Adiponectin is associated with lipid profile and insulin sensitivity in French adolescents

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Abstract

Aim. – The favourable relationship of adiponectin with the metabolic profile demonstrated in adults has been less studied in youths. The aim of this study was to examine cross-sectional and longitudinal associations between adiponectin and various metabolic risk factors in 12-year-old adolescents.

Methods. – Subjects were participants in a randomized controlled study to promote physical activity (PA). Cross-sectional associations were assessed at entry in 2002 among 647 PA-exposed and control first-level students (49% male, 11.6 ± 0.6 years of age). Longitudinal analyses involved 288 control students surveyed in 2002 and 2004. Baseline measurements included fasting serum adiponectin and anthropometric indices (body mass, waist size, body fat [BF] by bioimpedance), insulin concentration, homeostasis model assessment (HOMA), high-density lipoprotein (HDL) cholesterol, triglycerides (TG), soluble TNF-α receptor 1 (sTNF-α R1) and high-sensitivity C-reactive protein. Analyses were performed with generalized linear mixed-effects models, taking into account correlations among adolescents in the same school.

Results. – Cross-sectionally, plasma adiponectin was inversely associated with obesity indices, especially waist size (P < 10^−2), HOMA (P < 0.03), insulin (P < 0.04), TG (P < 10^−4) and sTNF-α R1 (P < 0.05), and positively related to HDL cholesterol (P < 10^−4), after adjusting for age, gender, sexual maturity, sports participation and adiposity when relevant. Longitudinally, a higher baseline adiponectin level was associated with a more favourable two-year change in TG (P < 0.05), even after accounting for baseline TG, and two-year BF and insulin changes.

Conclusion. – The findings of this study suggest a favourable relationship between adiponectin and both metabolic profile and subsequent changes in TG level in young adolescents.

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Résumé

L’adiponectine est associée au profil lipidique et à la sensibilité à l’insuline chez des adolescents français.

Objectifs. – Les associations favorables entre l’adiponectine et le profil métabolique démontrées chez l’adulte ont été moins étudiées chez le sujet jeune. Le but de cette étude était d’examiner les associations transversales et longitudinales entre l’adiponectine et différents facteurs de risque métaboliques chez des adolescents de 12 ans.

Méthodes. – Les sujets étaient des participants à une étude contrôlée et randomisée visant à promouvoir l’activité physique (AP). Les associations transversales ont été étudiées lors de l’inclusion en 2002 chez 647 élèves de sixième soumis à l’intervention ou témoins (49 % de garçons, âge moyen 11,6 ans ± 0,6). Les analyses longitudinales concernaient les 288 élèves témoins suivis en 2002 et 2004. Les mesures à l’inclusion comprenaient l’adiponectine et des paramètres anthropométriques (poids, tour de taille (TT), masse grasse par impédancemétrie), l’insuline, l’index Homeostasis Model Assessment (HOMA), le cholestérol HDL, les triglycérides, le récepteur soluble de type 1 du TNF (sTNF-α R1) et la protéine-C réactive. Les analyses ont été effectuées avec des modèles linéaires généralisés mixtes prenant en compte les corrélations entre adolescents d’un même collège.

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2.1. Study population

Adiponectin is an adipose-tissue-specific protein, which has been shown to enhance insulin action and to exert antiatherogenic effects. Plasma levels of adiponectin are reduced in obese and diabetic subjects [1], and in patients with metabolic syndrome or cardiovascular diseases [2,3]. Moreover, some epidemiological studies have shown an inverse relationship between higher levels of adiponectin and the occurrence of diabetes or cardiovascular events [4,5].

Adiponectin may also play a role in the development of diabetes mellitus, and its co-morbidities, because of its involvement in the regulation of carbohydrate and fat metabolism, as demonstrated in several animal and in vitro studies [6]. While these findings have generally been confirmed in adult populations, the relationship of adiponectin levels to metabolic risk factors has been less studied in adolescents. Also, most results were obtained in samples with a high representation of obese or diabetic children, or from unadjusted models. In addition, most associations came from cross-sectional studies, with only a few assessing the relationships between baseline adiponectin levels and changes in metabolic risk factors such as insulin resistance (IR) and lipid profile, and the majority involved in adult populations [4,7,8]. Only two studies, using very small samples, have reported longitudinal associations in young people [9,10].

The objectives of the present study were to examine the cross-sectional associations and two-year changes between plasma adiponectin and adiposity, lipid parameters, surrogate measures of IR and inflammation in healthy adolescents.

