Circulating levels of the adipokines vaspin and omentin in patients with juvenile idiopathic arthritis, and relation to disease activity

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Abstract

Objective

Vaspin and omentin are two recently discovered adipokines that have been involved in chronic inflammatory processes. The aims of our study were to evaluate their serum levels in patients affected by juvenile idiopathic arthritis (JIA), in comparison to healthy controls, and to correlate circulating levels to parameters of disease activity.

Methods

Serum levels of omentin and vaspin were assayed by enzyme-linked immunosorbent assay in 40 patients with JIA classified according to the ILAR criteria and 26 healthy controls.

Results

Serum omentin levels were significantly higher in JIA patients versus healthy controls ($p<0.0001$) whereas serum vaspin levels did not significantly differ between the two groups. JIA children with active joints showed higher omentin serum levels than JIA children without active joints ($p<0.001$) and omentin serum levels significantly correlated with the presence of active joints ($p<0.0001$). Omentin serum levels were also significantly related with the number of active joints ($p<0.002$). Vaspin serum level did not show statistical significant differences between JIA children with active joints and those with no active joints. There was no correlation between plasma vaspin levels and the presence of active joints, or the number of active joints

Conclusions

Our study is the first report on the new adipokines vaspin and omentin in patients with JIA, and it shows that omentin is significantly higher in JIA patients in comparison with healthy controls. In addition, we also report that omentin plasma levels are significantly correlated with the presence and the number of active joints.

Key words

juvenile idiopathic arthritis, adipokines, omentin, vaspin, inflammation

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Introduction

White adipose tissue produces more than 50 adipokines and other molecoles that partecipate through endocrine, paracrine, autocrine or juxtacrine mechanisms of action in a wide variety of physiopathological processes, including food intake, insulin sensitivity, vascular sclerotic processes, immunity and inflammation (1). Vaspin and omentin are two recently discovered adipokines, that have been involved in chronic inflammatory processes (2, 3).

Vaspin (visceral-adipose-tissue-derived serpin) was discovered by Hida *et al*. as a serpin (serine protease inhibitor) produced in rat visceral adipose (2). The induction of vaspin by adipose tissue may constitute a compensatory mechanism in response to obesity, severe insulin resistance and type 2 diabetes (2). Furthermore, vaspin is reported to likely have anti-inflammatory actions as it suppresses the productions of TNF-α and proinflammatory adipokines (2).

Omentin-1 is a novel 34 kDa adipokine that is more likely to be produced by visceral adipose tissue than by subcutaneous adipose tissue. Although its physiological roles remain largely unknown, recent data suggest that it might be involved in intestinal defense mechanisms in chronic inflammatory diseases (3-6). Omentin has recently been involved in chronic inflammatory processes and it can be hypothesized that its levels could be relevant to systemic inflammation (4-6).

Elevated vaspin and reduced omentin levels have been recently found in the synovial fluid of patients with rheumatoid arthritis, thus demonstrating their different regulations at the site of local inflammation (5).

The aims of our study were to evaluate serum vaspin and omentin levels in patients affected by juvenile idiopathic arthritis (JIA), in comparison to healthy controls, and to correlate circulating levels to parameters of disease activity

Materials and methods

Our study group included 40 children (7 male, 33 female) with JIA, classified according to the ILAR criteria (7); fifteen had polyarticular onset disease and 25 had oligoarticular onset disease; 3/25 patients with oligoarticular diseases had extended oligoarthritis. None of them were receiving or had received oral or intra-articular corticosteroids before the start of the study. Ten patients with polyarticular and 7 with the oligoarticular disease were treated with oral methotrexate; 2 patients with oligoarticular disease were receiving oral sulphasalazine. The remaining patients were treated with non-steroidal anti-inflammatory drugs (NSAIDs) only.

