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## The role of Alzheimer's disease risk genes in endolysosomal pathways

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### Abstract

There is ample pathological and biological evidence for endo-lysosomal dysfunction in Alzheimer's disease (AD) and emerging genetic studies repeatedly implicate endo-lysosomal genes as associated with increased AD risk. The endo-lysosomal network (ELN) is essential for all cell types of the central nervous system (CNS), yet each unique cell type utilizes cellular trafficking differently (see Fig. 1). Challenges ahead involve defining the role of AD associated genes in the functionality of the endo-lysosomal network (ELN) and understanding how this impacts the cellular dysfunction that occurs in AD. This is critical to the development of new therapeutics that will impact, and potentially reverse, early disease phenotypes. Here we review some early evidence of ELN dysfunction in AD pathogenesis and discuss the role of selected AD-associated risk genes in this pathway. In particular, we review genes that have been replicated in multiple genome-wide association studies (Andrews et al., 2020; Jansen et al., 2019; Kunkle et al., 2019; Lambert et al., 2013; Marioni et al., 2018) and reviewed in (Andrews et al., 2020) that have defined roles in the endo-lysosomal network. These genes include *SORL1*, an AD risk gene harboring both rare and common variants associated with AD risk and a role in trafficking cargo, including APP, through the ELN; *BINI*, a regulator of clathrin-mediated endocytosis whose expression correlates with Tau pathology; *CD2AP*, an AD risk gene with roles in endosome morphology and recycling; *PICALM*, a clathrin-binding protein that mediates trafficking between the trans-Golgi network and endosomes; and Ephrin Receptors, a family of receptor tyrosine kinases with AD associations and interactions with other AD risk genes. Finally, we will discuss how human cellular models can elucidate cell-type specific differences in ELN dysfunction in AD and aid in therapeutic development.

### Keywords

AD risk genes; Endolysosomal network; Human cellular model

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Declaration of Competing Interest

None.

## 1. Introduction

Alzheimer's disease (AD) is a progressive degenerative disease that affects upwards of 5 million people in the United States alone and is one of the top 10 causes of death (Alzheimer's disease facts and figures. *Alzheimers Dement*, 2020; Collaborators, 2019). Clinically, AD is characterized by progressive memory decline, language and speech defects, impairment of executive function and a progressive impairment of the execution of routine daily activities (Cappa, 2018; Graham et al., 2017; Masters et al., 2015; Querfurth and LaFerla, 2010; Scheltens et al., 2016). Neuropathologically, senile plaques and neurofibrillary tangles comprise the hallmark features of AD. Although several classes of medications are currently approved to treat AD symptoms, there are currently no treatments that substantially reverse or affect the progression of the disease, no reliable early diagnostic approaches for AD, and no effective preventative approaches (Alzheimer's disease facts and figures. *Alzheimers Dement*, 2020; Cummings et al., 2019). The lack of effective therapies is, at least in part, related to the fact that biology of AD is only incompletely understood.

Genome-wide association studies (GWAS) have identified dozens of loci associated with increased disease risk for developing AD (Bis et al., 2020; Gao et al., 2018; Jansen et al., 2019; Kunkle et al., 2019; Lambert et al., 2013; Marioni et al., 2018). These studies strongly point to aberrations in the endolysosomal pathway, immune response, and lipid processing as contributing to AD risk (Pimenova et al., 2018). Furthermore, recent and larger GWAS studies continue to identify additional risk loci. The genetic details and implications of GWAS to AD have been extensively reviewed and many of these reviews provide a broad overview of these biological pathways (Andrews et al., 2020; Karch and Goate, 2015; Pimenova et al., 2018). In this review, we aim to provide a focused discussion on key loci implicated in endolysosomal trafficking. The ELN is dynamic and its function overlaps with both immune response and lipid processing, therefore, there are many loci that may overlap between the ELN and other pathways. We have chosen to discuss five genes that have been extensively replicated in genetic studies with defined roles specifically in endolysosomal trafficking (Andrews et al., 2020; Jansen et al., 2019; Kunkle et al., 2019; Lambert et al., 2013; Marioni et al., 2018). In addition, one of these genes, *SORL1*, harbors rare coding variants that may be causal for AD (Scheltens et al., 2021).

We provide a detailed introduction to the biology of ELN trafficking and we summarize the literature related to the pathogenic role of the ELN dysfunction in AD. Within the cells of the brain, enlarged endosomes in neurons are an early cytopathological hallmark of AD (Cataldo et al., 2000) along with synaptic alterations and inflammatory pathologies in glial cells (Small and Petsko, 2020). We and others have recently shown that human induced pluripotent stem cells (iPSC) models of AD can recapitulate some of the endosomal trafficking defects phenotypes that impact these pathologies and we have observed that there are important cell-type specific differences in endolysosomal phenotypes (Hung et al., 2021; Knupp et al., 2020). Changes in cellular pathology may occur decades prior to the onset of clinical symptoms (Cataldo et al., 2000). Thus, it is important to dissect these phenotypes in cell-type specific models that can capture early stages of cellular dysfunction, therefore we end this review by commenting on human models to further study the ELN and other biological pathways implicated in AD genetics.

## 2. The endo-lysosomal network

The ELN consists of intracellular membranous organelles which interconvert in a dynamic fashion. Proteins associated with these organelles in the ELN can distinguish the main components such as early endosomes (EEs), recycling endosomes (REs), late endosomes (LEs), autophagosomes and lysosomes. These proteins are widely used as markers for discreet structures within the ELN, although the network itself is fluid and dynamic.

Cellular cargo are internalized into the cell in a process called endocytosis. Internalized cargo is then sorted through various vesicles or sorting complexes to either the cell surface (via recycling endosomes), to lysosomes for degradation (via late endosomes), or to the trans-Golgi for additional sorting (via the multiprotein complex retromer)(Bonifacino and Rojas, 2006; Luzio et al., 2007; Maxfield and McGraw, 2004; Poteryaev et al., 2010). Additionally, the ELN is involved in the release of selected cellular contents into the extracellular environment through a process called exocytosis(Chieriegatti and Meldolesi, 2005) and involved in the specialized degradative process, macroautophagy, wherein damaged cellular contents are ensconced in a doublemembrane bound organelle then fused with lysosomes and degraded (Gatica et al., 2018).

Endocytosis regulates several processes initiated at the plasma membrane including receptor mediated cell signaling and nutrient uptake. Here we will review clathrin-mediated endocytosis (CME), a common type of endocytosis that internalize a wide range of cargo, usually <200 nm in diameter. Clathrin-independent pathways also exist (Mayor et al., 2014; Sandvig et al., 2018; Shafaq-Zadah et al., 2020) but will not be reviewed here. CME starts with cargo recognition, which leads to membrane bending and the formation of specialized 'pits' called clathrin coated pits. Clathrin forms a complex lattice like structure around vesicles formed by membrane invaginations(Marsh and McMahon, 1999; Merrifield et al., 2005; Pearse, 1976; Ungewickell and Hinrichsen, 2007). Several AD risk genes are involved in the mechanisms of CME. Formation of vesicles and the linking of cargo to the clathrin coat requires adaptor and scaffold proteins. CD2-associated protein (CD2AP) supports an important link between vesicle formation and the actin cytoskeleton(Kadlecova et al., 2017; Kelly et al., 2014) and Phosphatidylinositol Binding Clathrin Assembly Protein (PICALM) is a monomeric adaptor protein that recruits clathrin to the cell membrane(Meyerholz et al., 2005; Miller et al., 2015). Scission of these clathrin coated pits from the plasma membrane is caused by a large GTPase, dynamin(van der Bliek et al., 1993) along with dynamin partners including Bin, amphiphysin and Rvs (BAR) domain proteins, including BIN1(Daumke et al., 2014; McPherson et al., 2013; Takei et al., 1999). These vesicles then fuse with pre-existing EEs for subsequent intracellular trafficking (Choudhury et al., 2005; Ungewickell et al., 2004).

