Statistical Model Fitted to Examine the Association between Vitamin D and Diabetes; Using Cross Sectional Study of NHANES Dataset from 2009 to 2010

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ABSTRACT

The main object of this study is to examine the association between vitamin D and diabetes. National Health Interview Survey (NHANES) dataset has been used from 2009 to 2010. The participants aged between 20 and 74 are included in the analysis. Logistic regression model is used. Unadjusted and adjusted models controlling by age, body mass index (BMI), serum cotinine level, C-reactive protein (CRP), total cholesterol (TC), High-density lipoprotein *(*HDL*)* and Low-density lipoprotein (LDL) are estimated. All analyses are stratified by gender to examine the specific-gender differences. In conclusion, there is an inverse association between vitamin D and diabetes. Gender differences have been shown significant difference.

Keywords

Vitamin D, Diabetes, gender differences, logistic regression model, NHANES dataset.

INTRODUCTION

The prevalence of diabetes is raised worldwide[1]. Obesity has been proven as one of the important risk factor of diabetes[2]. However, recent studies found the proper diet is one of the most challenging issue of treating patients with diabetes or preventing the disease[3]. Daily requirements of nutrient diet that satisfied a person needs might not be the best optional with all individuals[4]. Vitamins intake is also considered as one of the other option of sufficient individual needs beside the diet[5]. Recently, Vitamin D is pointed out as one of the important factor of health benefits[6-8]. Previous studies have shown an association between vitamin D and osteoporosis or cancer[9, 10]. However, vitamin D with other chronic diseases have also investigated[11]. Studies have shown that decreasing the level of vitamin D might be the cause of increasing haemoglobin A1c, hypertension, insulin resistance and developing of diabetes as well as cardiovascular disease[12-14]

Studies have proved that diabetes is related to complications (e.g. stroke disease). Stroke disease and renal impairment have been shown relation with vitamin D level[15]. However, studying whether the association between vitamin D, as an independent variable, and diabetes is still controversial. In this study, the aim is to investigate that association between vitamin D concentration and the risk of diabetes for National Health and Nutrition Examination Survey(NHANES from 2009 to 2010).

Unadjusted and adjusted model are fitted. Adjusted models are controlling by age, BMI, serum cotinine level(SC), C-reactive protein (CRP), Total cholesterol(TC), High-density lipoprotein *(*HDL*)* and Low-density lipoprotein (LDL). Stratified analysis by gender is used to examine the specific-gender differences between vitamin D concentrations and diabetes risk.

Methods

Study Population

In this study, National Health Interview Survey (NHANES) dataset was used for one wave from 2009 to 2010. The study protocol, methodology and ethics approval are given elsewhere[16, 17]. Individuals aged between 20 to 74 years old are included in this study. Missing data are estimated by using multiple imputation analysis. Extreme values for CRP are omitted from the analysis to avoid overestimating or underestimating the findings.

Measurements

Vitamin D was measured by using the LC-MS/MS method for the wave 2009 to 2010. In particular,*"25-hydroxyvitamin D3 (25OHD3), quantitative detection of 25-hydroxyvitamin D2 (25OHD2), and epi-25-hydroxyvitamin D3 (epi-25OHD3) was made by utilizing ultra-high performance liquid-chromatography mass spectrometer and 25OHD was used as the sum of 25OHD2 and 25OHD3"[18].* Vitamin D is measured in a continuous scale. Diabetes was measured by self-reported questionnaire. The question was *"Doctor told you have diabetes"* and the answers were in a binary scale (yes or no).

Laboratory variables

Serum cotinine level is a biochemical variable which used as an indicator for smoking status. The method that has been used to measure the serum cotinine level is given as follows " *an isotope dilution–high-performance liquid chromatography-atmospheric pressure chemical ionization-tandem mass spectrometry*", for more details see [19]. In previous studies, it is shown that CRP is one of the main major risk factors for many diseases such as cardiovascular disease, hypertension, obesity and diabetes. The method that used to measure CRP is given as *"The latex-enhanced nephelometry with high sensitivity by using a Dade Behring Nephelometer II Analyzer System "*, for more information see [16].Total cholesterol (TC), High-density lipoprotein *(*HDL*)* and Low-density lipoprotein (LDL) are risk factors for cardiovascular disease, which is one of the complicated disease that related to diabetes. The method of measuring these variables is as follows*"Hitachi 704 Analyzer which is serviced by Roche Diagnostics (formerly Boehringer-Mannheim Diagnostics), Indianapolis",* for more details see somewhere else[20, 21].