Twenty-six healthy children (14 male, 12 female) attending our outpatient clinic (Department of Paediatrics, Rheumatology Unit, Anna Meyer Children's Hospital, University of Florence, Italy) for arthralgias and/or musculoskeletal pain were enrolled as controls. All healthy controls underwent routine clinical, laboratory, and instrumental investigations in order to rule out possible rheumatic diseases, infections, endocrine and/or metabolic disorders. Nobody presented any signs of inflammation. Table I shows the baseline demographic and clinical characteristics of JIA patients and healthy controls.

For the purposes of this study, we excluded subjects with history of diabetes mellitus, instable weight history and those treated with medications known to affect body weight.

Blood samples (6 ml) were drawn from an antecubital vein with patient in the supine position in the morning after an overnight fast. The blood was immediately centrifuged and serum was stored at -80°C until analysed.

Plasma vaspin levels were determined by the enzyme-linked immunosorbent assay method using Vaspin (human) Elisa kit (AdipoGen Inc. Korea) (5). Sensitivity of samples was 12 pg/ml. Inter- and intra-assay coefficients of variation were 3–9% and 1.5–3.5%, respectively.

Plasma omentin levels were detected by the enzyme-linked immunosorbent assay method using Manual Omentin 1 (human) ELISA Kit (Apotech Corporation) (5). Sensitivity of samples was 0.4 ng/ml. Inter- and intra-assay coefficients of variation were 4-9% and 4–7%, respectively. Ethics committee approval and informed consent from parents or guardians were obtained.

Statistical analysis

All results are expressed as mean ± standard deviation (S.D) or median (range). Body mass index standard deviation score (BMI-SDS) is expressed as mean ± standard error of the mean (SEM). Mann-Whitney U-test, with Fisher's exact test, when appropriate, and analysis of covariance (ANCOVA) with least significant difference (LSD) correction were used to evaluate the mean differences (±SD) of vaspin and omentin between groups, considering the following covariates for ANCOVA: sex, age, age at disease onset, age at the time of collecting sample, weight, height, BMI, BMI-SDS, disease duration; along with the number of active joints, erytrocyte sedimentation rate (ESR), C-reactive protein (CRP), when the different onset types of JIA were compared.

The Spearman rank correlation test was used to determine correlation coefficients between Omentin and Vaspin levels and the above reported entered variables.

Non-parametric tests were used, where necessary, due to the small size of our groups and to the skewness of our data. Levels of $p<0.05$ were considered statistically significant. Analyses were performed on SPSS package for Windows, version 13.0 (SPSS, Inc., Chicago, IL, USA).

Results

Serum omentin levels were significantly higher in JIA patients versus healthy controls ($p<0.0001$, Fig. 1a) whereas plasma vaspin levels did not significantly differ between the two groups (p = NS) (Table I) (Fig. 1b).

JIA children with active joints showed higher serum levels than JIA children without active joints (10.64 ng/ml *vs.* 6.45 ng/ml, $p<0.001$ Fig. 2a), with omentin serum levels significantly correlated with the presence of active joints ($r_s=0.77$, $r^2=0.32$, $p<0.0001$) (Fig. 2b).

Omentin serum levels were also significantly related with the number of active joints ($r_s=0.52$, $r^2= 0.27$, $p<0.002$) (Fig. 2c). However, no significant correlation was found between serum omentin and BMI ($r_s=-0.42$, p=NS), BMI-SDS ($r_s=-1.72$, p=NS), disease

Table I. Baseline demographic and clinical characteristics of JIA patients.

Characteristics	JIA patients	Controls	*p*-value
n. of subjects	40	26	
Sex (female/male)	33/7	14/12	
Age (years) (m±SD)	6.81 ± 4.6	8.84 ± 3.8	
BMI (kg/m^2) (m±SD)	16.61 ± 1,84	17.04 ± 1.96	n.s
BMI-SDS (m±SEM)	0.08 ± 0.17	0.09 ± 0.21	n.s
NSAIDs	21		
MTX	17		
SSZ (n. of subjects)	2		
Vaspin levels (ng/ml) (m±SD)	2.704 ± 2.73	3.78 ± 4,00	n.s
Omentin levels (ng/ml) (m±SD)	8.516 ± 3,0620	6.123 ± 1,8796	0.0001

JIA: juvenile idiopathic arthritis; mean ± SD: mean ± standard deviation; m ± SEM: mean ± standard error of the mean; BMI: body mass index; SDS: standard deviation score; NSAIDs: non-steroidal anti-inflammatory drugs; MTX: methotrexate; SSZ: sulphasalazine.

