Published in final edited form as:

JAMA Neurol. 2016 July 01; 73(7): 867–874. doi:10.1001/jamaneurol.2016.0301.

Genomics of Alzheimer Disease:

A Review

Roger N. Rosenberg, MD [Editor, *JAMA Neurology*],

Department of Neurology and Neurotherapeutics, University of Texas Southwestern Medical Center, Dallas

Doris Lambracht-Washington, PhD [Editor, *JAMA Neurology*],

Department of Neurology and Neurotherapeutics, University of Texas Southwestern Medical Center, Dallas

Gang Yu, PhD,

Department of Neuroscience, University of Texas Southwestern Medical Center, Dallas

Weiming Xia, PhD

Geriatric Research, Education and Clinical Center, Bedford Veterans Hospital, Bedford, Massachusetts

Department of Pharmacology and Experimental Therapeutics, Boston University School of Medicine, Boston, Massachusetts

Abstract

IMPORTANCE—To provide a comprehensive review of knowledge of the genomics of Alzheimer disease (AD) and DNA amyloid β 42 (A β 42) vaccination as a potential therapy.

OBSERVATIONS—Genotype-phenotype correlations of AD are presented to provide a comprehensive appreciation of the spectrum of disease causation. Alzheimer disease is caused in part by the overproduction and lack of clearance of A β protein. Oligomer A β , the most toxic species of A β , causes direct injury to neurons, accompanied by enhanced neuroinflammation, astrogliosis and gliosis, and eventually neuronal loss. The strongest genetic evidence supporting this hypothesis derives from mutations in the amyloid precursor protein (*APP*) gene. A detrimental

Corresponding Author: Roger N. Rosenberg, MD, University of Texas Southwestern Medical Center, Department of Neurology and Neurotherapeutics, Dallas, TX 75390 (roger.rosenberg@utsouthwestern.edu).

Author Contributions: Dr Rosenberg had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Each of the authors has provided ideas and concepts and has contributed to the writing of the manuscript.

Study concept and design: Rosenberg, Yu, Xia.

Acquisition, analysis, or interpretation of data: Labracht-Washington.

Drafting of the manuscript: Rosenberg, Xia.

Critical revision of the manuscript for important intellectual content: All authors.

Obtained funding: Xia.

Study supervision: Xia.

Conflict of Interest Disclosures: Dr Rosenberg is director of the Alzheimer's Disease Center at the University of Texas Southwestern Medical Center at Dallas. He received a US patent for "Amyloid β Gene Vaccines" in 2009.

Disclaimer: Dr Rosenberg is the Editor of *JAMA Neurology* but was not involved in the editorial review or the decision to accept the manuscript for publication. The views expressed in this article are those of the authors and do not represent the views of the US Department of Veterans Affairs or the US Government.

APP mutation at the β -secretase cleavage site linked to early-onset AD found in a Swedish pedigree enhances A β production, in contrast to a beneficial mutation 2 residues away in *APP* that reduces A β production and protects against the onset of sporadic AD. A number of common variants associated with late-onset AD have been identified including apolipoprotein E, *BINI*, *ABC7*, *PICALM*, *MS4A4E/MS4A6A*, *CD2Ap*, *CD33*, *EPHA1*, *CLU*, *CRI*, and *SORL1*. One or 2 copies of the apolipoprotein E ϵ 4 allele are a major risk factor for late-onset AD. With DNA A β 42 vaccination, a Th2-type noninflammatory immune response was achieved with a downregulation of A β 42-specific effector (Th1, Th17, and Th2) cell responses at later immunization times. DNA A β 42 vaccination upregulated T regulator cells (CD4+, CD25+, and FoxP3+) and its cytokine interleukin 10, resulting in downregulation of T effectors.

CONCLUSIONS AND RELEVANCE—Mutations in *APP* and *PS-1* and *PS-2* genes that are associated with early-onset, autosomal, dominantly inherited AD, in addition to the at-risk gene polymorphisms responsible for late-onset AD, all indicate a direct and early role of A β in the pathogenesis of AD. A translational result of genomic research has been A β -reducing therapies including DNA A β 42 vaccination as a promising approach to delay or prevent this disease.

Alzheimer disease (AD) is characterized by 2 pathological hallmarks, amyloid β (A β) protein-containing neuritic plaques and hyperphosphorylated tau-containing paired helical filament in neurofibrillary tangles.¹ Amyloid β protein is generated by sequential cleavages of the amyloid precursor protein (APP) by β - and γ -secretases. First, APP is proteolytically processed by β -secretase and generates a 12-kDa C-terminal stub of the *APP* gene (C99); second, C99 is cleaved by γ -secretase to yield 2 major species of A β ending at residue 40 (A β 40) or residue 42 (A β 42).^{2,3} Multiple mutations in the gene encoding *APP* cause early-onset familial AD, and most of them either lead to an increase in A β production or in the ratio of A β 42 to A β 40, thus enhancing the aggregation of A β peptides. Compared with shorter A β peptides, such as A β 40 and A β 38, many studies have documented that the 42-residue A β 42 enhances aggregation propensity,⁴ leading to accelerated formation of small (low-n) A β oligomers.⁵ Mutations in presenilin genes have similar effect in increasing the ratio of A β 42 to A β 40. Cells bearing familial AD mutant genes (*APP* or *PS1/2*) produced higher levels of A β oligomers,⁶ which were observed in plasma and postmortem brains of patients with AD.⁷

Mutation in the tau gene causes frontotemporal dementia with parkinsonism linked to chromosome 17 (*FTDP-17*).⁸ In mice, A β accumulation can drive tau pathology in vivo.⁹ Transgenic mice expressing mutant tau show close association between tau mutation, neurofibrillary tangle formation, and neurodegeneration.^{10,11} Tau protein has been used as a marker for axonal damage, and tau levels in cerebrospinal fluids reflect these changes in the central nervous system. Tau protein levels (total tau, phosphorylated tau, and the ratio) in cerebrospinal fluid have been explored as potential markers for AD.

Translational genomic research has developed potential DNA A β 42 therapeutics for AD. A noninflammatory Th2-mediated immune response with effective levels of anti-A β 42 antibody has been developed in transgenic AD model mice with the DNA A β 42 vaccine and is included as a direct extension of advances in AD genomics.

Amyloid Cascade Hypothesis

Mechanisms for AD pathogenesis have been extensively explored, and the major hypothesis supported by genetic and neuropathological evidences is the amyloid cascade hypothesis.¹² It suggests that AD is caused in part by the overproduction and lack of clearance of A β . The most toxic species of A β , such as oligomer A β , cause direct injury to neurons, accompanied by enhanced neuroinflammation, astrogliosis and gliosis, and eventually neuronal loss. The strongest genetic evidence supporting this hypothesis derives from mutations in the *APP* gene. A detrimental *APP* mutation at the β -secretase cleavage site (the *APP*K670/671NL mutation), linked to early-onset AD found in a Swedish pedigree, enhances A β production,¹³ in contrast to a beneficial mutation 2 residues away (Icelandic mutation A673T) in *APP* that reduces A β production and protects against the onset of sporadic AD.¹⁴ The Swedish mutant *APP* is a better substrate for recognition and cleavage by the β -secretase, while the Icelandic mutant *APP* is a much poorer substrate for the β -secretase,¹⁴ and the Icelandic mutation greatly attenuates *APP*- β -secretase interaction.¹⁵

