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The relationship of serum klotho levels and triglyceride glucose index-related indicators

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Abstract

Background Klotho, an anti-aging protein, is linked to energy metabolism. There is limited research on the association of serum klotho and triglyceride glucose (TyG) index-related indicators. Our research aims to investigate the relationship of serum klotho with TyG-BMI (body mass index), TyG-WC (waist circumference), and TyG-WHtR (waist-to-height ratio).

Methods From 2007 to 2016, we examined 6,370 participants in the National Health and Nutrition Examination Survey (NHANES). The enzyme-linked immunosorbent assay (ELISA) was utilized to measure serum klotho. We calculated the TyG-BMI, TyG-WC, and TyG-WHtR based on fasting triglycerides, fasting glucose, BMI, WC, and WHtR. Multiple linear regression analysis was used to evaluate the association of serum klotho with TyG-BMI, TyG-WC, and TyG-WHtR. Additionally, generalized additive model (GAM) and smoothing curves were used to evaluate the linear and nonlinear relationships. A piecewise regression model was also utilized to test for threshold effects and determine the breakpoints. Finally, the potential independent associations of serum klotho with TyG-BMI, TyG-WC, and TyG-WHtR were further explored using subgroup analysis.

Results We observed a statistically significant difference in serum klotho levels across different quartiles of the population. Based on the multiple linear regression analysis, serum klotho levels were negatively associated with TyG-related indicators. There was a nonlinear relationship between the serum klotho and TyG-BMI, TyG-WC, and TyG-WHtR. The segmented regression analysis revealed that the breakpoints of TyG-BMI, TyG-WC, and TyG-WHtR were 5.42, 6.67, and 1.89, respectively. Subgroup analysis showed that TyG-related indicators interacted with gender and diabetes.

Conclusions In this study, a negative and nonlinear relationship was identified between serum klotho and TyG-related indicators. Further research is needed to clarify the potential mechanisms that may link serum klotho to TyG-BMI, TyG-WC, and TyG-WHtR.

Keywords Klotho, NHANES, Aging, Triglyceride-glucose (TyG) index, Insulin resistance (IR)

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Introduction

Insulin resistance (IR) is a metabolic dysfunction characterized by the body's reduced responsiveness to insulin, typically associated with metabolic disorders such as diabetes or obesity [1]. Aging is the gradual loss of function and adaptability in an organism, accompanied by decreased cellular function and organ degeneration [2]. Research indicates a close association between IR and aging. Prolonged IR increases oxidative stress and inflammatory responses, damaging cell structure and function, thus accelerating the aging process [3]. Additionally, IR affects the expression of genes related to aging, regulating cell apoptosis, proliferation and repair capacity, thereby influencing the overall pace of biological aging [4]. Conversely, aging can also lead to the onset and exacerbation of IR. As age advances, cells undergo changes that affect their sensitivity to insulin, which can disrupt insulin signaling pathways and contribute to the development of IR [5]. Furthermore, aging may induce changes in organismal energy metabolism, which can further exacerbate the degree of IR [6].

The klotho protein, encoded by the klotho gene, has been shown by recent research to play a significant role in modulating the aging process. Originally found in mice, the absence of the klotho gene in these animals can result in syndromes similar to those seen in human aging, including a shortened lifespan, infertility, skin atrophy, osteoporosis, etc [7]. In healthy individuals, serum klotho levels decrease with age, and this decline is further exacerbated in those with metabolic diseases [8], such as obesity [9], diabetes [1], hypertension [10], hyperlipidemia [11], and metabolic syndrome [12]. As research advances, klotho is gaining prominence for its crucial regulatory function in various physiological processes, including IR, glucose homeostasis, oxidative stress and inflammation, etc [13].

Based on the incorporation of triglycerides and fasting blood glucose (FBG), the triglyceride-glucose (TyG) index serves as a measure of IR and provides insight into an individual's sensitivity to insulin [14]. Researchers have recently begun exploring the integration of the TyG index with other physiological indicators, such as WC, BMI, WHtR, or other simple, cost-effective, and non-invasive anthropometric measures. Furthermore, research has demonstrated a close correlation between metabolic diseases and IR. There is promising potential in integrating the TyG index with indicators (BMI, WC, WHtR) to enhance the predictive accuracy of IR or other metabolic disorders. For example, Yan et al. [15] proposed that TyG-BMI, TyG-WC, and TyG-WHtR could effectively screen for non-alcoholic fatty liver disease (NAFLD) and metabolic-associated fatty liver disease (MAFLD) at an early stage. Additionally, incorporating the HOMA-IR index allowed for metabolic risk assessment and disease monitoring for individuals with NAFLD. Wang et al. [16] identified that increase in TyG, TyG-BMI and METS-IR were associated with a higher risk of hyperuricemia (HU). Additionally, both TyG-BMI and METS-IR exhibited enhanced discriminatory capabilities for HU across both genders. Li et al. [17] demonstrated that TyG-BMI, TyG-WC, and TyG-WHtR were significant predictors of metabolic syndrome among middle-aged to elderly Chinese people.