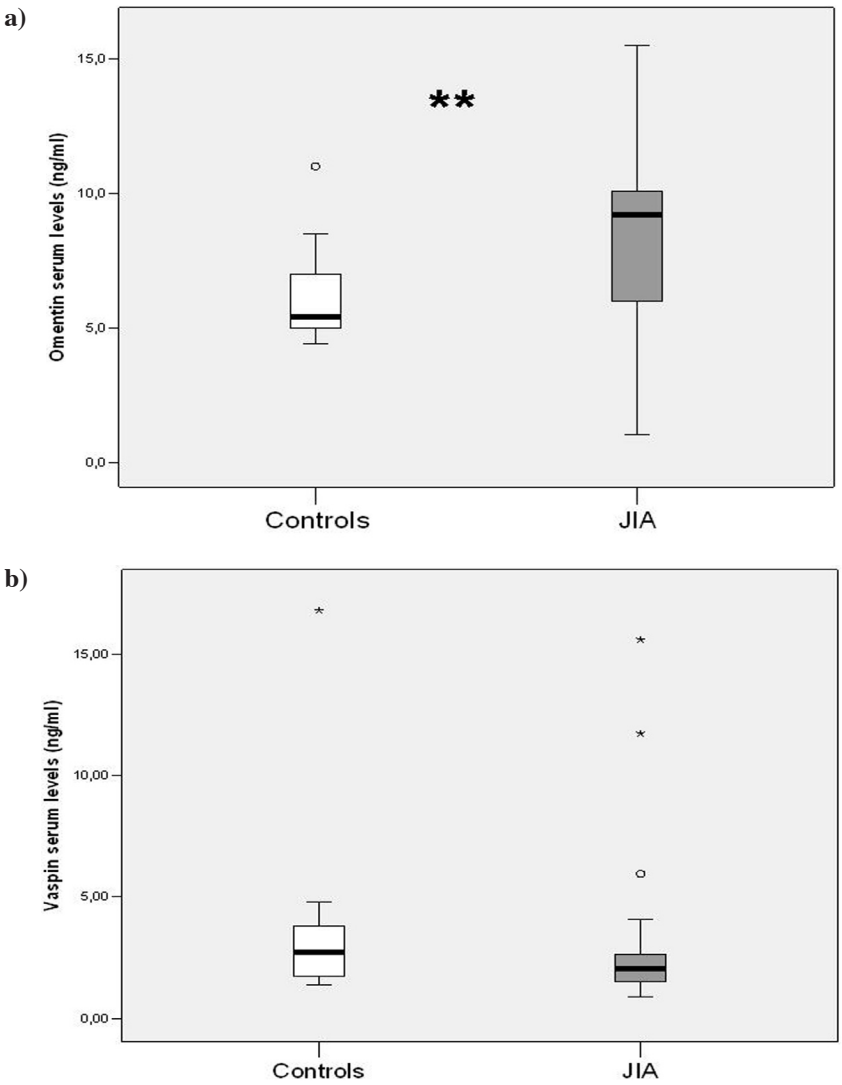

Fig. 1. a) Omentin serum levels in juvenile idiopathic arthritis (JIA) children and controls (**$p<0.0001$) The central line represents the distribution median, boxes span 25th to 75th percentiles, and error bars extend from 10th to 90th percentiles. Dots are values higher than the 90th percentile; **b)** Vaspin serum levels in juvenile idiopathic arthritis (JIA) children and controls. The central line represents the distribution median, boxes span 25th to 75th percentiles, and error bars extend from 10th to 90th percentiles. Dots are values higher than the 90th percentile, with proximal (°) and distal (*) outliers.