After endocytosis, the early endosome (EE) receives cargo and serves as a sorting hub(Huotari and Helenius, 2011). EEs are marked by the GTP-binding protein Rab5 and/or its effector protein, early endosome antigen 1 (EEA1)(Behnia and Munro, 2005; Zerial and McBride, 2001). After rapid sorting at the EE, cargo is either directed to late endosomes (LEs) and lysosomes(Luzio et al., 2007; Poteryaev et al., 2010) for degradation or recycled back to the plasma membrane(Maxfield and McGraw, 2004). Recycling can occur either

directly from the EE to the plasma membrane or via specialized vesicles called recycling endosomes (REs) marked by the GTPase Rab11 (Pasqualato et al., 2004; Sonnichsen et al., 2000). Conversion of EEs to downstream endosomal compartments in the ELN occurs through endosomal maturation that involves a gradual acidification of the lumen (Huotari and Helenius, 2011). LEs are a varied class of endosomes that transport selected cargo from EE into lysosomes (Rink et al., 2005), newly synthesized lysosomal hydrolases from the *trans*-Golgi network (TGN) into lysosomes, and selected cargo from LE to TGN (Zhang et al., 2009). Transport from LE and EE to the TGN occurs via the large multiprotein complex retromer (Rojas et al., 2008) or, alternatively via a LE class marked by Rab9 (Bonifacino and Rojas, 2006). LEs are typically marked by the GTPase Rab7 and biogenesis of these vesicles depends on activity of receptors of certain signaling pathways, suggesting that this process is subject to alterations depending on cellular activity (Research in Medical Education, 1992). LEs eventually form the most acidic intracellular compartments of the ELN, lysosomes, which are marked by membrane proteins such as lysosomal-associated membrane proteins 1 and 2 (LAMP1 and LAMP2) and have a lumen composition comprised of enzymes called acid hydrolases that are active only at acidic pH (Maxfield and Yamashiro, 1987). While LAMP1/2 are commonly used as lysosome markers, a recent study demonstrated that these markers alone do not always label degradative lysosomes and can be associated with LEs and EEs (Cheng et al., 2018). Therefore, when measuring degradative lysosomes, it is important to also consider organelles containing lysosomal hydrolases such as the cathepsins, the most abundant lysosomal proteases. Continuous endocytic trafficking of cargo to lysosomes is crucial to maintain lysosome morphology, pH and intracellular localization (Bucci et al., 2000).

In addition to endocytosis, compartments of the ELN also participate in formation of exosomes, involved in the process of exocytosis. Exosomes have an endosomal origin formed from multivesicular endosomes or multivesicular bodies (MVBs) with several intraluminal vesicles (ILVs) (Hessvik and Llorente, 2018; van Niel et al., 2018). Exosomes aid in maintaining cellular homeostasis by eliminating unwanted cellular contents and also contributing to intercellular communication, since exosomal contents of one cell can be taken up by neighboring cells (Mathieu et al., 2019; Yuyama and Igarashi, 2016). The ELN also contributes to macroautophagy, a pathway where internal substrates are enclosed within specific membranes and degraded. This process begins with the formation of autophagosomes that have an endosomal origin (Puri et al., 2018) and proceed to fuse with lysosomes to form autolysosomes that degrade any damaged cellular material (Nakatogawa, 2020).

Overall, through coordination of these multiple complex intracellular pathways, the ELN maintains cellular homeostasis and dysfunction of this network can result in disease phenotypes in distinct cell types. For more in-depth reviews of the ELN we refer to excellent reviews (Gautreau et al., 2014; Hu et al., 2015; Hyttinen et al., 2013; Inpanathan and Botelho, 2019; Klumperman and Raposo, 2014; Kumari et al., 2010; Luzio et al., 2014).

### 3. The endo-lysosomal network in Alzheimer's disease

Early reports of ELN dysfunction in AD were closely associated with the trafficking and cleavage of the amyloid precursor protein, APP. Studies showed that APP localizes within vesicle-like structures in cells (Ferreira et al., 1993) and these vesicles and endocytic pathways were identified as locations where APP cleavage could occur, increasing amyloid beta (A $\beta$ ) peptides and plaque formation (Roher et al., 1988). Early clinical studies suggested a pathogenic role for the ELN in AD. Schwagerl et al., measured the levels of Cathepsin D (Cat-D), a major lysosomal protease, in the cerebrospinal fluid (CSF) of patients with AD. Cat-D was presumed to enter the CSF following neuronal death and measured levels were approximately 4-times higher than healthy control levels, and also higher than the corresponding levels in patients with other neurodegenerative diseases such as Huntington's disease (Schwagerl et al., 1995). This study, and others performed at the time (Cataldo and Nixon, 1990; Cataldo et al., 1991), suggested that dysfunction of the lysosome could lead to neuronal cell death associated with various neurodegenerative diseases including AD (Nixon and Cataldo, 1995). Further work showed hippocampal and prefrontal cortex cells from AD patients contained 2-8-fold more hydrolase-positive vacuolar compartments than healthy control brains, suggestive of distinct physiologic differences in ELN components in AD that were unrelated to normal aging (Cataldo et al., 1996). More recent work has suggested that Cat-D is upregulated in the AD neocortex and that Cat-D immunoreactivity was colocalized with neurofibrillary tangles (Chai et al., 2019). Interestingly, this study compared Cat-D staining between AD, Parkinson's Disease Dementia and Lewy Body Dementias (LBD) but only observed an upregulation of Cat-D in AD (Chai et al., 2019). In contrast, Kim et al., recently reported decreased levels of Cat-D in plasma collected from AD patients (Kim et al., 2021). More recent studies have also looked at ELN proteins in biospecimens of AD patients. Morena et al., found a significant reduction of hexosaminidase isoenzymes A and B in the PBMCs of mild AD patients, along with an increase in the secretion of HexA and HexB in the plasma of severe AD patients. Higher levels of beta-galactosidase were also found in the plasma of severe AD patients, indicating a differential expression of lysosomal enzymes in both mild and severe cases of AD (Morena et al., 2017). Sjodin et al. analyzed the concentration of ELN proteins within the CSF of Alzheimer's and Parkinson's patients. They found an increase in the CSF concentrations of proteins such as AP2B1, CTSB, CTSE, LAMP1, and LAMP2 in AD patients and a subset set of these proteins were altered in PD patients (Sjodin et al., 2019). Taken together, these studies show that the ELN has long been and continues to be an attractive target for biomarker development in AD. However, these findings also underscore the complexity of ELN molecules and suggest that different processes may occur in the periphery versus the CNS.

Enlarged EEs, indicative of delayed maturation or a block in the endocytic pathway (Kaur and Lakkaraju, 2018), are also markers of ELN dysfunction. Elegant work in the late 1990s and early 2000s demonstrated that enlarged EEs preceded canonical AD pathology in patient brains, indicating that ELN dysfunction occurs early in the course of disease (Cataldo et al., 1997; Cataldo et al., 2004; Cataldo et al., 2000). Further studies have shown alterations in genes that regulate endosomal pathways in both autosomal dominant familial AD (FAD) and in late-onset sporadic AD (SAD). In FAD, mutations in the Amyloid Precursor Protein gene

(*APP*) and the catalytic components of the  $\gamma$ -secretase complex, Presenilin 1 and Presenilin 2 (*PSENI1*, and *PSENI2*) genes disrupt various components of the ELN. APP is trafficked throughout the ELN and its processing by  $\beta$  and  $\gamma$  secretases occurs in various ELN compartment (Haass et al., 2012; Thinakaran and Koo, 2008) and reviewed in (Winckler et al., 2018). FAD mutations in *APP* lead to enlarged EEs, can alter interactions between APP and BACE1 in endocytic compartments, affect intracellular sorting and ultimately increase amyloidogenic processing (Kwart et al., 2019). Presenilins, especially presenilin 2, are localized to endolysosomal vesicles and this localization may be altered in AD (Deaton and Johnson, 2020; Pasternak et al., 2003; Sannerud et al., 2016). *PSENI1/2* mutations disrupt lysosome function, alter autophagy, and also result in enlarged EEs (Bhalla et al., 2012; Das et al., 2016; Fedeli et al., 2019; Hung and Livesey, 2018; Kwart et al., 2019; La Rosa et al., 2015; Lee et al., 2010; Mecozzi et al., 2014; Nixon, 2017; Reddy et al., 2016; Sannerud et al., 2016; Zhang et al., 2012). As we mentioned in the introduction, for late-onset SAD, GWAS have consistently implicated endo-lysosomal loci with increased genetic risk for disease development, including the genes we discuss below (Bellenguez, 2020; Gao et al., 2018; Karch and Goate, 2015; Schwartzentruber et al., 2021; Van Acker et al., 2019). Thus, endosomal trafficking and recycling has been referred to as a “hub” that can reconcile multiple cellular pathologies in both FAD and SAD (Small and Petsko, 2020).