The amyloid cascade hypothesis is also supported by additional genetic studies with the other 2 genes associated with early-onset familial AD, the presenilin 1 and 2 genes (*PS1* and *PS2*). To our knowledge, only 3 genes have been identified to associate with familial AD, and all of them encode the key components of the A β synthesis pathway. The *APP* gene encodes the precursor of A β and is the substrate of the γ -secretase, and *PS1* and *PS2* genes encode the proteins that carry the active site of the γ -secretase.¹⁶ γ -Secretase is composed of *PS1* (or *PS2*), presenilin enhancer-2, anterior pharynx defective-1, and nicastrin.^{17–19} *PS1* itself has proteolytic activity,^{20,21} which is activated by presenilin enhancer-2.^{20,22} Nicastrin is the substrate receptor for *APP*.²³ A number of mutations in *APP*¹³ or *PS1*/^{26,24,25} that link to familial AD affect the γ -secretase cleavage of *APP* and shift A β production from A β 40 to the more toxic and aggregation-prone A β 42. Mutations that provide more copies of *APP* genelike duplications²⁶ and Down syndrome²⁷ generate more A β in the brains of those patients. Almost all pathogenic mutations found in the *APP* gene lead to increased generation of A β or the A β 42 to A β 40 ratio because A β 42 is more toxic than A β 40. The Swedish mutation at the β -secretase cleavage site allows *APP* to be more accessible to the enzyme and generates more A β .^{13,28} The Arctic mutation of *APP* (*APP*E693G) increases the aggregation propensity of A β and fibril formation.²⁹ Pathogenic V717F, V717I, and V717L mutations all increase the A β 42/A β 40 ratio.^{30–32}

Major AD Risk Genes and Their Involvement in A β Metabolism

A number of common variants associated with late-onset AD have been identified, including apolipoprotein E (*APOE*), *BIN1*, *ABCA7*, *PICALM*, *MS4A4E/MS4A6A*, *CD2AP*, *CD33*, *EPHA1*, *CLU*, *CRI*, and *SORL1*.^{33–36} One or 2 copies of the *APOE* ϵ 4 allele is a major risk factor for late-onset AD (LOAD).^{37,38} The *APOE* gene has 3 major isoforms, *APOE* ϵ 2, ϵ 3, and ϵ 4. Brains of patients with sporadic AD carrying the *APOE* ϵ 4 allele were found to have increased density of A β deposits, limited capability to clear A β , and enhanced neuroinflammation.³⁹ Binding of *APOE* loaded with A β to cell surface receptors, such as the low-density lipoprotein receptor-related protein-1 (*LRP1*), is one of the mechanisms for A β clearance.^{40,41}

Clusterin (encoded by *CLU*) directly interacts with soluble A β and forms a complex to cross the blood-brain barrier.⁴² This function is similar to *APOE*, which acts as a molecular chaperone for A β . Both clusterin and *APOE* directly influence A β during its aggregation and deposition. The presence of *APOE*/clusterin changes A β conformation and its toxicity.^{43,44} Therefore, *APOE* and clusterin may regulate the conversion of soluble A β into insoluble forms such as oligomers, thus suppressing A β toxicity and deposition. Because *APOE* and clusterin are involved in a complex formation with A β to cross the blood-brain barrier, both proteins regulate A β clearance.⁴⁵ Alternative pathways for A β clearance are mediated by microglia. When extracellular A β aggregates, such as A β , are engulfed by microglia, inflammasomes (such as NOD-like receptor family pyrin domain-containing3 [*NLRP3*]) are triggered, which activate caspases and promote interleukin 1 β release.^{46,47} These aggregates are known to activate innate immune responses via complement pathways, including complement receptor 1 (*CR1*), which has been associated with LOAD.⁴⁸ Those A β isoforms that bind to scavenger receptors expressed on microglia, such as CD36⁴⁹ and Scaral,⁵⁰ enter microglia and activate inflammation. Systems analysis of hundreds of brains with AD reveals changes in networks related to immunologic molecules and microglial cells including microglial protein TYROBP, which binds the microglial receptor TREM2 (triggering receptors expressed on myeloid cells 2) and may regulate *CD33* function.⁵¹ Genetic mutations found in TREM2 triple a person's risk for AD^{52,53} and increased expression of *CD33*, which functions to suppress AP uptake and clearance.^{54,55}

Another transporter protein associated with LOAD is ATP-binding cassette transporter (*ABCA7*), which belongs to the ATP-binding cassette transporter superfamily that transports many substrates across cell membranes. The *ABCA7* gene is involved in the efflux of lipids from cells to lipoprotein particles. In an *APP* transgenic mouse model (J20) that is deficient in *ABCA7*, levels of *APOE* were not changed.⁵⁶ However, both soluble and insoluble A β levels and thioflavine-S-positive plaques were increased in the *ABCA7*-deficient mice.^{56,57} In cultured cells, enhanced endocytosis of *APP* was observed in *ABCA7* knockout cells, leading to increased A β production.⁵⁷

Phosphatidylinositol-binding clathrin assembly protein (*PICALM*) plays a critical role in clathrin-mediated endocytosis and protein/lipid internalization.³³ When full-length APP at the cell surface is internalized by clathrin-mediated endocytosis, β - and γ -secretase cleavages occur, and a significant amount of A β is generated.⁵⁸ When endocytosis is promoted by increased synaptic activity, more *APP* is retrieved from the cell surface to endosomes, resulting in an increase of A β generation and secretion.⁵⁹ Involvement of *PICALM* with clathrin-mediated endocytosis directly affects *APP* processing and A β synthesis.

Bridging integrator 1 (*BINI*) is highly expressed in brain, and all *BINI* isoforms interact with clusterin and are involved in surface protein endocytosis. While it is possible that *BINI* influences APP endocytosis and A β production, its functional interaction with tau was demonstrated in a drosophila model where knockdown of the *BINI* ortholog *Amph* could suppress the rough eye phenotype caused by overexpression of human tau.⁶⁰

Genetic variants in the sortilin-related receptor *SORL1* gene are associated with LOAD.⁶¹ Reduction of *SORL1* expression was observed in vulnerable regions in brains with AD.^{62,63} In cultured cells, *SORL1* interacts with APP and both proteins colocalize in endosomal and Golgi compartments.^{61,63} They also interact with vacuolar protein sorting-associated protein 35 (*VPS35*); *VPS35* promotes cargo selection in the retromer through *SORL1*. When expression of *SORL1* is increased, *SORL1* regulates differential sorting of APP into the retromer recycling pathway, thus reducing A β production; when expression of *SORL1* is reduced, APP trafficking is directed toward endosome-lysosome compartments that facilitate A β production.⁶¹ Deficient *SORL1* expression in knockout mice resulted in increased levels of A β in animal brains.^{62,63}

The *PCDH11X* gene is a cell surface receptor molecule belonging to the protocadherin gene, which is a subfamily of the cadherin superfamily. Like other cadherins, it mediates cell-cell adhesion and is cleaved by γ -secretase.⁶⁴ Cadherins, such as E- and N-cadherins, form the complex with *PS1*/ γ -secretase⁶⁵ and regulate cell-cell interaction after the cleavage by γ -secretase, this process is inhibited by familial AD mutations in *PS1*.⁶⁶ Therefore, *PCDH11X* is believed to play a role in cell signaling that is critical in the development of the central nervous system.⁶⁷ Genetic variation in *PCDH11X* is associated with LOAD.⁶⁸

