Research in Molecular Medicine

Identification of Critical Genes and Underlying Diseases in Early-onset Alzheimer's Disease by Bioinformatics Approach

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ABSTRACT

Background: Alzheimer's disease (AD) is a neurodegenerative and multifactorial disorder. Investigating the key genes and metabolic pathways is important for understanding the mechanisms of AD. This study aimed to analyze the gene network and biological pathways of AD using bioinformatics approaches.

Materials and Methods: AD-related genes were identified, and a gene network was constructed using STRING. Network analysis was performed to identify functional modules and key genes related to AD using Cytoscape, and gene ontology was investigated using g:Profiler.

Results: Through network clustering, five functional modules were identified, which play important roles in amyloid-beta formation, protein metabolic processes, and responses to organic substances. Key genes in AD included *APOE*, *TREM2*, *SORL1*, *BIN1*, *PICALM*, *ABCA7*, *CD2AP*, *CD33*, *MS4A6A*, and *CLU*. These genes play roles in the negative regulation of amyloid precursor protein catabolic process. A significant function of these genes is amyloid-beta binding, and they are mostly localized in the somatodendritic compartment. Four key genes—*APOE*, *SORL1*, *ABCA7*, and *TREM2*—are associated with early-onset AD (EOAD) and play critical roles in AD. Additionally, the *APOE* and *SORL1* genes have an indirect role in AD at a young age through underlying diseases such as obesity, hypercholesterolemia, diabetes mellitus, and hypertension.

Conclusion: Some underlying diseases are related to EOAD, and controlling these conditions may prevent the early onset of AD. These results can provide novel insights into the pathogenesis and treatment of AD.

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Introduction

Alzheimer's disease (AD) is the most common neurodegenerative disorder and a complex brain disorder whose risk genes have not been fully identified [1]. Alois Alzheimer, a German psychiatrist, first identified the disease in 1907 [2]. As one of the main causes of dementia, AD is a major concern for global health, and its consequences have a significant impact on individuals and societies [3]. According to the World Health Organization (WHO), AD and other forms of dementia are among the top ten causes of death in the world, with 65% of deaths from this disease occurring in women. Approximately 24 million people worldwide suffer from dementia, with AD constituting the predominant form of this neurodegenerative condition [4]. With the rising average age of the population, it is predicted that the number of people with AD will exceed 81 million by 2040 [5]. The incidence of this disease exhibits exponential growth, and the risk of developing AD doubles every five years after the age of 65. The main risk factors for AD include genetics, aging, and neuroinflammation. The disease typically begins with memory loss and progresses to cognitive impairment, such as cognitive decline, disorientation, difficulty in independent functioning, and speech impairment, which can interfere with daily activities [6].

Research has shown that low birth weight, preterm birth, and diabetes are related to dementia and AD. Females show a disproportionately higher prevalence of AD for their age, suggesting the influence of additional risk factors beyond lifespan [7]. AD is categorized into two groups depending on the age at which it begins: Early-onset AD (EOAD), which occurs before the 65 age, and late-onset AD (LOAD), which occurs after the age of 65. AD is related to neurofibrillary tangles (NFTs) and senile plaques (SP). Amyloid-beta, a major component of SP, has various pathological effects on organelle function and cells. Mutations in three genes—*PSEN1*, *PSEN2*, and *APP*—are more commonly associated with EOAD, as they encode the amyloid beta precursor protein APP and contribute to the production of amyloid-beta [8]. These genes are involved in the amyloidogenic pathway, which leads to the formation of amyloid plaques, a hallmark of AD. In LOAD, factors, such as lifestyle, environment, genetics, and aging play significant roles, with about 10% of all patients in this group being influenced by these factors. The *APOE* gene is the strongest genetic risk factor for LOAD. Other genes, such as *BIN1*, *CD33*, and *PICALM*, have also been associated with an increased risk of developing LOAD. Previous research suggests that as people age, they become more susceptible to neurodegenerative disorders, characterized by a series of biological events, including severe inflammation, oxidative damage, compromised metabolism, endocrine disruption, and organ decline [9, 10]. As AD progresses, individuals experience a relentless erosion of cognitive function, manifesting as debilitating memory deficits, delusions, and confusion. Several pathophysiological mechanisms, including the contraction of blood vessels and muscles, dysfunction of mitochondria, and production of free radicals, are involved in memory loss [11].