## 4. AD risk genes in the endo-lysosomal network

### 4.1. SORL1

*SORL1* encodes the protein SORLA and was originally identified as a member of the LDL family of receptors (Yamazaki et al., 1996). Subsequent studies also classified SORLA as one of five mammalian sorting receptors that contain a vacuolar protein sorting domain (VPS10) (Jacobsen et al., 2001; Jacobsen et al., 1996; Willnow et al., 2008; Yamazaki et al., 1996). Prior to genetic associations with AD, early studies described loss of SORLA expression in neurons in SAD brains (Dodson et al., 2006; Scherzer et al., 2004). Further biochemical characterization revealed a direct interaction between SORLA and APP and characterized SORLA as a sorting receptor that protected APP from amyloidogenic processing (Andersen et al., 2005). Functionally, SORLA is now well-characterized as a receptor that serves as an adaptor protein for the retromer complex (Andersen et al., 2016; Fjorback et al., 2012). SORLA has also been reported to traffic A $\beta$  peptides to lysosomes (Caglayan et al., 2014), and manipulating SORLA expression in non-neuronal and neuronal cells directly impacts APP processing to A $\beta$  peptides (Rogaeva et al., 2007; Young et al., 2015). In addition to a direct role in APP processing, SORLA traffics receptors for the neurotrophins BDNF and GDNF (Geng et al., 2011; Glerup et al., 2013; Rohe et al., 2013) and SORLA expression may impact synaptic function. Synapsins are a family of proteins that are involved with the regulation of neurotransmitter release at synapses, and thus are essential in the processes of endo- and exocytosis. Deletion of SORLA in mice resulted in an accumulation of phosphorylated synapsins in various regions of the brain, indicating that SORLA may have a direct effect on the degradation of synapsins, which, in turn, could impact on synaptic vesicle endocytosis (Hartl et al., 2016). Additionally, Huang and colleagues used SORLA transgenic mice to demonstrate an interaction with the ephrin receptor EphA4, which is important for synaptic structure and function (Huang et al., 2017).



Aberrant EphA4 activation by A $\beta$  has been described in AD (Fu et al., 2007; Vargas et al., 2018) and SORLA association with EphA4 was shown to suppress synaptotoxic activation of this receptor (Huang et al., 2017).

In 2007, a candidate gene study implicated two haplotypes in *SORL1* that were associated with increased AD risk in several population groups (Rogaeva et al., 2007), indicating that *SORL1* might be considered an AD risk gene. Exome sequencing studies in 2012 identified rare coding variants in *SORL1* present in families with early-onset AD without known mutations in APP or PSEN1/2 and subsequent larger exome studies have revealed multiple coding variants in many domains of the protein (Holstege et al., 2017; Pottier et al., 2012). *SORL1* is a large gene and while this in-depth analysis has also found variants that occur in controls, frameshift variants that lead to premature stop codons and therefore haploinsufficiency of *SORL1* appear, thus far, to only occur in AD cases (Holstege et al., 2017; Raghavan et al., 2018). A large GWAS in 2013 associated *SORL1* as a susceptibility locus in AD and this association has been consistently replicated (Lambert et al., 2013; Miyashita et al., 2013; Raghavan et al., 2018).

Both genetic and functional studies have continued to indicate the important role *SORL1* plays in AD pathogenesis, supporting the further study of this gene. Recently, our group has investigated the effect of CRISPR/Cas9-mediated depletion of *SORL1* expression in human induced pluripotent stem cells in the context of AD pathogenesis. We found that the neurons lacking SORLA expression developed a significant degree of EE enlargement, which was concomitant with an altered APP localization within the ELN (Knupp et al., 2020), indicating a critical role for *SORL1* in endosomal trafficking. Taken together, the above studies clearly suggest that the *SORL1* gene and SORLA protein may have a direct role in ELN function and in AD pathogenesis and may have the potential to lead to future diagnostic or therapeutic approaches in AD.

#### 4.2. BIN1

*BIN1* (Bridging Integrator-1) was originally characterized as a mycinteracting protein, structurally related to amphiphysin (Sakamuro et al., 1996). Further studies showed that Bin1 was involved in dynamin recruitment to clathrin-coated pits at nerve terminals (Wigge et al., 1997). *BIN1* is repeatedly associated with increased AD risk in multiple genome-wide studies with an association second after APOE (Harold et al., 2009; Kunkle et al., 2019; Lambert et al., 2013; Seshadri et al., 2010). *BIN1* is part of the BAR domain protein family that regulates membrane dynamics (De Rossi et al., 2016; Prokic et al., 2014). It undergoes alternative splicing to generate several cell type specific isoforms. All the isoforms contain two functional domains, BAR and the Src-homology (SH3) domains and regulate membrane invaginations and interactions with dynamin. However, the neuronal isoform has an additional domain, called the clathrin and adaptor binding domain (CLAP) domain suggesting that *BIN1* participates in clathrin mediated endocytosis in neurons. Loss of *BIN1* in rodent cortical and hippocampal neurons results in increased endocytosis and enlarged Rab5-positive EEs, leading to endosomal membrane damage and tau leakage (Calafate et al., 2016). Loss of *BIN1* in rodent neurons also impaired trafficking of the type I transmembrane aspartyl protease enzyme, BACE1, required for A $\beta$  production

to lysosomes. (Miyagawa et al., 2016). Interestingly, studies in human neurons report that *BINI* overexpression resulted in increased EE size and loss of *BINI* resulted in decreased EE size (Lambert et al., 2021). Taken together, these studies suggest that the role of *BINI*, although different in rodents and humans, is crucial in regulating ELN function in neurons especially at the level of EEs. The BAR domain in *BINI* is responsible for vesicle budding and hence it is possible that BIN1 is required for maturation of EEs to LEs. Loss of *BINI* alters this process resulting in changes to EE size, which may affect downstream trafficking pathways.

Targeted, quantitative RT-PCR studies using RNA isolated from postmortem brain tissue demonstrated that levels of *BINI* expression are associated with AD progression and age of onset, while expression of neuronal isoforms of *BINI* were associated with duration of the disease (Karch et al., 2012). Further evidence in post-mortem brain tissue shows neuronal isoforms of *BINI* decreased in AD (Holler et al., 2014). Novel computational strategies enabled Marques-Coelho et al. to perform differential gene expression analysis of several genes including *BINI* and determine that *BINI* expression in different cell types is capable of isoform switching in early- and late-stage AD brains (Marques-Coelho et al., 2021).

*BINI* expression correlates with Tau pathology (Holler et al., 2014) and BIN1 protein may interact directly with Tau, although this interaction may be disrupted by Tau phosphorylation (Sottejeau et al., 2015). This further underscores a role for *BINI* in molecular regulation of Tau. Axonal sorting of BACE1 by BIN1 can alter whether A $\beta$  is generated in the somatodendritic or axonal compartments in neurons (Ubelmann et al., 2017). Together, this work suggests that *BINI* may impact both neuropathologies of AD, A $\beta$  and hyperphosphorylated Tau. However, other studies have reported no association between BIN1 with neurofibrillary tangles and senile plaques (De Rossi et al., 2016) and shown that reduction of *BINI* expression does not regulate A $\beta$  in 5XFAD mice (Andrew et al., 2019).