The *MS4A4E/MS4A6A* genes belong to the *MS4A* gene cluster on chromosome 11. The proteins encoded by these similar genes have a common transmembrane domain and likely are cell surface proteins. Their involvement in A β production is not known. The *EPHA1* gene is a member of the ephrin receptor family. Both ephrins and Eph receptors play important roles in cell and axon guidance, and another member of this family, *EphA4*, is cleaved by Y-secretase induced by synaptic activity.⁶⁹ The CD2-associated protein (*CD2AP*) is a scaffold/adaptor protein and mediates receptor-regulated endocytosis.⁷⁰ In cultured cells, reducing *CD2AP* expression results in decreased levels of A β and a lower A β 42/A β 40 ratio. Knockout of *CD2AP* in transgenic mice overexpressing *PS1* and *APP* (*PS1APP* mice) decreased the A β 42/A β 40 ratio in the brain, expressing 1 copy of *CD2APP* in *PS1APP* mice does not affect A β deposition.⁷¹

Imbalanced A β homeostasis, ie, increased A β production and/or decreased A β clearance, is an upstream event of neurodegeneration that is directly affected by AD risk gene products (Figure 1). The *SORL1*, *PICALM*, and *CD2AP* genes are involved in endocytic internalization of cell surface APP for β - and γ -secretase cleavages to generate A β , while *APOE*, *CLU*, *TREM2*, *ABCA7*, *PICALM*, *CD33*, *CD2AP*, and *CR1* are all involved in A β clearance through multiple mechanisms. These risk genes, in addition to familial AD-linked mutant genes *APP*, *PS1*, and *PS2*, all point to the direct and early role of A β in the pathogenesis of AD. Amyloid- β -reducing therapies, including DNA A β 42 vaccination, have a therapeutic potential to delay or prevent the disease (Table).

DNA and Peptide A β 42 Vaccination: Translational Therapy From Genomics

The amyloid cascade hypothesis postulates that A β deposition in the brain is a primary event necessary in the multifactoral pathogenesis of AD.^{12,89,90} It has been demonstrated that A β deposition precedes AD symptoms by at least 20 years.⁹¹ Therapeutic approaches using

active and passive immunizations against A β have a high possibility to be effective in removing amyloid from the brain and thereby delaying or preventing downstream pathologies. Since 2000, a number of clinical trials for AD immunotherapy have started, have failed, and are continuing.^{92–99} Amyloid β protein at residue 42 peptide vaccination with adjuvant in the clinical trial AN1792^{92,93,99} resulted in an autoimmune meningoencephalitis with T cells infiltrating the brain of affected vaccinated patients, and the study was stopped. A clinical trial^{94,100} with the monoclonal anti-amyloid antibody bapineuzumab reduced the level of amyloid accumulation as seen by amyloid imaging but had no clinical benefit in patients expressing clinical signs of AD.

New studies started between 2013 and 2015 focus on therapy in patient cohorts before the onset of clinical symptoms of AD.¹⁰¹ These include the Dominantly Inherited Alzheimer Network study,¹⁰² the Alzheimer Prevention Initiative, and the Treatment of Asymptomatic Alzheimer.¹⁰³ A major achievement will occur if passive A β immunotherapy for AD can be shown to have clinical benefit. If and when these new prevention studies using passive immunizations with anti-A β antibodies provide clinical benefit by delaying or preventing AD, active vaccination, potentially with a DNA A β 42 vaccine, will increase in interest because it induces an effective and noninflammatory immune response and is applied more efficiently and economically to large populations.

A translational result of genomic research has been the development of immunization, with a full-length DNA A β 42 plasmid in mice generating a polyclonal multivalent vaccine.¹⁰⁴ Studies in out-breed animals, such as rabbits and dogs, have shown that the immune response to fibrillar A β is diverse and reflects the polymorphic structure of the antigen itself. These studies also lead to the conclusion that single therapeutic monoclonal antibodies with 1 specific epitope may not be able to target all the different aggregates to have an effect on diminishing disease progression.^{105,106}

We have developed an A β 42 plasmid encoding 3 copies of the full-length A β 42 sequence. Transcription of A β 42 is initiated by the transcription factor Gal4, which is encoded by a second plasmid that is delivered simultaneously by cotransfection (Figure 2). This approach had been shown to increase the expression of the A β 42 sequence without causing A β 42 cytotoxicity in the transfected cells.¹⁰⁷

Immunization with A β 1–42 is strongly influenced by the fact that this is a self-antigen, and tolerance against self-antigens has to be broken. In wild-type mice, human A β 42 is a foreign antigen owing to 3 amino acid alterations in the N-terminal segment of the peptide, and wild-type mice produce high levels of anti-A β antibodies following immunization. Higher levels of antibody produced in the wild-type mouse are owing to the fact that the DNA in the plasmid coding for A β 42 peptide is of human type. This explains the differences in the comparison of the antibody responses in wild-type mice and human *APP* transgenic mice. As in the *APP* transgenic mouse receiving the DNA A β 42 plasmid, the expressed A β 42 peptide in the brain and in the vaccinated skin cells is both of human type. The eFigure in the Supplement shows the analysis of double transgenic and wild-type mice that have been immunized 9 times with the DNA A β 42 vaccine via gene gun administration. The wild-type mice in this group reached antibody levels with a mean (SD) of 63 (18.16) μ g/mL of plasma

and the transgenic mice had antibody levels with a mean (SD) of 13.51 (9.18) $\mu\text{g/mL}$ of plasma ($P < .001$).

In AD mouse models, we have demonstrated up to a 50% reduction in brain amyloid following full-length DNA A β 42 vaccination.^{108–110} In Figure 3, the results shown were from 2 double transgenic mouse cohorts, from which amyloid levels in the brain had been analyzed 2 weeks following the last immunization (Figure 3A) or 4 months after the last immunization (Figure 3B). Group A showed an amyloid reduction of 65% and group B showed a reduction of A β 1–42 brain levels of 25%. In both groups, the amyloid reduction was substantial in comparison with the control DNA (Luc)-immunized mice (Mann-Whitney test: $P = .006$ and $P = .007$, respectively). The marked difference between these 2 groups is explained by the time differences between final immunizations and brain level analyses (14 days and 4 months following the last immunization; 12 and 16 months of age) and the marked differences in total A β 42 levels in the brain owing to the 4-month age difference between groups A and B. Concentration of A β 42 in the brain increased from 10 $\mu\text{g/g}$ wet tissue to 50 $\mu\text{g/g}$ in the Luc control mice owing to increased AD pathology with the age progression in these mice.

With DNA A β 42 vaccination, a Th2-type noninflammatory immune response was achieved, with a downregulation of A β 42-specific T effector (Th1, Th17, and Th2) cell responses at later immunization times.^{111–116} On the other hand, DNA A β 42 vaccination upregulated T regulator cells (CD4+, CD25+, and FoxP3+) and the cytokine interleukin 10, resulting in downregulation of T effectors. In distinction, A β 42 peptide vaccination induced the opposite effect, with upregulation of T effectors (CD4+, CD25+, and FoxP3-) and no effect on T regulator cells. Thus, the immune response following DNA immunization differs quantitatively and qualitatively from the immune response elicited by A β 42 peptide immunization. DNA A β 42 immunization with downregulation of T effectors (Th1, Th17, and Th2 cells) would significantly reduce the opportunity for cytotoxic T cells from migrating and transiting through brain, which was evident in the A β 42 peptide vaccine clinical trial AN1792, resulting in meningoencephalitis.^{92,115}

Immunizations of 16 New Zealand white rabbits produced after 5 vaccinations anti-A β 42 antibodies at mean levels of 250 $\mu\text{g/mL}$ of serum. The isotyping of the serum antibody showed IgG, IgM, and IgA anti-A β 42 antibodies. In autopsies of 9 rabbits, there was no evidence of meningoencephalitis. Immunizations of 6 rhesus monkeys produced after 4 vaccinations of anti-A β 42 antibodies at mean levels of 112.6 $\mu\text{g/mL}$ of serum and they have remained healthy with no adverse effects (D.L.-W. and R.N.R., preliminary unpublished observations).