Amyloid-beta precursor protein (APP) is a membrane protein that penetrates the membrane of nerve cells and is essential for the growth, survival, and repair of nerve cells. The gene encoding the APP protein is located on chromosome 21 [12]. The *PS* gene is responsible for the production of an intramembrane protein, and mutation in this gene causes a change in the APP, leading to increased amyloid production [13]. Mutations in both the APP and PS proteins consistently elevate levels of amyloid-beta peptides, particularly the more aggregation-prone amyloid-beta42. Amyloid-beta forms Ca^{2+} permeable pores and modulates several synaptic proteins, including mGluR5, VGCC, and NMDAR, inducing the overloading of neurons with calcium ions. As a result, cellular Ca^{2+} disturbances lead to autophagy defects, neuronal apoptosis, mitochondrial abnormalities, synaptic vulnerability, defective neurotransmission, and neurodegeneration in AD. Increased oxidative stress and metabolic disorders in AD lead to endoplasmic reticulum (ER) stress and neuronal death. ER stress is a condition caused by an imbalance between protein production and development, leading to problems in protein reactions. FAD-linked PS1 mutation reduces the unfolded protein response and results in vulnerability to ER stress. The ApoE protein plays a role in the metabolism of body fats. This protein is involved in AD and cardiovascular disease and carries cholesterol into and out of cells. Maintaining an optimal cholesterol level in the blood and the balancing role that lipoproteins play in cholesterol transport in the nervous system are important for brain health and neuronal function [14, 15]. In addition to the aforementioned genes, the new genome-wide association study (GWAS) approach has identified several chromosomal regions and genetic loci related to the risk of AD. These regions are responsible for disease signaling and are involved in major pathophysiological pathways, such as APP metabolism [16]. In genomic studies, it was found that 19 gene regions—*CASS4*, *CELF1*, *FERMT2, HLA-DRB5, INPP5D, MEF2C, NME8, PTK2B, SORL1, ZCWPW1, SLC24A4, CLU, PICALM, CR1, BIN1, MS4A, ABCA7, EPHA1* and *CD2AP*— are associated with the risk of AD [17].

Multiple hypotheses have been proposed regarding the causes of AD. The amyloid-beta cascade hypothesis states that amyloid accumulation initiates AD, and evidence shows that disruption in Aβ clearance, rather than its overproduction causes the disease. This molecule is a primary factor in AD, and the results of mutations in the *APP*, *PSEN1*, and *APOE* genes also support this hypothesis. The amyloid oligomer hypothesis is another theory, which has led to significant progress in understanding the mechanisms of synapse destruction and neuronal death associated with it. Also, due to the role of lysosome destruction and autophagy in AD, other hypotheses regarding synapse destruction and neuronal death have been proposed [18]. Although amyloid-beta is at the forefront of the causative cascade of AD, it is also involved in the formation of hyperphosphorylated tau and NFT formation. Today, it is believed that amyloid beta and tau have mutual relationships and effects. Many molecular pathways leading to synapse destruction and neuronal death interact with each other, making it impossible to propose a single hypothesis for the molecular causes of AD [19].

AD is related to several diseases, such as type 2 diabetes, Down syndrome, bowel diseases, and kidney diseases. Research shows that diabetes and any other disease that affects blood sugar can affect brain and nervous system function. Increased blood sugar levels increase amyloid-beta levels in the patient's body, which can lead to AD [20]. Down syndrome is one of the most common genetic disorders that is caused by the complete or partial trisomy of chromosome 21. In this trisomy, people with Down syndrome have an extra copy of the APP gene and produce excessive amounts of amyloid-beta. Almost all adults with Down syndrome create amyloid plaques and NFTs, which are signs of AD. Furthermore, with improvements in healthcare and longer lifespans for individuals with Down syndrome, the population diagnosed with AD within this community is steadily increasing. These results show the need for further research and early intervention strategies to address the specific challenges of AD in people with Down syndrome [21]. Recently, the relationship between constipation and changes in the gut microbiome with AD has been identified. Changes in the gut microbiome have been reported as inflammatory responses in the development of AD. This inflammatory response leads to damage to the central nervous system. In addition, metabolites and cytokines are secreted from the intestinal cells, which affect the activity of the microbiome and affect the activity of neurons [22]. Recent studies have identified an association between chronic kidney disease (CKD), particularly in elderly populations, and an increased risk of developing various forms of cognitive decline, ranging from mild cognitive impairment (MCI) to dementia [23].

These findings have helped to better identify the biological pathways involved in the disease, and the promising results from this study suggest that future therapies might be developed to treat or prevent AD. Although there is no known way to stop or prevent the progression of this disease, some treatments can help improve its symptoms. Given the increasing number of people affected, it is crucial to identify the metabolic pathways involved in the disease and predict target genes and biomarkers to aid in the early diagnosis of AD. In this study, we aimed to use bioinformatics tools to investigate the genes identified for AD and analyze gene interactions and metabolic pathways.

Materials and Methods

Data collection

We performed a systematic search in January 2023, using the Gene dataset from the NCBI and DisGeNET databases for studies on AD [24]. The following keywords were used: ["Early-onset alzheimer's disease" and "Late-onset alzheimer's disease"]. This search was filtered for: "Organism: Homo sapiens.

