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Original Article

Vitamin D levels and insulin resistance in young Chinese obese men with normal glucose tolerance

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Abstract: This case-control study was aimed to evaluate serum 25-hydroxyvitamin D [25(OH)D] levels in young Chinese obese men with normal glucose tolerance (NGT) and its relationship with insulin resistance (IR). A total of 157 normal-NGT men aged 25 to 40 years were enrolled, including 57 obese men (BMI≥28.00 kg/m^2), 58 overweight men (24.00 kg/m^2≤BMI<28.00 kg/m^2), and 42 healthy normal-weight controls (18.50 kg/m^2≤BMI<24.00 kg/m^2). Fasting blood glucose, insulin and serum 25(OH)D were measured. Homeostatic model assessment of insulin resistance (HOMA-IR) was calculated. The largest hip circumference, highest fasting insulin oral glucose tolerance and HOMA-IR were found in obese men (P<0.003), and their HOMA-IR was significantly higher than that in normal-weight controls (P≤0.001). Normal-weight men had highest 25(OH)D_3 levels while obese men had significantly lower 25(OH)D by comparison (both P<0.001). Among all subjects, 5.1% (8/157) had sufficient 25(OH)D levels, 8.9% (14/157) had insufficient levels, 78.4% (123/157) had mild vitamin D deficiency and 7.6% (12/157) had severe vitamin D deficiency. In obese patients, waist circumference (WC) correlated positively with changes in HOMA-IR levels, while 25(OH)D_3 levels correlated negatively with HOMA-IR levels. In multivariable models, higher WC and lower 25(OH)D_3 in obese patients were associated with increased HOMA-IR. Vitamin D deficiency is common among young Chinese obese men with NGT, and serum 25(OH)D and IR are independently associated.

Keywords: Insulin resistance, obesity, vitamin D, waist circumference

Introduction

Vitamin D deficiency is present in epidemic proportions globally, with an estimated prevalence as high as 64% in the general population, including only minor differences associated with age and gender [1]. The deficiency can result from inadequate exposure to sunlight, low consumption of food containing ergocalciferol, malabsorption of calcium, and accelerated catabolism derived from taking certain medications [2, 3]. Besides acting as a regulatory hormone in calcium metabolism, vitamin D is essential for noncalciotropic effects such as cellular differentiation and replication in many organs. As such, deficiency of vitamin D is significantly associated with the development of diabetes mellitus, hypertension, hyperlipidemia and peripheral vascular disease [1, 2]. The effects of vitamin D deficiency on adiposity, metabolic syndrome, coronary heart disease, and renal disease have also been gaining attention [1, 4, 5]. Together, deficiencies in vitamin D and calcium have been shown to negatively influence glycemia and studies consistently show associations with type 2 diabetes mellitus, insulin resistance (IR), and metabolic syndrome [2, 4, 6]. In fact, the noted National Health and Nutrition Examination Survey (NHANES) study (2001-2006) revealed that abdominal obesity and insufficient serum 25(OH)D concentration jointly influence IR risk in diabetic subjects [7].

Previous studies have reported that 25-hydroxyvitamin D, or 25(OH)D, the commonly accepted indicator of vitamin D status, is inversely associated with adiposity and IR [8-12]. However, results of other studies failed to confirm this [13-17]. For example, a study among elderly subjects in the Netherlands found no significant association between 25(OH)D and the incidence of diabetes or with results of 2-hour oral glucose tolerance tests [13]. These

discrepancies in study results may be ascribed to different study populations with varying ethnicity, sex, and age, or other variables. A recent study, for example, reported that inverse associations between 25(OH)D and IR markers were demonstrated in males but not females [18].

Metabolic syndrome, including the component of abdominal adiposity, has become a widespread health problem in Asian countries, with prevalence increasing markedly during the past twenty years in response to rapid transitions in nutrition and lifestyle [19]. In China, one study reported that lower circulating 25(OH)D levels were inversely associated with higher fasting insulin and homeostasis model assessments of insulin resistance (HOMA-IR) in a middle-aged to elderly Chinese population, and also associated with increased risk of metabolic syndrome [12]. Another recent study found that vitamin D levels in a large cohort of healthy non-diabetic Chinese females were significantly and independently associated with IR and β-cell function [17]. Meanwhile, although studies in China and major studies in other populations, including a young adult population [18], confirm an association between vitamin D and IR, no such association has been demonstrated in younger Chinese adults with normal glucose tolerance (NGT), and it is also not known whether data from western studies can be extrapolated to Chinese populations. With known increasing trends in obesity and vitamin D deficiency in China, we hypothesized that the relationship between vitamin D and IR could possibly be demonstrated in younger Chinese men of different body weight status and with NGT. The significance and novelty of this study is to make contribution to relate important aspects of young Chinese obese men to the current literatures. The purpose of this study was to investigate serum 25(OH)D concentration and its association with IR in young Chinese obese men with NGT. Results of this study may increase awareness of a need to improve vitamin D nutritional status in the Chinese population, particularly for young Chinese obese men.

Subjects and methods

Study design

This prospective, case-control study enrolled 157 consecutive obese, overweight and normal-weight men who had received routine health examinations at the First Hospital of Qinhuangdao between December 2011 and February 2012 and who met the inclusion criteria (below). The study protocol was approved by the ethics committee of the Hebei Medical University. All subjects provided signed informed consent prior to study initiation.