Interestingly, Crotti et al., showed that AD-associated SNPs in *BINI* may promote increased BIN1 expression in microglia and that this, in turn, may affect expression of chaperones that participate in Tau clearance as well as increased release of tau in extracellular vesicles (Crotti et al., 2019). This data further implicates microglia in Tau spreading and could indicate a cell-type specific role for altered expression of *BINI*. Exosomes released from neurons may also be involved in cellular crosstalk between neurons and microglia, thereby influencing pathophysiological functions of microglia in the brain. Indeed, McAvoy et al. recently highlighted non-cell autonomous effects of neuronal BIN1 on microglia demonstrating that genetic ablation of *BINI* in excitatory neurons modulates the transcriptome of microglia, activating pathways involved in neuroinflammation and reactive-oxygen species production (McAvoy et al., 2019). However, whether this effect was mediated via exosomes was not studied. *BINI* is highly expressed in other brain cell types such as oligodendrocytes and plays a role in myelination (De Rossi et al., 2016). It is not known if this is mediated through an endocytic mechanism but this concept will be interesting to explore because intracellular trafficking of myelin through the ELN within oligodendrocytes is crucial for the process of myelination (Winterstein et al., 2008).



### 4.3. CD2AP

CD2AP was described in 1998 as an adaptor protein; initially its proposed role was to link specific adhesion receptors to the cytoskeleton of the cell (Dustin et al., 1998; Tao et al., 2019). Further studies demonstrated a role for *CD2AP* in endocytosis and endosome morphology in conjunction with RAB4, a marker of fast recycling endosomes (Cormont et al., 2003), growth-factor induced endocytosis (Kobayashi et al., 2004), and lysosomal trafficking (Furusawa et al., 2019). Beginning in 2011, multiple studies have linked *CD2AP* to increased AD risk (Hollingworth et al., 2011; Kamboh et al., 2012), including a recent GWAS of AD risk loci in African American individuals (Kunkle et al., 2021). Functionally, reduction of expression of *CD2AP* reduces A $\beta$  peptides and lowers the A $\beta$ 42:40 ratio in neuroblastoma cell lines and transgenic mice (Liao et al., 2015). Interestingly, *CD2AP* was also shown to impact APP degradation where reduced expression of *CD2AP* in neurons led to accumulation of APP in early endosomes (Ubelmann et al., 2017) preventing local degradation by lysosomes. Furthermore, overexpression of *CD2AP* increased APP colocalization in late endosomes, stimulating APP degradation (Furusawa et al., 2019). These findings are consistent with a model in which CD2AP protein could function to increase the degradation rate of APP, thus reducing amyloidogenic cleavage. *CD2AP* is ubiquitously expressed and is also implicated in non-neuronal mechanisms that contribute to AD pathogenesis. *CD2AP* expression is enriched in brain microvascular endothelial cells (Dustin et al., 1998) and Cochran et al., found reduced blood-brain barrier (BBB) integrity in *CD2AP* null mice (Cochran et al., 2015). *CD2AP* may also play a role in immune function and immune cell migration (Srivatsan et al., 2013). Microglia also express *CD2AP* (Tao et al., 2019) and, as endocytosis and phagocytosis proceed through similar cellular mechanisms, it is plausible that *CD2AP* could regulate microglial function as well, although this has not yet been directly tested. However, these studies indicate that *CD2AP* could have distinct roles in CNS cells that impact multiple aspects of AD pathogenesis.

### 4.4. PICALM

*PICALM* was identified in 1996 as CALM (Clathrin-assembly lymphoid myeloid), a gene involved in a chromosomal translocation found in leukemias (Dreyling et al., 1996). Further studies demonstrated that *PICALM* is a clathrin-binding protein that mediates endocytosis and trafficking between the TGN and endosomes (Tebar et al., 1999). Mutations in *PICALM* that result in truncated proteins led to defects in hematopoiesis, iron metabolism, and bone growth in mice, underscoring the importance of endocytic function in these processes (Klebig et al., 2003). GWAS first associated *PICALM* with AD in 2009 (Harold et al., 2009) and this association has been replicated in several studies (Corneveaux et al., 2010; Jun et al., 2010). In 2013, Ando et al., provided immunohistochemical evidence of increased *PICALM* expression in the AD brain. Interestingly *PICALM* was highly upregulated in both SAD and FAD neurons, but only upregulated in SAD microglia (Ando et al., 2013). Furthermore, they documented a high association of *PICALM* protein with neurofibrillary tangles but not with amyloid plaques (Ando et al., 2013). Recent work using both baker's yeast and human induced pluripotent stem cell (hiPSC) models has uncovered high conservation between *PICALM* and its yeast homolog Yap1802p. This study showed that expression of human APOE  $\epsilon$ 4 in yeast induced EE abnormalities including disrupted localization of the RAB5 homologs in yeast, YPT5 and VPS21. They also showed

that hiPSC-derived astrocytes expressing APOE  $\epsilon$ 4 had reduced EEA1 levels and reduced internalization of the cell surface ligands transferrin and the epidermal growth factor (EGF). In both models, the endosomal defects were rescued by increasing expression of PICALM/Yap1802p(Narayan et al., 2020). This study shows a functional interaction between AD-linked genes, APOE  $\epsilon$ 4 and PICALM, and highlights the biological conservation of endocytic processes across organisms.

#### 4.5. Ephrin receptors

Ephrin receptors belong to a large receptor tyrosine kinase family that is involved in interactions between cells and the extracellular matrix to regulate morphology and cellular behavior(Yamazaki et al., 2009). Ephrin receptors are endocytosed, sometimes as a complex, to regulate signaling events necessary for proper synapse formation and maturation in neurons(Pitulescu and Adams, 2010). Common variants in *EPHA1* have been associated with increase AD risk by GWAS(Hollingworth et al., 2011) and replicated in a case-control study(Carrasquillo et al., 2011). Several models show reduction of Ephrin receptors in AD mouse models(Simon et al., 2009) and increasing expression of these receptors may rescue A $\beta$ -induced neurotoxicity(Geng et al., 2013). Although not associated with AD by GWAS, EphA4 receptor levels were shown to be decreased in the hippocampus of patients with AD(Simon et al., 2009). Interestingly, a more recent study showed that the AD-risk gene *SORL1/SORLA* interacts with the EphA4 receptor and attenuates A $\beta$ -mediated EphA4 activation(Huang et al., 2017) thus impacting neurotoxicity and synaptic function. Together this work reinforces the connections between endocytic trafficking and neuronal synaptic function.

### 5. Human models for AD risk genes

For many genes implicated in AD risk, cell type specific expression and function will be both a biological phenomenon and a challenge for therapeutic development. Recent studies have identified cell-type specific expression quantitative trait loci in blood and brain(Patel et al., 2021)as well as AD risk enhancers that may regulate cell-type specific expression(Novikova et al., 2021). Gene expression is important to consider outside of the CNS as well, as many GWAS disease variants are enriched in the brain vasculature(Yang et al., 2021). The ELN is an essential cellular pathway that is present in every cell type. However, the function of the ELN and the expression of the genes and proteins that regulate it may be very different in the diverse cell types of the CNS. This challenge impacts both basic mechanistic studies, which need to perform experiments in relevant cell types, as well as pre-clinical drug testing studies, which need to apply potential therapeutic compounds to relevant cell types. To fully understand the biology of AD risk genes, including those that function in the ELN, we require dynamic cellular models to understand the mechanisms that lead to neurodegeneration. hiPSCs derived from patients, controls, or autopsy tissue have provided insights into disease pathogenesis in brain-relevant cell types. For the past ~15 years, hiPSC-derived neural cell models have uncovered important insights into disease mechanism in both familial AD (FAD) and sporadic AD (SAD) models(Mungenast et al., 2016; Penney et al., 2020; Qian and Tcw, 2021; Raman et al., 2020; Riemens et al., 2020). The inherent power in this model is the ability to finely manipulate genomic