2.2. Laboratory methods

Baseline venous blood samples were obtained in the morning after a 10-h fast at least. All blood samples were processed within three hours, and the serum aliquoted for immediate analysis or long-term storage at −80°C until assay. Levels of circulating adiponectin and soluble tumour necrosis factor (TNF)-α receptor 1 (sTNF-α R1) were measured by ELISA (RD Systems, Minneapolis, MN, USA), with intra- and interassay CV values of 2.9 and 6.5%, respectively, for adiponectin, and 4.4 and 6.1%, respectively, for sTNF-α R1. High-sensitivity
C-reactive protein (hs-CRP) was measured by immunoassay using the Synchron-LX system (Beckman Coulter, Galway, Ireland), with intra- and interassay CV values of 5 and 7.5%, respectively. Plasma insulin was determined by direct chemiluminescence immunoassay performed on an Advia Centaur apparatus (Bayer Diagnostics, Tarrytown, NY, USA) with intra- and interassay CV values of 4.5 and 6.5%, respectively. The homeostasis model assessment (HOMA), calculated as fasting insulin (µU/mL) * fasting glucose (mmol/L)/22.5, was used to estimate IR. Plasma high-density lipoprotein (HDL) cholesterol was analyzed using a cholesterol oxidase method (intra- and interassay CV values of 1.8 and 4.5%, respectively) in the supernatant fraction after precipitation of non-HDL lipoproteins using magnesium-dextran-precipitating reagent. Plasma triglyceride was determined using a standard glycerol-blanked enzymatic triglyceride method (intra- and interassay CV values of 1.4 and 4.5%, respectively).

2.3. Follow-up data

After two years of follow-up, biomedical data were again collected, using the same standardized methods as for study entry. The students’ participation in structured PA outside of school was also reassessed.

2.4. Statistical analyses

Participants’ baseline characteristics are presented as percentages or means (SEM) for continuous variables. Variables with a skewed distribution were natural log-transformed before analysis. Associations between adiponectin levels and adiposity parameters, lipid profile, IR, and inflammatory markers were assessed using generalized linear mixed-effects models. Interaction parameters were added to the model to test for a modifying effect of gender or adiposity on adiponectin levels. Not surprisingly, the metabolic profile was worse for overweight adolescents compared with normal-weight subjects after adjusting for gender.

3. Results

3.1. Baseline characteristics

Cross-sectional analyses were made of the 647 adolescents for whom we had an adiponectin level (329 girls and 318 boys). They were, on average, younger than non-participants in the ICAPS study and those without adiponectin measurements (11.6 ± 0.6 versus 11.8 ± 0.7 years, respectively; P < 10^{-4}). However, after adjusting for age and gender, the groups did not differ in terms of adiposity parameters such as BMI, BF or waist circumference (data not shown).

Clinical characteristics of the study subjects are presented in Table 1 by degree of adiposity. Of these 12-year-old adolescents, 22.8% were overweight (the same for girls and boys). As expected, leptin levels were higher in overweight subjects (P < 10^{-4}) whereas adiponectin levels were lower (P < 10^{-3}). Mean levels of adiponectin tended to be lower in boys than in girls (13.51 ± 5.77 versus 14.31 ± 5.79 mg/L, respectively; P = 0.08). There was no interaction between effects of gender and adiposity on adiponectin levels. Not surprisingly, the metabolic profile was worse for overweight adolescents compared with normal-weight adolescents. The former had lower HDL cholesterol, higher triglycerides, lower insulin sensitivity and more low-grade inflammation, as assessed by their elevated levels of hs-CRP and sTNF-α R1.

3.2. Plasma adiponectin and adiposity parameters

After adjusting for age, gender, sexual maturity and participation in structured PA, adiponectin levels correlated negatively

