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Galectin-1 is inversely associated with type 2 diabetes independently of obesity – A SCAPIS pilot study☆

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ABSTRACT

Objectives: Galectin-1 is a recently discovered adipokine that increases with obesity and increased energy intake in adipose tissue. Our aim was to assess whether serum galectin-1 is associated with type 2 diabetes (T2D) and other parameters of the metabolic syndrome independently of body mass index (BMI) in a cohort from the general population.

Methods: In this cross-sectional population-based cohort study from the western part of Sweden, we investigated associations between serum galectin-1, clinical characteristics and inflammatory markers in 989 women and men aged 50–65 years [part of the Swedish CArdioPulmonary bioImage Study (SCAPIS) pilot cohort].

Results: We showed in linear models that serum galectin-1 was independently and: (1) inversely associated with T2D ($p < 0.05$) and glucose ($p < 0.05$); and (2) positively associated with age ($p < 0.01$), sex ($p < 0.01$), BMI ($p < 0.01$), insulin ($p < 0.01$) and C-reactive protein ($p < 0.01$). Furthermore, galectin-1 demonstrated univariate correlations with triglycerides ($r = 0.20$, $p < 0.01$), homeostasis model assessment for insulin resistance ($r = 0.24$, $p < 0.01$), tumor necrosis factor-α ($r = 0.24$, $p < 0.01$), interleukin-6 (IL-6; $r = 0.20$, $p < 0.01$) and HbA1c ($r = 0.14$, $p < 0.01$).

Conclusion: In a cross-sectional study of a middle-aged population, we showed that serum galectin-1 is: (1) inversely associated with T2D independently of BMI; and (2) independently associated with other markers of the metabolic syndrome These results warrant prospective and functional studies on the role of galectin-1 in T2D.

1. Introduction

Galectin-1, which is part of a larger family of carbohydrate-binding proteins with an affinity for beta-galactosides, is known to have anti-inflammatory and proangiogenic effects [1]. Recent studies have shown that galectin expression is upregulated in adipose tissue of mice with high-fat-diet-induced obesity [2] and that serum galectin-1 levels is positively correlated with body fat in children with obesity [3]. Moreover, we have showed that galectin-1 gene expression in adipose tissue decreased during weight loss in obese patients and increased after overfeeding in healthy subjects [4]. In line, a study in overweight and obese participants showed

Abbreviations: ALAT, alanine aminotransferase; BMI, body mass index; CRP, C-reactive protein; ELISA, electrochemiluminescence immunoassay; HDL, high-density lipoprotein; HOMA, homeostasis model assessment; IFN-γ, interferon gamma; IL, interleukin; LDL, low-density lipoprotein; MSD, Meso Scale Diagnostics; SCAPIS, Swedish CArdioPulmonary bioImage Study; SEM, standard error of the mean; TNF-α, tumor necrosis factor-α; T2D, type 2 diabetes.

☆ Swedish CArdioPulmonary bioImage Study.

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that galectin-1 expression in abdominal adipose tissue is regulated by dietary interventions [5].

Our laboratory recently proposed a functional role of galectin-1 in subcutaneous adipose tissue in individuals with type 2 diabetes (T2D) [4]. However, it is unclear whether circulating galectin-1 associates with T2D independently of adipose tissue mass. Indeed, little is known about the association between circulating galectin-1 and T2D including correlations with components of the metabolic syndrome. Here, we investigated associations between serum galectin-1, clinical characteristics and inflammatory markers in a large population-based cohort of middle-aged subjects from the Swedish CArdioPulmonary bioImage Study (SCAPIS) pilot.

2. Methods

2.1. Participants

This study was conducted in participants of the SCAPIS pilot [6]. It was approved by the Ethical Committee of Gothenburg University and written informed consent was obtained from all participants. In brief, 1111 individuals residing in Gothenburg, Sweden, and aged 50–64 years at the time of invitation participated in the SCAPIS pilot. Serum galectin-1 was not measured in 24 individuals from whom we did not get consent for biomarker analysis or from whom we could not obtain venous blood. Because galectin-1 is associated with cancer, only participants without a medical history of cancer were enrolled. Thus, we excluded those with a medical history of cancer (n = 81) or when a medical history of cancer was not known (n = 12). Individuals with latent autoimmune diabetes of the adult or type 1 diabetes were also excluded (n = 5). In total, 989 individuals were enrolled in the study (Fig. 1).