Construction and analysis of the protein-protein interaction (PPI) network

The identified genes were uploaded to the STRING database version 11.5 and the PPI network of AD genes was constructed [25]. The PPI network was constructed based on protein homology, co-expression, and gene neighborhood criteria. Then, to analyze the topology of the AD network, key genes and functional modules were identified using the Cytoscape software. version 3.9.1 [26]. The molecular complex detection (MCODE) plug-in with default settings (K-core: 2, node score cutoff: 0.2, max depth: 100, Haircut: True, and degree cutoff: 2) was used for clustering the network and identifying functional modules. The Cytohubba plug-in, based on the MCC ranking method, was used to identify key genes in the AD PPI network.

Gene network enrichment

To identify gene ontology (GO) terms, including biological process (BP), cellular component (CC), and molecular function (MF), g:Profiler was utilized. Additionally, an investigation of the metabolic pathways associated with AD was conducted using the Kyoto encyclopedia of genes and genomes (KEGG) [27]. MicroRNAs (miRNAs) regulate many cellular processes and metabolic pathways and play a role in various dis-

eases, including neurological diseases, cancer, and inflammatory diseases. Therefore, in this study, we identified miRNAs associated with key genes and AD using the Mirwalk and MirNet web servers, respectively [28, 29]. The workflow of this study is shown in Figure 1.

Results

Construction and analysis of the protein network

A total of 200 genes were identified for AD using the NCBI and DisGeNET databases. Then, AD genes were submitted to the STRING database and constructed the AD network. This network contains 1,105 edges, an average node degree of 13.3, and a PPI enrichment P<1.0e-16, as shown in Figure 2.

GO enrichment analysis of modules

The AD PPI network was clustered using the MCODE plug-in, resulting in the placement of related genes into five functional modules. The functional enrichment analysis of BP for each module was conducted using g:Profiler. The results are presented in Table 1. The metabolic pathways of AD in KEGG are shown in Figure 3. Also, after the investigation of each module, the common genes between them and EOAD and LOAD were found and reported in Table 2.

Analysis of key genes

Ten key genes were identified in the AD gene network. These key genes include *APOE*, *TREM2, SORL1, BIN1, PICALM, ABCA7, CD2AP, CD33, MS4A6A,* and *CLU*. The interaction network of these ten key genes is shown in Figure 4.

Go enrichment analysis of key genes

Functional enrichment of key genes was investigated using the g: Profiler platform, and the results are presented in Table 3.

Identification of underlying diseases

An investigation of key genes in DisGeNET showed that five key genes, which included *BIN1*, *CLU*, *CD33*, *PICALM*, and *CD2AP* are related to LOAD, and four key genes, which included *APOE*, *SORL1*, *TREM2*, and *ABCA7* play a role in EOAD. *TREM2* and *ABCA7* genes play a critical role in the development of neurodegenerative diseases, such as AD, Parkinson's disease, memory impairment, dementia, and cognitive impairment. The *APOE* and *SORL1* genes, play a direct role in AD and also play an indirect role in the occurrence of this disease. *APOE* is one of the genes that cause obesity, hypercholesterolemia, diabetes mellitus, and hypertensive