Study subjects

Weight was classified according to the 2003 diagnostic criteria for Chinese obesity (Guide for Control and Prevention of Overweight and Obesity in Chinese Adults 2003) as follows: normal weight (18.50 kg/m^2≤BMI<24.00 kg/m^2), overweight (24.00 kg/m^2≤ BMI<28.00 kg/m^2), and obese (BMI≥28.0 kg/m^2). Subjects included 57obese men (mean age 33.54±4.62 years, BMI 30.51±2.60 kg/m^2), 58 overweight men (mean age 33.41±4.56 years, BMI 25.98±1.20 kg/m^2), and a control group of 42 healthy normal-weight men (mean age 33.02 ±3.97 years, BMI (22.41±1.46 kg/m^2). Blood samples were drawn from all subjects from December 2011 to February 2012. All subjects were of Han ethnicity, and each underwent an oral glucose tolerance test (OGTT) initiated at 8:00 am with administration of 75 g of oral anhydrous glucose. Peripheral venous blood samples were taken at 0 and 120 min after glucose loading. Inclusion criteria were: (i) subjects were clinically stable with no previous medical history of diabetes, hypertension, dyslipidemia, coronary artery diseases, or cerebral stroke; (ii) subjects were obese with no clinical evidence of endocrinopathy; (iii) subjects had fasting plasma glucose (FPG) levels <5.6 mmol/l and 2-h plasma glucose (2-h PG) levels <7.8 mmol/L after a 75 g OGTT, based on the 2008 diagnostic criteria of the American Diabetes Association; and (iv) subjects were not taking medications known to affect glucose metabolism, including glucocorticoids, thyroid hormones, or thiazide diuretics. Exclusion criteria were: (i) subjects with hepatic or renal dysfunction, (ii) subjects taking vitamin and/or mineral supplements, and (iii) subjects with acute and chronic inflammation,

Measurements

A detailed history was taken for all enrolled subjects, including lifestyle behaviors (e.g., smoking, alcohol intake), and a physical examination was performed, including body mass (kg),

Table 1. Subjects' baseline demographic, anthropometric and clinical characteristics

	Normal (n=42)	Overweight (n=58)	Obese (n=57)	*P*
Age, year	32.5 (31.0, 36.0)	34.0 (29.0, 37.0)	35.0 (31.0, 36.0)	0.736
BMI, kg/m^2	22.8 (21.8, 23.5)	25.9 (25.1, 27.0)*	29.6 (28.9, 30.5)*,†	<0.001
WC, cm	82.7±6.8	91.9±5.5	103.1±7.1*,†	<0.001
HC, cm	96 (92, 100)	98 (96, 100.5)*	105 (103, 110)*,†	<0.001
WHR	0.9±0.1	0.9±0.1	1.0±0.04*,†	0.001
SBP, mmHg	120 (110, 130)	129 (120, 140)	130 (130, 140)*,†	<0.001
DBP, mmHg	80 (75, 80)	80 (80, 90)	85 (80, 90)*,†	0.001
Scr, mmol/L	74.4 (70.0, 79.3)	71.8 (66.1, 76.6)	74.0 (69.7, 78.1)	0.077
Urea, umol/L	5.7 (4.6, 6.6)	5.6 (4.5, 6.1)	5.8 (4.8, 6.6)	0.373
UA, umol/L	304.5±67.7	342.5±100.9	335.8±96.6	0.106
TG, mmol/L	1.4 (0.8, 2)	1.6 (1.2, 2.4)	1.8 (1.3, 2.9)*	0.008
TC, mmol/L	4.4±1.3	4.3±1.3	5.2±1.0*,†	0.001
HDL-C, mmol/L	1.2 (1.1, 1.6)	1.1 (1, 1.3)	1.1 (1, 1.3)	0.086
LDL-C, mmol/L	2.3 (2.0, 3.1)	3.0 (2.1, 3.3)	3.0 (2.3, 3.9)*	0.019
FPG, mmol/L	4.5 (4.2, 5.3)	4.8 (4.5, 5.4)	4.6 (4.3, 5.2)	0.280
Plasma glucose 2-h OGTT, mmol/L	5.1±1.2	5.8±1.4*	5.8±1.1*	0.009
HbA_1c, %	5.3 (4.8, 5.8)	5.5 (5.0, 5.9)	5.6 (5.2, 5.9)	0.081
Fasting insulin OGTT, uIU/mL	3.3 (2.1, 6.2)	6.6 (3.3, 10.8)*	11.6 (8.3, 15.2)*,†	<0.001
HOMA-IR	0.6 (0.4, 1.4)	1.3 (0.7, 2.4)*	2.2 (1.7, 2.9)*,†	<0.001
Calcium, mmol/L	2.3 (2.2, 2.3)	2.3 (2.3, 2.3)	2.3 (2.2, 2.4)	0.813
Phosphorus, mmol/L	1.2 (1.2, 1.3)	1.2 (1.1, 1.3)	1.2 (1.1, 1.3)	0.368
PTH, pg/ml	36.7±7.5	37.8±9.3	39.6±11.8	0.335
$25(OH)D_3$, nmol/L	45 (35.1, 55)	37.2 (31.7, 40.9)*	32.1 (26.7, 37.1)*,†	<0.001
<25	0 (0)	2 (3.4)	10 (17.5)	<0.001
25-49	28 (66.7)	49 (84.5)	46 (80.7)	
50-74	7 (16.7)	6 (10.4)	1 (1.8)	
>74	7 (16.7)	1 (1.7)	0 (0)	
Smoking	16 (38.1)	27 (46.6)	28 (49.1)	0.535
Drinking	19 (45.2)	31 (53.4)	33 (57.9)	0.457