| Table 1 Baseline characteristics of study subjects according to weight |
|------------------------|------------------------|------------------------|
|                        | Normal weight           | Overweight             | Total (n = 647)       |
| Age (years)            | (n = 500)               | (n = 147)              |                       |
| Boys (%)               | 48.8                   | 50.3                   | 49.2                  |
| BMI (kg/m²)            | 17.26 (0.10)           | 23.99 (0.18)^c         | 18.79 (0.14)          |
| BMI Z-score            | 0.11 (0.04)            | 2.76 (0.08)^a          | 0.71 (0.06)           |
| BF (%)                 | 15.06 (0.27)           | 29.01 (0.49)^a         | 18.27 (0.34)          |
| Waist circumference (cm) | 60.92 (0.26)         | 75.95 (0.49)^a         | 64.32 (0.34)          |
| Adiponectin (mg/L)     | 14.33 (0.27)           | 12.54 (0.47)^b         | 13.91 (0.23)          |
| Leptin (ng/mL)         | 9.98 (0.46)            | 32.86 (0.85)^a         | 15.25 (0.57)          |
| Fasting insulin (µU/mL) | 6.93 (0.19)            | 12.53 (0.35)^a         | 8.27 (0.20)           |
| HOMA²                  | 1.57 (0.04)            | 2.90 (0.08)^a          | 1.89 (0.05)           |
| HDL cholesterol (g/L)  | 0.50 (0.01)            | 0.42 (0.01)^a          | 0.48 (0.01)           |
| Triglycerides (g/L)    | 0.68 (0.01)            | 0.91 (0.02)^a          | 0.73 (0.01)           |
| sTNF-α R1 (pg/mL)      | 949 (8)                | 1043 (15)^a            | 970 (7)               |
| hs-CRP (mg/L)          | 0.84 (0.10)            | 1.82 (0.19)^b          | 1.07 (0.09)           |

Values are percentages for categorical variables and means (SEM) for continuous variables. BMI, body mass index; BF, body fat; HOMA, homeostasis model assessment; sTNF-α R1, soluble tumour necrosis factor-α receptor 1; hs-CRP, high-sensitivity C-reactive protein.

a P < 10^{-4} (versus normal-weight subjects after adjusting for gender).
b P < 10^{-3}.
c Log-transformed variables to estimate statistical significance.
Table 2

<table>
<thead>
<tr>
<th></th>
<th>β</th>
<th>SE</th>
<th>P</th>
<th>R² (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Multivariate models</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI Z-score</td>
<td>-0.0221</td>
<td>0.0098</td>
<td>&lt;0.03</td>
<td>1.62</td>
</tr>
<tr>
<td>BF (%)</td>
<td>-0.1005</td>
<td>0.0560</td>
<td>0.08</td>
<td>10.66</td>
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<tr>
<td>Waist circumference (cm)</td>
<td>-0.1747</td>
<td>0.0561</td>
<td>&lt;10^-2</td>
<td>7.81</td>
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<tr>
<td>Multivariate models</td>
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<tr>
<td>BMI Z-score</td>
<td>-0.0064</td>
<td>0.0046</td>
<td>0.17</td>
<td>77.95</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>-0.0899</td>
<td>0.0313</td>
<td>&lt;10^-2</td>
<td>71.41</td>
</tr>
</tbody>
</table>

General linear mixed-effects models.

- Adjusted for age, gender, sexual maturity and participation in structured physical activity.
- Further adjusted for BF.

and significantly with the BMI Z-score ($P < 0.03$) and waist circumference ($P < 10^{-2}$), and showed borderline significance with BF ($P = 0.08$) (Table 2). In a multivariate model taking BF into account to explain 71.4% of the waist-size variability, a 5.80-mg/L increase in adiponectin (corresponding to one standard deviation) correlated with a 0.5 cm decrease of waist circumference. Interaction with gender was not significant in any of the models.

To investigate the contribution of central adiposity to differences in adiponectin concentration, overweight adolescents were separated into two groups according to waist circumference (< or ≥ 75 cm, the median value in our adolescent sample) and compared with normal-weight students (Fig. 1). After adjusting for the BMI Z-score, only the comparison between normal-weight and overweight subjects with a large waist circumference was significant ($P < 10^{-2}$).

### 3.3. Plasma adiponectin and lipid profile

Adiponectin was positively associated with HDL cholesterol, a relationship that persisted even after adjusting for BMI Z-score and BF (data not shown), waist size and triglyceride levels (Table 3). It remained significant after further adjustment for fasting insulin levels ($P < 0.02$; data not shown). In contrast, adiponectin correlated negatively and significantly with triglyceride levels. There was no interaction between effects of gender and adiponectin on lipid levels whatever the model tested.