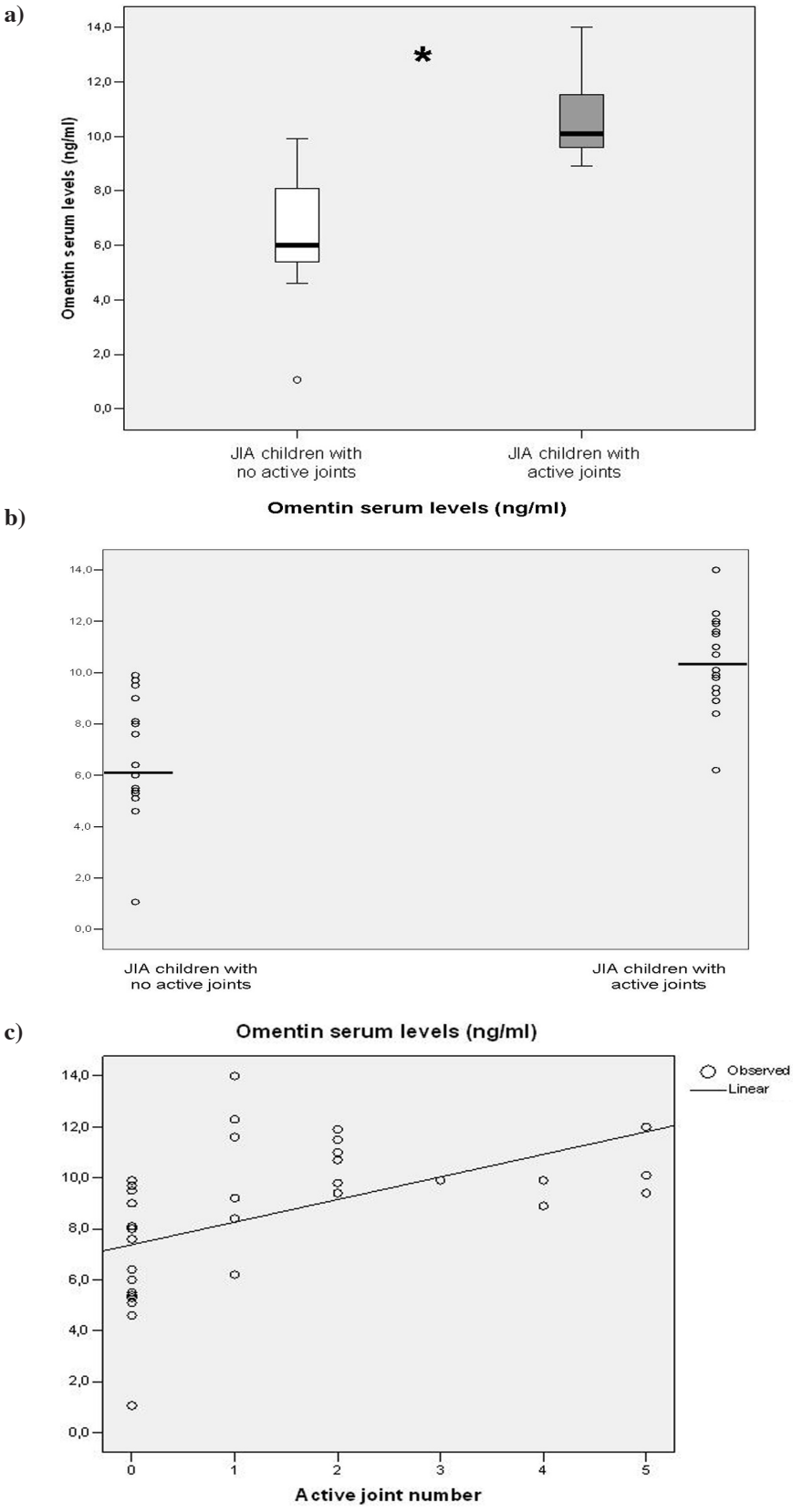

Fig. 2. **a)** Omentin serum levels in juvenile idiopathic arthritis (JIA) children with active joints and without active joints (*p<0.001) The central line represents the distribution median, boxes span 25th to 75th percentiles, and error bars extend from 10th to 90th percentiles. Dots are values higher than the 90th percentile; **b)** Correlation of the presence of active joints with omentin serum levels. r_s=0.77, r^2=0.32, p<0.0001 **c)** Correlation of the number of active joints with omentin serum levels. r_s=0.52, r^2=0.27, p<0.002.

