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Case report

Circulating galanin and galanin like peptide concentrations are correlated with increased triglyceride concentration in obese patients

Penghua Fang [a,c,1], Mei Yu [e,1], Xuewen Gu [d,1], Mingyi Shi [b,c], Yan Zhu [b,*], Zhenwen Zhang [b,*], Ping Bo [b,*]

[a] Department of Physiology, Nanjing University of Chinese Medicine Hanlin College, Taizhou 225300, China
[b] Department of Endocrinology, Clinical Medical College, Yangzhou University, Yangzhou 225001, China
[c] Jiangsu Key Laboratory of Integrated Traditional Chinese and Western Medicine for Prevention and Treatment of Senile Diseases, Medical College, Yangzhou University, Yangzhou, China 225001
[d] Department of Pathology, Clinical Medical College, Yangzhou University, Yangzhou, Jiangsu 225001, China
[e] Taizhou Hospital of Traditional Chinese Medicine, Nanjing University of Chinese Medicine, Taizhou, Jiangsu 225300, China

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ABSTRACT

Background: Obesity is strongly linked to metabolic complications, including type 2 diabetes mellitus and hyperlipidemia. Experimental evidences indicate that galanin (GAL) and galanin-like peptide (GALP) are involved in the regulation of feeding behavior and energy metabolism. We evaluated the possible relationships between both peptide concentrations and blood fat indexes in obese and normal subjects.
Methods: The study groups consisted of 41 obese subjects (age 35.17 ± 12.29 year, BMI 30.97 ± 2.75 kg/m^2) and 38 healthy controls (age 38.47 ± 11.63 year, BMI 22.83 ± 3.00 kg/m^2). Plasma GAL and GALP concentration was determined using ELISA.
Results: Plasma GAL and GALP concentration was significantly higher in obese subjects than healthy controls ($P < 0.001$). In addition, the positive correlations were found between: GAL and triglyceride (TG) concentrations ($r = 0.636$; $P < 0.001$), GALP and TG concentrations ($r = 0.362$; $P = 0.020$) in obese subjects. *Conclusions:* Our results indicated that obese individuals have higher plasma GAL and GALP concentrations and both peptide concentrations were positively correlative to TG concentrations in obese human. GAL and GALP concentrations may be taken as potential biomarkers to predict development of obesity.

1. Introduction

The rapid increase in overweight and obesity is becoming an important health problem worldwide [1]. Obesity may cause several metabolic complications, including type 2 diabetes mellitus, hyperlipidemia, high cholesterol, coronary artery disease as well as hypertension [1]. Prevention and treatment of obesity will benefit the treatment of these related diseases. Despite extensive investigations into obesity entity, the precise molecular mechanisms of obesity have not been fully elucidated. Thus, further exploration of the precise molecular mechanisms of obesity is urgently needed.

Several peptides in brain and adipose tissue have been identified as potential biomarkers to forecast development of obesity [2]. Experimental evidences display that galanin (GAL) and galanin like peptide (GALP) are involved in regulation of feeding behavior and energy metabolism of animals [3]. GAL, a 29/30 amino-acid peptide, is widely distributed throughout the central and peripheral nervous systems and other tissues [4]. The GAL protein and mRNA concentrations were increased in the paraventricular nucleus (PVN) of obesity-prone rats fed with high-fat diet, in comparison with the obesity-prone rats fed with high-carbohydrate diet or obesity-resistant rats fed with high-fat diet [5,6]. Administration of GAL can increase food intake and fat preference, and reduce energy metabolism, resulting in enhanced risk for development of obesity, dyslipidemia and metabolic syndrome [7]. These roles of GAL in modulating energy metabolism mediates by acting GAL1 in hypothalamic neuronal circuits [2]. The homozygous galanin transgenic C57BL/6J mice exhibited reduced energy expenditure and increased body weight [8]. In addition, a significant positive correlation was found between GAL concentration and body mass index (BMI) in women with gestational diabetes mellitus [9].