### 3.4. Plasma adiponectin and IR

Associations between adiponectin and IR are presented in Table 3. Adiponectin negatively and significantly correlated with the HOMA index and insulin, even after adjusting for measures of obesity such as the BMI Z-score and waist circumference ($P < 0.03$ and $P < 0.04$, respectively), and BF ($P < 10^{-2}$; data not shown). Further adjustment for leptin only slightly modified this relationship ($P < 0.02$), whereas adiposity had a modifying effect on the relationship between adiponectin and IR. While adiponectin was unrelated to HOMA and insulin in normal-weight subjects ($P = 0.20$ and $P = 0.25$, respectively), a significant inverse relationship was found in overweight adolescents ($P < 0.02$ for both parameters).

### 3.5. Plasma adiponectin and low-grade inflammation

Adiponectin levels were inversely associated with sTNF-α R1 after adjusting for age, gender, sexual maturity and participation in structured PA ($β = -3.56$; $P < 10^{-2}$), and further adjustments for BMI Z-score and waist size ($β = -2.36$; $P < 0.05$) or BF ($β = -3.05$; $P < 0.02$) (data not shown). Gender had no modifying effect on these relationships. Adiponectin was not associated with hs-CRP in neither univariate nor multivariate models.

### 3.6. Plasma adiponectin and changes in metabolic factors

A baseline adiponectin level was obtained in 339 control students. Of these, 51 were lost to follow-up in 2004 mostly because of changes in schools. These students were, on average, older ($P < 0.03$) than those re-surveyed in 2004 ($n = 288$). After adjustment for age and gender, surveyed and non-surveyed adolescents did not differ in terms of the major variables considered (BMI, BF, waist circumference, adiponectin, triglycerides and insulin levels in 2002; results not shown).

Table 4 shows the results of the generalized linear mixed-effects models with Δ triglycerides as the dependent variable. This longitudinal analysis was performed after adjusting for baseline triglycerides, and the two-year BF and insulin changes. Adolescents with higher baseline adiponectin had more favourable changes in triglycerides between 2002 and 2004.
### Table 3

<table>
<thead>
<tr>
<th>Variables in the model</th>
<th>( \beta )</th>
<th>SE</th>
<th>( R^2 )</th>
<th>( P )</th>
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</thead>
<tbody>
<tr>
<td>Adiponectin (mg/L)</td>
<td>-0.0692</td>
<td>0.0350</td>
<td>&lt; 0.05</td>
<td></td>
</tr>
<tr>
<td>Triglycerides (g/L)</td>
<td>-0.3065</td>
<td>0.0438</td>
<td>&lt; 10^{-4}</td>
<td></td>
</tr>
<tr>
<td>( \Delta ) BF</td>
<td>0.0114</td>
<td>0.0041</td>
<td>&lt; 10^{-2}</td>
<td></td>
</tr>
<tr>
<td>( \Delta ) Insulin</td>
<td>0.0114</td>
<td>0.0029</td>
<td>&lt; 10^{-4}</td>
<td></td>
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</tbody>
</table>

General linear mixed-effects model with \( \Delta \) triglycerides as the dependent variable (\( \Delta \) triglycerides = triglyceride level in 2004 minus triglyceride level in 2002).

\( a \) Adjusted for age, gender, sexual maturity and participation in structured physical activity in 2002 and 2004.

\( b \) Adiponectin and triglyceride were natural log-transformed.

\( c \) \( \Delta \) BF: two-year body fat change.

\( d \) \( \Delta \) Insulin: two-year insulin change.

### Table 4

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\( a \) Adjusted for age, gender, sexual maturity and participation in structured physical activity in 2002 and 2004.

\( b \) Adiponectin and triglyceride were natural log-transformed.

\( c \) \( \Delta \) BF: two-year body fat change.

\( d \) \( \Delta \) Insulin: two-year insulin change.

\( P < 0.05 \) as demonstrated by the negative \( \beta \) coefficient. Baseline adiponectin was not significantly associated with subsequent changes in HDL cholesterol, IR or BMI (data not shown).

### 4. Discussion

In this sample of 12-year-old adolescents, adiponectin correlated negatively with adiposity, IR and low-grade inflammation, and was favourably associated with the lipid profile. Indeed, this study demonstrates for the first time that higher baseline adiponectin levels are associated with more favourable two-year changes in triglycerides, independent of BMI, insulin and changes in HDL cholesterol.

The study results – showing unfavourable associations between low levels of adiponectin and various factors known to be involved in the mechanisms of atherosclerosis – are supportive of anti-inflammatory and anti-atherogenic effects of adiponectin being already present in young subjects. These findings are in line with two recent papers, which reported that low adiponectin levels are associated with early atherosclerosis, as assessed by an increased carotid intima-media thickness in obese children [15,16].

