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Genetic risk factors of Alzheimer's disease

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ABSTRACT

Introduction. Alzheimer's disease (AD) is one of the most common neurodegenerative diseases, which is a serious health problem for societies that live longer. Spontaneous dominant mutations and polymorphisms of selected genes play an important role in development of AD.

Aim. Several polymorphisms in selected genes strongly associated with development of Alzheimer's disease were highlighted in this review: *APOE, CYP46, APP, PSEN1, PSEN2, UBQLN1, BACE1, PRND, APBB2, TOMM 40*. These gene polymorphisms have a significant role in the development of Alzheimer's disease and they have potential to be biomarkers. Researchers combine efforts to find significant polymorphisms that would ensure that a person is predisposed to the occurrence of disease symptoms. This topic is often taken up by scientists seeking to develop effective genetic tests for diagnosing AD.

Material and methods. Analysis of literature from web of knowledge: Web of Science (all database), NCBI and PubMed.

Results. We reviewed the selected important genes and polymorphisms which are most often associated with development of AD.

Conclusion. It should be noted that nowadays scientists strive not to focus on only one polymorphism in the gene but on several polymorphisms in different genes concomitantly and above all on interactions between them to the diagnosis of this disease. Only this approach to AD will contribute to the creation of appropriate identification methods. Moreover, we should use the new generation tools - the platform for collecting data and personalized medicine.

Keywords. autosomal genetic mutations, early-onset Alzheimer Disease, genetic polymorphisms, late-onset Alzheimer Disease

Introduction

Alzheimer's disease general characteristics

Alzheimer's disease is the most common form of dementia. It affects 24.3 million people around the world. To date, over twenty genetic loci have been associated with AD and a significant number of genetic variants were mapped within these loci. A large part of important genetic variants lies outside the coding region (in introns). However, the reliable function of these variants is still under unexplored.[1-3]

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Alzheimer's disease is one of many neurodegenerative diseases that appear in a population of societies that live longer. In spite of many years of intense search, no unequivocal genetic and biochemical markers have been found that would allow for the in vivo differentiation of AD among many other dementia syndromes. Due to the variety of symptoms and the course of AD, and along with the development of molecular techniques, a different approach to the disease began to be looked at: as a polygenic disease (polymorphisms), methylation of histone proteins of selected genes, and splicing methods.

In Verheijen's review, particular attention was paid to GWAS (genome-wide association studies), TWAS (transcriptome-wide association studies) and EWAS (epigenome-wide association studies).[4]

The review of scientific articles provides us with a wider spectrum of opinions on this health problem and sees them as an opportunity to faster diagnose and prevent the development of AD.

Several forms of Alzheimer's disease are distinguished due to family conditions and the age of the patient during the first symptoms of the disease. Classification based on the number of people affected by dementia in the patient's family. We can distinguish the family form Alzheimer disease (at least 2 people in the patient's family – FAD) and the sporadic form Alzheimer disease (without family conditions – SAD). It is estimated that in 15-40% of cases the disease may have family conditions. Only a few percent of all FAD cases show a clear autosomal dominant AD inheritance pattern. The sporadic form predominates in the patient population (60-85%) with a more complex type of inheritance and a multifactorial etiology.

Depending on whether the first symptoms occurred in the patient before 65 years of age or after 65 years of age there is an early form (early onset Alzheimer disease: EOAD; ≤65 years) and late form (late onset Alzheimer disease: LOAD; >65 years). EOAD is revealed in 2% of cases, characterized by impaired dexterity, impaired ability to interpret sensory impressions, memory disorders, associative problems, personality changes and degeneration of the frontal and parietal lobes.[5] In 98% of people with LOAD, memory disorders prevail, while the dexterity and the ability to interpret sensory impressions are less damaged.[6] In LOAD, clinical symptoms result from extensive cortical atrophy. The duration of the disease is usually 5-12 years. Death occurs as a result of non-cerebral complications (pneumonia, decubitus ulcers). Research focuses not only on the family form of the disease but also on spontaneous, dominant mutations, which were formerly ignored in scientific research. It is noted that they can lead to the first symptoms of the disease, even before the age of 30 named young onset Alzheimer disease (YOAD).[7,8]