Data on continuous variables with normal distributions are presented as mean ± standard deviation and tested by analysis of variance; other interval variables with skewed data distributions are expressed as medians (interquartile range) and tested by Kruskall-Wallis test. Abbreviations: BMI, body mass index; WC, waist circumference; HC, hip circumference; WHR, waist hip ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure; Scr, serum creatinine; UA, uric acid; TG, triglyceride; TC, total cholesterol; HDL-C, high density lipoprotein cholesterol; LDL-C, low density lipoprotein cholesterol; FPG, fasting plasma glucose; OGTT, oral glucose tolerance test; HOMA-IR, homeostasis model assessment insulin resistance; PTH, parathyroid hormone; $25(OH)D_3$, 25-Hydroxyvitamin D. *Indicates significantly different from normal group, $P<0.017$. †Indicates significantly different from normal group, $P<0.017$.

height (cm), and body mass index (BMI) calculated as kilograms per square meter (kg•m^{-2}), waist circumference (WC), hip circumference (HC), waist/hip ratio (WHR, WC/HC) and blood pressure (BP). Weight in light clothing was measured, height was measured to the nearest 0.05 cm, WC and HC evaluated to the nearest 0.01 cm. Diastolic blood pressure (DBP) and systolic blood pressure (SBP) measurements were recorded twice on the right arm using a standard mercury sphygmomanometer after 15 min rest while subjects were seated, and the average of the two measurements was used for analysis. All aforementioned physical examinations were done while patients admitted to hospital.

Plasma glucose levels were measured using the glucose oxidase method. Total cholesterol (TC), high-density lipoprotein cholesterol (HDL), low-density lipoprotein cholesterol (LDL), and triglycerides (TG) were measured, as well as

Table 2. Univariate analysis of HOMA-IR in three groups by BMI

	Normal (n=42)	Overweight (n=58)	Obese (n=57)
	γ_s (P)	γ_s (P)	γ_s (*P*)
Age, years	0.033 (0.837)	0.019 (0.887)	0.031 (0.817)
BMI, kg/m^2	0.184 (0.244)	0.082 (0.542)	0.217 (0.105)
WC, cm	0.177 (0.262)	0.143 (0.284)	**0.299 (0.024)**
HC, cm	0.285 (0.067)	0.159 (0.233)	0.238 (0.075)
WHR	-0.046 (0.771)	0.042 (0.753)	0.199 (0.139)
SB, mmHg	0.081 (0.610)	0.078 (0.559)	-0.204 (0.128)
DBP, mmHg	0.222 (0.157)	0.096 (0.473)	-0.141 (0.296)
SCr, mmol/L	0.045 (0.778)	0.012 (0.927)	0.138 (0.305)
Urea, umol/L	-0.043 (0.786)	-0.025 (0.855)	0.004 (0.977)
UA, umol/L	-0.069 (0.666)	0.113 (0.398)	-0.005 (0.970)
TG, mmol/L	-0.041 (0.795)	0.142 (0.287)	0.226 (0.091)
TC, mmol/L	**-0.311 (0.045)**	-0.081 (0.544)	0.205 (0.127)
HDL-C, mmol/L	0.049 (0.759)	-0.101 (0.451)	-0.006 (0.966)
LDL-C, mmol/L	-0.112 (0.479)	-0.128 (0.339)	0.149 (0.270)
Plasma glucose 2-h OGTT, mmol/L	0.029 (0.855)	-0.033 (0.805)	0.020 (0.880)
HbA_1c, %	-0.056 (0.725)	0.193 (0.146)	-0.022 (0.873)
Calcium, mmol/L	0.046 (0.773)	0.058 (0.663)	0.033 (0.807)
Phosphorus, mmol/L	0.108 (0.497)	0.064 (0.633)	0.004 (0.976)
PTH, pg/ml	0.107 (0.498)	0.154 (0.249)	-0.004 (0.975)
25(OH)D_3, nmol/L	-0.020 (0.901)	-0.078 (0.561)	**-0.328 (0.013)**

Abbreviations: BMI, body mass index; WC, waist circumference; HC, hip circumference; WHR, waist hip ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure; SCr, serum creatinine; UA, uric acid; TG, triglyceride; TC, total cholesterol; HDL-C, high density lipoprotein cholesterol; LDL-C, low density lipoprotein cholesterol; FPG, fasting plasma glucose; OGTT, oral glucose tolerance test; PTH, parathyroid hormone; 25(OH)D_3, 25-Hydroxyvitamin D. Bold values indicate statistical significance, $P<0.005$.

hepatic and renal function, using enzymatic assays with a Hitachi autoanalyzer (Hitachi, Tokyo, Japan). Serum 25(OH)D levels were estimated by radioimmunoassay procedure. The intra- and inter-assay coefficients of variation were 5.3% and 4.6%, respectively (IDS Corporation, Hampshire, UK). Plasma concentrations of insulin were measured by radioimmunological assay using a commercially available kit (North Institute of Biological Technology, Beijing, China). Insulin resistance was estimated from fasting plasma measurements using HOMA-IR (insulin (mU/L) × glucose (mmol/L)/22.5), as described previously [20]. Vitamin D status was classified as severely deficient, mildly deficient, insufficient, or sufficient [serum 25(OH)D: <25 nmol/L, 25~49 nmol/L, 50~74 nmol/L and ≥75 nmol/L, respectively], as described previously [21, 22].