At present, research on the potential association between serum klotho and TyG-related indicators is limited. Therefore, this study utilized the NHANES database to explore the association between serum klotho and the TyG-related indicators.

Materials and methods

Data source

The research utilized data from the National Health and Nutrition Examination Survey (NHANES), a cross-sectional study conducted by the National Center for Health Statistics (NCHS) and the Centers for Disease Control and Prevention (CDC) [18]. NHANES employed a complex stratified sampling methodology to ensure the representativeness of the sample. NHANES has been approved by the NCHS Ethics Review Board (ERB), with all participants furnishing written informed consent [19]. Data for this study was sourced from https://www.cdc.gov/nchs/nhanes/.

Study population

Our research incorporated data from five NHANES cycles (2007 to 2016), including demographics data, examination results, laboratory data, and questionnaire data. Overall, NHANES collected data from 50,588 qualified participants from 2007 to 2016. Based on the research criteria, the following participants were excluded: (1) absence of klotho data; (2) missing triglyceride and glucose data; and (3) lacking data on BMI, WC, and body height data. Ultimately, 6,370 participants were included for analysis. The screening flowchart is shown in Fig. 1.

Exposure and outcome variable

In this study, the exposure variables included TyG-BMI, TyG-WC, and TyG-WHtR. The calculation procedure is outlined below: TyG = Ln [fasting triglycerides (mg/dL) × fasting glucose (mg/dL)/2] [20]; BMI = body mass (kg)/height^2 (m^2); WHtR = WC (cm)/height (cm); TyG-BMI = TyG × BMI [21]; TyG-WC = TyG × WC [21]; TyG-WHtR = TyG × WHtR [22].

Serum klotho were the outcome variable. Blood samples were obtained from individuals aged over 40, preserved in dry ice at -80 °C, and then sent to the University of Washington's Northwest Lipid Metabolism