Other Variables

Self-reported questionnaire has been used to measure the demographic variables (age, gender, and race). Age is measured in a continuous scale. Gender is measured in a binary scale (zero for male and one for female). Race is measured in five categorizes (Mexican American, Non-Hispanic Black, Non-Hispanic White, Other-Hispanic, and others). BMI is used as an indicator for obesity and it is measured in a continuous scale. It is measured by weight (kg)/height (m^2) [22].

Statistical Analysis

Logistic regression model is used for the statistical analysis. Unadjusted and adjusted models are fitted to examine whether vitamin D is associated with diabetes disease. Four models are fitted;(i) Model1 is an unadjusted model which examine the association between vitamin D and participants who had diabetes; (ii) Model 2 is an adjusted model which is mainly examine the association between vitamin D concentration and patients with diabetes after controlling by age, BMI and race; (iii) Model 3 is model 2 after controlling by serum cotinine level and CRP (iv) Model 4 is model 3 after controlling by TC, HDL, and LDL. All models are fitted for all data. The analyses have been repeated after stratified by gender to examine the effect of the gender differences. Imputation analysis is used to estimate the missing data for diabetes. All statistical analyses are running by Stata version 13.

Results

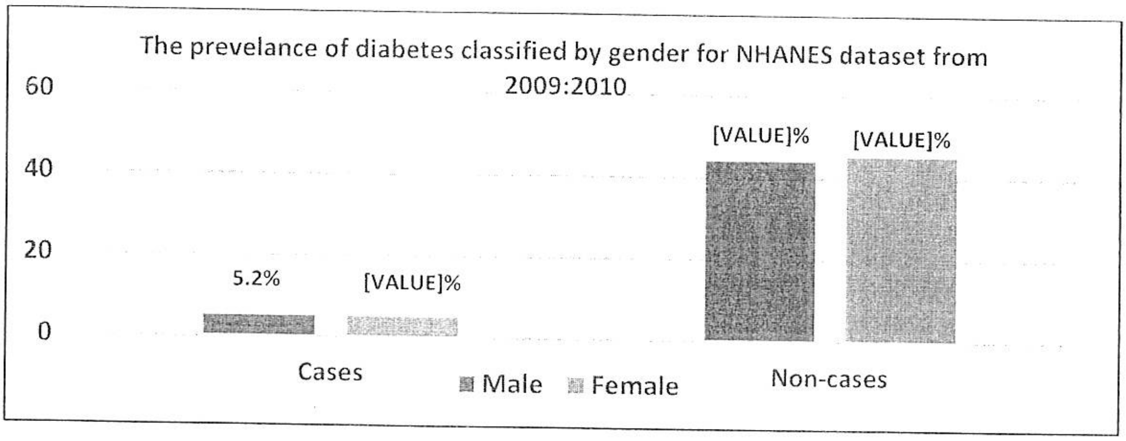
Descriptive statistics for the participants who diagnosed with diabetes (cases) versus participants without diabetes(non-cases)in NHANES dataset from 2009 to 2010 are summarized in Table 1. All participants in wave 2009 to 2010 are included in the study(n=10507. Three-hundred fifty-eight participants are excluded from the analyses as they reported that they did not know whether they diagnosis diabetes or not. The net population size is 10149 participants divided by 1046 (10.30%) patients with diabetes and 9103(89.70%) participants without diabetes.The average mean for vitamin D are less in patients with diabetes (57.67nmol/L) compared to non-cases participants (61.4 nmol/L). The average mean for age for who diagnosed diabetes is larger than who non-diagnosed diabetes. The average meansfor BMI, C-reactive protein, TC and LDL are more in patients with diabetes compared to non-cases group, see Table 1. While, the average mean for the serum cotinine level and LDL are smaller in patients with diabetes compared to non-cases group.