2.2. Blood analysis

Galectin-1 was measured using the Human Galectin-1 Quantikine ELISA Kit (R&D Systems, Minneapolis, MN, USA) according to the manufacturers' instructions. Intra-assay and inter-assay coefficient of variation were 7.1% and 9.5%, respectively. Cytokines were measured in serum using the ultrasensitive multiplex electrochemiluminescence immunoassay (ELISA) (Meso Scale Diagnostics (MSD), Rockville, MD, USA). The human V-PLEX Pro-Inflammatory Panel 1 Human Kit (#K15049) was used to quantify interferon gamma (IFN-γ), interleukin (IL)-1β, IL-2, IL-4, IL-6, IL-8, IL-10, IL-12p70, IL-13 and tumor necrosis factor-alpha (TNF-α) according to the manufacturer's instructions.

Routine analyses of glucose, insulin, and cholesterol were performed at the Department of Clinical Chemistry, Sahlgrenska University Hospital, on a Cobas instrument (Roche Molecular Diagnostics). The homeostasis model assessment for insulin resistance (HOMA-IR) was calculated using the formula [glucose]*[insulin]/22.5 [7].

2.3. Statistics

Multiple comparisons between groups were tested with Kruskal-Wallis test and Dunn's multiple comparisons test post hoc. Single comparisons between groups on continuous variables were calculated using Mann-Whitney *U* test using GraphPad Prism 7 (GraphPad Software, La Jolla, CA, USA). Chi^2 -test and Fisher's exact test were used for comparing categorical data. Spearman correlations were performed between galectin-1 levels and clinical characteristics of the subjects. To segregate the impact of covariates, two linear models were calculated using SPSS Statistics 25 (IBM, New York, USA). Non-Gaussian distributed variables according to Kolmogorov–Smirnov were log-transformed for near normal distribution in the linear model. *P* values below 0.05 were considered statistically significant for all statistical tests.

3. Results

3.1. Demographics

Clinical characteristics of the population are presented in Table 1. The average participant was overweight and men (n = 473) had a higher BMI than women (n = 516). Measures of glucose homeostasis including glucose and insulin, but not HbA1c, were higher in men than women. Total cholesterol and high-density lipoprotein (HDL) cholesterol were higher in women whereas triglyceride levels were higher in men. Low-density lipoprotein (LDL) cholesterol levels were similar in men and women.

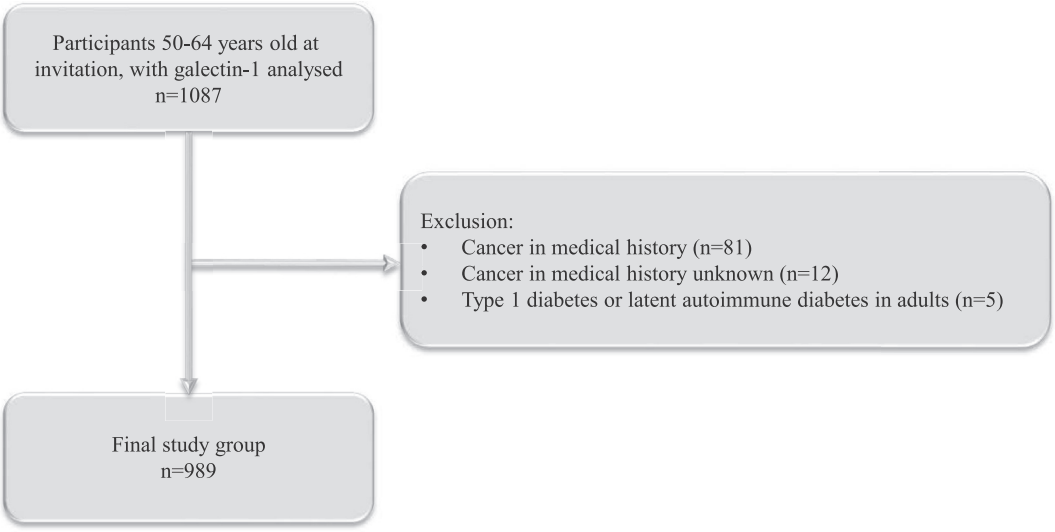

Fig. 1. Flowchart of participant selection for the study.

Table 1
Clinical characteristics of the study population.