There are several reasons for the occurrence of AD disease: genetic determinants, lifestyle, diet, interpersonal contacts. It was found that the more we lead an active lifestyle (we undertake to solve problems, we educate ourselves, we run a social life, we have a family, we develop our interests, for example: we solve crosswords, read books) the later we can observe the development of the disease. In Alzheimer's disease we distinguish various stages of disease: mild cognitive impairment (MCI), and: AD I, AD II, AD III.[9] Clinical diagnosis of MCI and different stages of AD is based on laboratory tests and medical history combined with the exclusion of symptoms typical of other dementias (vascular dementia (VaD), frontotemporal dementia (FTD), pseudo dementia).[10] Changes in Alzheimer's disease relate to cognitive function disorders, memory, disorders in the field orientation, coping with stress and problems with everyday life. One of the symptoms of the disease is isolation from the environment. Morphologically the pathological changes of brain are observed: in the brain size (losses in the amount of tissue 1-2% per year), changes in the hippocampus and sometimes in the thickness of the gray matter.[11]

Usually diagnostic research uses cerebrospinal fluid (CSF) to determine typical markers in AD. Morphologically in the diseased tissue are observed: amyloid plaques, intraneuronal deposits of spirally Tau protein fibers, granular-filamentous degeneration (hyperphosphorylated Tau protein) and hirano bodies (e.g. cytoplasmic, eosinophilic, rod-shaped structures found inside nerve cells witch are built from actin and proteins). Rarely are observed: neuron atrophy, synapse degeneration, brain atrophy (cerebral tissue rupture). Changes confirming the diagnosed form of Alzheimer's disease are often observed *post mortem* - after the patient's death.[2]

Genetic polymorphism

The genetic determinants play an important role in the development of AD disease. Over 20 genes responsible for the development of this disease have been detected. Knowledge of genetic predisposition may affect a person at risk of developing Alzheimer's disease to take appropriate steps in a lifestyle change and early treatment to delay the development of the disease. Genetic polymorphisms that cause changes leading to dementia concern codons of genes (exons) and non-coding sequences (introns). Modern research also focuses on mitochondrial inheritance. In genetics, polymorphism means the occurrence of differences DNA in the population. However, polymorphism is not defined as rare phenomenon (1% or higher frequency in the population).[12]

Polymorphism can be divided into:
– SNP (Single Nucleotide Polymorphism) - a single nucleotide polymorphism consisting in exchanging

one base for another in the DNA strand. The phenomenon of single nucleotide polymorphisms is identified by the RFLP PCR (Restriction Fragments Length Polymorphism PCR) or HRM PCR (High Resolution Melting PCR) technique;

– polymorphism of tandem repeats – variable number of tandem repeats (VNTR) (e.g. in the mitochondrial gene (mtDNA) – e.g. gene *Tomm 40*);

– insertion/deletion (I/D) polymorphism of larger fragment nucleotides (e.g. in the *ACE, VEGF* genes);

– polymorphism of mini- and microsatellite sequences (short tandem repeats – STR) – it is estimated that there are approximately several thousand loci containing human-genome sequences in the human genome, accumulated mainly in telomere chromosomal regions, containing from 7–100 base pairs in one repetition and about 2 – to several hundred sequentially ordered repeats in one loci. The level of mutations in mini satellite sequences is very high and amounts to about 5%. Microsatellite sequences are a different type of polymorphism, consisting in the presence of a variable number (5–100) of tandem repeats composed of 1 to 6 nucleotides (e.g. (CATG) n or (CA) n), distributed evenly every 6-10,000 base pairs. Different STR alleles usually occur in a population of several to a dozen times, which significantly increases the resolving power of these methods compared to the SNP variability analysis. In the human genome, they are most often located in sequences flanking genes and introns (noncoding areas).