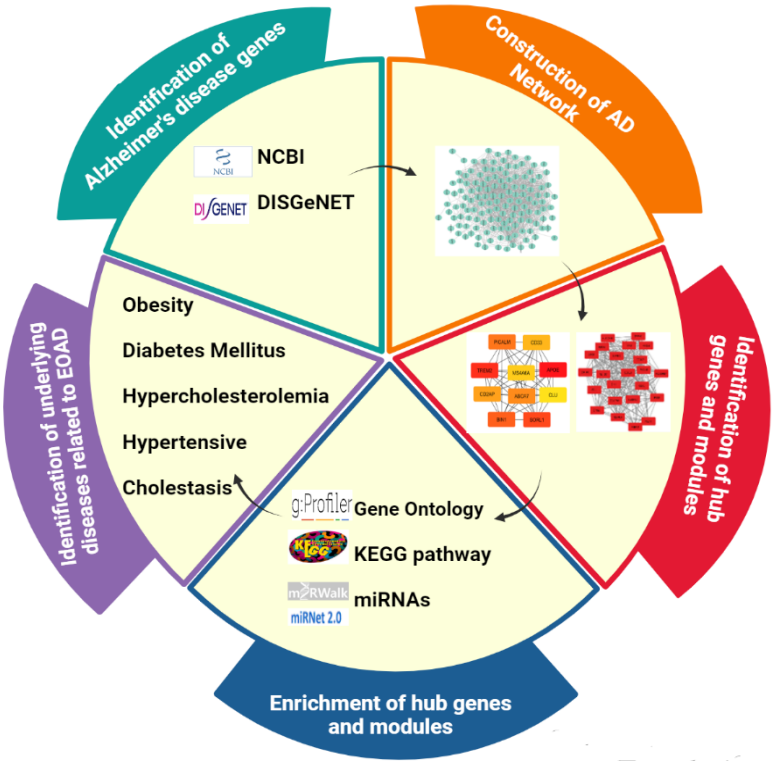


Figure 1. Research workflow

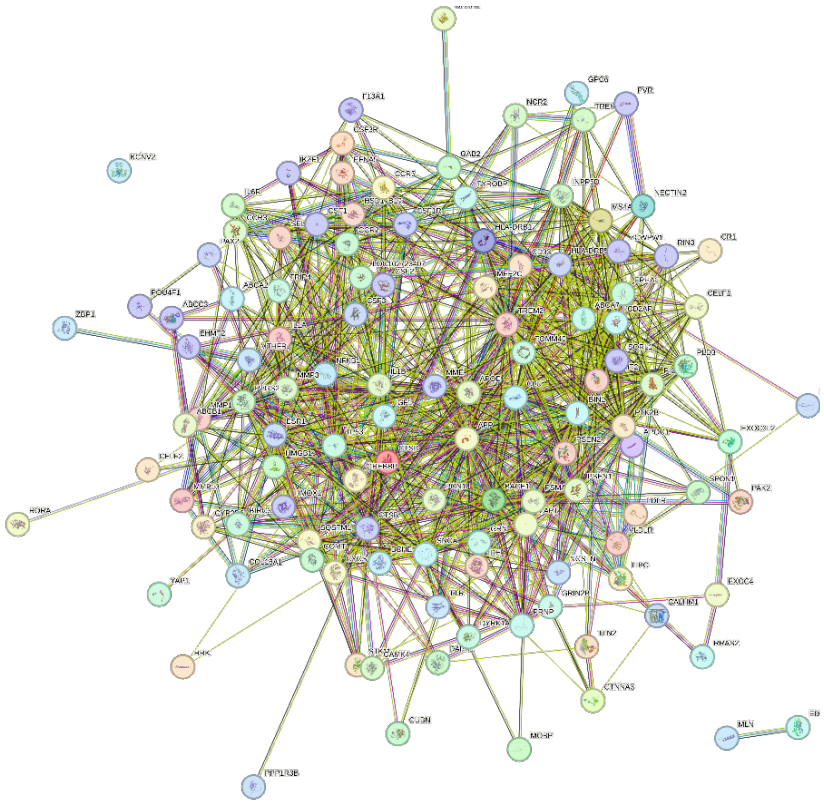


Figure 2. Gene network of the AD

disease through the metabolism, signal transduction, transport of small molecules, metabolism of proteins, vesicle-mediated transport, gene expression (transcription), and sensory perception pathways. Additionally, *SORL1* is implicated in obesity, cholestasis, non-insulin-dependent diabetes mellitus, and hypercholesterolemia through protein metabolism pathways.

miRNA-key gene interaction network

miRNAs are short RNA molecules that consist of 23–17 nucleotides. They play a key role in regulating gene function. These molecules interact with target molecules that have a critical role in protein synthesis. In this study, the miRNAs associated with key genes and AD were extracted using the miRWalk and MirNet platforms, respectively. A total of 156 miRNAs were found to be shared between AD and the key genes. The miRNA-key gene interaction network was constructed using Cytoscape (Figure 5). This result highlights the importance of the key genes in AD.

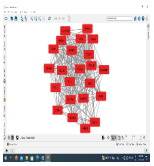
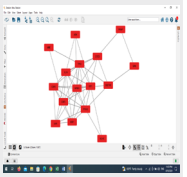
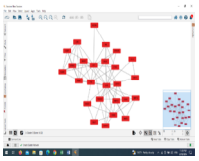
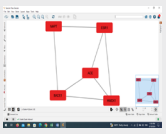
Discussion

After analyzing the AD gene network, the functional module and key genes were identified and then the functional enrichment analysis of them was performed. Five functional modules were identified, each playing an important role in different processes related to AD. The functions of each module are as follows:

Module M1 consists of 23 nodes and all ten key genes were found to be located within this module. M1 has a critical role in the amyloid-beta formation, regulation of multicellular organismal processes, positive regulation of ERK1 and ERK2 cascade, regulation of neuron apoptotic process, and learning or memory. In this module, three genes—*PSEN2*, *PLD3*, and *RIN3*—are associated with EOAD, while seven genes—*BIN1*, *CLU*, *CD33*, *PICALM*, *EPHA1*, *CD2AP*, and *PTK2B*—are associated with LOAD. Also, five genes, including *PSEN1*, *APOE*, *SORL1*, *TREM2*, and *ABCA7*, are shared between EOAD and LOAD. This result shows the importance of this module in AD.