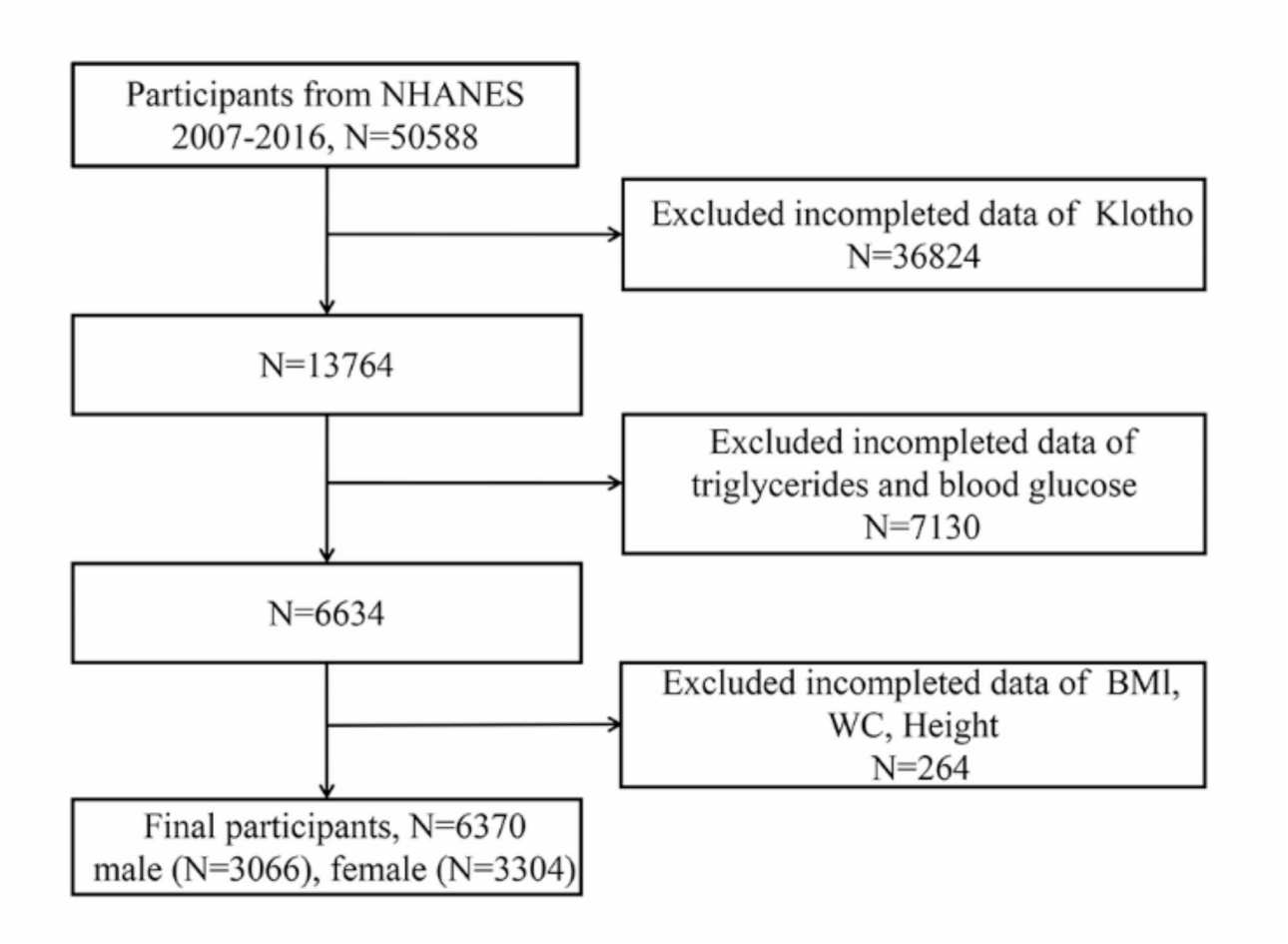

Fig. 1 Flowchart of participant selection. NHANES: National Health and Nutrition Examination Survey; BMI: body mass index; WC: waist circumference

and Diabetes Research Laboratory for examination. Each sample was analyzed twice, and the mean value was used as the final result. More detailed testing methods for serum klotho can be obtained on the NHANES website.

Covariables

Covariables included demographic information, examination results, laboratory data, and questionnaire data. Demographics information included gender, age, race, educational level, marital status, and poverty income ratio (PIR). Examination data included body mass, WC, body height, and hypertension. Laboratory data included triglycerides, glycosylated hemoglobin, FBG, and serum klotho. Questionnaire data included information on smoking (the average number of cigarettes per day over the past 30 days) and alcohol intake (the average number of drinks per day over the past 12 months). The assessment of hypertension: in a resting state, mean systolic pressure≥140 mmHg or mean diastolic pressure≥90 mmHg on three consecutive measurements, or the use of antihypertensive medications. Diabetes is diagnosed with glycosylated hemoglobin (HbA1c)≥6.5%, FBG≥7.0 mmol/L (126 mg/dL) [23], or the use of hypoglycemic medications. Hyperlipidemia is diagnosed if the total cholesterol≥200 mg/dL, triglyceride≥150 mg/dL, low density lipoprotein≥130 mg/dL, high density lipoprotein≤40 mg/dL for males and ≤50 mg/dL for females [24], or the use of cholesterol-lowering medications.

Statistical analysis

In accordance with CDC guidelines, this study used a complex sampling design to analyze the relationship between serum klotho and TyG-BMI, TyG-WC, and TyG-WHtR. Initially, the study population was divided into four groups based on quartiles (Q1-Q4) of serum klotho. Rao-Scott $\chi 2$ and non-parametric tests was employed to assess the statistical differences in the four groups for categorical variables. The continuous variables were presented as mean±standard error. The missing values were imputed by multiple imputation (e.g., smoking, alcohol intake) (Supplement material S1)) [25]. Considering the skewed distribution, the natural logarithm transformation of serum klotho (Inklotho), TyG-BMI (InTyG-BMI), TyG-WC (InTyG-WC), and TyG-WHtR

(InTyG-WHtR) were performed when assessing the relationship between serum klotho and TyG-related indicators. Therefore, three types of weighted linear regression models were used to investigate the relationship of serum klotho with TyG-BMI, TyG-WC, and TyG-WHtR. Subsequently, we generated a smoothed curve using a generalized additive model. Furthermore, a segmented linear regression model was used to assess the potential nonlinear relationship between serum klotho and TyG-related indicators, determine the threshold effects, and calculate the inflection points. Finally, we conducted subgroup analysis to explore the potential factors affecting the association between serum klotho and TyG-related indicators. The statistical analysis was conducted using R software (version 4.3.1) and SPSS 27.0. A *P*-value less than 0.05 was considered statistically significant.

Results

Participant baseline characteristics

Our research included a sample of 6,370 individuals. We observed that gender, age, race, educational level, marriage status, PIR, smoking, alcohol intake, hypertension, diabetes, hyperlipidemia, BMI, WC, and height were statistically significant in serum klotho at different quartiles. With the increase in serum klotho concentration, BMI, WC, height, TyG-BMI, TyG-WC, and TyG-WHtR showed a decreasing trend (Table 1).