Statistical analysis

Continuous variables with normal distributions (i.e., WC, WHR, uric acid (UA), TC, 2 h-PG-OGTT, and parathyroid hormone (PTH)) are reported as mean ± standard deviation, and other continuous variables are expressed as median (interquartile range, P_{25}-P_{75}). Categorical data are shown as frequency (%). Differences in interval variables with normal distributions, non-normal distributions and categorical parameters were analyzed by analysis of variance (ANOVA), Kruskal-Wallis, and chi-square test, respectively. When significant differences were revealed among any of the aforementioned statistical methods, post-hoc tests were then implemented with Bonferroni correction for normally distributed variables, Mann-Whitney U test for non-normally distributed interval parameters, and chi-square test for categorical variables, respectively.

Spearman's rank correlation was used to identify effectors of HOMA-IR in univariable analysis. Square root transformation was applied to HOMA-IR before performing linear regression analysis. Significant variables with $P<0.05$ were then placed in multiple linear regression mod-

Table 3. Multiple regression analysis for effectors of homeostasis model assessment insulin resistance (HOMA-IR)[1] in obese patients

	Obese (n=57)	
	β (95% confidence interval)	*P*
Waist circumference, cm	0.018 (0.006, 0.030)	**0.004**
25-Hydroxyvitamin D, nmol/L	-0.016 (-0.026, -0.005)	**0.004**

[1]Square root transformation was applied to data on HOMA-IR before proceeding linear regression with backward selection. R^2 was 0.323 for model of obese group respectively. Bold values indicate statistical significance, *P*<0.005.

els and backward selection was applied. *P*< 0.05 was considered statistically significant. Significance level was adjusted to 0.017 (0.05/3) or 0.008 (0.05/6) as appropriate. All analyses were two-sided and performed using SPSS 22 statistical software (IBM Corp., Armonk, NY, USA).

Results

Baseline characteristics of 157 subjects

A total of 157 males between 25 and 40 years were enrolled in this study and classified into three groups based on BMI, including an overweight group of 58 men, an obese group of 57 men, and a control group of 42 normal-weight men. Subjects' baseline characteristics are summarized in **Table 1**. The three groups of patients shared similarities in age, serum creatinine, urea, UA, HDL-C, fasting plasma glucose (FPG), HbA_1c, calcium, phosphorus, PTH, and proportions of patients who reported smoking and drinking.

The largest HC, fasting insulin OGTT, and HOMA-IR, were found in the obese group (*P*≤0.003). The highest $25(OH)D_3$ level was found in the normal group, followed by overweight group and obese group (all *P*≤0.001). The WC, WHR, SBP, DBP, and TC in the normal and overweight groups were similar and all of these values in the obese group were higher than in both of the other groups (both *P*<0.009). The overweight group and obese group had similar high levels of 2-h G-OGTT in contrast to those of the normal group, which had low levels of this parameter (both *P*≤0.004). Obese patients also had higher levels of TG and LDL-C than those with normal BMI (*P*=0.003 and *P*=0.006, respectively).

Effectors of HOMA-IR

In univariate analysis, no statistically significant results were found in the overweight group, but HOMA-IR levels were influenced significantly by one factor in the normal group and by two factors in the obese group. In patients with normal BMI, HOMA-IR levels increased as TC levels were reduced. In obese patients, WC correlated positively with changes in HOMA-IR levels, while $25(OH)D_3$ levels correlated negatively with HOMA-IR levels (**Table 2**). According to results reported in **Table 2**, only one factor was relative to the HOMA-IR in subjects with normal BMI, and no related factor was found in the overweight group, therefore, multivariable analysis was merely done for the obese group. As the results of univariate linear regression analyses should be similar to those of correlation analyses, only the factors with *P*<0.05 explored by the correlation analyses were further used in multivariable analysis. Results were similar in the multivariable models: higher WC (β=0.018, 95% CI: 0.006 to 0.030, *P*=0.004) and lower $25(OH)D_3$ in obese patients (β=-0.016, 95% CI: -0.026 to -0.005, *P*=0.004) were associated with increases in HOMA-IR (**Table 3**).

Factors related to vitamin D status

Figure 1 shows the $25(OH)D_3$ levels, anthropometric data and HOMA-IR for the three groups. Four levels of vitamin D status were defined based on 25(OH)D levels: severe deficiency (<25 nmol/L), mild deficiency (25-49 nmol/L), insufficient (50-74 nmol/L), and sufficient (≥75 nmol/L). In the whole study population, only 5.1% of subjects (8/157) had sufficient 25(OH)D levels, 8.9% (14/157) had vitamin D insufficiency, 78.4% (123/157) had mild deficiency in vitamin D, and 7.6% (12/157) had severe deficiency in vitamin D. Notably, as compared with subjects with sufficient vitamin D, those with vitamin D deficiency were more likely to be overweight or obese (all *P*<0.007, **Table 1**). Moreover, larger WC, HC, WHR, and higher HOMA-IR levels were found in subjects with vitamin D deficiency as compared with those with insufficient or sufficient vitamin D (all *P*<0.007).

