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Circulating leptin and adiponectin concentrations in healthy exceptional longevity

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ABSTRACT

People reaching exceptional longevity free of major age-related diseases represent the paradigm of successful aging. Adipose tissue function declines as we age, potentially resulting in changes of circulating adipokines (e.g., leptin and adiponectin). Here, we measured circulating levels of leptin and adiponectin in healthy centenarians (n=81; 100–104 years) and younger elderly controls (n=46; 70–80 years). Centenarians had significantly higher serum levels of leptin compared with controls (p<0.001), whereas no significant differences were observed for adiponectin. Further research including also other blood variables will be needed to elucidate whether high leptin levels could serve as a hallmark of healthy exceptional longevity.

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renal or hepatic failure, and major psychiatric disorders). The study complied with the tenets of the Declaration of Helsinki and was approved by the local ethics committee. Written informed consent was obtained from all participants.

Serum leptin (DRG GmbH, Marburg, Germany) and adiponectin (Biovendor, Brno, Czech Republic) levels were measured using commercially available enzyme-linked immunosorbent assays according to the manufacturers’ instructions. For all assays, the intra- and inter-assay coefficients of variation were <6% and <9%, respectively. Each sample was analyzed in duplicate, and the mean value of the two measures was used for analyses. Laboratory personnel were blinded with regard to subjects’ group (centenarians or controls).

Because data were skewed, the Mann-Whitney U test was applied to compare serum adipokine levels between groups. Receiver operating characteristic (ROC) curves were used to determine the optimal cut-off point for the association with the likelihood of being a centenarian. The Youden index was used to define the optimal cut-off point and the analysis was completed by calculating areas under the curve (AUC) and 95% confidence interval; AUC values ≥0.90, 0.80–0.90, 0.70–0.79 and <0.70 were considered as ‘excellent’, ‘good’, ‘fair’, and ‘poor’ predictor, respectively (Trost et al., 2012). A leave-one-out cross validation was performed for assessing if the optimal cut-off point could be generalized to an independent data set.

Centenarians had higher leptin serum levels compared with elderly controls (p < 0.001, Table 1; see also dot plot in Fig. 1A), although no significant differences for adiponectin were noted (p = 0.185, Table 1; Fig. 1B). The ROC curve results are shown in Table 2 and Fig. 2. The AUC for being a centenarian according to serum leptin levels was 0.68 (sensitivity = 80%; specificity = 70%).

Controversy still exists on the relationship between leptin and longevity (Arai et al., 2008; Gulcelik et al., 2013; Kuo and Halpern, 2011). To our knowledge, no research has assessed this issue in healthy centenarians as we did in our study. This methodological difference could explain the discrepant findings. Adipose tissue dysfunction (as reflected by a dysregulation in leptin and adiponectin) is associated with poor prognosis in centenarians (Arai et al., 2011). The hypoxic state that characterizes aging leads to a SIRT1 reduction in different tissues (Poulose and Raju, 2015) including the hypothalamus, where it induces leptin resistance and thereby increased adiposity (Sasaki, 2015). Arai et al. analyzed the association between circulating adipokines (leptin and adiponectin, among others) and all-cause mortality in 252 lean centenarians who were not disease-free (Arai et al., 2008). In contrast with our results, these centenarians exhibited low plasma levels of leptin, with the lowest tertile of leptin being associated with increased all-cause mortality. These authors concluded that the cumulative dysregulation of adipokines could represent a marker of poor prognosis in centenarians (Arai et al., 2011). These results were supported by Meazza and co-workers, who found lower blood leptin levels in centenarians compared with healthy elderly subjects (Meazza et al., 2011). Paolillo et al. found higher and lower blood leptin concentration in centenarians compared with adults (≥50 years) or old people (75–99 years) (Paolillo et al., 1997).

Interestingly in centenarians leptin was negatively correlated with the ratio of plasma insulin-like–growth factor I (IGF-1)/IGF-binding protein-3 (IGFBP-3 levels), with the latter being hypothesized to be positively linked with improved insulin action and plasma lipid concentration in centenarians (Paolillo et al., 1997). Recently, Mishra et al. studied the association of serum leptin with mortality in older adults (70–79 years). They found that moderately elevated leptin concentrations were correlated with lower risk of all-cause mortality and CVD-related mortality among older women, whereas no association was observed in men (Mishra et al., 2015).