	Women	Men	*p value*
n (%)	473 (48)	516 (52)	
Age, years	57 [54; 61]	57 [54; 62]	ns.
BMI, kg/m^2	26.0 [23.5; 29,2]	27.2 [25.1; 29.5]	**<0.01**
Waist, cm	89 [81; 98]	99 [94; 105]	**<0.01**
Systolic blood pressure, mmHg	120 [111; 130]	125 [116; 136]	**<0.01**
Diastolic blood pressure, mmHg	72 [66; 78]	77 [72; 83]	**<0.01**
Current smoker, n (%)	93 (20)	93 (18)	ns.
Current employment, n (%)	355 (75)	391 (76)	ns.
Galectin-1, ng/mL	22.7 [19.8; 26.1]	21.5 [18.9; 24.8]	**<0.05**
Glucose, mmol/L	5.5 [5.1; 5.8]	5.8 [5.5; 6.3]	**<0.01**
Insulin, mU/L	6.4 [4.3; 9.1]	7.7 [5.2; 11.0]	**<0.01**
HbA1c, mmol/mol	35 [33; 37]	35 [33; 38]	ns.
Total Cholesterol, mmol/L	5.9 [5.3; 6,6]	5.6 [4.8; 6.3]	**<0.01**
LDL-C, mmol/L	3.8 [3.1; 4.5]	3.8 [3.1; 4.4]	ns.
HDL-C, mmol/L	1.8 [1.5; 2.3]	1.4 [1.2; 1.7]	**<0.01**
Triglycerides, mmol/L	1.0 [0.74; 1.4]	1.2 [0.91; 1.8]	**<0.01**
Creatinine, μmol/L	70 [63; 77]	86 [79; 94]	**<0.02**
ALT, μkat/L	0.39 [0.31; 0.50]	0.48 [0.38; 0.63]	**<0.01**
Hypertension, n (%)	135 (29)	172 (33)	ns.
T2D, n (%)	19 (4)	49 (9)	**<0.01**
Prevalent cardiovascular disease[a], n (%)	9 (2)	21 (4)	**<0.05**
Rheumatic disease, n (%)	40 (8)	23 (4)	**<0.05**
Any prescribed drug, n (%)	217 (46)	209 (41)	ns.
Pharmacological diabetes treatment, n (%)	9 (2)	31 (6)	**<0.01**
Statins, n (%)	34 (7)	49 (9)	ns.
Betablockers, n (%)	32 (7)	35 (7)	ns.
ACE inhibitors or ARB, n (%)	54 (11)	59 (11)	ns.

BMI: body mass index; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol; ALT: alanine aminotransferase; ACE inhibitor: angiotensin II converting enzyme inhibitor; ARB: angiotensin II receptor blocker. HbA1c, ref value: 31–46 mmol/mol. Data are presented as n (%) or median [1st; 3rd quartile] for categorical data and continuous data respectively. [a] Medical history of acute myocardial infarction, stroke, percutaneous coronary intervention, or radiological examinations demonstrating coronary artery stent or graft.

3.2. Correlations between galectin-1, clinical, metabolic and inflammatory variables

We observed significantly higher levels of galectin-1 in women than in men (Table 1). Galectin-1 displayed a weak correlation with age and systolic blood pressure (Fig. 2 A and B) but a stronger correlation with waist and BMI circumference (Fig. 2C and D) and increased step-wise with BMI in both sexes (Fig. 2 E). There was no difference in galectin-1 between participants with (n = 68) or without (n = 921) T2D (Appendix Table A). Markers associated with the metabolic syndrome were consistently higher in participants with T2D than in participants without T2D (Appendix Table A). Galectin-1 showed positive correlations with glucose, insulin, HOMA, HbA1c, and triglycerides but an inverse relation with HDL (Fig. 3A–F). Galectin-1 levels also correlated positively with C-reactive protein (CRP), IL-6, TNF-α, and IFN-γ (Table 2).

3.3. Independent predictors of galectin-1

To adjust for covariates associated with galectin-1, a linear model was used. We first tested the impact of age, sex, BMI and T2D and showed that they were all significant independent predictors of galectin-1 (Table 3). Interestingly, participants with T2D were predicted to have lower galectin-1 levels than non-diabetic participants and women were predicted to have higher galectin-1 levels than men, when confounding variables were accounted for. Furthermore, galectin-1 was predicted to increase with age and for every unit increase in BMI.