Gene polymorphism means the occurrence of various variants of a given gene, which in consequence may lead to differences in the structure and action of the protein encoded by this gene (non-synonymous mutation - it involves changing the amino acid in the protein). In the case of a silent mutation - synonymous, there is no change in the amino acid in the protein, this is due to the fact that the genetic code is degenerate. On the other hand, changes in non-coding sequences (in introns) may lead to changes in protein expression or may not affect their expression and structure.[13]

Genes that play a major role in AD

Polymorphism of the *APOE* gene (apolipoprotein E) is the first risk factor identified in AD discovered in 1993.[14] In the brain, it plays a role in the metabolism of lipoproteins and cholesterol homeostasis. The gene is located on chromosome 19ql3.2 and occurs in the form of multiple alleles ε2, ε3, ε4 encoding individual isoforms, ApoE2, ApoE3 and ApoE4, respectively. The presence of three *APOE* alleles determines the occurrence of six genotypes in the human population, including three homozygous (2/2, 3/3, 4/4) and three heterozygous (2/3, 2/4, 3/4). The human population is dominated by a 3/3

homozygous genotype (approximately 60%), while Apo 3/4 heterozygotes account for approximately 20% and Apo 3/2 approximately 13%. The remaining genotypes homozygous and heterozygous are a minority, Apo 4/4 2-3%, Apo 2/2 1%, Apo 2/4 1-2%.[15]

The best known variant of the apolipoprotein E (*APOE4*) gene is responsible for the development of the AD. It is a gene coding for a molecule (apolipoprotein) that carries cholesterol in the blood.

– ApoE2 – reduces the chances of getting sick. It occurs in about 10% of healthy people and in 2% with Alzheimer's disease. If a person with ApoE2 becomes ill, it will happen later than in a person without this variant,

– ApoE3 – the most common variant, most likely without affecting the risk of disease,

– ApoE4 – increases the likelihood of developing Alzheimer's disease. It is present in 40% of patients with late form and in about 25% of healthy people. There are genetic tests to detect this variant, but their use is very controversial.[16]

The individual isoforms of ApoE are not evenly distributed among the world's population. Isoforms ε4 and ε2 are more common in Africa and relatively rare in Asia.[17] In Europe, ε4 isoform is more common in the northern countries, unlike in the Mediterranean region, where it is rare.[18] The main place of ApoE synthesis are cells liver, where most of the apolipoprotein E present in the plasma is formed (66-75%). Smaller amounts of ApoE are synthesized outside the liver, in many different peripheral cells, mainly macrophages, astrocytes, but also in the lungs, kidneys, spleen, and muscles. The concentration of apolipoprotein E in healthy people's plasma ranges from 0.016-0.17 g/l.

ApoE compound with pathological disorders

Levels of ApoE are usually measured in plasma and CSF. Peripheral blood ApoE levels have been proposed as biomarkers of AD, but tend to be lower in patients with AD than in healthy individuals. Such findings remain controversial. For this reason Wang *et al.* in their meta-analysis re-examined the potential role of peripheral ApoE in AD diagnosis. Wang *et al.* supports a lowered level of blood ApoE in AD patients. They reportered that value of ApoE as an important risk factor for AD.[19]

The main role plays presence of the ε4 allel, recognized such a risk factor for Alzheimer's disease.[15,20,21] The presence of at least one ε4 allele was found in 80% of familial and 64% of sporadic Alzheimer's disease, while in healthy people, ε4 is approximately 31%. It is estimated that in people homozygous ε4/ε4 symptoms of the disease appear on average 16 years earlier than in non-carriers of this allele.[22] The increased frequency of the ε4 allele was also found in patients with other neurodegenerative diseases, including AD with Lewy bodies, mixed dementia

and mild cognitive impairment.[23-25] There was a consistent association between the presence of an ε4 allele and both the clinical diagnosis of dementia and cognitive decline. These findings confirm a genetic heterogeneity in late onset sporadic AD and prompt caution in the use of ApoE genotype to predict an elderly individual's susceptibility to either dementia or cognitive decline. In addition to the *APOE* gene and its product - apolipoprotein, the Tau protein and the phosphorylated tau protein also play an important role in AD.[26]