In contrast, adiponectin levels seem to be unaltered in healthy centenarians according to hour data. However, the literature available is again controversial. Some authors have reported an association of high adiponectin levels with longevity (Bik et al., 2013; Gulcelik et al., 2013), whereas increased adiponectin could reflect a higher mortality risk in younger individuals or CVD patients (Bik and Baranowska, 2009). In contrast with our results, Meazza et al. found that centenarians had higher adiponectin levels compared with elderly controls (Meazza et al., 2011). This finding was in line with those reported by Bik et al., who studied the serum levels of all adipokine isoforms in centenarians and elderly controls and found that the concentrations of all isoforms of this adipokine were higher in the former (Bik et al., 2013). In addition, they showed that centenarians’ adipokine levels positively correlated with age and HDL-cholesterol whereas high-molecular weight (HMW)-adiponectin was negatively associated with insulin and triglycerides (Bik et al., 2013). As we have previously detailed elsewhere, none of the centenarians of our study had hypercholesterolemia (total cholesterol ≥ 200 mg/dL) or hypertriglyceridemia (Bik et al., 2013) and (triglycerides ≥ 150 mg/dL) (Emanuele et al., 2014). In this regard, however, one of the limitations of our investigation is that we did not report data on glucose homeostasis or insulin resistance, e.g., insulin resistance index using homeostatic model assessment (HOMA)-insulin resistance (IR). Dysregulation of adiponectin in the elderly can be caused by loss of function of this circulating adipokine or a response to an inflammatory alteration (Gulcelik et al., 2013).

In summary, our findings suggest that high leptin levels might be associated with healthy exceptional longevity. Further research is needed to corroborate these preliminary findings in larger, independent cohorts.

Fig. 1. Values for leptin (A) and adiponectin (B) by group.
**Fig. 2.** Results of receiver operating characteristic (ROC) curves for leptin serum levels associated with successful aging.

**Table 1**
Serum levels of biomarkers (mean ± SD; median and range) and age by group. Biomarkers data did not follow a normal distribution. *Statistical post-hoc power analysis to detect differences between group means with a significance level (α) of 0.05 (2-tailed). ** Estimated optimal sample size (enrollment ratio 1:1) to obtain a statistical power ≥90% for detecting a difference between group means with a significance level (α) of 0.05 (2-tailed).

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>Centenarians n=81</th>
<th>Controls n=46</th>
<th>p value</th>
<th>Statistical power*</th>
<th>Optimal total sample size (n)**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leptin (ng/mL)</td>
<td>867.8 ± 264.0 909.2 (67.7, 1703.5)</td>
<td>760.8 ± 468.8 593.3 (61.2, 1899.2)</td>
<td>0.001</td>
<td>30%</td>
<td>256</td>
</tr>
<tr>
<td>Adiponectin (μg/mL)</td>
<td>18.5 ± 8.1 17.32 (7.2, 38.4)</td>
<td>20.3 ± 7.9 18.87 (6.81, 36.3)</td>
<td>0.185</td>
<td>23%</td>
<td>852</td>
</tr>
</tbody>
</table>

**Table 2**
Receiver operating characteristic (ROC) results for leptin as independent variables associated with successful aging. Abbreviations: CI: confidence interval; AUC, area under the curve.

<table>
<thead>
<tr>
<th>Optimal cut-off for leptin levels</th>
<th>AUC</th>
<th>95% CI</th>
<th>p value</th>
<th>Standard error</th>
<th>Sensitivity (%)</th>
<th>Specificity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;737.9 μg/L</td>
<td>0.676</td>
<td>0.561–0.791</td>
<td>0.001</td>
<td>0.059</td>
<td>80%</td>
<td>70%</td>
</tr>
</tbody>
</table>

**Conflict of interests**
The funding organization(s) played no role in the study design; in the collection, analysis, and interpretation of data; in the writing of the report; or in the decision to submit the report for publication.

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**Author contributions**
All the authors have accepted responsibility for the entire content of this submitted manuscript and approved submission.

**References**

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