Conclusions

Research into the genetics and genomics of AD has made major strides in the past decade. The *PS1*, *PS2*, and *APP* gene mutations have been documented to result in aggressive, early-onset autosomal dominant disease. Polymorphisms in multiple genes have been shown to increase the risk for the disease. A polymorphism in the *APP* gene, preventing binding of β -secretase to *APP* in *APOE4* persons and impairing production of A β 42 peptide being

processed from the parent *APP* molecule, prevents the clinical expression of AD and is in strong support of the amyloid cascade hypothesis. Amyloid imaging has provided the accurate means to identify asymptomatic at-risk persons accumulating amyloid to test anti-amyloid therapies.¹¹⁷ Active vaccination with a DNA A β 42 vaccine offers a potential effective and noninflammatory approach and is one of several active vaccination approaches after demonstration of clinical benefit from passive anti-amyloid antibody therapy.¹¹⁸ The magnitude of defining the genomic and epigenomic factors involved in the pathogenesis of AD and new therapeutic strategies that will occur as a result are large in number and complexity. The Human Alzheimer Disease Project, an undertaking at the level of commitment and funding equivalent to the Human Genome Project, is necessary and required to stem the crescendo of anticipated increases in the prevalence of this disease in the foreseeable future.¹¹⁹

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Funding/Support: Dr Rosenberg is principal investigator of the National Institutes of Health/National Institute on Aging grant P30AG12300–16. He receives funding from the Rudman Partnership, AWARE Group in Dallas, Freiburger Family Fund, McCune/Losinger Fund, Presbyterian Foundation, North Foundation, the Darryl K Royal Foundation, and the Zale Foundation. Dr Xia received funding from award I21BX002215 from the Biomedical Laboratory Research and Development Service of the Veterans Affairs Office of Research and Development.

Role of the Funder/Sponsor: The funding sources had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

REFERENCES

1. Selkoe DJ; American College of Physicians; American Physiological Society. Alzheimer disease: mechanistic understanding predicts novel therapies. *Ann intern Med.* 2004;140(8):627–638. [PubMed: 15096334]
2. Haass C, Schlossmacher MG, Hung AY, et al. Amyloid beta-peptide is produced by cultured cells during normal metabolism. *Nature.* 1992;359(6393):322–325. [PubMed: 1383826]
3. Shoji M, Golde TE, Ghiso J, et al. Production of the Alzheimer amyloid beta protein by normal proteolytic processing. *Science.* 1992;258(5079):126–129. [PubMed: 1439760]
4. Jarrett JT, Berger EP, Lansbury PT Jr. The carboxy terminus of the beta amyloid protein is critical for the seeding of amyloid formation: implications for the pathogenesis of Alzheimer's disease. *Biochemistry.* 1993;32(18):4693–4697. [PubMed: 8490014]
5. Walsh DM, Selkoe DJ. Deciphering the molecular basis of memory failure in Alzheimer's disease. *Neuron.* 2004;44(1):181–193. [PubMed: 15450169]
6. Xia W, Zhang J, Kholodenko D, et al. Enhanced production and oligomerization of the 42-residue amyloid beta-protein by Chinese hamster ovary cells stably expressing mutant presenilins. *J Biol Chem.* 1997;272(12):7977–7982. [PubMed: 9065468]
7. Xia W, Yang T, Shankar G, et al. A specific enzyme-linked immunosorbent assay for measuring beta-amyloid protein oligomers in human plasma and brain tissue of patients with Alzheimer disease. *Arch Neurol.* 2009;66(2):190–199. [PubMed: 19204155]
8. Hutton M. Molecular genetics of chromosome 17 tauopathies. *Ann NY Acad Sci.* 2000;920:63–73. [PubMed: 11193178]

9. Lee VM, Trojanowski JQ. Progress from Alzheimer's tangles to pathological tau points towards more effective therapies now. *J Alzheimers Dis.* 2006;9(3)(suppl):257–262. [PubMed: 16914864]
10. Lewis J, McGowan E, Rockwood J, et al. Neurofibrillary tangles, amyotrophy, and progressive motor disturbance in mice expressing mutant (P301L) tau protein. *Nat Genet.* 2000;25(4):402–405. [PubMed: 10932182]
11. Götz J, Chen F, van Dorpe J, Nitsch RM. Formation of neurofibrillary tangles in P301Ltau transgenic mice induced by Abeta 42 fibrils. *Science.* 2001;293(5534):1491–1495. [PubMed: 11520988]
12. Hardy J, Selkoe DJ. The amyloid hypothesis of Alzheimer's disease: progress and problems on the road to therapeutics. *Science.* 2002;297(5580):353–356. [PubMed: 12130773]
13. Citron M, Oltersdorf T, Haass C, et al. Mutation of the beta-amyloid precursor protein in familial Alzheimer's disease increases beta-protein production. *Nature.* 1992;360(6405):672–674. [PubMed: 1465129]
14. Jonsson T, Atwal JK, Steinberg S, et al. A mutation in APP protects against Alzheimer's disease and age-related cognitive decline. *Nature.* 2012;488(7409):96–99. [PubMed: 22801501]
15. Das U, Wang L, Ganguly A, et al. Visualizing APP and BACE-1 approximation in neurons yields insight into the amyloidogenic pathway. *Nat Neurosci.* 2016;19(1):55–64. [PubMed: 26642089]
16. Wolfe MS, Xia W, Ostaszewski BL, Diehl TS, Kimberly WT, Selkoe DJ. Two transmembrane aspartates in presenilin-1 required for presenilin endoproteolysis and gamma-secretase activity. *Nature.* 1999;398(6727):513–517. [PubMed: 10206644]
17. Sherrington R, Rogaev EI, Liang Y, et al. Cloning of a gene bearing missense mutations in early-onset familial Alzheimer's disease. *Nature.* 1995;375(6534):754–760. [PubMed: 7596406]
18. Francis R, McGrath G, Zhang J, et al. aph-1 And pen-2 are required for Notch pathway signaling, gamma-secretase cleavage of betaAPP, and presenilin protein accumulation. *Dev Cell.* 2002;3(1):85–97. [PubMed: 12110170]
19. Yu G, Nishimura M, Arawaka S, et al. Nicastrin modulates presenilin-mediated notch/glp-1 signal transduction and betaAPP processing. *Nature.* 2000;407(6800):48–54. [PubMed: 10993067]
20. Ahn K, Shelton CC, Tian Y, et al. Activation and intrinsic gamma-secretase activity of presenilin 1. *Proc Natl Acad Sci US A.* 2010;107(50):21435–21440.
21. Lessard CB, Wagner SL, Koo EH. And four equals one: presenilin takes the gamma-secretase role by itself. *Proc Natl Acad Sci USA.* 2010;107(50):21236–21237.
22. Li YM, Xu M, Lai MT, et al. Photoactivated gamma-secretase inhibitors directed to the active site covalently label presenilin 1. *Nature.* 2000;405(6787):689–694. [PubMed: 10864326]
23. Shah S, Lee SF, Tabuchi K, et al. Nicastrin functions as a gamma-secretase-substrate receptor. *Cell.* 2005;122(3):435–44. [PubMed: 16096062]
24. Citron M, Westaway D, Xia W, et al. Mutant presenilins of Alzheimer's disease increase production of 42-residue amyloid beta-protein in both transfected cells and transgenic mice. *Nat Med.* 1997;3(1):67–72. [PubMed: 8986743]
25. Scheuner D, Eckman C, Jensen M, et al. Secreted amyloid beta-protein similar to that in the senile plaques of Alzheimer's disease is increased in vivo by the presenilin 1 and 2 and APP mutations linked to familial Alzheimer's disease. *Nat Med.* 1996;2(8):864–870. [PubMed: 8705854]
26. Rovelet-Lecrux A, Hannequin D, Raux G, et al. APP locus duplication causes autosomal dominant early-onset Alzheimer disease with cerebral amyloid angiopathy. *Nat Genet.* 2006;38(1):24–26. [PubMed: 16369530]
27. Wisniewski KE, Wisniewski HM, Wen GY. Occurrence of neuropathological changes and dementia of Alzheimer's disease in Down's syndrome. *Ann Neurol.* 1985;17(3):278–282. [PubMed: 3158266]
28. Haass C, Lemere CA, Capell A, et al. The Swedish mutation causes early-onset Alzheimer's disease by beta-secretase cleavage within the secretory pathway. *Nat Med.* 1995;1(12):1291–1296. [PubMed: 7489411]
29. Nilsberth C, Westlind-Danielsson A, Eckman CB, et al. The "Arctic" APP mutation (E693G) causes Alzheimer's disease by enhanced Abeta protofibril formation. *Nat Neurosci.* 2001;4(9):887–893. [PubMed: 11528419]

