How closely is the gene expression in blood of Alzheimer’s disease patients associated with the known biology of the disease?

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Summary

A new blood test, ADtect®, has been developed that can aid the early detection of Alzheimer’s Disease (AD). The test is based on measuring the expression of selected genes in blood and is defined as the AD-specific gene signature. ADtect® comprises a low density array of 96 selected gene assays using RNA extracted from a blood sample. The performance of each of the 96 gene assays is calculated with an algorithm resulting in a positive or negative test score value indicating the presence or absence of AD. In a multicenter study of 412 subjects the test is able to discriminate AD subjects from cognitively healthy controls with a 72% overall agreement with the clinical diagnosis, an important gold standard. The test performance is confirmed in two independent validation studies and shows a similar and consistent good performance.

It was found that 32% (39/120) of the 84 genes in ADtect® encode proteins with a biological function associated with AD, brain or neurological function. Four of them are also listed in the current AlzGene database. The identity of the 12 remaining genes in the test could not be found in available data bases.

Introduction

Early and accurate detection of AD is critical for implementing active management strategies which may delay the onset of the more debilitating symptoms of the disease. A convenient blood test to define AD specific management strategy of those subjects with memory complaints presenting at clinics. Several independent studies have indicated that a peripheral blood based test could be used for diagnostic profiling in neurological diseases(14). The potential use of a blood-based expression profiling in diagnosis of brain pathology has been described15 and further studies also demonstrate a significant degree of co-variability in gene expression between brain tissue and peripheral blood cells16,17. Lew et al.17 indicate that about 80% of the genes expressed in brain samples are also expressed in blood cells and that at least some of the differential regulation of gene expression are regulated in a similar way in the two tissues. This opens an alternative approach to find useful biomarkers for the early detection of AD using blood as an alternative sampling tissue. Gene expression is able to detect subtle changes and have the potential to detect even minor alterations in biology associated with a disease like AD. Gene expression studies for detection of AD have been described by others15,16 but these studies were performed on relatively small sample sizes and no models for AD prediction were developed.

With the new blood test, ADtect®, intended to aid the detection of AD it is now for the first time possible to examine if there are similarities in the set of genes known to be associated with AD in brain tissue and in the genes found to be informative for AD in blood.

Results

AD is associated with profound biochemical and pathologic alterations in the brain, including aberrant amyloid precursor protein (APP), amyloid β (Aβ) protein metabolism, tau protein phosphorylation, oxidative stress, inflammation, cellular Ca2+-homeostasis, and lipid dysregulation. In AD, the main cause of dementia is assumed to result from the progressive loss of synaptic function and neurologic degeneration. Here we show that among the genes included in the ADtect® test we find genes associated with essentially all biochemical alterations that has been related with AD.

AlzGene

Four of the genes (Box 1) are included in the most recent AlzGene list with TNF ranked as 3#11. Another gene in ADtect® but not listed in the AlzGene table, CALM3, maps closely to ApoE and it has been discussed if also this gene is linked to an increased risk of developing AD.

Amyloid-β

The four genes listed (Box 1) have all been associated with processing and metabolism of APP and Aβ. It has been shown that APP and Presenilin1 interact with the adaptor GRB2 and modulate ERK 1,2 signaling16 and that there is a correlation between decreased PEBP1 expression and accumulation of Aβ15. Almost all $100(Aβ) immune-reactivity in brain is concentrated to astrocytes surrounding the Aβ plaques18. Also, SELM has been shown to co-localize with Aβ plaques as well as to tau containing tangles19.

Tau/Microtubules

There are seven genes listed with an association with Tau and/or microtubules. DNDH1, KIF13B, MAP1S and PLEC1 are all proteins associated with microtubules.

Mitochondria

All four of the genes that are associated with mitochondrial function are nuclear encoded. While COX8B is involved in ATP production the remaining HDAC1, MAP1S, and PLEC1 are all essential proteins for transportation and aggregation of mitochondria in neurons.

Calcium Regulation

As a primary calcium signal transducer, calmodulin; CALM3, responds to cytosolic calcium fluxes by binding to and regulating the activity of target proteins. S100A6, a Ca2+-binding protein, is believed to stabilize intracellular (Ca2+) homeostasis23. Also SYT13 binds to calcium and appear to function as a calcium sensor in the regulation of neurotransmitter release.

Oxidative stress

There are three genes listed in this table (Box 1) but others, like ANKR58, CALM3 and S100A6, could equally well have been listed here as calcium regulation is believed to be essential to mitigate oxidative stress.

Infammation

Inflammation is a process related with the onset of several neurodegenerative disorders, including AD, where interleukins like TNF seem to display a neuroprotective activity. DELENDD2 is essential in TNF-signaling24.

Assayed with AD, Other Neurological Diseases, Brain and Neurone Function:

As shown in these tables (Box 1) several genes are listed and they cannot all be presented here. It is worth mentioning TARBP that is the major pathological protein of FTLD with ubiquitin-immunoreactive inclusions (FTLDU) with or without argyrophilic grain-like (ALS) and sporadic ALS but at the same time it is also associated with AD27. The presence of the three proteins, UBE3A, UBE4B, and UBL3 indicate that ubiquitin-dependent protein degradation might be affected.

Discussion

AD is multifactorial and heterogeneous in both its clinical and histopathological appearance. The clinical heterogeneity of the disease means that the diagnosis remains uncertain until post mortem, when a histopathological examination can be performed. The selection of the genes included in ADtect® has been based entirely to the informative value of their expression to predict the disease in blood. It is interesting to note that so many of the genes encode proteins involved in bioenergetics and cellular processes that are associated with both cell death and brain tissue. The results suggest that many of the features associated with AD is not only localized to brain tissue but also to blood and maybe other tissues as well. It would be of interest to explore if also some of the ADtect® genes where no links to AD have been found today actually have some connection to the biology of the disease. The exploration of these genes may find additional insight into the relative pathoscopical basis of AD.

References

1. Bougreen, M. & Stenberg, A. J., Pharmacological Pharma. 7. 351-360 (2002);
2. Bougreen, M. & Stenberg, A. J., Pharmacological Pharma. 13. 1711-1717 (2005);
4. Lundeberg, M., et al. European Neuroimmunology Research. 29. 30 (2003);
6. Tung, S., et al. Journal of Neuroimmunology 243. 8-12 (2011);

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Box 1

Box 1. List of all genes associated with AD, and brain or neuron function that are included in the ADtect® test. The different lines indicate to which tables the genes are listed.

ADtect® Gene List

<table>
<thead>
<tr>
<th>Gene Name</th>
<th>Gene List</th>
<th>Associated with AD, Other Neurological Diseases, Brain and Neurone Function</th>
</tr>
</thead>
<tbody>
<tr>
<td>TARDBP</td>
<td>PDE4A</td>
<td>Mitochondria, Calcium Regulation, Oxidative stress, Inflammation, Tau/Microtubules, Mitochondria</td>
</tr>
<tr>
<td>GRB2</td>
<td>SYT13</td>
<td>Mitochondria, Calcium Regulation, Oxidative stress, Inflammation, Tau/Microtubules, Mitochondria</td>
</tr>
<tr>
<td>KIF13B</td>
<td>MAP1S</td>
<td>Mitochondria, Calcium Regulation, Oxidative stress, Inflammation, Tau/Microtubules, Mitochondria</td>
</tr>
<tr>
<td>PLEC1</td>
<td>S100A6</td>
<td>Mitochondria, Calcium Regulation, Oxidative stress, Inflammation, Tau/Microtubules, Mitochondria</td>
</tr>
</tbody>
</table>

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