We found adiponectin to be more closely related to waist circumference, a surrogate measure of central adiposity, than with total adiposity as assessed by BMI. Although not reported in all studies [17], these results confirm previous observations in children and in adult populations. Huang et al. [18] demonstrated a stronger correlation between adiponectin and waist circumference than between adiponectin and BMI in adolescents. In a Canadian study [19] assessing fat accumulation with computed tomography (CT), only visceral fat was inversely and independently associated with adiponectin levels in a multivariate model, including various obesity indices. In Japanese children, adiponectin correlated negatively with waist circumference and visceral adipose tissue as measured by CT [20]. These findings all highlight the role of central fat in predicting plasma adiponectin levels.

We found associations between adiponectin and markers of IR that, unlike data reported by Vikram et al. [21], remained significant after adjusting for BMI and BF. Other studies in young people failed to show any independent association between adiponectin and measures of IR [10,22]. However, our results...
are consistent with previous reports in children [18,23] and in adults [24,25], and are in agreement with animal studies reporting that adiponectin stimulates glucose utilization and fatty-acid oxidation by activating 5′-AMP-activated protein kinase, which leads to improvement in insulin sensitivity [26]. Discrepancies across studies may be due to characteristics of the sampled populations in terms of age, adiposity or pubertal stage, to variables accounted for or to the method of adiponectin measurement. Above all, we observed, as did Kantartzis et al. [27], that the connection between adiponectin and IR depends on the degree of adiposity, the association being present only in overweight adolescents.

Few studies have been published on the link between adiponectin and low-grade inflammation in young people [21,28,29]. Although inverse relationships have been reported between adiponectin and CRP levels in Japanese women, after adjusting for BMI and percentage fat mass [30], and in male Japanese patients [31], in two studies involving children – as well as our present study – there was no association found between adiponectin and CRP [21,28]. In fact, CRP levels are very low in children, and this may not allow such associations to be observed. In contrast, we found that higher adiponectin levels are strongly associated with lower sTNF-α R1 levels, which may be more appropriate markers of low-grade inflammation than TNF-α itself. Other work confirms this relationship – for instance, in human immunodeficiency virus-infected patients with a lipodystrophy syndrome [32] or in healthy subjects [33]. These results are in agreement with recent in vitro and in vivo studies showing that TNF-α and adiponectin inhibit each other [34], and lend further support to the anti-inflammatory actions of adiponectin, which may already be, in effect, in adolescents.

We found significant links between high adiponectin levels and high HDL cholesterol or low triglycerides, independent of BF, an association not entirely explained by IR. This result, in line with previously reported data, suggests an independent effect of adiponectin on lipid metabolism. Although the underlying mechanisms are, as yet unknown, adiponectin has been shown to be an important determinant of ApoA-I catabolism independent of insulin sensitivity [35]. Also, an inverse association between adiponectin and hepatic lipase activity has been reported [36]. In the longitudinal analysis of the present study, we demonstrated for the first time that adiponectin predicts a favourable change in triglyceride levels independent of BF and insulin changes. There are only two previous longitudinal studies of the effects of adiponectin on metabolic factors in children. In a study of Pima Indian children, baseline adiponectin levels were not associated with either fat-mass or insulin evolution [10]. In 88 African-American children and adolescents, baseline adiponectin did not predict changes in insulin sensitivity over two years [9]. In adults, previous studies failed to reveal any association between baseline adiponectin and subsequent lipid or weight changes [7,37]. In contrast, although not seen in our student sample, lower baseline adiponectin concentrations were independently associated with future IR or diabetes in nested case-control [4,38] or longitudinal studies [7,8] in adults. The lower variability of IR in adolescents compared with adults may explain some of the discrepancy.

The limitations of the present study need to be mentioned. Although covering a greater number of participants than did previously reported longitudinal data in children, our longitudinal analysis may have suffered from a lack of power to detect significant associations. Also, we did not differentiate among the various molecular forms of circulating adiponectin. Recent studies in adults [39] and children [40], have pointed out that favourable associations between adiponectin and metabolic factors depend especially on the proportion of high-molecular-weight adiponectin.

The strengths of the present study are its longitudinal design and the adjustments for several potential confounders such as sexual maturity and adiposity.

In conclusion, our findings suggest a favourable relationship between adiponectin and the metabolic profile as well as a subsequent lowering of triglyceride levels in young adolescents. The mechanisms underlying these associations, however, remain to be clarified.

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References