30. Goate A, Chartier-Harlin M-C, Mullan M, et al. Segregation of a missense mutation in the amyloid precursor protein gene with familial Alzheimer's disease. *Nature*. 1991;349(6311):704–706. [PubMed: 1671712]
31. Murrell JR, Hake AM, Quaid KA, Farlow MR, Ghetti B. Early-onset Alzheimer disease caused by a new mutation (V717L) in the amyloid precursor protein gene. *Arch Neurol*. 2000;57(6):885–887 [PubMed: 10867787]
32. Murrell J, Farlow M, Ghetti B, Benson MD. A mutation in the amyloid precursor protein associated with hereditary Alzheimer's disease. *Science*. 1991;254(5028):97–99. [PubMed: 1925564]
33. Harold D, Abraham R, Hollingworth P, et al. Genome-wide association study identifies variants at CLU and PICALM associated with Alzheimer's disease [published correction appears in *Nat Genet* 2013;45(6):712]. *Nat Genet*. 2009;41(10):1088–1093. [PubMed: 19734902]
34. Naj AC, Jun G, Beecham GW, et al. Common variants at MS4A4/MS4A6E, CD2AP, CD33 and EPHA1 are associated with late-onset Alzheimer's disease. *Nat Genet*. 2011;43(5):436–441. [PubMed: 21460841]
35. Hollingworth P, Harold D, Sims R, et al.; Alzheimer's Disease Neuroimaging Initiative; CHARGE consortium; EADI1 consortium. Common variants at ABCA7, MS4A6A/MS4A4E, EPHA1, CD33, and CD2AP are associated with Alzheimer's disease. *Nat Genet*. 2011;43(5):429–435. [PubMed: 21460840]
36. Lambert JC, Heath S, Even G, et al.; European Alzheimer's Disease Initiative Investigators. Genome-wide association study identifies variants at CLU and CR1 associated with Alzheimer's disease. *Nat Genet*. 2009;41(10):1094–1099. [PubMed: 19734903]
37. Strittmatter WJ, Saunders AM, Schmechel D, et al. Apolipoprotein E: high-avidity binding to beta-amyloid and increased frequency of type 4 allele in late-onset familial Alzheimer disease. *Proc Natl Acad Sci USA*. 1993;90(5):1977–1981. [PubMed: 8446617]
38. Rebeck GW, Reiter JS, Strickland DK, Hyman BT. Apolipoprotein E in sporadic Alzheimer's disease: allelic variation and receptor interactions. *Neuron*. 1993;11(4):575–580. [PubMed: 8398148]
39. Castellano JM, Kim J, Stewart FR, et al. Human apoE isoforms differentially regulate brain amyloid- β peptide clearance. *Sci Transl Med*. 2011;3(89):89ra57.
40. Sagare A, Deane R, Bell RD, et al. Clearance of amyloid-beta by circulating lipoprotein receptors. *Nat Med*. 2007;13(9):1029–1031. [PubMed: 17694066]
41. Deane R, Wu Z, Sagare A, et al. LRP/amyloid beta-peptide interaction mediates differential brain efflux of A β isoforms. *Neuron*. 2004;43(3):333–344. [PubMed: 15294142]
42. Zlokovic BV, Martel CL, Matsubara E, et al. Glycoprotein 330/megalin: probable role in receptor-mediated transport of apolipoprotein J alone and in a complex with Alzheimer disease amyloid beta at the blood-brain and blood-cerebrospinal fluid barriers. *Proc Natl Acad Sci USA*. 1996;93(9):4229–4234. [PubMed: 8633046]
43. Ma J, Yee A, Brewer HB Jr, Das S, Potter H. Amyloid-associated proteins alpha 1-antichymotrypsin and apolipoprotein E promote assembly of Alzheimer beta-protein into filaments. *Nature*. 1994;372(6501):92–94. [PubMed: 7969426]
44. DeMattos RB, O'dell MA, Parsadanian M, et al. Clusterin promotes amyloid plaque formation and is critical for neuritic toxicity in a mouse model of Alzheimer's disease. *Proc Natl Acad Sci USA*. 2002; 99(16):10843–10848.
45. Bell RD, Sagare AP, Friedman AE, et al. Transport pathways for clearance of human Alzheimer's amyloid beta-peptide and apolipoproteins E and J in the mouse central nervous system. *J Cereb Blood Flow Metab*. 2007;27(5):909–918. [PubMed: 17077814]
46. Halle A, Hornung V, Petzold GC, et al. The NALP3 inflammasome is involved in the innate immune response to amyloid-beta. *Nat Immunol*. 2008;9(8):857–865. [PubMed: 18604209]
47. Heneka MT, Kummer MP, Stutz A, et al. NLRP3 is activated in Alzheimer's disease and contributes to pathology in APP/PS1 mice. *Nature*. 2013;493(7434):674–678. [PubMed: 23254930]