Multivariable regression analysis

The association between serum klotho and TyG-related indicators was examined using a weighted multivariable regression model (Table 2). A significant negative association was found between serum klotho and TyG-BMI, TyG-WC, and TyG-WHtR. In model 1, serum lnklotho decreased by 0.063 pg/ml, 0.137 pg/ml, and 0.115 pg/ml

Table 1 Baseline characteristics of participants in the NHANES from 2007–2016

Characteristic	Klotho (pg/ml)					*P*-value
	Overall (*n* = 6370)	Q1 (< 657.20) (*n* = 1593)	Q2 (657.20-807.40) (*n* = 1593)	Q3 (807.40-1002.90) (*n* = 1593)	Q4 (> 1002.90) (*n* = 1591)	
Gender						< 0.001
Male	3066 (47.36%)	846 (12.69%)	820 (13.51%)	733 (11.50%)	667 (9.67%)	
Female	3304 (52.64%)	747 (12.43%)	773 (12.85%)	860 (13.68%)	924 (13.68%)	
Age						< 0.001
40–60	3714 (65.67%)	859 (15.62%)	930 (17.31%)	922 (16.19%)	1003 (16.53%)	
> 60	2656 (34.33%)	734 (9.50%)	663 (9.04%)	671 (8.99%)	588 (6.81%)	
Race						< 0.001
Mexican American	990 (6.46%)	247 (1.55%)	250 (1.61%)	254 (1.70%)	239 (1.60%)	
Non-Hispanic Black	2805 (72.53%)	749 (18.61%)	754 (19.84%)	712 (18.48%)	590 (15.59%)	
Non-Hispanic White	1198 (9.48%)	293 (2.31%)	241 (1.93%)	259 (2.07%)	405 (3.18%)	
Other	1377 (11.54%)	304 (2.66%)	348 (2.98%)	368 (2.93%)	357 (2.97%)	
Educational level						< 0.001
Below high school	1785 (16.71%)	450 (3.97%)	449 (4.50%)	444 (4.07%)	442 (4.17%)	
High school or above	4585 (83.29%)	1143 (21.15%)	1144 (21.86%)	1149 (21.11%)	1149 (19.17%)	
Marriage status						< 0.001
Married/Living with partner	4140 (71.22%)	1033 (17.55%)	1035 (19.01%)	1048 (17.88%)	1024 (16.78%)	
Widowed/Separated/Divorced/Never married	2230 (28.78%)	560 (7.58%)	558 (7.35%)	545 (7.30%)	567 (6.56%)	
PIR (%)	3.24 ± 0.00	3.24 ± 0.00	3.23 ± 0.00	3.25 ± 0.00	3.25 ± 0.00	< 0.001
Smoke (cigarettes/day)	(5, 20)	(5, 20)	(5, 20)	(5, 20)	(5, 20)	0.834
Alcohol intake (alcoholic drinks/day)	(1, 3)	(2, 4.5)	(2, 5)	(1, 5)	(1, 4)	< 0.001
Hypertension	1416 (18.70%)	366 (4.67%)	356 (5.23%)	324 (4.35%)	370 (4.45%)	< 0.001
Diabetes	1271 (14.99%)	305 (3.47%)	282 (3.71%)	298 (3.69%)	386 (4.13%)	< 0.001
Hyperlipidemia	4378 (68.96%)	1107 (17.84%)	1110 (18.62%)	1095 (17.41%)	1066 (15.08%)	< 0.001
BMI (kg/m^2)	29.52 ± 0.00	29.69 ± 0.00	29.45 ± 0.00	29.66 ± 0.00	29.27 ± 0.00	< 0.001
WC (cm)	101.82 ± 0.00	102.86 ± 0.00	101.93 ± 0.00	102.09 ± 0.00	100.27 ± 0.00	< 0.001
Height (cm)	168.50 ± 0.00	168.95 ± 0.00	168.71 ± 0.00	168.46 ± 0.00	167.83 ± 0.00	< 0.001
TyG-BMI	258.07 ± 0.00	260.64 ± 0.01	257.84 ± 0.01	259.05 ± 0.01	254.51 ± 0.01	< 0.001
TyG-WC	889.47 ± 0.01	902.37 ± 0.02	891.64 ± 0.02	891.03 ± 0.02	871.43 ± 0.03	< 0.001
TyG-WHtR	5.29 ± 0.00	5.35 ± 0.00	5.29 ± 0.00	5.30 ± 0.00	5.20 ± 0.00	< 0.001