As type 2 diabetes is a multifactorial disease characterized by both low-grade inflammation and metabolic aberrations, a second linear model was used to determine which characteristic features of T2D could interact with galectin-1. Glucose, insulin and CRP were all independently associated with galectin-1. Of note, glucose was the only parameter in addition to T2D that demonstrated an inverse relationship with galectin-1 (Table 3).

4. Discussion

This study investigated the association between galectin-1 and variables of the metabolic syndrome in a large cohort of middle-aged Swedish subjects. We show that galectin-1 is inversely associated with type T2D independent of obesity and that age, sex, BMI, insulin, CRP and glucose were all independent predictors of galectin-1.

The correlations observed for galectin-1 and clinical parameters in this cohort are in agreement with previously observed associations between galectin-1 and variables of the metabolic syndrome [3,4]. Variables showing the strongest correlations with galectin-1 in this study were markers of obesity and insulin resistance. BMI, waist, insulin and triglycerides had a stronger correlation with galectin-1 than markers of glycemia such as glucose and HbA1c. In line with the observed associations, recent animal studies have indicated a role for galectin-1 in obesity [2,8–10]. In rats, treatment with the galectin-1 inhibitor thiodigalactoside induced weight loss and suppressed triglyceride synthesis and lipid storage, suggesting that the inhibitor interferes with lipid droplet formation [8]. Additional animal studies on galectin-1 and adiposity also support the relevance of galectin-1 in adipose tissue physiology [2]. However, further studies are required to determine the functional role of galectin-1 in lipid metabolism.

Surprisingly, galectin-1 was predicted to be lower in participants with T2D compared with non-diabetic participants after adjusting for age, sex and BMI. The paradoxical finding of an inverse association between galectin-1 and T2D could indicate a down-regulation of galectin-1 by genetic factors or glucose per se [3,11] that may happen in predisposed individuals. The inverse