48. Jun G, Naj AC, Beecham GW, et al.; Alzheimer's Disease Genetics Consortium. Meta-analysis confirms CR1, CLU, and PICALM as alzheimer disease risk loci and reveals interactions with APOE genotypes. *Arch Neurol*. 2010;67(12):1473–1484. [PubMed: 20697030]
49. Sheedy FJ, Grebe A, Rayner KJ, et al. CD36 coordinates NLRP3 inflammasome activation by facilitating intracellular nucleation of soluble ligands into particulate ligands in sterile inflammation. *Nat Immunol*. 2013;14(8):812–820. [PubMed: 23812099]
50. Frenkel D, Wilkinson K, Zhao L, et al. Scara1 deficiency impairs clearance of soluble amyloid- β by mononuclear phagocytes and accelerates Alzheimer's-like disease progression. *Nat Commun*. 2013;4(2030):2030. [PubMed: 23799536]
51. Zhang B, Gaiteri C, Bodea LG, et al. Integrated systems approach identifies genetic nodes and networks in late-onset Alzheimer's disease. *Cell*. 2013;153(3):707–720. [PubMed: 23622250]
52. Guerreiro R, Wojtas A, Bras J, et al.; Alzheimer Genetic Analysis Group. TREM2 variants in Alzheimer's disease. *N Engl J Med*. 2013;368(2):117–12. [PubMed: 23150934]
53. Jonsson T, Stefansson H, Steinberg S, et al. Variant of TREM2 associated with the risk of Alzheimer's disease. *N Engl J Med*. 2013;368(2):107–116. [PubMed: 23150908]
54. Griciuc A, Serrano-Pozo A, Parrado AR, et al. Alzheimer's disease risk gene CD33 inhibits microglial uptake of amyloid beta. *Neuron*. 2013;78(4):631–643. [PubMed: 23623698]
55. Bradshaw EM, Chibnik LB, Keenan BT, et al.; Alzheimer Disease Neuroimaging Initiative. CD33 Alzheimer's disease locus: altered monocyte function and amyloid biology. *Nat Neurosci*. 2013;16(7):848–850. [PubMed: 23708142]
56. Kim WS, Li H, Ruberu K, et al. Deletion of Abca7 increases cerebral amyloid- β accumulation in the J20 mouse model of Alzheimer's disease. *J Neurosci*. 2013;33(10):4387–4394. [PubMed: 23467355]
57. Satoh K, Abe-Dohmae S, Yokoyama S, St George-Hyslop P, Fraser PE. ATP-binding cassette transporter A7 (ABCA7) loss of function alters Alzheimer amyloid processing. *J Biol Chem*. 2015;290(40):24152–24165. [PubMed: 26260791]
58. Perez RG, Squazzo SL, Koo EH. Enhanced release of amyloid beta-protein from codon 670/671 "Swedish" mutant beta-amyloid precursor protein occurs in both secretory and endocytic pathways. *J Biol Chem*. 1996;271(15):9100–9107 [PubMed: 8621560]
59. Cirrito JR, Kang JE, Lee J, et al. Endocytosis is required for synaptic activity-dependent release of amyloid-beta in vivo. *Neuron*. 2008;58(1):42–51. [PubMed: 18400162]
60. Chapuis J, Hansmannel F, Gistelink M, et al.; GERAD consortium. Increased expression of BIN1 mediates Alzheimer genetic risk by modulating tau pathology. *Mol Psychiatry*. 2013;18(11):1225–1234. [PubMed: 23399914]
61. Rogaeva E, Meng Y, Lee JH, et al. The neuronal sortilin-related receptor SORL1 is genetically associated with Alzheimer disease. *Nat Genet*. 2007;39(2):168–17. [PubMed: 17220890]
62. Offe K, Dodson SE, Shoemaker JT, et al. The lipoprotein receptor LR11 regulates amyloid beta production and amyloid precursor protein traffic in endosomal compartments. *J Neurosci*. 2006;26(5):1596–1603. [PubMed: 16452683]
63. Andersen OM, Reiche J, Schmidt V, et al. Neuronal sorting protein-related receptor sorLA/LR11 regulates processing of the amyloid precursor protein. *Proc Natl Acad Sci USA*. 2005;102(38):13461–13466. [PubMed: 16174740]
64. Haas IG, Frank M, Véron N, Kemler R. Presenilin-dependent processing and nuclear function of gamma-protocadherins. *J Biol Chem*. 2005;280(10):9313–9319. [PubMed: 15611067]
65. Georgakopoulos A, Marambaud P, Efthimiopoulos S, et al. Presenilin-1 forms complexes with the cadherin/catenin cell-cell adhesion system and is recruited to intercellular and synaptic contacts. *Mol Cell*. 1999;4(6):893–902. [PubMed: 10635315]
66. Marambaud P, Wen PH, Dutt A, et al. A CBP binding transcriptional repressor produced by the PS1/epsilon-cleavage of N-cadherin is inhibited by PS1 FAD mutations. *Cell*. 2003;114(5):635–645. [PubMed: 13678586]
67. Blanco P, Sargent CA, Boucher CA, Mitchell M, Affara NA. Conservation of PCDHX in mammals; expression of human X/Y genes predominantly in brain. *Mamm Genome*. 2000;11(10):906–914. [PubMed: 11003707]

68. Carrasquillo MM, Zou F, Pankratz VS, et al. Genetic variation in PCDH11X is associated with susceptibility to late-onset Alzheimer's disease. *Nat Genet.* 2009;41(2):192–198. [PubMed: 19136949]
69. Inoue E, Deguchi-Tawarada M, Togawa A, et al. Synaptic activity prompts gamma-secretase-mediated cleavage of EphA4 and dendritic spine formation. *J Cel Biol.* 2009;185(3):551–564.
70. Lynch DK, Winata SC, Lyons RJ, et al. A Cortactin-CD2-associated protein (CD2AP) complex provides a novel link between epidermal growth factor receptor endocytosis and the actin cytoskeleton. *J Biol Chem.* 2003;278(24):21805–21813. [PubMed: 12672817]
71. Liao F, Jiang H, Srivatsan S, et al. Effects of CD2-associated protein deficiency on amyloid- β in neuroblastoma cells and in an APP transgenic mouse model. *Mol Neurodegener.* 2015;10:12. [PubMed: 25887956]
72. Mullan M, Crawford F, Axelman K, et al. A pathogenic mutation for probable Alzheimer's disease in the APP gene at the N-terminus of beta-amyloid. *Nat Genet.* 1992;1(5):345–34. [PubMed: 1302033]
73. Citron M, Oltersdorf T, Haass C, et al. Mutation of the beta-amyloid precursor protein in familial Alzheimer's disease increases beta-protein production. *Nature.* 1992;360(6405):672–674. [PubMed: 1465129]
74. Chen WT, Hong CJ, Lin YT, et al. Amyloid-beta (A β) D7H mutation increases oligomeric A β 42 and alters properties of A β -zinc/copper assemblies. *PLoS One.* 2012;7(4):e35807.
75. Wakutani Y, Watanabe K, Adachi Y, et al. Novel amyloid precursor protein gene missense mutation (D678N) in probable familial Alzheimer's disease. *J Neurol Neurosurg Psychiatry.* 2004;75(7):1039–1042. [PubMed: 15201367]
76. Zhou L, Brouwers N, Benilova I, et al. Amyloid precursor protein mutation E682K at the alternative β -secretase cleavage β' -site increases A β generation. *EMBO Mol Med.* 2011;3(5):291–302. [PubMed: 21500352]
77. Kaden D, Harmeier A, Weise C, et al. Novel APP/A β mutation K16N produces highly toxic heteromeric A β oligomers. *EMBO Mol Med.* 2012;4(7):647–659. [PubMed: 22514144]
78. Hendriks L, van Duijn CM, Cras P, et al. Presenile dementia and cerebral haemorrhage linked to a mutation at codon 692 of the beta-amyloid precursor protein gene. *Nat Genet.* 1992;1(3):218–221. [PubMed: 1303239]
79. Kamino K, Orr HT, Payami H, et al. Linkage and mutational analysis of familial Alzheimer disease kindreds for the APP gene region. *Am J Hum Genet.* 1992;51(5):998–1014. [PubMed: 1415269]
80. Levy E, Carman MD, Fernandez-Madrid IJ, et al. Mutation of the Alzheimer's disease amyloid gene in hereditary cerebral hemorrhage, Dutch type. *Science.* 1990;248(4959):1124–1126. [PubMed: 2111584]
81. Grabowski TJ, Cho HS, Vonsattel JP, Rebeck GW, Greenberg SM. Novel amyloid precursor protein mutation in an Iowa family with dementia and severe cerebral amyloid angiopathy. *Ann Neurol.* 2001;49(6):697–705. [PubMed: 11409420]
82. Kumar-Singh S, De Jonghe C, Cruts M, et al. Nonfibrillar diffuse amyloid deposition due to a gamma(42)-secretase site mutation points to an essential role for N-truncated A beta(42) in Alzheimer's disease. *Hum Mol Genet.* 2000;9(18):2589–2598. [PubMed: 11063718]
83. Cruts M, Dermaut B, Rademakers R, Van den Broeck M, Stögbauer F, Van Broeckhoven C. Novel APP mutation V715A associated with presenile Alzheimer's disease in a German family. *J Neurol.* 2003;250(11):1374–1375. [PubMed: 14648157]
84. Ancolio K, Dumanchin C, Barelli H, et al. Unusual phenotypic alteration of beta amyloid precursor protein (betaAPP) maturation by a new Val-715 -> Met betaAPP-770 mutation responsible for probable early-onset Alzheimer's disease. *Proc Nat Acad Sci USA.* 1999;96(7):4119–4124. [PubMed: 10097173]
85. Eckman CB, Mehta ND, Crook R, et al. A new pathogenic mutation in the APP gene (I716V) increases the relative proportion of A beta 42(43). *Hum Mol Genet.* 1997;6(12):2087–2089. [PubMed: 9328472]
86. Guerreiro RJ, Baquero M, Blesa R, et al. Genetic screening of Alzheimer's disease genes in Iberian and African samples yields novel mutations in presenilins and APP. *Neurobiol Aging.* 2010;31(5):725–731. [PubMed: 18667258]