variants that are associated with disease risk via genome editing as well as generate cell lines that encompass the genomes of AD patients, which can begin to ascertain the biological effects of polygenic disease risk. hiPSCs can be differentiated into all cell types of the central nervous system and periphery, thus generating dynamic living models to visualize human disease pathogenesis (See Fig. 1). This technology is increasingly important as we uncover more evidence of cell-type specific expression of genes. Multiple protocols exist to generate cell types of interest, although as with any lab model, certain limitations exist. These limitations include immaturity of cells and lack of exact ratios of cell types to mimic the human brain. However, advances in differentiation technology, direct conversion or transdifferentiation of cells, and organoid technology are moving to overcome these challenges (D'Souza et al., 2021). For SAD in particular, given its genetic complexity, understanding the biological underpinnings of how ELN genes increase AD risk is crucial. Although GWAS-identified loci are common and have low-effect sizes in a population, subtle changes in ELN function may have strong cellular effects in human neural cells over the course of a decades-long disease such as AD. Cellular systems, both patient-derived and gene-edited, can identify early phenotypes due to AD risk genes, prior to modification by the environment. This is particularly important in understanding early stages of AD pathogenesis and identifying which processes are potentially modifiable for therapeutic development.

## 6. Conclusion

Understanding the genetic underpinnings of late-onset, sporadic AD and related dementias (ADRD) is a challenging and rapidly progressing field. Recently, large GWAS study has reported 42 novel loci linked to increased risk for ADRD (Bellenguez, 2020) which include novel hits in ELN-related genes, including SORT1 (Sortilin), SNX1 (Sorting nexin 1) and CTSB (Cathepsin B) among others. We fully expect that future studies will continue to implicate the ELN as a relevant biological pathway. The ELN is a highly conserved network, essential for proper function of all cell types. Disruptions to the ELN are apparent in neurodegenerative disorders, underscoring the importance of protein trafficking in the brain. As we have discussed above, ELN genes associated with increased AD risk have complex and often cell-type specific roles, some of which are integrated with AD neuropathology and some of which are independent of amyloid or Tau pathology. Despite this complexity, alteration in ELN function are likely early events in disease pathogenesis, making it an attractive target for development of disease-modifying therapeutics.

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## References

- Alzheimer's disease facts and figures. *Alzheimers Dement*, 2020.
- Andersen OM, Reiche J, Schmidt V, Gotthardt M, Spoelgen R, Behlke J, von Arnim CA, Breiderhoff T, Jansen P, Wu X, et al. , 2005. Neuronal sorting protein-related receptor sorLA/LR11 regulates processing of the amyloid precursor protein. *Proc. Natl. Acad. Sci. U. S. A* 102, 13461–13466. [PubMed: 16174740]

- Andersen OM, Rudolph IM, Willnow TE, 2016. Risk factor SORL1: from genetic association to functional validation in Alzheimer's disease. *Acta Neuropathol.* 132, 653–665. [PubMed: 27638701]
- Ando K, Brion JP, Stygelbout V, Suain V, Authelet M, Dedecker R, Chanut A, Lacor P, Lavour J, Sazdovitch V, et al. , 2013. Clathrin adaptor CALM/PICALM is associated with neurofibrillary tangles and is cleaved in Alzheimer's brains. *Acta Neuropathol.* 125, 861–878. [PubMed: 23589030]
- Andrew RJ, De Rossi P, Nguyen P, Kowalski HR, Recupero AJ, Guerbette T, Krause SV, Rice RC, Laury-Kleintop L, Wagner SL, et al. , 2019. Reduction of the expression of the late-onset Alzheimer's disease (AD) risk-factor BIN1 does not affect amyloid pathology in an AD mouse model. *J. Biol. Chem* 294, 4477–4487. [PubMed: 30692199]
- Andrews SJ, Fulton-Howard B, Goate A, 2020. Interpretation of risk loci from genome-wide association studies of Alzheimer's disease. *Lancet Neurol.* 19, 326–335. [PubMed: 31986256]
- Behnia R, Munro S, 2005. Organelle identity and the signposts for membrane traffic. *Nature* 438, 597–604. [PubMed: 16319879]
- Bellenguez CEA, 2020. New insights on the genetic etiology of Alzheimer's and related dementia. medRxiv. 10.1101/2020.10.0120200659.
- Bhalla A, Vetanovetz CP, Morel E, Chamoun Z, Di Paolo G, Small SA, 2012. The location and trafficking routes of the neuronal retromer and its role in amyloid precursor protein transport. *Neurobiol. Dis* 47, 126–134. [PubMed: 22516235]
- Bis JC, Jian X, Kunkle BW, Chen Y, Hamilton-Nelson KL, Bush WS, Salerno WJ, Lancour D, Ma Y, Renton AE, et al. , 2020. Whole exome sequencing study identifies novel rare and common Alzheimer's-associated variants involved in immune response and transcriptional regulation. *Mol. Psychiatry* 25, 1859–1875. [PubMed: 30108311]
- Bonifacino JS, Rojas R, 2006. Retrograde transport from endosomes to the trans-Golgi network. *Nat. Rev. Mol. Cell Biol* 7, 568–579. [PubMed: 16936697]
- Bucci C, Thomsen P, Nicoziani P, McCarthy J, van Deurs B, 2000. Rab7: a key to lysosome biogenesis. *Mol. Biol. Cell* 11, 467–480. [PubMed: 10679007]
- Caglayan S, Takagi-Niidome S, Liao F, Carlo AS, Schmidt V, Burgert T, Kitago Y, Fuchtbauer EM, Fuchtbauer A, Holtzman DM, et al. , 2014. Lysosomal sorting of amyloid-beta by the SORLA receptor is impaired by a familial Alzheimer's disease mutation. *Sci. Transl. Med* 6, 223ra220.
- Calafate S, Flavin W, Verstreken P, Moechars D, 2016. Loss of Bin1 promotes the propagation of tau pathology. *Cell Rep.* 17, 931–940. [PubMed: 27760323]
- Cappa SF, 2018. The quest for an Alzheimer therapy. *Front. Neurol* 9, 108. [PubMed: 29599741]
- Carrasquillo MM, Belbin O, Hunter TA, Ma L, Bisceglia GD, Zou F, Crook JE, Pankratz VS, Sando SB, Aasly JO, et al. , 2011. Replication of EPHA1 and CD33 associations with late-onset Alzheimer's disease: a multi-Centre case-control study. *Mol. Neurodegener* 6, 54. [PubMed: 21798052]
- Cataldo AM, Nixon RA, 1990. Enzymatically active lysosomal proteases are associated with amyloid deposits in Alzheimer brain. *Proc. Natl. Acad. Sci. U. S. A* 87, 3861–3865. [PubMed: 1692625]
- Cataldo AM, Paskevich PA, Kominami E, Nixon RA, 1991. Lysosomal hydrolases of different classes are abnormally distributed in brains of patients with Alzheimer disease. *Proc. Natl. Acad. Sci. U. S. A* 88, 10998–11002. [PubMed: 1837142]
- Cataldo AM, Hamilton DJ, Barnett JL, Paskevich PA, Nixon RA, 1996. Properties of the endosomal-lysosomal system in the human central nervous system: disturbances mark most neurons in populations at risk to degenerate in Alzheimer's disease. *J. Neurosci* 16, 186–199. [PubMed: 8613784]
- Cataldo AM, Barnett JL, Pieroni C, Nixon RA, 1997. Increased neuronal endocytosis and protease delivery to early endosomes in sporadic Alzheimer's disease: neuropathologic evidence for a mechanism of increased beta-amyloidogenesis. *J. Neurosci* 17, 6142–6151. [PubMed: 9236226]
- Cataldo AM, Peterhoff CM, Troncoso JC, Gomez-Isla T, Hyman BT, Nixon RA, 2000. Endocytic pathway abnormalities precede amyloid beta deposition in sporadic Alzheimer's disease and down syndrome: differential effects of APOE genotype and presenilin mutations. *Am. J. Pathol* 157, 277–286. [PubMed: 10880397]