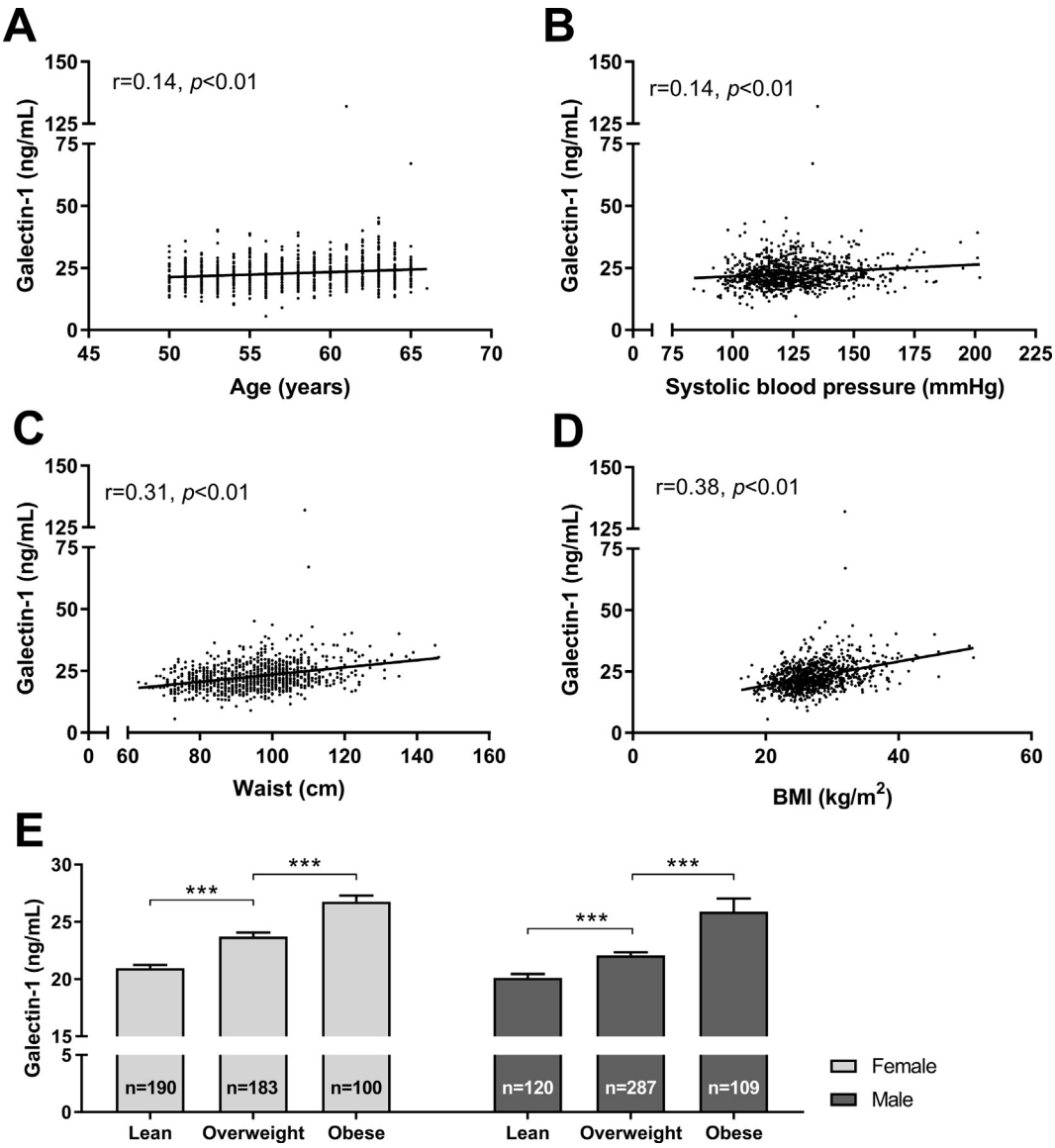

Fig. 2. Spearman correlations between galectin-1 and clinical characteristics. (A) Age, n = 989; (B) systolic blood pressure, n = 979; (C) waist, n = 988 and (D) BMI, n = 989. (E) Galectin-1 in women and men divided according to BMI presented as mean ± standard error of the mean (SEM). ***p < 0.001.

association between glucose and galectin-1 in the linear model further strengthens such a concept. In line with this hypothesis, it has been reported that women with a genetic predisposition in chromosome 22 who develop gestational diabetes present an inability to increase galectin-1 in the later parts of pregnancy, in contrast to healthy pregnant women [11]. Of note, in this study women displayed higher serum galectin-1 compared with men, and sex predicted galectin-1 in the linear models. Similar observations were made in a small sample of 38 healthy controls in a recent study in cancer patients [12]. Longitudinal studies of large cohorts are required to address whether the association between galectin-1 and T2D differs between women and men.

Despite the inverse association between T2D and galectin-1 it is possible that associations between galectin-1 and facets of the metabolic syndrome shown in this study indicate increased diabetes risk. It is well known that subclinical inflammation measured as IL-6 and TNF-α in the adipose tissue links obesity to insulin resistance [13,14]. Therefore, it was interesting that serum levels of these cytokines also presented the closest correlation with galectin-1 in this study. Results from the linear model that included CRP (chosen because it was the inflammatory marker with the strongest correlation to galectin-1) showed that CRP associated with galectin-1 independent of glucose and insulin levels. This is important as galectin-1 has been widely investigated in inflammatory conditions including different cancers [15], and inhibitors of galectin-1 are undergoing clinical trials within this field [16].

In conclusion, galectin-1 was associated with T2D independent of obesity in a cross-sectional study of middle-aged subjects. Furthermore, galectin-1 was associated with several different markers of metabolic disease. These results warrant prospective and functional studies on the role of galectin-1 in T2D.

Author contributions

LS and AL analysed the samples. LB, LMH, GB and PAJ planned the study. EF and MS analysed the data. EF and PAJ wrote the paper. All authors gave constructive comments and approved the final article.