Another important genes in pathology AD

Identification of specific risk genes in AD is problematic because the increase risk conferred by a single gene is small. In addition, there is need to identify combinations of dementia risk alleles not just individual genes. The complicating factor is also the heterogeneity of the underlying pathological changes, particularly concurrent cerebrovascular disease.[27] Multiple genetic and environmental factors regulate the susceptibility to AD. There is also a common belief that, apart from the best characterized risk factor, the APOE polymorphism, some other genes are involved in sporadic AD susceptibility for example *CYP46, APP, PSEN1, PSEN2, UBQLN1, BACE1, PRND, APBB2, TOMM 40*. To date, numerous genes have been tested for their putative influence on AD; the investigations have been focused mainly on polymorphisms located in particular genes of interest and in intron sequences. Current data on case-control genetic studies are being collected and updated in the AlzGene Database.[28,29]

A gene encoding a cholesterol degrading enzyme of the brain, called 24-hydroxylase (CYP46A1) is located on chromosome 14q32.1 and it has been linked with risk for AD. The single nucleotide polymorphism (T/C) there is in intron 2 of *CYP46* gene. The product of the CYP46 gene is the water soluble 24(S)-hydroxycholesterol. The elevated plasma and CSF hydroxycholesterol concentrations have been found in AD. This is associated with increased cholesterol turnover in brain or neurodegenerative process. Results of analyses of the connection between *CYP46* polymorphisms and AD carried out so far have been incredibly divergent. Both CC and TT genotypes of the rs754203 polymorphic site were proposed as genetic risk factors for AD; whereas other analyses showed no association between this polymorphism and AD.[30-32]

In the research of Golanska et al., Juhasz et al. and Combarros *at al.* the authors pointed to the *CYP 46* gene (cholesterol 24S-hydroxylase) as a potential marker for the identification of AD. The *CYP 46* gene is responsible for the removal of excess brain cholesterol by hydroxylation. The effect of *CYP46* gene polymorphisms on AD has been mainly focused on the known intronic single nucleotide polymorphism (SNP) rs754203.

In the study Golanska *at al.* were found a relationship between polymorphism *CYP 46* rs754203 (CC) and APOE ε4.[30] They analyzed polymorphisms in 215 Polish AD cases and 173 healthy individuals. It was noticed that the CC genotype of the known rs754203 polymorphic site might be a risk factor for AD, especially in at least one APOE ε4 carriers.

These observations were in contrast to results of other authors, who did not find any significant difference in *CYP46* genotypes frequencies between AD and controls after stratification by *APOE* status, although they found an association between the *CYP46* CC genotype and higher risk of AD.

Other conclusions are presented by Anna Juhasz *et al.* in their research. A case–control study was performed on 125 AD and 102 age- and gender-matched control subjects from Hungary, to test the association of *CYP46* T/C and apolipoprotein E (ApoE) gene polymorphisms in AD. They indicate that the intron 2 T/C polymorphism of *CYP46* gene (neither alone, nor together with the ε4 allele) does not increase the susceptibility to late-onset sporadic AD in the Hungarian population.[31]

In another studies Golanska *et al.* (2009) were found that the levels of phosphorylated tau protein and p-amyloid peptide in CSF were increased in AD patients carrying the rs754203 TT genotype. Analysis of polymorphism distribution carried out in various ways up to now, however, does not reveal a clear relationship between *CYP46* and AD genotypes.[29]

A case-control study Combarros's *et al.* utilizing a group of 321 sporadic AD patients and 315 control subjects was performed to examine the relationship between different genotypes in gene *CYP 46*. Their results indicated too that the intron 2 *CYP46* CC genotype may predispose to AD, and this association is independent of the apolipoprotein E genotype.[32]

For familial form of early-onset Alzheimer's disease, mutations in 3 genes have been described: amyloid precursor protein (*APP*), presenilin proteins (*PSEN1* and *PSEN2*) but they account only a small fraction of all AD cases. Mutations in these three genes maybe cause autosomal dominant forms of EOAD.[33] Although these genes were identified in the 1990s, variant classification remains a challenge, highlighting the need to colligate mutations from large series. Mutations in the gene for *APP* account for about 10-15% of all cases of familial, autosomal dominant form of Alzheimer's, in *PSEN1* - 20-70%, and mutations in *PSEN2* have been described so far only in single families and are considered very rare.