- Cataldo AM, Petanceska S, Terio NB, Peterhoff CM, Durham R, Mercken M, Mehta PD, Buxbaum J, Haroutunian V, Nixon RA, 2004. Abeta localization in abnormal endosomes: association with earliest Abeta elevations in AD and down syndrome. *Neurobiol. Aging* 25, 1263–1272. [PubMed: 15465622]
- Chai YL, Chong JR, Weng J, Howlett D, Halsey A, Lee JH, Attems J, Aarsland D, Francis PT, Chen CP, et al. , 2019. Lysosomal cathepsin D is upregulated in Alzheimer's disease neocortex and may be a marker for neurofibrillary degeneration. *Brain Pathol.* 29, 63–74. [PubMed: 30051532]
- Cheng XT, Xie YX, Zhou B, Huang N, Farfel-Becker T, Sheng ZH, 2018. Characterization of LAMP1-labeled nondegradative lysosomal and endocytic compartments in neurons. *J. Cell Biol.* 217, 3127–3139. [PubMed: 29695488]
- Chieregatti E, Meldolesi J, 2005. Regulated exocytosis: new organelles for non-secretory purposes. *Nat. Rev. Mol. Cell Biol* 6, 181–187. [PubMed: 15688003]
- Choudhury R, Diao A, Zhang F, Eisenberg E, Saint-Pol A, Williams C, Konstantakopoulos A, Lucocq J, Johannes L, Rabouille C, et al. , 2005. Lowe syndrome protein OCRL1 interacts with clathrin and regulates protein trafficking between endosomes and the trans-Golgi network. *Mol. Biol. Cell* 16, 3467–3479. [PubMed: 15917292]
- Cochran JN, Rush T, Buckingham SC, Roberson ED, 2015. The Alzheimer's disease risk factor CD2AP maintains blood-brain barrier integrity. *Hum. Mol. Genet* 24, 6667–6674. [PubMed: 26358779]
- Collaborators GBDD, 2019. Global, regional, and national burden of Alzheimer's disease and other dementias, 1990-2016: a systematic analysis for the global burden of disease study 2016. *Lancet Neurol.* 18, 88–106. [PubMed: 30497964]
- Cormont M, Meton I, Mari M, Monzo P, Keslair F, Gaskin C, McGraw TE, Le Marchand-Brustel Y, 2003. CD2AP/CMS regulates endosome morphology and traffic to the degradative pathway through its interaction with Rab4 and c-Cbl. *Traffic* 4, 97–112. [PubMed: 12559036]
- Corneveaux JJ, Myers AJ, Allen AN, Pruzin JJ, Ramirez M, Engel A, Nalls MA, Chen K, Lee W, Chewning K, et al. , 2010. Association of CR1, CLU and PICALM with Alzheimer's disease in a cohort of clinically characterized and neuropathologically verified individuals. *Hum. Mol. Genet.* 19, 3295–3301. [PubMed: 20534741]
- Crotti A, Sait HR, McAvoy KM, Estrada K, Ergun A, Szak S, Marsh G, Jandreski L, Peterson M, Reynolds TL, et al. , 2019. BIN1 favors the spreading of tau via extracellular vesicles. *Sci. Rep* 9, 9477. [PubMed: 31263146]
- Cummings J, Lee G, Ritter A, Sabbagh M, Zhong K, 2019. Alzheimer's disease drug development pipeline: 2019. *Alzheimers Dement (N Y)* 5, 272–293. [PubMed: 31334330]
- Das U, Wang L, Ganguly A, Saikia JM, Wagner SL, Koo EH, Roy S, 2016. Visualizing APP and BACE-1 approximation in neurons yields insight into the amyloidogenic pathway. *Nat. Neurosci* 19, 55–64. [PubMed: 26642089]
- Daumke O, Roux A, Haucke V, 2014. BAR domain scaffolds in dynamin-mediated membrane fission. *Cell* 156, 882–892. [PubMed: 24581490]
- De Rossi P, Buggia-Prevot V, Clayton BL, Vasquez JB, van Sanford C, Andrew RJ, Lesnick R, Botte A, Deyts C, Salem S, et al. , 2016. Predominant expression of Alzheimer's disease-associated BIN1 in mature oligodendrocytes and localization to white matter tracts. *Mol. Neurodegener* 11, 59. [PubMed: 27488240]
- Deaton CA, Johnson GVW, 2020. Presenilin 1 regulates membrane homeostatic pathways that are dysregulated in Alzheimer's disease. *J. Alzheimers Dis* 77, 961–977. [PubMed: 32804090]
- Dodson SE, Gearing M, Lippa CF, Montine TJ, Levey AI, Lah JJ, 2006. LR11/ SorLA expression is reduced in sporadic Alzheimer disease but not in familial Alzheimer disease. *J. Neuropathol. Exp. Neurol* 65, 866–872. [PubMed: 16957580]
- Dreyling MH, Martinez-Climent JA, Zheng M, Mao J, Rowley JD, Bohlander SK, 1996. The t(10;11)(p13;q14) in the U937 cell line results in the fusion of the AF10 gene and CALM, encoding a new member of the AP-3 clathrin assembly protein family. *Proc. Natl. Acad. Sci. U. S. A* 93, 4804–4809. [PubMed: 8643484]