GALP is a 60 amino-acid neuropeptide that shares same sequence homology with GAL (1−13) in position 9–21 [4]. There is growing evidence supporting that GALP can regulate feeding behavior, body weight and energy homeostasis in animals [7]. Different from GAL, the impact of GALP on of obesity is still controversial. Some studies found that an acute injection of GALP into intracerebroventricles can decrease food intake and body weight of murine [10,11]. The chronic administration of GALP for 14 days may decrease body weight and food intake in ob/ob mice too [12]. However, other studies reported that the central

* Corresponding authors at: Department of Endocrinology, Clinical Medical College, Yangzhou University, Yangzhou 225001, China.
E-mail address: physiolpep@sina.cn (Z. Zhang).
[1] The authors contributed equally to this work.

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injection of GALP may increase glucose uptake, lipid metabolism and GLUT4 mRNA expression concentrations, but inhibit gluconeogenesis and fatty acid synthesis in mice [13]. After fasting, the GALP-knockout mice consumed less food and gained less weight than the wild-type controls as feeding with a high fat diet [14].

2. Materials and methods

The present study was conducted in the Clinical Medical College, Yangzhou University. The present study consisted of 41 obese volunteers and 38 age- and gender-matched healthy volunteers with normal weight according to a physical examination and routine laboratory tests. In this study each participant gave official written consent, had normal exercise and eating behavior, without fat preference or fat aversion. Individuals with diabetes, active hepatitis/liver cirrhosis, hypertension, chronic renal failure on hemodialysis, congestive heart failure or other known major disease were precluded from the study. According to the criteria of the Working Group on Obesity in China (WGOC) [15], general obesity was defined as BMI ≥ 28.0 kg/m^2 and normal weight was defined as 19.0 < BMI < 24.0 kg/m^2.

For each case BMI was calculated at the time of blood collection as weight in kilograms divided by height in meters squared. HOMA-IR index was calculated for each participant using the formula [fasting glucose (mmol/L) × fasting insulin (mIU/L)/22.5]. The protocol of the study was approved by the Ethics Committee of Clinical Medical College, Yangzhou University.

After an overnight fast, blood samples were collected from each participant at 08:00 A.M. and immediately centrifuged (4 °C) as described previously [9]. In brief, the blood samples (2 ml) were collected in prechilled EDTA tubes containing 100 μl aprotinin (1 μg/ml) and were immediately centrifuged for 15 min at 1000 ×*g* 4 °C within 30 min of collection. Plasma was separated into vials and stored at −80 °C until measurement. Plasma insulin concentrations were measured by radioimmunoassay. Regular biochemical tests included glucose, triglyceride (TG), total cholesterol (TC), high-density (HDL-C) and low-density lipoprotein cholesterol (LDL-C) were performed on the Olympus AU2700 chemistry analyzer. After routine analysis, left over serum was immediately stored at −80 °C for GAL and GALP determination.

Plasma concentration of human GAL and GALP was analyzed using an enzyme-linked immunosorbent assay (CUSABIO, Inc.). According to the manufacturer's specification, the assay range for GAL was 4.7–300 pg/ml, and the average sensitivity was 1.17 pg/ml and intra-assay precision CV% <8% and inter-assay precision CV% <10%, as well as the assay range for GALP was 0.156–10 ng/ml, and the average sensitivity was 0.039 ng/ml, Intra-assay precision CV% < 8% and inter-assay precision CV% < 10%. All measurements were performed in duplicate, and the mean of the two measurements was considered.

2.1. Statistical analysis

The statistical analyses were performed with SPSS 17.0 for Windows. All data were presented as mean ± SD. The differences between the groups were analyzed with independent *t*-test. Correlations were evaluated using the multiple linear regression method. Statistical significance was considered to be P <0.05.