Module M2 contains 16 genes. M2 is involved in the positive regulation of protein metabolic processes, response to organic substances, regulation of catalytic activity, positive regulation of organelle organization, and Ras protein signal transduction. Six genes—*CSF1, CSF3, IL1A, CSF2, PRNP,* and *NFKB1*—are associated with EOAD, while one gene, A2M, is found in the LOAD gene list. These results indicate that M2 is primarily involved in EOAD.

Table 1. GO enrichment of functional modules M1 to M5

Module Name	Node No.	Node Name	GO ID	GO Function	Image of the Module
M1	23	*FERMT2, PSEN1, SORL1, EPHA1, PLD3, APOE, PSEN2, CD2AP, RIN3, PTK2B, ZCWPW1, PICALM, CELF1, BIN1, MEF2C, HLA-DRB5, MS4A6A, ABCA7, CD33, CLU, INPP5D, HLA-DRB1, TREM2*	GO:0034205 GO:0051239 GO:0070374 GO:0043523 GO:0007611	Amyloid-beta formation; regulation of multicellular organismal processes; positive regulation of the ERK1 and ERK2 cascade; regulation of the neuron apoptotic process; learning or memory	
M2	16	*TP53, SQSTM1, PTGS2, CSF3, BCHE, A2M, IL6R, CSF2, CSF1, IL1A, CYCS, NFKB1, GRN, MME, PRNP, IGF1*	GO:0051247 GO:0010033 GO:0050790 GO:0010638 GO:0007265	Positive regulation of protein metabolic processes; response to organic substances; regulation of catalytic activity; positive regulation of organelle organization; ras protein signal transduction	
M3	26	*SNCA, LIPC, GRIN2B, PON1, TTR, BCL3, HMGB1, NCSTN, CTSD, LDLR, CSF3R, ABCA1, MMP3, APP, CCR2, IDE, CSF1R, MMP12, IL1B, CCR3, SELP, APOC1, TYROBP, VLDLR, MMP13, BIRC3*	GO:0010033 GO:0008203 GO:0097006 GO:0006897 GO:0030301	Response to organic substances; cholesterol metabolic processes; regulation of plasma lipoprotein particle levels; endocytosis; cholesterol transport	
M4	5	*HMOX1, ESR1, ACE, MAPT, BAC1*	GO:1901700 GO:0010288 GO:0140448 GO:1990000 GO:0009628	Response to oxygen-containing compounds; response to lead ions; signaling receptor ligand precursor processing; amyloid fibril formation; response to abiotic stimuli	
M5	3	*MTHFR, ABCB1, COMT*	GO:0009410	Response to xenobiotic stimulus	

Module M3 contains 26 genes and plays an important role in the response to organic substances, cholesterol metabolic processes, regulation of plasma lipoprotein particle levels, endocytosis, and cholesterol transport. In this module, five genes—*SNCA, TYROBP, CSF1R, CSF3R*, and *NCSTN*— are associated with EOAD, while three genes—*IDE, LDLR,* and *ABCA1*—are associated with LOAD. Additionally, the APP gene is common between EOAD and LOAD. This suggests that this module is related to EOAD.

Module M4 contains five genes. This module is related to in response to oxygen-containing compounds, response to lead ions, signaling receptor ligand precursor processing, amyloid fibril formation, and response to abiotic stimuli. Two genes—*ACE* and *BACE1*—are associated with LOAD, while *MAPT* genes are common between EOAD and LOAD.

Module M5 consists of three genes and is involved in the response to xenobiotic stimuli. The *MTHFR* gene in this module is one of the genes associated with LOAD.

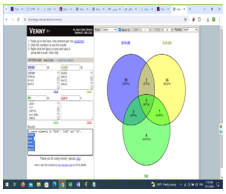
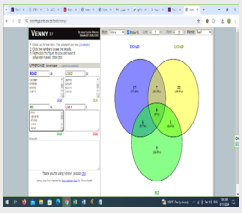
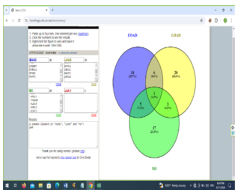
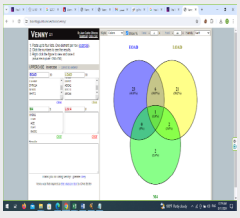
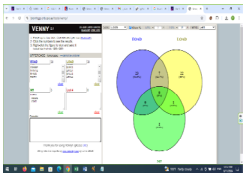
Considering these results, it seems that M2 and M3 are related to EOAD and M4 and M5 are associated with LOAD.