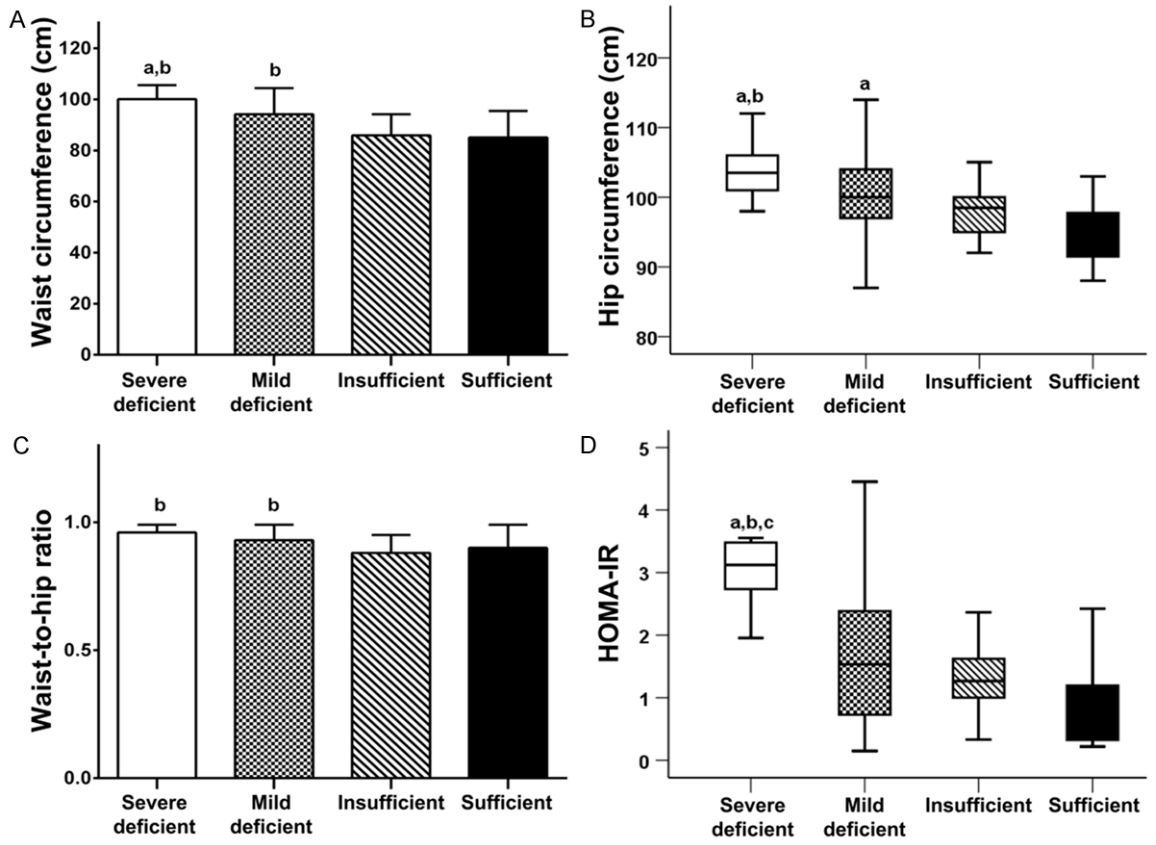

Figure 1. Difference in waist circumference (A), hip circumference (B), waist-to-hip ratio (C), and homeostasis model assessment insulin resistance (HOMA-IR, D) among four levels of vitamin D deficiency. Mean and standard deviation are expressed for waist and waist-to-hip ratio and tested by analysis of variance. Box plots (middle line in the box, top and bottom of the box indicate median, P_{25} and P_{75} respectively) are illustrated for hip circumference and HOIA-IR and Kruskal-Wallis test followed by Mann-Whitney U test for post-hoc tects were performed. Letters denote significantly different from (P<0.008) sufficient[a], insufficient[b], or mildly deficient[c] group.

Discussion

In the present study, we investigated vitamin D status in obese, overweight and normal-weight young Chinese men with NGT and evaluated whether independent associations existed between serum vitamin D levels and IR. Among the three groups of subjects, 25(OH)D levels were significantly lower in obese men compared to the levels in normal weight controls. Only 5.1% of subjects had sufficient vitamin D levels, primarily among normal-weight controls, and mild (78.4%) or severe (7.6%) deficiency was found predominately in the obese or overweight groups. We also found that men with severe deficiency in Vitamin D had significantly higher BMI, WC and HC than did men with sufficient vitamin D. Notably, while WC, WHR, SBP, DBP, and TC were similar in the normal and overweight groups, all of these values were higher in the obese group than in both of the other groups. In addition, we found that WC correlated positively with changes in HOMA-IR levels, while 25(OH)D_3 levels correlated negatively with HOMA-IR levels. In multivariable models, higher WC and lower 25(OH)D_3 in obese patients were associated with increases in HOMA-IR. Our hypothesis that the relationship between vitamin D and IR is accepted and demonstrated in younger Chinese men of different body weight status and with NGT.

The incidence of vitamin D deficiency has been shown in previous studies to be increased in obese individuals [23, 24]. Obesity itself is a global epidemic and associated with insulin resistance and other metabolic syndrome components [25]. The link between vitamin D deficiency and obesity is not coincidental, which can be explained in part by the fact that vitamin

D deficiency decreases bioavailability of adipose tissue and increases deposition of adipose tissue, particularly in the abdominal compartment [25-27]. Low serum 25(OH)D has also shown effects on lipogenesis and adipogenesis in adipose tissue, which may be the main contributing factor [25]. A recent study of 191 obese patients reported a high prevalence of vitamin D deficiency (48.7%) and vitamin D insufficiency (33.0%), with correlations shown between 25(OH)D levels and the degree of adiposity; 25(OH)D levels also correlated inversely with BMI, body weight, and WC [28]. Tzotzas et al. also found that 25(OH)D levels were lower in obese subjects compared with control subjects, and vitamin D levels increased after a 10% non-bariatric surgery-induced weight loss. Importantly, this increase was significantly associated with improvement in IR [29]. Such results suggest that adipose tissue may sequester vitamin D and during weight loss it is again released into circulation. This potential mechanism needs further investigation.

