CROSSTALK BETWEEN GENETICS, GENE EXPRESSION AND BIOCHEMICAL MARKERS SUPPORTS SYSTEMIC IRON HOMEOSTASIS DYSREGULATION IN ALZHEIMER DISEASE

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INTRODUCTION

The distinction between normal aging and Alzheimer’s disease (AD) is a first and relevant step to combat this disease efficiently. Because of the clinical interest in predicting patient evolution and prognosis, the identification of biomarkers and the unravelling of genetic factors underlying AD are of crucial importance.

Several lines of evidence implicate an imbalance of the redox-active biomacromolecules, copper and iron in AD. Metal-catalyzed hydroxyl radicals are potent mediators of cellular injury and are central to the oxidative injury hypothesis of AD pathogenesis [1,2].

In this study, we seek to further investigate this hypothesis through the identification of serum biomarkers/endoenzymes related to Fe/Cu metabolism and candidate genes involved in Fe/Cu homeostasis.

In fact, an important genetic component has been recognized for AD [3]. Several genetic variants involved in Fe/Cu metabolism were evaluated for their contribution to AD susceptibility and to the assessed biological marker distributions.

This integrative approach is planned to deal with heterogeneity in this complex disorder, and will power phenotypic data expressed in AD to identify susceptibility loci, and further elucidate the contribution of Fe/Cu metabolism disruption to the etiopathogenesis of AD.

METHODS

SUBJECTS AND SAMPLE COLLECTION

A total of 116 AD patients (76 male, 40 female) and 92 healthy volunteers (68.2 7.7, 38 male and 54.1 women) were recruited at Hospital de Santa Maria, Hospital Fomento Fonseca and Hospital Magalhães Lemos. Blood samples were collected by venipuncture under no fasting conditions in serum gel and EDTA tubes.

The study was submitted and approved by the local ethics committee and each donor or legal representative signed an informed consent before blood collection.

BIOCHEMICAL MEASUREMENTS

Serum iron (Fe), transferrin (Tf), ferritin (Fi) concentration and transferrin saturation (Tf Sat) were measured. PASSW Statistics 11.0 (SPSS Inc.) software was used for MANCOVA and logistic regression analysis of all biochemical data.

GENETIC ANALYSIS

SNPs were evaluated by high-throughput genotyping in APOE (apolipoprotein E) and Fe/Cu metabolism-related genes: CYB5R1 (cytochrome b reductase 1), HAMP (hepcidin), HFE (hemochromatosis gene) HFE2 (iron responsive element binding protein 1), SLCO1A2 (divalent metal transporter 1), SLCT4A1 (ferroportin), TF (transferrin), TRZ2 (transferrin receptor 2).

SNPassc package for R 2.10.10 (1999-2006 R Development Core Team) was used for logistic regression analysis of the genotyping data for association with AD (adjusted for age and gender).

RESULTS

To determine whether Fe metabolism-related gene variants were associated with susceptibility to AD in a Portuguese group, allelic, and genotypic frequencies were compared between patients with AD and healthy control subjects.

The results show that specific iron metabolism gene variants are associated with AD susceptibility.

IRON METABOLISM BIOMARKERS IN ALZHEIMER’S DISEASE

<table>
<thead>
<tr>
<th>Parameters</th>
<th>AD cases n=116</th>
<th>Controls n=92</th>
<th>Mean±SD</th>
<th>Normal range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iron (µg/dL)</td>
<td>76.6±26.36</td>
<td>84.6±25.18</td>
<td>37.0-150.0</td>
<td></td>
</tr>
<tr>
<td>Transferin (mg/dL)</td>
<td>250.9±43.23</td>
<td>267.8±44.28</td>
<td>200.0-400.0</td>
<td></td>
</tr>
<tr>
<td>Ferritin (ng/µL)</td>
<td>123.4±17</td>
<td>23.5±7.91</td>
<td>25.0-50.0</td>
<td></td>
</tr>
</tbody>
</table>

SPECIFIC IRON METABOLISM GENETIC VARIANTS IN AD

<table>
<thead>
<tr>
<th>Gene</th>
<th>SNP Reference Position (Mb)</th>
<th>Assoc Allele</th>
<th>Chi Square</th>
<th>p value</th>
<th>OR [95%]</th>
</tr>
</thead>
<tbody>
<tr>
<td>APOE</td>
<td>rs42958</td>
<td>G/T</td>
<td>11.37</td>
<td>7.00E-4</td>
<td>2.64 [1.48-4.71]</td>
</tr>
<tr>
<td>HFE</td>
<td>rs12979862</td>
<td>G/A</td>
<td>4.97</td>
<td>0.0258</td>
<td>1.66 [1.06-2.59]</td>
</tr>
<tr>
<td>TF</td>
<td>rs3785804</td>
<td>G/A</td>
<td>7.716</td>
<td>0.0055</td>
<td>1.79 [1.19-2.71]</td>
</tr>
<tr>
<td>SLC40A1</td>
<td>rs1439386</td>
<td>G/A</td>
<td>4.355</td>
<td>0.0415</td>
<td>1.55 [1.03-2.30]</td>
</tr>
</tbody>
</table>

We thank the Unidade de Laboratório Integrado (ULI) Departamento de Promoção da Saúde e Doenças Crônicas, Instituto Nacional de Saúde Dr. Ricardo Jorge (INSALUD) for the measurement of biochemical parameters.

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CONCLUSIONS

We hypothesize that the low systemic Fe status profile observed in AD patients could be due to impaired regulation of cellular Fe efflux.

The intracellular accumulation of Fe, particularly in the brain would lead to a rise in oxidative damage, contributing to the AD pathophysiology.

REFERENCES


Table 1: Mean, standard deviation and normal range of each biochemical parameter measured in serum from AD cases and controls, after removal of outliers.

Table 2: SNPs associated with high risk of AD.