Identifying key genes and their vital pathways can enhance our understanding of diseases. In this study, we identified 10 key genes, and the results are as follows:

APOE gene: The APOE protein is one of the most important apolipoproteins in the brain and an amyloid-associated protein in AD. It is involved in the transport of cholesterol and other lipids. The *ApoE* gene exists in three forms: *ApoE2*, *ApoE3*, and *ApoE4*. The *ApoE4* gene leads to an increased risk of developing AD and is an important factor in the formation and accumulation of amyloid plaques in the brain [30-32].

BIN1 gene: This gene encodes a protein that is essential for the formation of cellular structures. Variants of the *BIN1* gene, in combination with *APOE*, are associated with an increased risk of AD. The *BIN1* gene plays a role in endocytosis and intracellular trafficking, particularly

Table 2. Common genes between each module and EOAD and LOAD

Module Name	EOAD Genes	LOAD Genes	Common EOAD and LOAD Genes	Images
M1	*PSEN2, PLD3, RIN3*	*BIN1, CLU, CD33, PICALM, EPHA1, CD2AP, PTK2B*	*PSEN1, APOE, SORL1, TREM2, ABCA7*	
M2	*CSF1, CSF3, IL1A, CSF2, PRNP, NFKB1*	*A2M*	-	
M3	*SNCA, TYROBP, CSF1R, CSF3R, NCSTN*	*IDE, LDLR, ABCA1*	*APP*	
M4	-	*ACE, BACE1*	*MAPT*	
M5	-	*MTHFR*	-	

in the NFTs and formation of β-amyloid plaques, which are important factors in the pathology of AD [33]. *BIN1* gene: This gene encodes a protein that is essential for the formation of cellular structures.

PICALM gene: PICALM is an amyloid-associated protein that is involved in cellular endocytosis processes. Mutation in this gene is associated with an increased risk of developing AD. GWAS studies have identified the *PICALM* gene as the most important source of genetic susceptibility after *APOE* and *BIN1*. This gene encodes a protein essential for endocytosis, and its role as an AD risk gene has been confirmed across variant populations through independent genetic studies. The progression of tau pathology has also been confirmed in AD models [34]. Interestingly, the predominant expression of *PICALM* in the brain's small blood vessels suggests a unique and potentially crucial role in cerebrovascular function and AD pathogenesis [35].

ABCA7 gene: This gene is an amyloid-associated protein that is responsible for the transport of phospholipids in cells. It has a significant role in early-onset dementia and amyloid deposition. Also, recent GWAS studies show that the *ABCA7* variants are risk factors for LOAD [36].

Table 3. Top ten GO terms of key genes

GO ID	GO Terms	Function
GO:0001540	MF	Amyloid-beta binding
GO:0048156	MF	Tau protein binding
GO:0050750	MF	Low-density lipoprotein particle receptor binding
GO:0071813	MF	Lipoprotein particle binding
GO:0019828	MF	Aspartic-type endopeptidase inhibitor activity
GO:0019899	MF	Enzyme binding
GO:0005543	MF	Phospholipid binding
GO:1902992	BP	Negative regulation of amyloid precursor protein catabolic process
GO:0097242	BP	Amyloid-beta clearance
GO:0051050	BP	Positive regulation of transport
GO:0061024	BP	Membrane organization
GO:1902532	BP	Negative regulation of intracellular signal transduction
GO:0051241	BP	Negative regulation of multicellular organismal process
GO:0097006	BP	Regulation of plasma lipoprotein particle levels
GO:0007611	BP	Learning or memory
GO:0061518	BP	Microglial cell proliferation
GO:0019216	BP	Regulation of lipid metabolic process
GO:0036477	CC	Somatodendritic compartment
GO:0032994	CC	Protein-lipid complex
GO:0009986	CC	Cell surface
GO:0005794	CC	Golgi apparatus
GO:0097418	CC	Neurofibrillary tangle
GO:0071944	CC	Cell periphery
GO:0030054 GO:0005771	CC	Cell junction
GO:0099020	CC	Multivesicular bodymultivesicular body
	CC	Perinuclear ER lumen

CD2AP gene: CD2AP is an amyloid-associated protein that is involved in calcium production, synaptic structure, and brain function. Cell-to-cell interactions (specific phospholipids) and β-amyloid production limit the regulation of endocytosis or the processing of *APP*. Evidence has shown that the protein associated with *CD2AP* is involved in signal transduction and regulation of cellular skeleton molecules. Variations within this gene, known as single nucleotide polymorphisms (SNPs), are related to a higher risk of developing AD [37].