87. Kwok JB, Li QX, Hallupp M, et al. Novel Leu723Pro amyloid precursor protein mutation increases amyloid beta42(43) peptide levels and induces apoptosis. *Ann Neurol*. 2000;47(2):249–253. [PubMed: 10665499]
88. Theuns J, Marjaux E, Vandenbulcke M, et al. Alzheimer dementia caused by a novel mutation located in the APPC-terminal intracytosolic fragment. *Hum Mutat*. 2006;27(9):888–896. [PubMed: 16917905]
89. Selkoe DJ. Amyloid beta-protein and the genetics of Alzheimer's disease. *J Biol Chem*. 1996;271(31):18295–18298.
90. Hardy J New insights into the genetics of Alzheimer's disease. *Ann Med*. 1996;28(3):255–258. [PubMed: 8811169]
91. Bateman RJ, Xiong C, Benzinger TL, et al.; Dominantly Inherited Alzheimer Network. Clinical and biomarker changes in dominantly inherited Alzheimer's disease. *N Engl J Med*. 2012;367(9):795–804. [PubMed: 22784036]
92. Orgogozo JM, Gilman S, Dartigues JF, et al. Subacute meningoencephalitis in a subset of patients with AD after Abeta42 immunization. *Neurology*. 2003;61(1):46–54. [PubMed: 12847155]
93. Fox NC, Black RS, Gilman S, et al.; AN1792(QS-21)-201 Study. Effects of Abeta immunization (AN1792) on MRI measures of cerebral volume in Alzheimer disease. *Neurology*. 2005;64(9):1563–1572. [PubMed: 15883317]
94. Salloway S, Sperling R, Gilman S, et al.; Bapineuzumab 201 Clinical Trial Investigators. A phase 2 multiple ascending dose trial of bapineuzumab in mild to moderate Alzheimer disease. *Neurology*. 2009;73(24):2061–2070. [PubMed: 19923550]
95. Adolfsson O, Pihlgren M, Toni N, et al. An effector-reduced anti- β -amyloid (A β) antibody with unique A β binding properties promotes neuroprotection and glial engulfment of A β . *J Neurosci*. 2012;32(28):9677–9689. [PubMed: 22787053]
96. Bohrmann B, Baumann K, Benz J, et al. Gantenerumab: a novel human anti-A β antibody demonstrates sustained cerebral amyloid- β binding and elicits cell-mediated removal of human amyloid- β . *J Alzheimers Dis*. 2012;28(1):49–69. [PubMed: 21955818]
97. Farlow M, Arnold SE, van Dyck CH, et al. Safety and biomarker effects of solanezumab in patients with Alzheimer's disease. *Alzheimers Dement*. 2012; 8(4):261–271. [PubMed: 22672770]
98. Relkin NR, Szabo P, Adamiak B, et al. 18-Month study of intravenous immunoglobulin for treatment of mild Alzheimer disease. *Neurobiol Aging*. 2009;30(11):1728–1736. [PubMed: 18294736]
99. Gilman S, Koller M, Black RS, et al.; AN1792(QS-21)-201 Study Team. Clinical effects of Abeta immunization (AN1792) in patients with AD in an interrupted trial. *Neurology*. 2005;64(9):1553–1562. [PubMed: 15883316]
100. Blennow K, Zetterberg H, Rinne JO, et al.; AAB-001201/202 Investigators. Effect of immunotherapy with bapineuzumab on cerebrospinal fluid biomarker levels in patients with mild to moderate Alzheimer disease. *Arch Neurol*. 2012;69(8):1002–1010. [PubMed: 22473769]
101. Miller G Alzheimer's research: stopping Alzheimer's before it starts. *Science*. 2012;337 (6096):790–792. [PubMed: 22903991]
102. Morris JC, Aisen PS, Bateman RJ, et al. Developing an international network for Alzheimer research: The Dominantly Inherited Alzheimer Network. *Clin Investig (Lond)*. 2012;2(10):975–984.
103. Reiman EM, Langbaum JB, Fleisher AS, et al. Alzheimer's Prevention Initiative: a plan to accelerate the evaluation of presymptomatic treatments. *J Alzheimers Dis*. 2011;26(suppl 3):321–329. [PubMed: 21971471]
104. Lambracht-Washington D, Rosenberg RN. DNA A β 42 immunization generates a multivalent vaccine: antibodies in plasma of active full-length DNA A β 42 immunized mice show polyclonal A β 42 peptide binding. Paper presented at: Alzheimer's Association International Conference; 2015; Washington, DC.
105. Vasilevko V, Pop V, Kim HJ, et al. Linear and conformation specific antibodies in aged beagles after prolonged vaccination with aggregated Abeta. *Neurobiol Dis*. 2010;39(3):301–310. [PubMed: 20451612]