PIR: poverty income ratio; BMI: body mass index; WC: waist circumference; TyG: triglyceride-glucose; WHtR = waist circumference (cm)/height (cm); TyG-BMI = TyG × BMI; Tyg-WC = TyG × WC; TyG-WHtR = TyG × WHtR

Table 2 Association between serum klotho and TyG-BMI, TyG-WC, TyG-WHtR

	Model 1	Model 2	Model 3
	β (95%CI) *P*-value	β (95%CI) *P*-value	β (95%CI) *P*-value
lnTyG-BMI	-0.063 (-0.114, -0.012) 0.015	-0.065 (-0.118, -0.012) 0.017	-0.064 (-0.118, -0.011) 0.018
Q1: < 5.37	Ref	Ref	Ref
Q2 (5.37–5.52)	-0.047 (-0.076, -0.017) 0.002	-0.035 (-0.064, -0.006) 0.020	-0.035 (-0.065, -0.006) 0.018
Q3 (5.52–5.68)	-0.052 (-0.084, -0.020) 0.002	-0.041 (-0.074, -0.008) 0.015	-0.041 (-0.074, -0.008) 0.015
Q4 > 5.68	-0.044 (-0.077, -0.010) 0.012	-0.044 (-0.080, -0.008) 0.016	-0.044 (-0.079, -0.008) 0.017
P for trend	0.006	0.051	0.050
lnTyG-WC	-0.137 (-0.207, -0.066) 0.000	-0.117 (-0.196, -0.038) 0.004	-0.116 (-0.195, -0.037) 0.005
Q1: < 6.65	Ref	Ref	Ref
Q2 (6.65–6.78)	-0.060 (-0.091, -0.030) < 0.001	-0.047 (-0.077, -0.016) 0.003	-0.047 (-0.077, -0.016) 0.003
Q3 (6.78–6.90)	-0.072 (-0.106, -0.039) < 0.001	-0.054 (-0.089, -0.019) 0.003	-0.054 (-0.089, -0.020) 0.003
Q4 > 6.90	-0.074 (-0.109, -0.040) < 0.001	-0.062 (-0.099, -0.025) 0.001	-0.062 (-0.099, -0.025) 0.001
P for trend	< 0.001	0.002	0.002
lnTyG-WHtR	-0.115 (-0.184,-0.045) 0.002	-0.127 (-0.204, -0.049) 0.002	-0.126 (-0.203, − 0.049) 0.002
Q1: < 1.53	Ref	Ref	Ref
Q2 (1.53–1.66)	-0.049 (-0.081, -0.017) 0.003	-0.038 (-0.070, -0.007) 0.018	-0.039 (-0.070, -0.007) 0.018
Q3 (1.66–1.79)	-0.062 (-0.094, -0.029) 0.000	-0.053 (-0.085, -0.022) 0.001	-0.054 (-0.085, -0.022) 0.001
Q4 > 1.79	-0.056 (-0.092, -0.020) 0.003	-0.058 (-0.099, -0.017) 0.006	-0.057 (-0.098, -0.017) 0.007
P for trend	< 0.001	0.004	0.004

Model 1 adjusted for no covariables. Model 2 adjusted for gender, age, race, educational level, marriage status, PIR, hypertension, diabetes, and hyperlipidemia. Model 3 adjusted for gender, age, race, education level, marriage status, PIR, hypertension, diabetes, hyperlipidemia, smoking, and alcohol intake

with each one-unit increase in lnTyG-BMI, lnTyG-WC, and lnTyG-WHtR, respectively. In model 2, after adjustments for gender, age, race, educational level, marital status, PIR, hypertension, diabetes, and hyperlipidemia, serum lnklotho decreased by 0.065 pg/ml, 0.117 pg/ml, and 0.127 pg/ml in lnTyG-BMI, lnTyG-WC, and lnTyG-WHtR, respectively. In model 3, with all variables adjusted, serum lnklotho decreased by 0.064 pg/ml, 0.116 pg/ml, and 0.126 pg/ml in lnTyG-BMI, lnTyG-WC, and lnTyG-WHtR, respectively.

lnTyG-BMI, lnTyG-WC, and lnTyG-WHtR were categorized into four groups (Q1 to Q4) based on their quartile, with the first quartile group (Q1) serving as the reference category (Table 2). We observed a nonlinear relationship between serum klotho and InTyG-BMI, InTyG-WC, and InTyG-WHtR, characterized by an initial decrease followed by an increase.