CD33 gene: CD33 is known as a type I transmembrane protein belonging to the immunoglobulin-like lectins that bind to sialic acid, which mediates cell-cell interac-

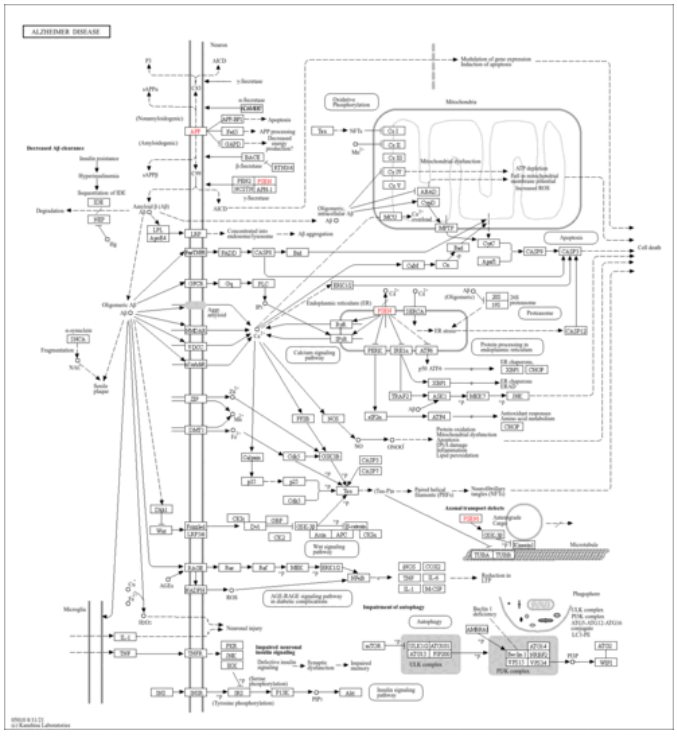


Figure 3. Metabolic pathways of AD in KEGG–map05010

Note: In the metabolic pathways of AD, the *APP, APOE, PS1*, and *PS2* genes, which are shown in red, play a role in the formation of amyloid plaques, oxidative stress, inflammation, calcium disturbances, and other metabolic disorders.

tions and inhibits the normal function of immune cells. This gene is mainly expressed in microglial cells. Increased expression of *CD33* is observed in the brains of patients with AD, and this increase is positively related to amyloid plaque load and disease severity. In addition, *CD33* overexpression disrupts Aβ metabolism and promotes the formation of amyloid plaques in the brain [18].

SORL1 gene: The *SORL1* gene is one of the genes that plays a critical role in developing AD. Recent studies show that loss of *SORL1*, as well as mutations in the *APP* and *PSEN1*/2 genes, are associated with AD [38]. The *SORLA* gene is expressed in neurons of the central nervous system, including the cortex, cerebellum, hippocampus, and spinal cord. The SORLA mechanism acts

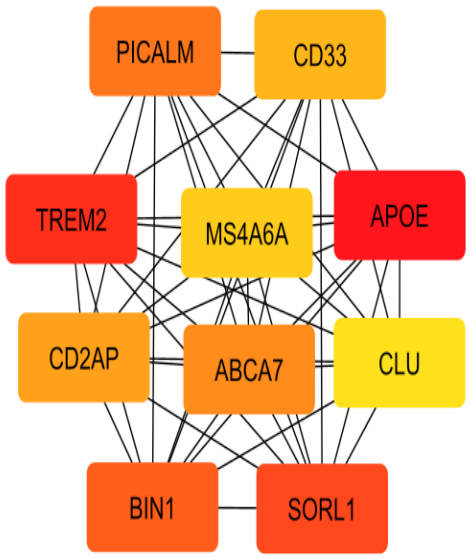


Figure 4. The network of AD key genes

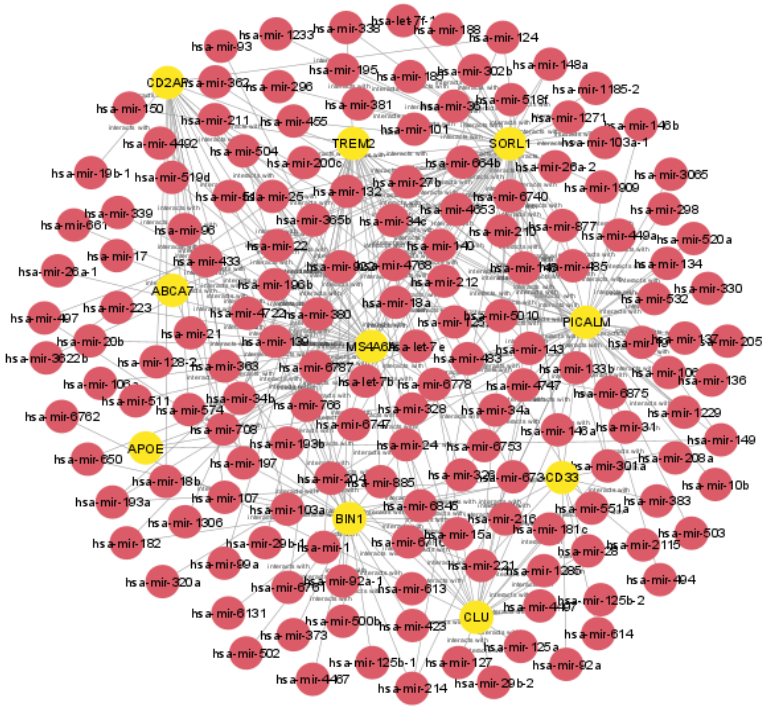


Figure 5. miRNA-key gene interaction network in AD

as a neuronal sorting receptor that regulates amyloidogenic processes in the brain. Recent studies indicate that *SORL1* is considered a biomarker for AD [39].