PSEN1 a gene was found on chromosome 14 (gene ID: 5663 NCBI), gene *PSEN2* on chromosome 1 (gene ID:5664 NCBI) and gene *APP* (amyloid beta precursor protein) on chromosome 21(gene ID: 351 NCBI). Patients with Alzheimer's disease (AD) with an inherited

form of the disease carry mutations in PSEN1, PSEN2 or the APP. These disease-related mutations cause increased production of the longer form of amyloid beta (Aβ) the main component of amyloid deposits found in AD brains). Presenilins are thought to regulate the processing of APP through their effects on gamma-secretase, an enzyme that cleaves APP.

APP amyloid beta precursor protein gene encodes a cell surface receptor and transmembrane precursor protein that is cleaved by secretases to form a number of peptides. Some of these peptides are the protein basis of the amyloid plaques found in the brains of patients with Alzheimer disease. Mutations in this gene have been implicated in autosomal dominant Alzheimer disease and cerebroarterial amyloidosis (cerebral amyloid angiopathy). Multiple transcript variants encoding several different isoforms have been found for this gene.

It is also believed that presenilins participate in the cleavage of the Notch receptor, such that they either directly regulate gamma secretase activity or act as protease enzymes themselves. Two alternatively spliced transcript variants encoding different isoforms of *PSEN2* have been identified. The mentioned genes are therefore further candidates as markers of Alzheimer's disease.[34] In the Lanoiselée *et al.* work we read that *PSEN1*, *PSEN2* and *APP* mutations are autosomal dominant forms of EOAD. Although these genes were identified in the 1990s, but need to colligate mutations from large series of familial and sporadic case. Their findings suggest that a unnoticeable part of *PSEN1* mutations occurs *de novo*, which is very importance for genetic counseling, as *PSEN1* mutational screening in autosomal dominant AD because this test is currently performed in familial cases only.[35] The *PSEN1* c.236C>T, p.(Ala79Val) substitution is currently considered pathogenic and to be associated with a LOAD compared to the other *PSEN1* variants. This substitution was also found in subjects with EOAD so its frequency is of importance for diagnosis in autosomal dominant form of AD. They identified only one novel *PSEN2* mutation, c.850A>G, p.(Arg284Gly), and a previously known mutation, p.(Thr122Pro), during this screen. In the *APP* gene, no novel mutation was found too. Researchers also identified a previously reported mutation from autosomal dominant EOAD families. The most frequent one was the c.2149G>A, p.(Val717Ile) substitution and the c.2137G>A, p.(Ala713Thr) mutation was found in other patients from unrelated families. In addition, Lanoiselée *et al.* found families carried mutations located within the coding sequence of the Aβ peptide: "Flemish" *APP* mutation c.2075C>G, p.(Ala692Gly), the "Italian" mutation c.2077G>A, p.(Glu693Lys), and the "Iowa" mutation c.2080G>A, p.(Asp694Asn). In the case of *APP* gene duplication, it was noticed that all patients exhibited

progressive cognitive impairment in autosomal dominant EOAD families and in the sporadic cases.[35] Similar results the effects of mutations of the *PSEN1* gene were shown in another research. Bagyinszky *et al.* reports that *PSEN1* mutation (c.335C>T), p.(Thr116Ile) was observed in two Korean families with autosomal dominant inheritance. The personality changes occurred in their 30 years old. This mutation (c.335C>T), p.(Thr116Ile) was first discovered in an Italian patient and two French families with EOAD with similar age of onset. The possible pathogenic mechanisms of mutation were verified. These were changes in the presenilin protein. In addition pathogenic mutation, *PSEN1* (Thr116Asn), was also found where the patient presented young onset AD (YOAD). Its mean that first symptoms appeared before the age of 30.[8]

Similar results were presented in study Giau *et al.* They carried out research on a 37-year-old man, a patient from Korea carrying the mutation *PSEN*1 (p.Gly-417Ala) mutation with exceptionally early and severe presentations, including a wide range of atypical symptoms of rapid cognitive decline with a stooped posture, rigidity, and bradykinesia.