106. Hatami A, Albay R III, Monjaze S, Milton S, Glabe C. Monoclonal antibodies against A β 42 fibrils distinguish multiple aggregation state polymorphisms in vitro and in Alzheimer disease brain. *J Biol Chem.* 2014;289(46):32131–32143.
107. Qu BX, Lambracht-Washington D, Fu M, Eagar TN, Stüve O, Rosenberg RN. Analysis of three plasmid systems for use in DNA A beta 42 immunization as therapy for Alzheimer's disease. *Vaccine.* 2010;28(32):5280–528 . [PubMed: 20562015]
108. Qu B, Boyer PJ, Johnston SA, Hynan LS, Rosenberg RN. Abeta42 gene vaccination reduces brain amyloid plaque burden in transgenic mice. *J Neurol Sci.* 2006;244(1–2):151–158. [PubMed: 16556449]
109. Qu BX, Xiang Q, Li L, Johnston SA, Hynan LS, Rosenberg RN. Abeta42 gene vaccine prevents Abeta42 deposition in brain of double transgenic mice. *J Neurol Sci.* 2007;260(1–2):204–213. [PubMed: 17574274]
110. Lambracht-Washington D, Rosenberg RN. Anti-amyloid beta to tau-based immunization: Developments in immunotherapy for Alzheimer disease. *Immunotargets Ther.* 2013;2013(2): 105–114. [PubMed: 24926455]
111. Lambracht-Washington D, Qu BX, Fu M, et al. A peptide prime-DNA boost immunization protocol provides significant benefits as a new generation A β 42 DNA vaccine for Alzheimer disease. *J Neuroimmunol.* 2013;254(1–2):63–68. [PubMed: 23036592]
112. Lambracht-Washington D, Qu BX, Fu M, et al. DNA immunization against amyloid beta 42 has high potential as safe therapy for Alzheimer's disease as it diminishes antigen-specific Th1 and Th17 cell proliferation. *Cell Mol Neurobiol.* 2011;31(6):867–874. [PubMed: 21625960]
113. Lambracht-Washington D, Qu BX, Fu M, Eagar TN, Stüve O, Rosenberg RN. DNA beta-amyloid(1–42) trimer immunization for Alzheimer disease in a wild-type mouse model. *JAMA.* 2009;302(16):1796–1802. [PubMed: 19861672]
114. Lambracht-Washington D, Rosenberg RN. Advances in the development of vaccines for Alzheimer's disease. *Discov Med.* 2013;15(84):319–326. [PubMed: 23725605]
115. Lambracht-Washington D, Rosenberg RN. Co-stimulation with TNF receptor superfamily 4/25 antibodies enhances in-vivo expansion of CD4+CD25+Foxp3+ T cells (Tregs) in a mouse study for active DNA A β 42 immunotherapy. *J Neuroimmunol.* 2015;278:90–99. [PubMed: 25595257]
116. Lambracht-Washington D, Rosenberg RN. A noninflammatory immune response in aged DNA A β 42-immunized mice supports its safety for possible use as immunotherapy in AD patients. *Neurobiol Aging.* 2015;36(3):1274–1281. [PubMed: 25725942]
117. Rosenberg RN. Defining amyloid pathology in persons with and without dementia syndromes: making the right diagnosis. *JAMA.* 2015;313(19):1913–1914. [PubMed: 25988459]
118. Rosenberg RN, Lambracht-Washington D. DNA A β 42 vaccination as possible alternative immunotherapy for Alzheimer disease. *JAMA Neurol.* 2013;70(6):772–773. [PubMed: 23700123]
119. Rosenberg RN, Petersen RC. The Human Alzheimer Disease Project: a new call to arms. *JAMA Neurol.* 2015;72(6):626–628. [PubMed: 25844883]

Key Points**Question**

What is the current genomic understanding of amyloid β protein-directed therapies for Alzheimer disease?

Findings

In this review, mutations associated with Alzheimer disease, polymorphisms increasing risk, and amyloid β protein-reducing therapies are defined. The magnitude of defining the genomic and epigenomic factors involved in the pathogenesis of Alzheimer disease and new therapeutic strategies as a result are large in number and complexity.

Meaning

Effective therapy to delay or prevent Alzheimer disease may be based on a defined genomic understanding.

Figure 1. Proteolytic Processing of Amyloid Precursor Protein (APP) by β - and γ -Secretases to Generate Amyloid β (A β) Protein

When APP undergoes nonamyloidogenic proteolytic cleavage by α - and γ -secretases, p3 instead of A β is generated. A small number of APP molecules is proteolytically processed by β -secretase and generates an N-terminal soluble APP and a 12-kDa C-terminal stub of APP, which is cleaved by γ -secretase to yield A β and amyloid intracellular domain. The *PS1* and *PS2* genes carry the active site of γ -secretase complex. Proteins encoded by multiple risk genes associated with late-onset Alzheimer disease are involved in A β clearance. The *SORL1*, *PICALM*, and *CD2AP* genes regulate APP endocytosis (vs retromer-mediated APP recycling) and A β generation in endosome-lysosomes.

Figure 2. Double DNA Plasmid System Incorporating GAL4/Upstream Activating Sequences to Increase Amyloid β 42(A β 42) Expression

It consists of 2 plasmids, an activator plasmid encoding the yeast GAL4 transcription factor and a responder plasmid encoding A β 1–42 for which its expression is driven by binding of GAL4 to upstream activating sequences (UAS sites) in front of a minimal promoter. The open reading frame for A β 42 was synthesized 3 times in a row, encoding DNA A β 42 trimer. A β 42-trimer/UAS responder plasmid and GAL4 activator plasmid are mixed in a ratio of 1 to 0.75 for preparation of the DNA-coated gold particles. The GAL4 protein binds as a homodimer to UAS sites present on the responder plasmid. GAL4 binding enhances transcription of the A β 42 trimer sequence, which is cloned into a DNA fragment containing an adenovirus E3 leader sequence and an endosomal targeting sequence derived from the mouse MHC class II gene, *H2-DM*. The final protein product has a size of about 21 kD.²⁰

Figure 3. Reductions of Amyloid β 42 (A β 42) in DNA A β 42 Trimer Immunized Amyloid Precursor Protein/Presenilin 1 (APP/PS1) Double Transgenic Mice and the Respective Control DNA Immunized Mice

Shown is the amount of A β 42 peptide per gram of wet brain tissue (μ g/g). Immunization was started in both groups in 3-month-old mice and was continued for 11 immunizations until the mice were 12 months old. Group A was killed for final analyses (plasma antibody levels, brain A β histology and biochemistry) 14 days following the last immunization, while mice in group B were killed 4 months following the 11th immunization. In both groups, a significant reduction of A β 1–42 levels in the brain was found in comparison with the respective control groups (DNA Luc control DNA immunized mice). Group A showed an amyloid reduction of 65% to 53% while group B showed a reduction of A β 1–42 brain levels of 25%. Reproduced with permission from *ImmunoTargets and Therapy*.¹¹⁰

Table.

Genotype to Phenotype Relationship Between Mutant Amyloid Precursor Protein (*APP*) and Amyloid β ($A\beta$) Generation

Mutation	$A\beta$ Generation	Reference
Swedish pathogenic <i>KM1670/671NL</i>	Better substrate for β -secretase and increase $A\beta$ generation	72,73
Icelandic Protective <i>A673T</i>	Attenuate APP- β -secretase interaction and reduce $A\beta$ production	14,15
Pathogenic <i>D678H</i> and Tottori <i>D678N</i>	Increase $A\beta$ production and $A\beta_{42}/A\beta_{40}$ ratio; increase oligomerization	74,75
Pathogenic <i>E682K</i>	Increase in total $A\beta$ and in $A\beta_{42}/A\beta_{40}$ ratio	76
Pathogenic <i>K687N</i>	Increase $A\beta_{40}$ and $A\beta_{42}$ and toxic heteromeric $A\beta$ oligomer	77
Flemish pathogenic <i>A692G</i>	Increase $A\beta_{40}$ and $A\beta_{42}$	78
Arctic, Dutch, and Italian pathogenic <i>E693G</i> , <i>E693Q</i> , and <i>E693K</i>	Increase the aggregation propensity of $A\beta$	79,80
Pathogenic <i>D694N</i>	Increase the aggregation propensity of $A\beta$	81
Pathogenic <i>T714I</i>	Increase $A\beta_{42}/A\beta_{40}$ ratio	82
Pathogenic <i>V715A</i> and <i>V715M</i>	Increase $A\beta_{42}/A\beta_{40}$ ratio	83,84
Florida Pathogenic <i>I716F</i> and <i>I716V</i>	Increase $A\beta_{42}/A\beta_{40}$ ratio	85,86
Pathogenic <i>V717F</i> and London <i>V717I</i> and <i>V717L</i>	Increase $A\beta_{42}/A\beta_{40}$ ratio and increase $A\beta_{42}$, decrease $A\beta_{40}$	30–32
Pathogenic <i>L723P</i>	Increase $A\beta_{42}$	87
Pathogenic <i>K724N</i>	Increase $A\beta_{42}$, decrease $A\beta_{40}$, and increase $A\beta_{42}/A\beta_{40}$	88