In the present study, lower circulating levels of 25(OH)D levels correlated negatively with HOMA-IR and remained significant after adjusting for age and BMI, which agrees with the results of previous studies conducted among Chinese people [11, 16]. Our results also agree with those of a study among healthy, young adult Caucasians, in which a significant inverse association was found between serum 25(OH)D levels and IR [18]. IR is a major underlying mechanism for development of arterial stiffness, diabetes, cardiovascular and metabolic syndrome, and a review and meta-analysis reports that most cross-sectional population studies have noted associations between 25(OH)D and insulin sensitivity [2]. Results of a large prospective study of non-diabetic high-risk Asian subjects suggest that vitamin D metabolism appears to play some role in the pathogenesis of type 2 diabetes [30]. An independent association has also been shown between vitamin D and glucose tolerance status with insulin sensitivity, insulin secretion and beta cell function, but that associations between serum 25(OH)D and these markers of type 2 diabetes do not correspond to OGTT status [31]. The role of vitamin D in glucose metabolism is to increase insulin secretion from pancreatic β-cells and increase insulin sensitivity in peripheral target tissues. Vitamin D deficiency also involves changes in oxidative stress, which may affect vitamin D receptor activity on pancreatic β-cells. These mechanisms are not fully understood, however, and previous studies assessing the association between vitamin D and IR have yielded inconsistent results. For example, while higher serum 25(OH)D is consistently associated with adiposity, triglycerides, HDL cholesterol and metabolic syndrome, it was not associated with HOMA-IR or beta cell function in healthy post-menopausal women [23]. Also, no association was found between serum vitamin D concentration and insulin action or insulin secretion in Europeans with metabolic syndrome or in postmenopausal Indian women [32, 33]. A recent study conducted in a rural Korean adult population also reported that vitamin D levels were not independently associated with IR [34]. Although we know that inconsistent results may result from different sample sizes, genetic factors, or different study populations with varying gender, age, or ethnicity, and also among healthy subjects or subjects with metabolic syndrome, clearly additional large, prospective trials are needed to establish the relationship between vitamin D deficiency and IR.

In the present study, we observed that up to 95% of the younger adult males studied had 25(OH)D deficiency or insufficiency. Poor vitamin D status was even shown in earlier studies of middle-aged and older Chinese individuals in Beijing [35] and Shenyang [36], respectively. Although little is known regarding to what extent the high prevalence of vitamin D deficiency in our population may be explained by certain environmental and/or genetic factors, geographic location and season may be factors in vitamin D status [37]. St. Petersburg, a northwestern region of Russia, and many other regions of Russia, are located higher than 42° North latitude and have approximately 62 sunny days per year, which may deprive the population of sufficient sunlight and predispose people to vitamin D deficiencies. In Spain, vitamin D levels were measured in older women (72±1.6 years) in winter and summer, with significantly lower levels shown for winter than for summer, specifically severely deficient levels (<25 nmol/L) were found in roughly 28% of subjects, about twice the percentage found in summer [38]. In the present study, participants were selected during winter months, December

2011 to February 2012, from Qinhuangdao, a northern city in China with a relatively high latitude (39° North latitude). In that region, compared to low latitude cities, daily sun exposure time is short, and ultraviolet light is weak. Therefore, this may help to explain why only 5.10% subjects had normal 25(OH)D levels. However, this was a small study and the study population may not be suitable for evaluating the prevalence of vitamin D deficiency, only that vitamin D deficiency may be common among young Chinese men in winter. A large-scale population study must be done for this purpose. However, our results do indicate that greater attention should be paid to improving vitamin D nutritional status in the Chinese population, particularly for those who are obese, less physically active, and living in high latitudes and urban areas.

We must also mention that several interventional trials have evaluated the effect of vitamin D supplementation in non-diabetic insulin-resistant subjects, demonstrating significant improvements in insulin secretion and IR after daily supplementation compared to placebo [39-41]. Although the present study was non-interventional, the benefits of supplementation must be evaluated for the Chinese population with vitamin D deficiency and its associations with adiposity, type 2 diabetes mellitus, IR, and metabolic syndrome.

The present study has several limitations. First, the sample was small and we applied cross-sectional design, which precludes showing a causal relationship between serum 25(OH)D and IR. Second, we did not include the subjects' degree of smoking and drinking in the analysis of variables. Furthermore, Vitamin D mainly stores in adipose tissue and the 25(OH) D level will have a direct impact to the Vitamin D level in blood stream. We did not provide any data to clarify this question. Third, Vitamin D is related to the amount of day light received. If there is a difference in the amount of day light received between obese, overweight, and normal weight individuals, the results of "obese individual's blood 25(OH)D level was lower" may be due to "total amount of daylight illumination was less in obese individual". We lack any evidence to explain it. For plasma glucose levels, we did not perform intra and inter-assay coefficient of variation. In **Table 3**, the results for linear regression with backward selection is one of the major basis for the present study. Finally, larger prospective studies are needed to further investigate both the prevalence of vitamin D deficiency in Chinese populations and also the associations between vitamin D, IR and BMI or other variables such as WC associated with overweight and obese status.