MS4A6A gene: The *MS4A6A* gene is part of the *MS4A* family, which is involved in the function of brain cells. Variants of this gene, in combination with the *APOE* gene, affect signaling related to AD. Extensive genome-wide studies have identified new loci and functional pathways that effectively reduce AD risk, which are associated with the effects of this gene [40].

TREM2 gene: This gene can increase an individual's risk of developing AD by up to three times. TREM2 is a transmembrane receptor expressed in amyloid cells, and its relationship to AD is linked to immune and inflammatory pathways. *TREM2* plays a critical role in changing the behavior of microglial cells, including their response to amyloid plaques [41].

CLU gene: The *CLU* gene encodes an amyloid-associated protein that protects neurons. The protein encoded by this gene acts as a chaperone and can also be found in cells under stressful conditions. It is involved in several fundamental biological processes, including tumor progression, cell death, and neurological disorders [42, 30].

GO enrichment analysis showed that the most important biological processes associated with these key genes include the negative regulation of the APP catabolic process, amyloid-beta clearance, positive regulation of transport, and membrane organization. A significant function of these genes involves amyloid-beta binding, tau protein binding, and low-density lipoprotein particle receptor binding. Additionally, these proteins are predominantly localized in the somatodendritic compartment, protein-lipid complexes, and the cell surface. The miRNA-key gene interaction network showed that half of the identified miRNAs related to AD influence the disease by controlling the expression of the key genes, indicating the important role these genes play in the development of AD.

According to the underlying diseases related to EOAD, many studies have been conducted. For example, there are several studies investigating the metabolic and cognitive effects of obesity and the *APOE* gene; however, the relationship between *APOE* and obesity in regulating the pathogenesis of AD remains unclear. The study by Bellenguez et al. shows that the *APOE4* gene and obesity increase the risk of developing AD. Also, Zhao et al. showed that *APOE* is a strong genetic risk factor for AD, and obesity is associated with AD [30, 43]. Sparks suggests that hypertension is one of the factors that cause SP in NFTs, and SP is related to AD. Also, *APOE* increases the risk of hypertension and AD [44]. Xu et al. showed

that hypercholesterolemia increases AD pathology by the accumulation of amyloid-β, and conversely, *APOE* is related to hypercholesterolemia [45]. Patel et al. showed that AD and type 2 diabetes are related, and diabetes and *ApoE* increase the risk of AD through neuritic plaques, aggregation of NFTs, and amyloid-β [46].

Recent studies have shown an association between *SORL1* and obesity. Schmidt et al. showed that *SORL1* expression affects obesity and glucose tolerance. This study shows that the relationship between *SORL1* and obesity leads to neurodegeneration and metabolism that cause AD [47]. Yu et al. showed that type 2 diabetes mellitus is related to an increased risk of dementia, including AD. Also, this study reported that the *SORL1* gene is a critical gene for AD [48]. Feng et al. declared that the *SORL1* gene is a candidate gene in lipid metabolic pathways associated with AD [49]. Nisha et al. indicated that SORLA is an underlying cause of AD and is related to metabolic pathologies, like obesity, diabetes, and hypercholesterolemia [50].

Conclusion

The findings of this study showed that bioinformatics tools and systems biology approaches are capable of investigating multi-gene diseases, such as AD. In addition, key genes, vital pathways, and functional modules can aid in the identification of diseases like EOAD, helping to discover biomarkers, drug targets, and related conditions. The results of our study showed that obesity, hypercholesterolemia, diabetes mellitus, hypertension, and cholestasis play a critical role in the occurrence or development of EOAD. Therefore, the treatment and management of these diseases can play an important role in preventing EOAD. However, more experimental studies are necessary to confirm the roles of the identified key genes and underlying diseases in the occurrence of AD.

Ethical Considerations

Compliance with ethical guidelines

There were no ethical considerations to be considered in this research.

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Authors contribution's

Conceptualization: Ali Kazemipour, Maryam Zangi Darestani, and Zahra Ziaastani; Data collection, investigation, formal analysis and writing the original draft: Maryam Zangi Darestani, and Zahra Ziaastani; Review and editing: Ali Kazemipour, Mehdi Abbasnejad, and Behjat Kalantari Khandani.

Conflict of interest

The authors declared no conflict of interest.

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