- D'Souza GX, Rose SE, Knupp A, Nicholson DA, Keene CD, Young JE, 2021. The application of in vitro-derived human neurons in neurodegenerative disease modeling. *J. Neurosci. Res* 99, 124–140. [PubMed: 32170790]
- Dustin ML, Olszowy MW, Holdorf AD, Li J, Bromley S, Desai N, Widder P, Rosenberger F, van der Merwe PA, Allen PM, et al. , 1998. A novel adaptor protein orchestrates receptor patterning and cytoskeletal polarity in T-cell contacts. *Cell* 94, 667–677. [PubMed: 9741631]
- Fedeli C, Filadi R, Rossi A, Mammucari C, Pizzo P, 2019. PSEN2 (presenilin 2) mutants linked to familial Alzheimer disease impair autophagy by altering ca(2+) homeostasis. *Autophagy* 15, 2044–2062. [PubMed: 30892128]
- Ferreira A, Caceres A, Kosik KS, 1993. Intraneuronal compartments of the amyloid precursor protein. *J. Neurosci* 13, 3112–3123. [PubMed: 8331388]
- Fjorback AW, Seaman M, Gustafsen C, Mehmedbasic A, Gokool S, Wu C, Militz D, Schmidt V, Madsen P, Nyengaard JR, et al. , 2012. Retromer binds the FANSHY sorting motif in SorLA to regulate amyloid precursor protein sorting and processing. *J. Neurosci* 32, 1467–1480. [PubMed: 22279231]
- Fu WY, Chen Y, Sahin M, Zhao XS, Shi L, Bikoff JB, Lai KO, Yung WH, Fu AK, Greenberg ME, et al. , 2007. Cdk5 regulates EphA4-mediated dendritic spine retraction through an ephexin1-dependent mechanism. *Nat. Neurosci* 10, 67–76. [PubMed: 17143272]
- Furusawa K, Takasugi T, Chiu YW, Hori Y, Tomita T, Fukuda M, Hisanaga SI, 2019. CD2-associated protein (CD2AP) overexpression accelerates amyloid precursor protein (APP) transfer from early endosomes to the lysosomal degradation pathway. *J. Biol. Chem* 294, 10886–10899. [PubMed: 31138646]
- Gao S, Casey AE, Sargeant TJ, Makinen VP, 2018. Genetic variation within endolysosomal system is associated with late-onset Alzheimer's disease. *Brain* 141, 2711–2720. [PubMed: 30124770]
- Gatica D, Lahiri V, Klionsky DJ, 2018. Cargo recognition and degradation by selective autophagy. *Nat. Cell Biol* 20, 233–242. [PubMed: 29476151]
- Gautreau A, Oguievetkaia K, Ungermann C, 2014. Function and regulation of the endosomal fusion and fission machineries. *Cold Spring Harb. Perspect. Biol* 6.
- Geng Z, Xu FY, Huang SH, Chen ZY, 2011. Sorting protein-related receptor SorLA controls regulated secretion of glial cell line-derived neurotrophic factor. *J. Biol. Chem* 286, 41871–41882. [PubMed: 21994944]
- Geng D, Kang L, Su Y, Jia J, Ma J, Li S, Du J, Cui H, 2013. Protective effects of EphB2 on Abeta1-42 oligomer-induced neurotoxicity and synaptic NMDA receptor signaling in hippocampal neurons. *Neurochem. Int* 63, 283–290. [PubMed: 23831214]
- Glerup S, Lume M, Olsen D, Nyengaard JR, Vaegter CB, Gustafsen C, Christensen EI, Kjolby M, Hay-Schmidt A, Bender D, et al. , 2013. SorLA controls neurotrophic activity by sorting of GDNF and its receptors GFRalpha1 and RET. *Cell Rep.* 3, 186–199. [PubMed: 2333276]
- Graham WV, Bonito-Oliva A, Sakmar TP, 2017. Update on Alzheimer's disease therapy and prevention strategies. *Annu. Rev. Med* 68, 413–430. [PubMed: 28099083]
- Haass C, Kaether C, Thinakaran G, Sisodia S, 2012. Trafficking and proteolytic processing of APP. *Cold Spring Harb. Perspect Med* 2, a006270. [PubMed: 22553493]
- Harold D, Abraham R, Hollingworth P, Sims R, Gerrish A, Hamshere ML, Pahwa JS, Moskva V, Dowzell K, Williams A, et al. , 2009. Genome-wide association study identifies variants at CLU and PICALM associated with Alzheimer's disease. *Nat. Genet.* 41, 1088–1093. [PubMed: 19734902]
- Hartl D, Nebrich G, Klein O, Stephanowitz H, Krause E, Rohe M, 2016. SORLA regulates calpain-dependent degradation of synapsin. *Alzheimers Dement.* 12, 952–963. [PubMed: 27021222]
- Hessvik NP, Llorente A, 2018. Current knowledge on exosome biogenesis and release. *Cell. Mol. Life Sci* 75, 193–208. [PubMed: 28733901]
- Holler CJ, Davis PR, Beckett TL, Platt TL, Webb RL, Head E, Murphy MP, 2014. Bridging integrator 1 (BIN1) protein expression increases in the Alzheimer's disease brain and correlates with neurofibrillary tangle pathology. *J. Alzheimers Dis* 42, 1221–1227. [PubMed: 25024306]
- Hollingworth P, Harold D, Sims R, Gerrish A, Lambert JC, Carrasquillo MM, Abraham R, Hamshere ML, Pahwa JS, Moskva V, et al. , 2011. Common variants at ABCA7, MS4A6A/MS4A4E,



EPHA1, CD33 and CD2AP are associated with Alzheimer's disease. *Nat. Genet* 43, 429–435. [PubMed: 21460840]

Holstege H, van der Lee SJ, Hulsman M, Wong TH, van Rooij JG, Weiss M, Louwersheimer E, Wolters FJ, Amin N, Uitterlinden AG, et al. , 2017. Characterization of pathogenic SORL1 genetic variants for association with Alzheimer's disease: a clinical interpretation strategy. *Eur. J. Hum. Genet* 25, 973–981. [PubMed: 28537274]

Hu YB, Dammer EB, Ren RJ, Wang G, 2015. The endosomal-lysosomal system: from acidification and cargo sorting to neurodegeneration. *Transl. Neurodegener* 4, 18. [PubMed: 26448863]

Huang TY, Zhao Y, Jiang LL, Li X, Liu Y, Sun Y, Pina-Crespo JC, Zhu B, Maslah E, Willnow TE, et al. , 2017. SORLA attenuates EphA4 signaling and amyloid beta-induced neurodegeneration. *J. Exp. Med* 214, 3669–3685. [PubMed: 29114064]

Hung COY, Livesey FJ, 2018. Altered gamma-secretase processing of APP disrupts lysosome and autophagosome function in monogenic Alzheimer's disease. *Cell Rep.* 25 (3647–3660), e3642.

Hung C, Tuck E, Stubbs V, van der Lee SJ, Aalfs C, van Spaendonk R, Scheltens P, Hardy J, Holstege H, Livesey FJ, 2021. SORL1 deficiency in human excitatory neurons causes APP-dependent defects in the endolysosome-autophagy network. *Cell Rep.* 35, 109259. [PubMed: 34133918]

Huotari J, Helenius A, 2011. Endosome maturation. *EMBO J.* 30, 3481–3500. [PubMed: 21878991]

Hyttinen JM, Niittykoski M, Salminen A, Kaarniranta K, 2013. Maturation of autophagosomes and endosomes: a key role for Rab7. *Biochim. Biophys. Acta* 1833, 503–510. [PubMed: 23220125]

Inpanathan S, Botelho RJ, 2019. The lysosome Signaling platform: adapting with the times. *Front. Cell Dev. Biol* 7, 113. [PubMed: 31281815]

Jacobsen L, Madsen P, Moestrup SK, Lund AH, Tommerup N, Nykjaer A, Sottrup-Jensen L, Gliemann J, Petersen CM, 1996. Molecular characterization of a novel human hybrid-type receptor that binds the alpha2-macroglobulin receptor-associated protein. *J. Biol. Chem.* 271, 31379–31383. [PubMed: 8940146]

Jacobsen L, Madsen P, Jacobsen C, Nielsen MS, Gliemann J, Petersen CM, 2001. Activation and functional characterization of the mosaic receptor SorLA/LR11. *J. Biol. Chem* 276, 22788–22796. [PubMed: 11294867]

Jansen IE, Savage JE, Watanabe K, Bryois J, Williams DM, Steinberg S, Sealock J, Karlsson IK, Hagg S, Athanasiu L, et al. , 2019. Genome-wide metaanalysis identifies new loci and functional pathways influencing Alzheimer's disease risk. *Nat. Genet* 51, 404–413. [PubMed: 30617256]

Jun G, Naj AC, Beecham GW, Wang LS, Buros J, Gallins PJ, Buxbaum JD, Ertekin-Taner N, Fallin MD, Friedland R, et al. , 2010. Meta-analysis confirms CR1, CLU, and PICALM as Alzheimer disease risk loci and reveals interactions with APOE genotypes. *Arch. Neurol* 67, 1473–1484. [PubMed: 20697030]

Kadlecova Z, Spielman SJ, Loerke D, Mohanakrishnan A, Reed DK, Schmid SL, 2017. Regulation of clathrin-mediated endocytosis by hierarchical allosteric activation of AP2. *J. Cell Biol* 216, 167–179. [PubMed: 28003333]

Kamboh MI, Demirci FY, Wang X, Minster RL, Carrasquillo MM, Pankratz VS, Younkin SG, Saykin AJ, Alzheimer's Disease Neuroimaging I, Jun G, et al. , 2012. Genome-wide association study of Alzheimer's disease. *Transl. Psychiatry* 2, e117. [PubMed: 22832961]

Karch CM, Goate AM, 2015. Alzheimer's disease risk genes and mechanisms of disease pathogenesis. *Biol. Psychiatry* 77, 43–51. [PubMed: 24951455]

Karch CM, Jeng AT, Nowotny P, Cady J, Cruchaga C, Goate AM, 2012. Expression of novel Alzheimer's disease risk genes in control and Alzheimer's disease brains. *PLoS One* 7, e50976. [PubMed: 23226438]

Kaur G, Lakkaraju A, 2018. Early endosome morphology in health and disease. *Adv. Exp. Med. Biol* 1074, 335–343. [PubMed: 29721961]

Kelly BT, Graham SC, Liska N, Dannhauser PN, Honing S, Ungewickell EJ, Owen DJ, 2014. Clathrin adaptors. AP2 controls clathrin polymerization with a membrane-activated switch. *Science* 345, 459–463. [PubMed: 25061211]