The dose-response association of serum lnklotho levels with lnTyG-BMI, lnTyG-WC, and lnTyG-WHtR

Generalized additive models are very sensitive in identifying both linear and nonlinear relationships. We generated three smooth curves to illustrate the potential relationships between serum Inklotho and InTyG-BMI, InTyG-WC, and InTyG-WHtR, based on model 3 (Fig. 2). In addition, we used a piecewise regression model to calculate the breakpoints of InTyG-BMI, InTyG-WC, and InTyG-WHtR, which were 5.42, 6.67, and 1.89, respectively (Table 3). The results showed that the piecewise regression model is more suitable for clarifying the relationship between serum Inklotho and InTyG-WC, and InTyG-WHtR.

Subgroup analysis

Subgroup analyses were conducted based on gender, age, diabetes, hypertension, and hyperlipidemia (Table 4). The analysis results showed that the association between serum klotho and TyG-related indicators was influenced by gender and diabetes (*P* for interaction < 0.05).

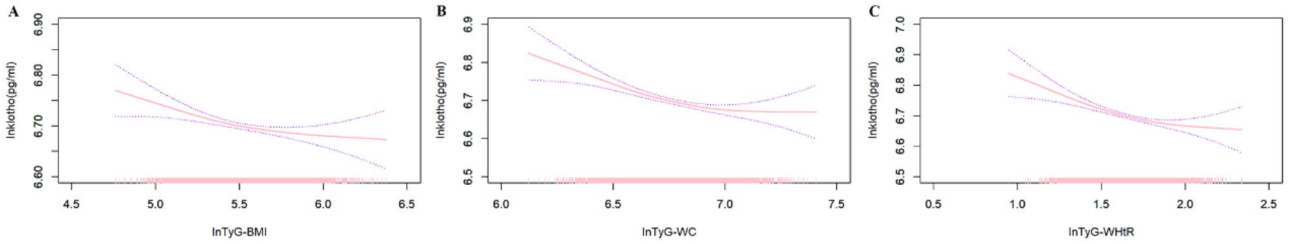

Fig. 2 The smooth curve fitting for the associations between serum klotho and TyG-related indicators: **(A)** the relationship between serum lnklotho and lnTyG-BMI; **(B)** the relationship between serum lnklotho and lnTyG-WC; **(C)** the relationship between serum lnklotho and lnTyG-WHtR

Table 3 Threshold effect analysis of the association between serum lnklotho and lnTyG-BMI, lnTyG-WC, TyG-WHtR in NHANES 2007–2016

Models	lnTyG-BMI	lnTyG-WC	lnTyG-WHtR
	β (95%CI) *P*-value	β (95%CI) *P*-value	β (95%CI) *P*-value
One linear effect	-0.060 (-0.097, -0.022) 0.002	-0.128 (-0.178, -0.078) < 0.001	-0.135 (-0.185, -0.084) < 0.001
Breakpoint (k)	5.42	6.67	1.89
≤ k	-0.145 (-0.240, -0.051) 0.003	-0.268 (-0.393, -0.142) < 0.001	-0.169 (-0.226, -0.112) < 0.001
> k	-0.021 (-0.075, 0.034) 0.456	-0.067 (-0.138, 0.004) 0.063	0.163 (-0.080, 0.407) 0.189
LRT test	0.053	0.017	0.014

Adjusting for gender, age, race, education level, marriage status, PIR, hypertension, diabetes, and hyperlipidemia. LRT test: Log likelihood ratio test

Discussion

Population aging poses a major public health challenge worldwide at present. The aging population often experiences declines in organ function and metabolic disorders. The potential link between the anti-aging protein klotho and metabolic diseases is also gaining attention [12]. Therefore, we used NHANES samples from 2007 to 2016 to explore the relationship between serum klotho and TyG-related indicators. After analyzing data from 6,370 participants, the key findings were as follows: (1) There were significant differences in levels of serum klotho according to the quartiles. (2) It showed a significant negative nonlinear relationship between the serum klotho and TyG-BMI, TyG-WC and TyG-WHtR. (3) The relationship between serum klotho and the TyG-related indicators was influenced by gender and diabetes.

The negative association between klotho and TyG-related indicators may suggest a potential role for klotho in regulating energy metabolism and slowing down the aging process. The klotho family comprises three subtypes: α, β, and γ. In addition, klotho can also be categorized into membrane-bound (mKL), secretory or soluble (sKL), and intracellular forms, with different forms of klotho involved in different physiological processes [26]. Studies have shown that the serum klotho levels in patients with diabetes are significantly lower than those in non-diabetic individuals [27]. Deficiency of soluble klotho in circulating blood leads to increased IR, diabetic phenotype, and hepatic lipid accumulation in T2D. Soluble klotho can target the IGF1R/PI3K/AKT/mTORC1 signaling pathway and up-regulate peroxisome proliferator-activated receptor α (PPARα) in T2D, thereby improving insulin sensitivity, hepatic glucose production, and lipid homeostasis [28]. The kidney is the main expression site for klotho, which affects glucose metabolism by influencing glucose reabsorption and filtration within the kidney [29]. Furthermore, klotho can alleviate podocyte damage in diabetic kidney disease (DKD) by modulating the IGF-1R/RAC1/OLR1 signaling pathway, thereby improving high-glucose-induced glomerular deposition of oxidized low-density lipoprotein (ox-LDL) [30]. Therefore, klotho has potential in the prevention and treatment of diabetes and its complications.