A targeted next-generation sequencing was performed on the patient which revealed a new nucleotide substitution (c1250G> C) in exon 12 of the *PSEN1* gene, changing glycine to alanine at position 417 (p.Gly-417Ala). This mutation may cause disturbances in the 8th transmembrane region, disrupting its functions from the increased hydrophobicity and amount of alanine with reduced elasticity. Since several glycine> alanine substitutions in other transmissive *PSEN1* helices have revealed aggressive phenotypes of Alzheimer's disease, *PSEN1* Gly417Ala may have a common pathogenic mechanism.[36]

Haapasalo *et al.* also reported that key events in the pathogenesis of different neurodegenerative diseases are abnormal protein aggregation and intracellular or extracellular accumulation of misfolded and deposited proteins. Additionally, stress in endoplasmic reticulum and debility of the ubiquitin–proteasome system probably contribute to neurodegeneration in these diseases also in AD. Evidence shows that the AD-associated presenilin also creates aggregates under certain conditions and that ubiquilin-1, controls protein aggregation and their deposition.[37]

The *UBQLN1* gene was found on chromosome 9 and encodes an ubiquitin-like protein (ubiquilin) containing an N-terminal ubiquitin-like domain and a C-terminal ubiquitin-associated domain. The studies suggested that a single intronic C/T polymorphism, UBQ-8i (rs12344615), contribute to AD risk. An association was found between a functional ubiquitination machine and a proteasome to affect protein degradation *in vivo*. This ubiquilin has also been shown to modulate

accumulation of presenilin proteins, and it was found in lesions associated with AD. Two transcript variants encoding different isoforms have been found for this gene. (gene ID: 29979 NCBI)

Single nucleotide polymorphisms in the ubiquilin-1 gene may confer risk for LOAD. El Ayadi et al. have shown previously that ubiquilin-1 functions as a molecular chaperone for the amyloid precursor protein (APP) and that protein levels of ubiquilin-1 are decreased in the brains of AD patients. They have recently found that ubiquilin-1 regulates APP trafficking and subsequent secretase processing by stimulating non-degradative ubiquitination of a single lysine residue in the cytosolic domain of APP. Thus, ubiquilin-1 plays a central role in regulating APP biosynthesis, trafficking and ultimately toxicity. As ubiquilin-1 and other ubiquilin family members have now been implicated in the pathogenesis of numerous neurodegenerative diseases, these findings provide mechanistic insights into the central role of ubiquilin proteins in maintaining neuronal proteostasis.[38] Results from Zhang et al. from a meta-analysis suggest that the UBQ-8i polymorphism may be associated with AD development. But further studies with larger group of respondents are needed to further estimate the presence of an association.[39] The same results were shown by Chuo et al. in their research.[40]

An important role in AD also played gene BACE1. Aβ is generated from amyloid precursor protein (APP) by β-site APP-cleaving enzyme 1 (BACE1) and γ-secretase-mediated cleavages. Ubiquilin-1, a ubiquitin-like protein, genetically associates with AD and affects APP trafficking, processing and degradation. Natunen et al. have investigated ubiquilin-1 expression in human brain in relation to AD-related neurofibrillary pathology and the effects of ubiquilin-1 overexpression on BACE1, tau, neuroinflammation, and neuronal viability in vitro in co-cultures of mouse. Taken together, these results suggest that ubiquilin-1 may mechanistically participate in AD molecular pathogenesis by affecting BACE1 and thereby APP processing and Aβ accumulation.[41]