Table 1: Baseline characteristics for patients with diabetes (cases) versus participants without diabetes(non-cases) for NHANES dataset from 2009 to 2010

	Diabetes	
	Cases	Non-cases
Diabetes: $n(\%)$	1,046(10.30%)	9,103(89.70%)
Vitamin D: mean$\pm SD$	57.67$\pm$23.3	61.4$\pm$24.4
Gender		
Male: $n(\%)$	532(5.2%)	4496(44.3%)
Female: $n(\%)$	514(5.1%)	4607(45.4%)
Race		
Mexican American: $n(\%)$	266(2.6%)	2023(19.9%)
Non-Hispanic Black: $n(\%)$	118(1.2%)	975(9.6%)
Non-Hispanic White: $n(\%)$	375(3.7%)	3888(38.3%)
Other-Hispanic: $n(\%)$	227(2.2%)	1663(16.4%)
Others: $n(\%)$	60(0.6%)	554(5.5%
Age: mean$\pm SD$	58.51$\pm$10.93	44.33$\pm$15.21
BMI: mean$\pm SD$	28.53$\pm$9.40	25.81$\pm$7.46
Cotinine: mean$\pm SD$	35.58(101.3)	43.15(109.8)
CRP: mean$\pm SD$	0.41(0.64)	0.33(0.72)
TC: mean$\pm SD$	194.57$\pm$41.43	184.93$\pm$43.86
HDL: mean$\pm SD$	47.91$\pm$14.48	52.56$\pm$16.22
LDL: mean$\pm SD$	115.91$\pm$34.79	103.52$\pm$33.69

Figure 1 and 2 shows the percentage of patients with diabetes (cases) versus individuals without diabetes classified by gender and race, respectively. In Figure 1, the percentages of diabetes for men and women samples are almost the same with 5.2% and 5.1%, respectively.

Figure 1: The percentage of patients with diabetes (cases) versus participants without diabetes (non-cases) for men and women samples in NHANES dataset from 2009 to 2010



In Figure 2, the highest percentages of patients with diabetes are reported in Non-Hispanic white and Mexican American with 3.7% and 2.6%, respectively; while the lowest percentages of diabetes are in others and non-Hispanic black with 0.6% and 1.2%, respectively.

Figure 2: The percentage of patients with diabetes (cases) versus participants without diabetes (non-cases) stratified by race in NHANES dataset from 2009 to 2010