Elevated plasma lipoprotein levels and decreased lipid clearance lead to lipid accumulation in the body. Klotho can increase the body's energy expenditure, reduce fat mass, and play a beneficial role in regulating lipid accumulation in the liver and adipose tissue. For example, a-klotho can improve obesity status in DIO mice by reducing lipid accumulation in the liver and adipose tissue [31]. Recombinant klotho can reduce lipid accumulation in foam cells and promote increased cholesterol efflux by inhibiting the Wnt/β-catenin pathway [32]. Furthermore, a-klotho promotes lipid oxidation and reduces adipogenesis by regulating gene expression in the liver and white adipose tissue (WAT) of DIO mice [26]. Soluble klotho can alleviate hepatic steatosis by inhibiting genes associated with lipid synthesis and uptake

Table 4 Subgroup analysis for the relationship of serum Inklotho with InTyG-BMI, InTyG-WC, TyG-WHtR

Subgroup		β (95%CI)	*P*-value	*P* for interaction
InTyG-BMI				
Gender	Male	0.001 (-0.052, 0.054)	0.966	0.044
	Female	-0.071 (-0.116, -0.026)	0.002	
Age	40–60	-0.049 (-0.092, -0.006)	0.027	0.469
	> 60	-0.023 (-0.079, 0.034)	0.431	
Diabetes	Yes	0.026 (-0.061, 0.112)	0.564	0.015
	No	-0.089 (-0.128, -0.049)	< 0.001	
Hypertension	Yes	0.008 (-0.069, 0.084)	0.844	0.152
	No	-0.053 (-0.091, -0.014)	0.007	
Hyperlipidemia	Yes	-0.026 (-0.068, 0.017)	0.235	0.382
	No	-0.059 (-0.124, 0.005)	0.070	
InTyG-WC				
Gender	Male	-0.025 (-0.091, 0.041)	0.456	0.003
	Female	-0.159 (-0.220, -0.099)	< 0.001	
Age	40–60	-0.122 (-0.178, -0.067)	< 0.001	0.753
	> 60	-0.108 (-0.180, -0.035)	0.004	
Diabetes	Yes	0.000 (-0.118, 0.117)	0.996	< 0.001
	No	-0.221 (-0.271, -0.170)	< 0.001	
Hypertension	Yes	-0.054 (-0.156, 0.048)	0.299	0.098
	No	-0.146 (-0.195, -0.097)	< 0.001	
Hyperlipidemia	Yes	-0.113 (-0.167, -0.059)	< 0.001	0.383
	No	-0.156 (-0.239, -0.074)	< 0.001	
InTyG-WHtR				
Gender	Male	-0.021 (-0.088, 0.045)	0.532	0.002
	Female	-0.166 (-0.224, -0.107)	< 0.001	
Age	40–60	-0.091 (-0.147, -0.035)	0.002	0.618
	> 60	-0.067 (-0.140, 0.006)	0.072	
Diabetes	Yes	-0.004 (-0.122, 0.114)	0.950	0.006
	No	-0.177 (-0.229, -0.126)	< 0.001	
Hypertension	Yes	-0.025 (-0.127, 0.078)	0.636	0.115
	No	-0.113 (-0.162, -0.064)	< 0.001	
Hyperlipidemia	Yes	-0.082 (-0.137, -0.027)	0.003	0.458
	No	-0.119 (-0.202, -0.036)	0.005	

while promoting genes involved in lipid oxidation [28]. Klotho has a protective effect against high-fat diet (HFD) induced hepatic steatosis, reduces the weight of WAT and liver, and enhances the expression of thermogenic genes in brown adipose tissue (BAT) [33].