It should be noted that for several years scientists have been some similarities between the background of neurodegenerative diseases such as AD or Parkinson's disease (PD) and prion diseases due to the fact that in both cases protein conformation and aggregation change are observed.[42] PRND gene was found on chromosome 20, mutations in this gene may lead to neurological disorders (gene ID: 23627 NCBI). The protein encoded by this gene is a membrane glycosylphosphatidylinositol-anchored glycoprotein. It there is approximately 20 kbp downstream of the gene encoding cellular prion protein, to which it is biochemically and structurally similar.[43]

APBB2 an amyloid beta precursor protein binding family B was found on chromosome 4. The protein interacts with the cytoplasmic domains of amyloid beta precursor protein (APP) and amyloid beta (Aβ) precursor-like protein 2 (APLP2). This protein which is thought to function in signal transduction, contains two phosphotyrosine binding (PTB) domains. Polymorphisms in this gene have been associated with AD.[44] Alternative splicing results in multiple transcript variants. (gene ID: 323 NCBI)

TOMM 40 also known as PEREC1-mitochondrial gene is responsible for translocase of outer mitochondrial membrane 40 is located on 19 chromosome (NCBI gene ID: 10452). Mitochondria are semiautonomous organelles. They are responsible for energy production (adenosine triphosphate - ATP). In addition mitochondria play a crucial role in mediating amino acid biosynthesis, in metabolism of lipid, intermediate metabolic pathways, calcium homeostasis and free radical scavenging. These organelles are semi-conservative, and their divisions do not depend on cell division. Their metabolism is important for cell survival and proper functioning. Mitochondrial dysfunction leads, among other things, to neurodegenerative diseases, including Alzheimer's disease. In AD, we first observe mitochondrial dysfunction and then the detectable amyloid pathology.[45,46] TOMM 40 is the gene which plays an increasingly important role in the pathogenesis of AD. The protein encoded by this gene is localized in the outer membrane of the mitochondria. It is the channel-forming subunit of the translocase of the mitochondrial outer membrane (TOM) complex that is essential for import of protein precursors into mitochondria. Alternatively spliced transcript variants have been found for this gene. In the TOMM40 gene, the rs10524523 ("523") variable length poly-T repeat polymorphism has more recently been associated with similar phenotypes AD.[47] A variable-length poly-T variant in intron 6 of the TOMM40 gene, is associated with risk and age of onset of sporadic LOAD. In Caucasians, are distinguish the three predominant alleles at locus in intron 6: Short (S), Long (L) or Very long (V) On an APOE ε3/3 background, the S/VL and VL/VL genotypes are more protective than S/S.[45] Research have shown that impaired mitochondrial function leads to reduced glucose uptake in older individuals, and can lead to insulin resistance, thus perpetuating the cycle linking TOMM40, diabetes mellitus (DM), and dementia. Compared to the TOMM40 short (S) allele, the very long (VL) allele is associated with earlier onset of Alzheimer's dementia, smaller brain volumes, and poorer cognitive performance. Based on the research results we can say mitochondria plays the role in inherited neurodegenerative diseases and they are inherited from the maternal line.[46,48]

In table 1 are included genes polymorphisms very important in increased risk of developing Alzheimer disease.