In conclusion, results of this study suggest that vitamin D deficiency is common among young Chinese obese men with NGT and that IR correlates with 25(OH)D and WC in this population. Regardless of the cause-effect relationship between vitamin D deficiency and obesity, and confirmation of possible mechanisms explaining vitamin D deficiency and IR risk, it is clear that the relationship affects many different parameters of public health in China and that results of our study are pertinent to improving both vitamin D status and related public health policy.

Disclosure of conflict of interest

None.

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References

[1] Anderson JL, May HT, Horne BD, Bair TL, Hall NL, Carlquist JF, Lappé DL, Muhlestein JB; Intermountain Heart Collaborative (IHC) Study Group. Relation of vitamin D deficiency to cardiovascular risk factors, disease status, and incident events in a general healthcare population. Am J Cardiol 2010; 106: 963-968.

[2] Pittas AG, Lau J, Hu FB, Dawson-Hughes B. The role of vitamin D and calcium in type 2 diabetes. A systematic review and meta-analysis. J Clin Endocrinol Metab 2007; 92: 2017-2129.

[3] Bikle D. Nonclassic actions of vitamin D. J Clin Endocrinol Metab 2009; 94: 26-34.

[4] Pittas AG, Chung M, Trikalinos T, Mitri J, Brendel M, Patel K, Lichtenstein AH, Lau J, Balk EM. Systematic review: vitamin D and cardiometabolic outcomes. Ann Intern Med 2010; 152: 307-314.

[5] Wolf M, Shah A, Gutierrez O, Ankers E, Monroy M, Tamez H, Steele D, Chang Y, Camargo CA Jr, Tonelli M, Thadhani R. Vitamin D levels and

early mortality among incident hemodialysis patients. Kidney Int 2007; 72: 1004-1013.
[6] Mitri J, Muraru MD, Pittas AG. Vitamin D and type 2 diabetes: a systematic review. Eur J Clin Nutr 2011; 65: 1005-1015.
[7] Kabadi SM, Lee BK, Liu L. Joint effects of obesity and vitamin D insufficiency on insulin resistance and type 2 diabetes: results from the NHANES 2001-2006. Diabetes Care 2012; 35: 2048-2054.
[8] Choi HS, Kim KA, Lim CY, Rhee SY, Hwang YC, Kim KM, Kim KJ, Rhee Y, Lim SK. Low serum vitamin D is associated with high risk of diabetes in Korean adults. J Nutr 2011; 141: 1524-1528.
[9] Rhee SY, Hwang YC, Chung HY, Woo JT. Vitamin D and diabetes in Koreans: analyses based on the Fourth Korea National Health and Nutrition Examination Survey (KNHANES), 2008-2009. Diabet Med 2012; 29: 1003-1010.
[10] Laway BA, Kotwal SK, Shah ZA. Pattern of 25 hydroxy vitamin D status in North Indian people with newly detected type 2 diabetes: A prospective case control study. Indian J Endocrinol Metab 2014; 18: 726-730.
[11] Pham NM, Akter S, Kurotani K, Nanri A, Sato M, Hayabuchi H, Yasuda K, Mizoue T. Serum 25-hydroxyvitamin D and markers of insulin resistance in a Japanese working population. Eur J Clin Nutr 2012; 66: 1323-1328.
[12] Lu L, Yu Z, Pan A, Hu FB, Franco OH, Li H, Li X, Yang X, Chen Y, Lin X. Plasma 25-hydroxyvitamin D concentration and metabolic syndrome among middle-aged and elderly Chinese individuals. Diabetes Care 2009; 32: 1278-1283.
[13] Pilz S, van den Hurk K, Nijpels G, Stehouwer CD, Van't Riet E, Kienreich K, Tomaschitz A, Dekker JM. Vitamin D status, incident diabetes and prospective changes in glucose metabolism in older subjects: the Hoorn study. Nutr Metab Cardiovasc Dis 2012; 22: 883-889.
[14] Ardabili HR, Gargari BP, Farzadi L. Vitamin D supplementation has no effect on insulin resistance assessment in women with polycystic ovary syndrome and vitamin D deficiency. Nutr Res 2012; 32: 195-201.
[15] Del Gobbo LC, Song Y, Dannenbaum DA, Dewailly E, Egeland GM. Serum 25-hydroxyvitamin D is not associated with insulin resistance or beta cell function in Canadian Cree. J Nutr 2011; 141: 290-295.
[16] Kositsawat J, Freeman VL, Gerber BS, Geraci S. Association of A1C levels with vitamin D status in U.S. adults: data from the National Health and Nutrition Examination Survey. Diabetes Care 2010; 33: 1236-1238.
[17] Tao MF, Zhang Z, Ke YH, He JW, Fu WZ, Zhang CQ, Zhang ZL. Association of serum 25-hydroxyvitamin D with insulin resistance and β-cell function in a healthy Chinese female population. Acta Pharmacol Sin 2013; 34: 1070-1074.
[18] Moore A, Hochner H, Sitlani CM, Williams MA, Hoofnagle AN, de Boer IH, Kestenbaum B, Siscovick DS, Friedlander Y, Enquobahrie DA. Plasma vitamin D is associated with fasting insulin and homeostatic model assessment of insulin resistance in young adult males, but not females, of the Jerusalem Perinatal Study. Public Health Nutr 2014; 22: 1-8.
[19] Cornier MA, Dabelea D, Hernandez TL, Lindstrom RC, Steig AJ, Stob NR, Van Pelt RE, Wang H, Eckel RH. The metabolic syndrome. Endocr Rev 2008; 29: 777-822.
[20] Matthews DR, Hosker JP, Rudenski AS. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985; 28: 412-419.
[21] Hathcock JN, Shao A, Vieth R, Heaney R. Risk assessment for vitamin D. Am J Clin Nutr 2007; 85: 6-18.
[22] Holick MF. Vitamin D deficiency. N Engl J Med 2007; 357: 266-281.
[23] Chacko SA, Song Y, Manson JE, Van Horn L, Eaton C, Martin LW, McTiernan A, Curb JD, Wylie-Rosett J, Phillips LS, Plodkowski RA, Liu S. Serum 25-hydroxyvitamin D concentrations in relation to cardiometabolic risk factors and metabolic syndrome in postmenopausal wemon. Am J Clin Nutr 2011; 94: 209-217.
[24] Cheng S, Massaro JM, Fox CS, Larson MG, Keyes MJ, McCabe EL, Robins SJ, O'Donnell CJ, Hoffmann U, Jacques PF, Booth SL, Vasan RS, Wolf M, Wang TJ. Adiposity, cardiometabolic risk, and vitamin D status: the Framingham Heart Study. Diabetes 2010; 59: 242-248.
[25] Earthman CP, Beckman LM, Masodkar K, Sibley SD. The link between obesity and low circulating 25-hydroxyvitamin D concentrations: considerations and implications. Int J Obes (Lond) 2012; 36: 387-396.
[26] Wortsman J, Matsuoka LY, Chen TC, Lu Z, Holick MF. Decreased bioavailability of vitamin D in obesity. Am J Clin Nutr 2000; 72: 690-693.
[27] Blum M, Dolnikowski G, Seyoum E, Harris SS, Booth SL, Peterson J, Saltzman E, Dawson-Hughes B. Vitamin D(3) in fat tissue. Endocrine 2008; 33: 90-94.
[28] Boonchaya-Anant P, Holick MF, Apovian CM. Serum 25-hydroxyvitamin D levels and metabolic health status in extremely obese individuals. Obesity (Silver Spring) 2014; 22: 2539-2543.
[29] Tzotzas T, Papadopoulou FG, Tziomalos K, Karras S, Gastaris K, Perros P, Krassas GE. Rising