Although our study primarily investigated the association between serum klotho and TyG-related indicators, it offers novel insights into the prevention and treatment of metabolic diseases from the perspective of aging. Studies have shown that the level of s-klotho in the plasma of diabetic NOD mice is significantly lower than that in the non-diabetic mice [34]. A meta-analysis has demonstrated that individuals with chronic kidney disease (CKD) exhibiting reduced serum klotho levels are at an elevated risk of all-cause mortality [35]. Furthermore, polymorphisms G395A and C1818T in the klotho gene have been correlated with an elevated risk of developing T2D [36]. Klotho exerts a protective effect against diabetic retinopathy by attenuating the senescence of retinal macrophages, which is achieved by downregulating HECTD1 and diminishing IRS1 ubiquitination, and subsequent proteasomal degradation [37]. Numerous studies have also confirmed the potential mechanisms of α-klotho in alleviating diabetes and its complications [36], NAFLD [38], obesity [39], and other metabolic diseases. Therefore, serum klotho can not only serve as a potential biomarker for metabolic diseases but also as a therapeutic target.

In the subgroup analysis, we observed the impact of gender and diabetes on the relationship between serum klotho and TyG-related indicators, particularly among female and non-diabetic patients. Our study results are consistent with previous research findings [40, 41]. Gender differences may be related to variations in hormone levels, fat distribution, and differences in insulin sensitivity. Women in menopause experience a decrease in estrogen levels, which can reduce sensitivity to insulin [42]. Our study subjects are primarily over the age

of 40. Furthermore, the reduction in estrogen levels can also lead to the accumulation of fat in the visceral area, thereby increasing metabolic risk [43]. Therefore, the gender differences observed in our study may be due to the influence of hormonal levels on serum klotho expression. Moreover, relevant research has also confirmed that estrogen can upregulate the expression of serum klotho [44]. Qiu S [41] investigated the relationship between klotho and TyG index in populations with and without diabetes, and the results indicated that klotho is positively associated with the TyG index in diabetic populations compared to those without diabetes. This result is consistent with our study findings. Wang C [40] speculated that as the level of IR increases, there may be a saturation effect in the association between klotho and TyG. In summary, the biological mechanisms underlying the influence of the relationship between serum klotho and TyG-related indicators in the non-diabetic female population still require further research for elucidation.

Our research has some advantages. Firstly, it represents the preliminary attempt to explore the association between serum klotho and indicators such as TyG-BMI, TyG-WC, and TyG-WHtR. This investigation may provide new insights into the prevention and treatment of both age-related conditions and metabolic disorders. Secondly, our study benefits from the weighted analysis of a population sample drawn from NHANES, enhancing the representativeness of the results. However, this study also has limitations. Firstly, this study is limited by its cross-sectional design, which hinders the ability to determine causality between klotho and TyG-BMI, TyG-WC, and TyG-WHtR. Secondly, our study included only the common complications of non-diabetic, hypertension, and hyperlipidemia, without incorporating other related chronic diseases. Future research should encompass a broader range of chronic conditions to fully understand the impact of different health status on the study outcomes. Finally, despite adjusting for potential confounding variables, there remains a possibility of unidentified factors that could impact the relationship between serum klotho and the TyG-BMI, TyG-WC, and TyG-WHtR.

Conclusion

Based on the analysis of nationally representative samples, our research revealed a negative nonlinear relationship between serum klotho and TyG-BMI, TyG-WC, and TyG-WHtR in U.S. adults. The findings indicate that the relationship between serum klotho and TyG-related indicators could offer a new perspective for anti-aging and prevention of metabolic diseases.

Abbreviations

TyG	Triglyceride glucose index
NHANES	National Health and Nutrition Examination Survey
BMI	Body mass index
WC	Waist circumference
WHtR	Waist-to-height ratio
ELISA	Enzyme-linked immunosorbent assay
GAM	Generalized additive model
IR	Insulin resistance
FBG	Fasting blood glucose
NAFLD	Non-alcoholic fatty liver disease
MAFLD	Metabolic-associated fatty liver disease
NCHS	National Center for Health Statistics
CDC	Centers for Disease Control and Prevention
PIR	Poverty income ratio
HbA1c	Glycosylated hemoglobin
mKL	Membrane-bound
sKL	Secretory or soluble
PPARα	Peroxisome proliferator-activated receptor α
DKD	Diabetic kidney disease
ox-LDL	Oxidized low-density lipoprotein
WAT	White adipose tissue
HFD	High fat diet
BAT	Brown adipose tissue
CKD	Chronic kidney disease

Supplementary Information

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Supplementary Material 1

Author contributions

YY: Writing – original draft, Conceptualization, Visualization, Formal Analysis, Methodology. YQ: Writing – original draft, Formal Analysis, Software, Methodology. FL: Formal Analysis, Software, Methodology. YM: Writing – review & editing, Formal Analysis, Software, Methodology. TT: Writing – review & editing, Formal Analysis, Software, Methodology. YS: Writing – review & editing, Supervision, Project administration, Funding acquisition, Conceptualization.

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Data availability

No datasets were generated or analysed during the current study.

Declarations

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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