Table 1. Genome significant representative SNPs reported for AD

NA* – Not available

No.	Genes	ID	located on chromosome	protein coding	single nucleotide polymorphisms (rs) ID	description of the mutation	Role in pathomechanism
	APOE	348	chromosome 19q13.2	Apolipoprotein E	rs429358 rs7412	three alleles APOE: ε2, ε3, ε4	amyloid beta plaques, which accumulate in the brains of human; familial and sporadic Alzheimer's disease ApoE4 increases the likelihood of developing AD
	CYP46A1	10858	chromosome 14q32.1	a member of the cytochrome P450 superfamily of enzymes	rs754203 rs4900442 (only in the Chinese population)	T/C	Increased cholesterol turnover in brain or neurodegenerative process
	APP	352	chromosome 21q21.3	amyloid precursor protein	NA* NA* NA* NA* NA* rs2830077	G/A p.(Val717Ile) G/A p.(Ala713Thr) G/A p.(Glu693Lys) G/A p.(Asp694Asn) C/G p.(Ala692Gly) C/A	longer form of amyloid beta (Aβ); inherited form of the disease: autosomal dominant EOAD/YOAD
	PSEN1	5663	chromosome 14q24.2	presenilin proteins are component of gamma secretase	rs63750730 rs63750231 rs63750306 rs63750083 rs63751037 rs63750577 NA*	C/T A/C A/C C/A A/G C/T G/C (p.Gly417Ala)	longer form of amyloid beta (Aβ); inherited form of the disease: autosomal dominant EOAD/YOAD
	PSEN2	5664	chromosome 1q42.13	presenilin proteins are component of gamma secretase	rs28936379 rs28936380 rs367855127 rs533813519 rs574125890 rs63750215 rs63750666 NA*	A/G C/G C/T A/C G/T A/T C/T A/G p.(Arg284Gly)	longer form of amyloid beta (Aβ); inherited form of the disease: autosomal dominant EOAD/YOAD
	UBQLN1	29979	chromosome 9q21.32; 9q21.2-q21.3	ubiquitin-like protein (ubiquilin)	rs12344615	C/T	a central role in regulating APP biosynthesis, ubiquilin has been shown to modulate accumulation of presenilin proteins; risk for LOAD
	BACE1	23621	chromosome 11q23.3	APP beta-secretase	rs4938369 rs638405	T/C C/G	the first step in the formation of amyloid beta peptide from amyloid precursor protein
	PRND	23627	chromosome 20p13	membrane glycoprotein	NA*	C/T 3'UTR (untranslated region)	behavioral abnormalities (an elevated risk for delusions, anxiety, agitation/aggression, apathy and irritability/emotional ability)
	APBB2	323	chromosome 4p14-p13	amyloid beta precursor protein binding family B	rs13133980	C/G	LOAD disease onset before 75 years of age characterized pathologically by neurofibrillary tangles and amyloid plaques and clinically by progressive impairment of mental functioning
	TOMM 40	10452	chromosome 19q13.32	Mitochondrial gene; translocase of outer mitochondrial membrane 40	rs10524523	poliT	EOAD and sporadic familiar LOAD

In addition to the polymorphisms of selected genes, histone proteins, DNA methylation and ncRNA also play an important role, which are often omitted in research as mechanisms associated with AD. Yu et al. noticed in their study that despite single biomarkers for AD having been determined on a genome-wide scale, the differential co-expression in gene pairs between regions and interactions with other types of cellular molecules, particularly non-coding ncRNAs, are often overlooked in studies investigating the underlying mechanisms associated with AD.[49]

Conclusions

Meta-analysis and comparative analysis of the gene complex responsible for AD

Looking at multiple genes together rather than analyzing them individually, may improve identification of AD risk alleles.[4,50] Moreover, there may be multiple sufficient risk sets for AD. LOAD is highly polygenic with 30 loci identified in human DNA by GWAS and early whole exome sequencing (WES) study.[51] Recent advances in technology and Internet development have led to new methods for studying various diseases called the "omics era." It allows the collection of large amounts of data and information at the molecular and protein level, along with the development of novel computational methods and the use of statistical tools that are able to analyzing and filtering such data, advances in the genotyping matrix, next generation sequencing (NGS), mass spectrometry technology and bioinformatics have enabled the simultaneous large-scale studies of thousands of genes, epigenetic factors, RNAs, metabolites and proteins, with the possibility of integrating many types of omics. All of these technological innovations have modified the approach to research on complex diseases, including Alzheimer's (AD). All above methods and GWAS, TWAS, and EWAS are a promising tool to study the relationship between several molecular pathways in AD and other diseases.[2,52]

In conclusion, to gain further understanding of the role of described genes in risk of AD scientists should have deeper sequencing approaches (Whole-Genome Sequencing -WGS) to identify variants in larger insertions and deletions and signals from non-coding regions of DNA. As of today, it is known that about 67 autosomal genes are strongly associated with LOAD. So far, there are no drugs aimed at alleviating the effects of polymorphisms and mutations in these genes.

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