duration (r_s=-0.08, p=NS), age (r_s=-0.14, p=NS), age at disease onset (r_s=-0.16, p=NS), gender (r_s=0.18, p=NS), ESR (r_s=0.22 , p=NS), CRP levels (r_s=-0.03, p=NS) and with the type of JIA (r_s=0.31, p=NS). Vaspin serum level did not shown statistical significant differences between JIA children with active joints and those with no active joints. (3.03±3.2 ng/ml *vs*. 2.44±2.2 ng/ml). There was no correlation between plasma vaspin levels and the presence of active joints (r_s=0.15, p=NS), the number of active joints (r_s=0.12, p=NS), BMI (r_s=-12, p=NS), BMI-SDS (r_s=- 0.22, p=NS), disease duration (r_s=0.11, p=NS), age (r_s=0.06, p=NS), age at disease onset (r_s=0.04, p=NS), gender (r_s=0.18, p=NS), ESR (r_s=0.19, p=NS), CRP (r_s=0,15, p=NS) and with the type of JIA (r_s=0.009, p=NS).

Discussion

In the last years the modulation of immunological and inflammatory pathways by adipokines has been estensively studied (1, 8-10).

Our study is the first report on the new adipokines vaspin and omentin in patients with JIA, and showed that omentin was significantly higher in comparison with healthy controls. In addition, we demonstrated that omentin plasma levels were significantly correlated with the presence and with the number of active joints.

In a previous study, synovial fluid concentrations of vaspin and omentin in rheumatoid arthritis (RA) patients were compared with those in osteoarthritis (OA) patients in order to characterise their potential association with the severity of RA (5). The authors first reported elevated levels of vaspin and reduced levels of omentin in the synovial fluid of patients with RA compared with those with OA. In addition, synovial fluid vaspin had a tendency to correlate with DAS28 in RA patients, although the correlation was not statistically significant.

Other authors recently reported that serum vaspin level patients are significantly increased in RA patients in comparison to healthy controls and patients with Behçet's disease. These increase remained significant even after adjust-

ments for age, gender, and BMI. In addition vaspin level was positively correlated with disease duration (11).

This discrepancy with our results may be due both to different patterns of adipokine distribution in the joint and the circulating compartment, as recently demonstrated for leptin and adiponectin in OA (12) and for resistin in RA in comparison to patients with OA and spondyloarthritis (13) and to the different age of patients and/or to different pathogenetic mechanism between RA and JIA.

It has been hypothesized that serum adipokine levels could be more relevant to systemic inflammation and/or disease activity (as we describe here), whereas synovial fluid adipokines might reflect the particular inflammatory process of the affected joint (13).

Recently, the spectrum of adipokines involved in arthritis has been rapidly widening (8). Further studies are needed in order to clarify possible omentin involvement in the regulation of the immune and/or inflammatory response. Our study failed to find correlations between serum vaspin levels and parameters of disease activity in JIA. On the contrary, omentin showed to be strongly correlated both to the presence of active joints and with the number of active joints. Our data suggest that omentin may be involved in the regulation of the inflammatory response in JIA. Although many issues still remain hazy, the increasing research effort in the area of adipokines is gradually revealing the intricate adipokine-mediated interplay among white adipose tissue, metabolic disorders and chronic inflammatory disorders. Understanding of the actions of the newer adipokines is still incomplete to generate well-supported therapeutic hypothesis, however the rate at which their roles are being clarified makes it probable that they might be central to pharmacotherapeutic approaches in immune disorders.

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