serum 25-hydroxy-vitamin D levels after weight loss in obese women correlate with improvement in insulin resistance. J Clin Endocrinol Metab 2010; 95: 4251-4257.

[30] Lim S, Kim MJ, Choi SH, Shin CS, Park KS, Jang HC, Billings LK, Meigs JB. Association of vitamin D deficiency with incidence of type 2 diabetes in high-risk Asian subjects. Am J Clin Nutr 2013; 97: 524-530.

[31] Morisset AS, Tardio V, Weisnagel J, Lemieux S, Bergeron J, Gagnon C. Associations between serum 25 hydroxivitaminD and insulin sensitivity, insulin secretion, and ß-cell function according to glucose tolerance status. Met Syndr Rel Dis 2015; 13: 208-213.

[32] Gulseth HL, Gjelstad IM, Tierney AC, Lovegrove JA, Defoort C, Blaak EE, Lopez-Miranda J, Kiec-Wilk B, Risérus U, Roche HM, Drevon CA, Birkeland KI. Serum vitamin D concentration does not predict insulin action or secretion in European subjects with the metabolic syndrome. Diabetes Care 2010; 33: 923-925.

[33] Agarwal N, Mithal A, Kaur P, Dhingra V, Godbole MM, Shukla M. Vitamin D and insulin resistance in postmenopausal Indian women. Indian J Endocrinol Metab 2014; 18: 89-93.

[34] Song BM, Kim HC, Choi DP, Oh SM, Suh II. Association between serum 25-hydroxyvitamin D level and insulin resistance in a rural population. Yonsei Med J 2014; 55: 1036-1041.

[35] Xue Y. Serum levels of 25-hydroxyvitamin D in normal Beijing subjects. China Prev Med 1991; 25: 177-179.

[36] Yan L, Zhou B, Wang X, D'Ath S, Laidlaw A, Laskey MA, Prentice A. Older people in China and the United Kingdom differ in the relationships among parathyroid hormone, vitamin D, and bone mineral status. Bone 2003; 33: 620-627.

[37] Lagunova Z, Porojnicu AC, Lindberg F, Hexeberg S, Moan J. The dependency of vitamin D status on body mass index, gender, age and season. Anticancer Res 2009; 29: 3713-3720.

[38] Rodríguez Sangrador M, Beltran de Miquel B, Quintanilla Murillas L, Cuadrado Vives C, Moreiras Tuny O. The contribution of diet and sun exposure to the nutritional status of vitamin D in elderly Spanish women: the five countries study (OPTIFORD Project). Nutr Hosp 2008; 23: 567-576.

[39] von Hurst PR, Stonehouse W, Coad J. Vitamin D supplementation reduces insulin resistance in South Asian women living in New Zealand who are insulin resistant and vitamin D deficient - a randomised, placebo-controlled trial. Br J Nutr 2010; 103: 549-555.

[40] Wood AD, Secombes KR, Thies F, Aucott L, Black AJ, Mavroeidi A, Simpson WG, Fraser WD, Reid DM, Macdonald HM. Vitamin D3 supplementation has no effect on conventional cardiovascular risk factors: a parallel-group, double-blind, placebo-controlled RCT. J Clin Endocrinol Metab 2012; 97: 3557-3568.

[41] Davidson MB, Duran P, Lee ML, Friedman TC. High-dose vitamin D supplementation in people with prediabetes and hypovitaminosis D. Diabetes Care 2013; 36: 260-266.