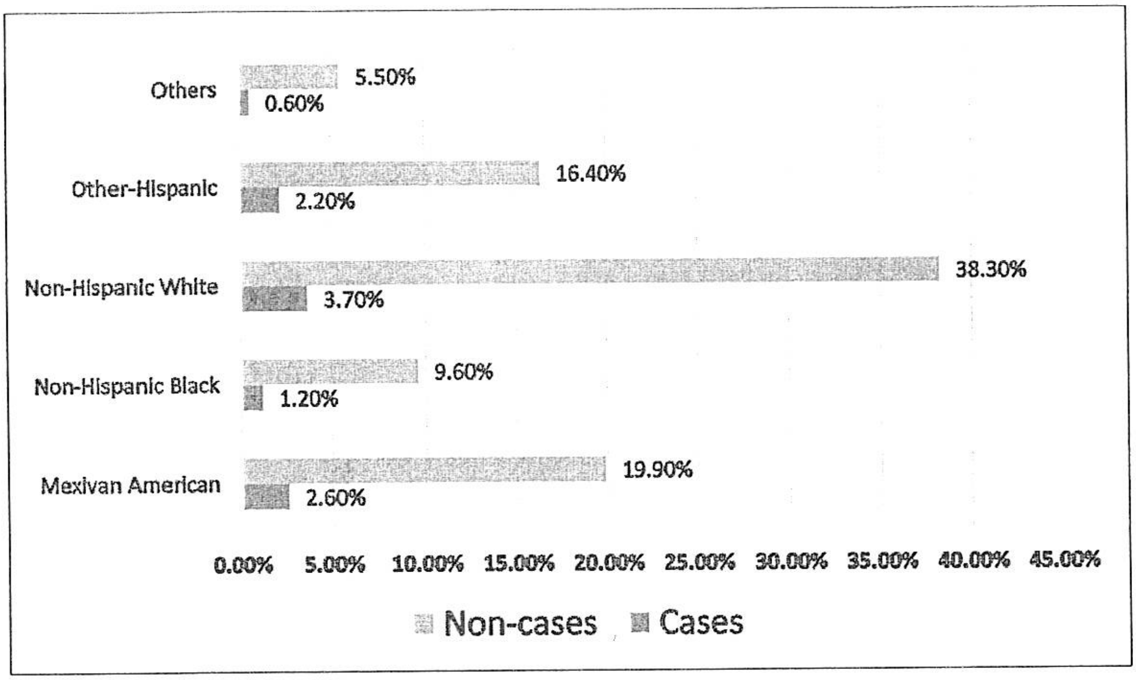


Table 2 gives the odds ratio with 95 % confidence interval for the association between vitamin D, as an independent variable, and participants who diagnosed diabetes for all NHANES dataset from 2009 and 2010. Unadjusted and adjusted models controlling by age, BMI, race, serum cotinine level, CRP, TC, LDL and HDL are fitted. There is an inverse association between Vitamin D and patients with diabetes. This means for one unit increase in vitamin D, the risk probability for diabetes is decreased by 1% for an unadjusted and all adjusted models. In addition, Age, BMI, and TC have been reported a positive association with patients with diabetes. This means for one unit increase for Age, BMI, or TC, the risk of diagnosed diabetes will increase, see Table 2. However, serum cotinine level and CRP does not show any significant association with diabetes. Only Non-Hispanic White shows a negative significant association with patients who diagnosed diabetes for all models, see Table 2.

Table 2:gives the odds ratio (OR) with 95% confidence interval (CI) for the association between participant who diagnosed diabetes and vitamin D for unadjusted and adjusted models for all participants in NHANES dataset from 2009 to 2010

	Model 1 OR:95%CI	Model2 OR:95%CI	Model3 OR:95%CI	Model3 OR:95%CI
Vitamin D	0.99 (0.98:0.995)	0.99 (0.98:0.99)	0.99 (0.98:0.99)	0.97 (0.96:01.01)
Age		1.07 (1.06:1.08)	1.07 (1.06:1.08)	1.07 (1.05:1.08)
BMI		1.08 (1.06:1.09)	1.08 (1.07:1.10)	1.06 (1.041.08)
Race				
Mexican American		-----------	----------	-----------
Non-Hispanic Black		1.07(0.76:1.49)	1.07(0.76:1.49)	1.25(0.76:2.01)
Non-Hispanic White		0.47(0.36:0.62)	0.46(035:0.60)	0.54(0.36:0.81)
Other-Hispanic		0.78(0.58:1.05)	0.78(0.58:1.05)	0.93(0.59:1.47)
Others		1.39(0.90:2.13)	1.38(0.90:2.11)	1.27(0.68:2.39)
Cotinine			1.00(0.99:1.01)	1.00(0.99:1.02)
CRP			0.89(0.78:1.03)	0.79(0.61:1.01)
TC				1.02(1.01:1.03)
HDL				0.96(0.95:0.97)
LDL				0.96(0.94:0.97)

Table 3 gives the odds ratio with 95 % confidence interval for the association between vitamin D and participants who diagnosed diabetes in men sample in NHANES dataset from 2009 and 2010. The unadjusted model shows an inverse significant association between vitamin D and patients who diagnosed diabetes. The same findings are found for model 2 and model 3. While in model 4, there is insignificant association between vitamin D and patients who diagnosed diabetes(OR:1.00; 95%CI 0.99:1.01) after controlling by the confounding factors (TC, HDL and LDL). This indicates that vitamin D is not the main risk factor for men with diabetes. LDL or TC might be one of the main risk factors for diabetes, see Table 3. The same findings are obtained for all other variables.

Table 3:gives the odds ratio (OR) with 95% confidence interval (CI) for the association between participant who diagnosed diabetes and vitamin D for unadjusted and adjusted models for men sample in NHANES dataset from 2009 to 2010

	Model 1	Model2	Model3	Model4
Vitamin D	0.99 (0.98:0.994)	0.99 (0.98:0.998)	0.99 (0.98:0.997)	1.00 (0.99:1.01)
Age		1.07 (1.06:1.08)	1.07(1.06:1.08)	1.06(1.04:1.07)
BMI		1.09 (1.07:1.12)	1.09(1.07:1.12)	1.09(1.04:1.12)
Race				
Mexican American		---------	-----------	-----------
Non-Hispanic Black		1.02(0.63:1.65)	1.03(0.63:1.67)	1.30(0.66:2.58)
Non-Hispanic White		0.47(0.32:0.68)	0.47(0.32:0.68)	0.50(0.28:0.89)
Other-Hispanic		0.64(0.42:0.97)	0.65(0.43:0.98)	0.61(0.31:1.21)
Others		1.19(0.64:2.21)	1.19(0.69:2.22)	1.52(0.65:3.57)
Cotinine			1.00(0.99:1.01)	1.00(0.99:1.02)
CRP			0.87(0.70:1.09)	0.71(0.46:1.08)
TC				1.01(0.99:1.02)
HDL				0.98(0.96:1.01)
LDL				0.97(0.95:0.98)

