated, since the glucosuria associated with hyperglycemia would be expected to

lead to increased urinary excretion of calcium. The higher glucose concentrations in our female subjects with diabetes may be a reflection of hyperinsulinemia resulting from decreased insulin sensitivity.

Hyperinsulinemia may also increase bone density by its negative association with sex-hormone-binding globulin (SHBG) which is the major sex hormone binding protein in plasma (45,46). Lower levels of SHBG would result in increases in the concentrations of free androgens that promote bone synthesis. A negative correlation between SHBG and bone mineral density in women with polycystic ovary disease has been reported (47). An unexpected but interesting observation we made in the present study and which may be related to SHBG was the gender-specific effect disease duration seemed to have on SI. We found a strong positive relation between SI and duration of disease in the female subjects with type 2 diabetes (Fig. 2), suggesting that in women at least, the disease provides a significant degree of benefit to skeletal health. We speculate that this apparent gender-specific advantage may be due to both prolonged hyperinsulinemia and a persistently low SHBG level in the women with type 2 diabetes.

Because of the agreement between our data and the results of other studies of the bone properties of diabetics that were conducted using dual-photon absorptiometry or DEXA, our findings indicate that calcaneal ultrasound measurements are useful to monitor the bone quality of patients with diabetes. Calcaneal ultrasound is a less expensive and portable alternative to DEXA or other absorptiometry methods for estimating bone mineral density, making it suitable for use in both developed and developing countries. It would be informative to conduct a long-term longitudinal study of patients with type 2 diabetes using calcaneal ultrasound to assess bone quality as they progress through the clinical course of their disease.

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