3. Results

The main indexes of body weight, BMI, glucose, insulin, TG, TC, HDL-C and LDL-C in obese group and healthy non-obese group were listed in Table 1. The plasma GAL and GALP concentrations were significantly higher in obese subjects compared to healthy controls (GAL, women 34.68 ± 11.48 vs. 16.50 ± 5.78 pg/ml, P < 0.001; men 27.33 ± 10.70 vs. 13.66 ± 4.12 pg/ml, P < 0.001; GALP, women 0.96 ± 0.34 vs. 0.70 ± 0.31 ng/ml, P < 0.001; men 1.12 ± 0.27 vs. 0.76 ± 0.30 ng/ml, P < 0.001) (Figs. 1 and 2). Additionally, the positive correlations were found between: GAL and TG (r = 0.636; P < 0.001), GALP and TG (r = 0.362; P = 0.020) in the obese group.

Table 1
Biochemical and demographic characteristics of two groups.

	normal control	Obese subject	P value
N	38 (male, 16; female 22)	41 (male, 18; female 23)	
Age (y)	38.47 ± 11.63	35.17 ± 12.29	0.224
Body weight (kg)	62.16 ± 8.90	81.91 ± 9.90	<0.001
BMI (kg/m2)	22.83 ± 3.00	30.97 ± 2.75	<0.001
Fasting glucose (mmol/l)	4.84 ± 0.50	5.77 ± 1.20	<0.001
Fasting Insulin (mIU/l)	6.75 ± 2.80	11.14 ± 3.47	<0.001
TG (mmol/l)	1.54 ± 0.82	2.36 ± 0.89	<0.001
TC (mmol/l)	4.70 ± 0.97	5.47 ± 0.97	0.001
HDL-C (mmol/l)	1.37 ± 0.23	1.26 ± 0.20	0.030
LDL-C (mmol/l)	2.67 ± 0.54	3.06 ± 0.53	0.002
HOMA-IR	1.48 ± 0.72	2.83 ± 0.90	<0.001

Results are shown as means ± SD; N, number of cases; statistical significance P < 0.05.

4. Discussion

The increasing evidence showed that appetite and satiety close associated with food intake and energy metabolism in animals through the actions of GAL and GALP [2]. The dysfunction of both peptides in orexigenic and anorexigenic regulation played a key role in the pathogenesis of obesity and metabolic syndrome. The circulating GAL concentrations were increased in obese women [16,17] but decreased in thin women compared to controls. GAL plays an important signaling role in hypothalamus to regulate feeding and energy homeostasis [7]. In line with these studies, our study showed that circulating GAL concentrations were significantly increased in obese subjects compared with controls with normal weight.

Data display there is a interaction between plasma TG concentrations and GAL protein and mRNA expression in the PVN of rats [5,6]. On the one hand, a fatty diet may enhance the central galanin protein and mRNA concentration of animals. In a chow test, the diet rich in saturated fatty acids increased not only circulating TG concentration, but also GAL protein and mRNA contents in the PVN compared with unsaturated fatty acids in rats [18]. The GAL mRNA concentration in the PVN was consistently and positively correlated with the TG concentrations [19]. Blocking fatty acid metabolism by intraperitoneal injection of mercaptoacetate, the galanin concentration in the anterior PVN was reduced in rats [20]. On the other hand, an acute and chronic increase in

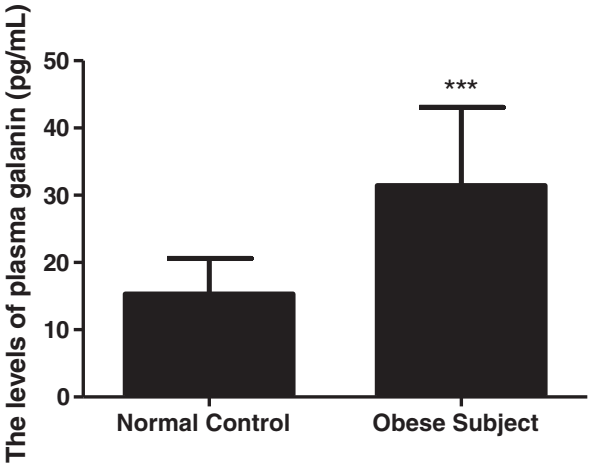

Fig. 1. The concentrations of plasma GAL in normal control and obese subject. Data are expressed as mean ± SD, ***P < 0.001 vs. normal control.