In female sample, there is indirect significant relationship between vitamin D and patients with diabetes in an unadjusted model, see Table 4. The results remain the same for the three adjusted models. Again, the other variables (age, BMI, TC, HDL, and LDL) show a positive significant association with participants who diagnosed diabetes. The other variables (such as serum cotinine level and C-reactive protein) did not show any significance difference with women who diagnosed diabetes (same as male sample). Only Non-Hispanic white group gives a significant negative association with women who diagnosed diabetes (same as the results that obtained from all data).

Table 4: gives the odds ratio (OR) with 95% confidence interval (CI) for the association between participant who diagnosed diabetes and vitamin D for unadjusted and adjusted models for women sample in NHANES dataset from 2009 to 2010

	Model 1	Model2	Model3	Model4
Vitamin D	0.99 (0.98:0.997)	0.99 (0.98:0.997)	0.99 (0.98:0.997)	0.98 (0.98:0.99)
Age		1.08(1.7:1.09)	1.08(1.07:1.09)	1.08(1.06:1.10)
BMI		1.07(1.05:1.9)	1.07(1.07:1.10)	1.05(1.02:1.08)
Race				
Mexican American		----------	-----------	
Non-Hispanic Black		1.14(0.72:1.80)	1.12(0.70:1.79)	1.23(0.63:2.44)
Non-Hispanic White		0.47(0.32:0.70)	0.47(0.30:0.68)	0.55(0.31:0.98)
Other-Hispanic		0.96(0.64:1.45)	0.94(0.62:1.42)	1.54(0.82:2.93)
Others		1.69(0.93:3.05)	1.67(0.92:3.03)	1.03(0.38:2.76)
Cotinine			1.00(0.99:1.01)	1.00(0.99:1.02)
CRP			0.94(0.78:1.13)	0.83(0.61:1.14)
TC				1.03(1.01:1.05)
HDL				0.94(0.93:0.97)
LDL				0.96(0.93:0.98)

Discussions

In this study, it has been shown that there is an inverse association between vitamin D and the risk of diabetes. The relative contribution of gender differences is estimated by evaluating the change in vitamin D on the diabetes risk for men compared with women. Further adjustment for age, BMI, race, serum cotinine level, and C-reactive Protein has been undertaken. The inverse relationship between vitamin D and diabetes risk remains in both male and female sample. However, in male sample after adjustment for TC, LDL, and HDL, the significance inverse association between vitamin D and diabetes is disappeared; while it remains without any change in female sample. The confounding factors such as age, BMI, and TC shows positive significant association with the risk of diabetes. Further variables such as HDL and LDL yield negative significant relationship with diabetes risk, while serum cotinine level and CRP does not show any significant association. On female sample, only non-Hispanic white shows a significance association with diabetes risk. However, in male sample non-Hispanic black and non-Hispanic white yield significant association with diabetes risk in all three adjusted models. In this study, our findings are consistent with previous studies[23-28].

The strength points of this paper are listed as follows: (i) vitamin D is measured in a continuous scale, (ii) all biochemical variables such as TC, LDL, HDL, C-reactive protein and serum cotinine level are measured in continuous scale. (ii) Unadjusted and adjusted models are fitted. (iii) Gender differences is measured for all unadjusted and adjusted models. (iv) Imputation analysis has been used to estimate the missing data in the diabetes. However, the weakness points in this study are listed as(i) the diagnosis of diabetes was obtained from self-reported questionnaire; the question was *"Doctor told you have diabetes*" and the answers were in a binary scale (yes or no); this might due to bias (ii) The findings of this study are based on US participants from NHANES dataset; different findings might be obtained from other population. (iii) the findings are also obtained from cross-sectional studies, other studies such as cohort studies might yield different findings.

Conclusion

There is an inverse association between vitamin D and diabetes risk. Gender differences have been shown significant difference. Age, BMI, and TC have been shown positive significance association with risk of diabetes.

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