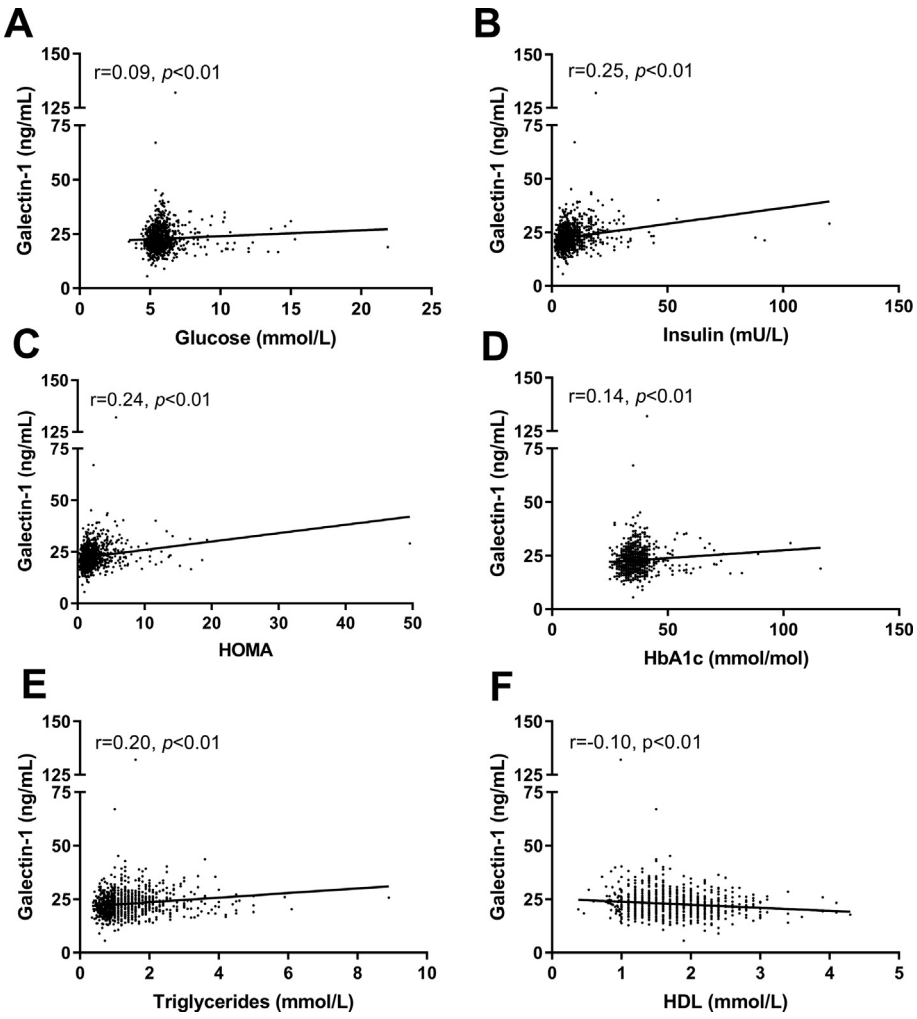

Fig. 3. Spearman correlations between galectin-1 and variables indicating insulin resistance. (A) glucose, n = 982; (B) insulin, n = 989; (C) HOMA, n = 982; (D) HbA1c, n = 987; (E) triglycerides, n = 988 and (F) HDL, n = 989.

Table 2
Spearman correlations between galectin-1 and inflammatory markers.

	r	*p value*	n
CRP	0.27	**<0.001**	989
TNF-α	0.24	**<0.001**	989
IFN-γ	0.08	**<0.05**	989
IL-1β	−0.00	ns.	989
IL-2	0.05	ns.	989
IL-4	−0.01	ns.	989
IL-6	0.20	**<0.001**	989
IL-8	0.02	ns.	989
IL-10	0.03	ns.	989
IL-12 p70	0.04	ns.	989
IL-13	−0.01	ns.	989

IFN- γ: interferon gamma; IL: interleukin; TNF-α: tumor necrosis factor alpha.

Table 3
Linear model predicting Ln Galectin-1 based on T2D, sex, age and BMI (Model 1), and a second model predicting Ln Galectin-1 based on sex, age, BMI, and markers of insulin resistance and subclinical inflammation (Model 2).

Parameter	β	SE	95% CI		Sig.
			Lower	Upper	
Model 1					
Female sex	0.06	0.01	0.04	0.09	**<0.01**
Ln Age (years)	0.50	0.09	0.33	0.67	**<0.01**
Ln BMI	0.61	0.04	0.53	0.70	**<0.01**
T2D	−0.05	0.03	−0.11	0.00	**<0.05**
Model 2					
Female sex	0.06	0.01	0.03	0.08	**<0.01**
Ln Age (years)	0.50	0.09	0.33	0.67	**<0.01**
Ln BMI	0.48	0.06	0.37	0.59	**<0.01**
Ln Glucose	−0.13	0.05	−0.22	−0.04	**<0.01**
Ln S-Insulin	0.04	0.01	0.01	0.07	**<0.01**
Ln CRP	0.02	0.01	0.01	0.04	**<0.01**

BMI: body mass index; CRP: C-reactive protein. Galectin-1, age, BMI, glucose, insulin and CRP were log transformed with the natural logarithm (ln).

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Duality of interest

The authors have no conflicting interests to disclose.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.metop.2019.100017.

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