Kim JW, Jung SY, Kim Y, Heo H, Hong CH, Seo SW, Choi SH, Son SJ, Lee S, Chang J, 2021. Identification of Cathepsin D as a plasma biomarker for Alzheimer's disease. *Cells* 10. [PubMed: 35011571]

- Klebig ML, Wall MD, Potter MD, Rowe EL, Carpenter DA, Rinchik EM, 2003. Mutations in the clathrin-assembly gene *Picalm* are responsible for the hematopoietic and iron metabolism abnormalities in *fit1* mice. *Proc. Natl. Acad. Sci. U. S. A* 100, 8360–8365. [PubMed: 12832620]
- Klumperman J, Raposo G, 2014. The complex ultrastructure of the endolysosomal system. *Cold Spring Harb. Perspect. Biol* 6, a016857. [PubMed: 24851870]
- Knupp A, Mishra S, Martinez R, Braggin JE, Szabo M, Kinoshita C, Hailey DW, Small SA, Jayadev S, Young JE, 2020. Depletion of the AD risk gene *SORL1* selectively impairs neuronal endosomal traffic independent of amyloidogenic APP processing. *Cell Rep.* 31, 107719. [PubMed: 32492427]
- Kobayashi S, Sawano A, Nojima Y, Shibuya M, Maru Y, 2004. The c-Cbl/CD2AP complex regulates VEGF-induced endocytosis and degradation of Flt-1 (VEGFR-1). *FASEB J.* 18, 929–931. [PubMed: 15001553]
- Kumari S, Mg S, Mayor S, 2010. Endocytosis unplugged: multiple ways to enter the cell. *Cell Res.* 20, 256–275. [PubMed: 20125123]
- Kunkle BW, Grenier-Boley B, Sims R, Bis JC, Damotte V, Naj AC, Boland A, Vronskaya M, van der Lee SJ, Amlie-Wolf A, et al. , 2019. Genetic meta-analysis of diagnosed Alzheimer’s disease identifies new risk loci and implicates *Abeta*, tau, immunity and lipid processing. *Nat. Genet* 51, 414–430. [PubMed: 30820047]
- Kunkle BW, Schmidt M, Klein HU, Naj AC, Hamilton-Nelson KL, Larson EB, Evans DA, De Jager PL, Crane PK, Buxbaum JD, et al. , 2021. Novel Alzheimer disease risk loci and pathways in African American individuals using the African genome resources panel: a meta-analysis. *JAMA Neurol.* 78, 102–113. [PubMed: 33074286]
- Kwart D, Gregg A, Scheckel C, Murphy EA, Paquet D, Duffield M, Fak J, Olsen O, Darnell RB, Tessier-Lavigne M, 2019. A large panel of isogenic APP and PSEN1 mutant human iPSC neurons reveals shared endosomal abnormalities mediated by APP beta-CTFs. *Not. Abeta. Neuron* 104 (256–270), e255.
- La Rosa LR, Perrone L, Nielsen MS, Calissano P, Andersen OM, Matrone C, 2015. Y682G mutation of amyloid precursor protein promotes Endo-lysosomal dysfunction by disrupting APP-SorLA interaction. *Front. Cell. Neurosci* 9, 109. [PubMed: 25904844]
- Lambert JC, Ibrahim-Verbaas CA, Harold D, Naj AC, Sims R, Bellenguez C, DeStafano AL, Bis JC, Beecham GW, Grenier-Boley B, et al. , 2013. Meta-analysis of 74,046 individuals identifies 11 new susceptibility loci for Alzheimer’s disease. *Nat. Genet.* 45, 1452–1458. [PubMed: 24162737]
- Lambert ESO, Landeira B, Melo de Farias A, Hermant X, Carrier A, Pelletier A, Davoine L, Dupont C, Amouyel P, Lafone F, Adelfettah F, Verstreken P, Chapuis J, Barois N, Delahaye F, Dermaut B, Lambert JC, Costa M, Dourlent P, 2021. The Alzheimer’s disease genetic risk factor *bin1* induces isoform-dependent neurotoxicity through early endosome defects. *Biorxiv.* 10.1101/2021.04.02.438184.
- Lee JH, Yu WH, Kumar A, Lee S, Mohan PS, Peterhoff CM, Wolfe DM, Martinez-Vicente M, Massey AC, Sovak G, et al. , 2010. Lysosomal proteolysis and autophagy require presenilin 1 and are disrupted by Alzheimer-related PS1 mutations. *Cell* 141, 1146–1158. [PubMed: 20541250]
- Liao F, Jiang H, Srivatsan S, Xiao Q, Lefton KB, Yamada K, Mahan TE, Lee JM , Shaw AS, Holtzman DM, 2015. Effects of CD2-associated protein deficiency on amyloid-beta in neuroblastoma cells and in an APP transgenic mouse model. *Mol. Neurodegener* 10, 12. [PubMed: 25887956]
- Luzio JP, Pryor PR, Bright NA, 2007. Lysosomes: fusion and function. *Nat. Rev. Mol. Cell Biol* 8, 622–632. [PubMed: 17637737]
- Luzio JP, Hackmann Y, Dieckmann NM, Griffiths GM, 2014. The biogenesis of lysosomes and lysosome-related organelles. *Cold Spring Harb. Perspect. Biol* 6, a016840. [PubMed: 25183830]
- Marioni RE, Harris SE, Zhang Q, McRae AF, Hagenaars SP, Hill WD, Davies G, Ritchie CW, Gale CR, Starr JM, et al. , 2018. GWAS on family history of Alzheimer’s disease. *Transl. Psychiatry* 8, 99. [PubMed: 29777097]
- Marques-Coelho D, Iohan L, Melo de Farias AR, Flaig A, Brainbank Neuro CEBNN , Lambert JC, Costa MR, 2021. Differential transcript usage unravels gene expression alterations in Alzheimer’s disease human brains. *NPJ Aging Mech. Dis* 7, 2. [PubMed: 33398016]
- Marsh M, McMahon HT, 1999. The structural era of endocytosis. *Science* 285, 215–220. [PubMed: 10398591]

- Masters CL, Bateman R, Blennow K, Rowe CC, Sperling RA, Cummings JL, 2015. Alzheimer's disease. *Nat. Rev. Dis. Primers* 1, 15056. [PubMed: 27188934]
- Mathieu M, Martin-Jaular L, Lavieu G, Thery C, 2019. Specificities of secretion and uptake of exosomes and other extracellular vesicles for cell-to-cell communication. *Nat. Cell Biol* 21, 9–17. [PubMed: 30602770]
- Maxfield FR, McGraw TE, 2004. Endocytic recycling. *Nat. Rev. Mol. Cell Biol* 5, 121–132. [PubMed: 15040445]
- Maxfield FR, Yamashiro DJ, 1987. Endosome acidification and the pathways of receptor-mediated endocytosis. *Adv. Exp. Med. Biol* 225, 189–198. [PubMed: 2839960]
- Mayor S, Parton RG, Donaldson JG, 2014. Clathrin-independent pathways of endocytosis. *Cold Spring Harb. Perspect. Biol* 6.
- McAvoy KM, Rajamohamed Sait H, Marsh G, Peterson M, Reynolds TL, Gagnon J, Geisler S, Leach P, Roberts C, Cahir-McFarland E, et al. , 2019. Cell-autonomous and non-cell autonomous effects of neuronal BIN1 loss in vivo. *PLoS One* 14, e0220125. [PubMed: 31408457]
- McPherson PS, Ritter B, Wendland B, 2013. Clathrin-mediated endocytosis. In: *Madame Curie Bioscience Database-NCBI*.
- Mecozzi VJ, Berman DE, Simoes S, Vetanovetz C, Awal MR, Patel VM, Schneider RT, Petsko GA, Ringe D, Small SA, 2014. Pharmacological chaperones stabilize retromer to limit APP processing. *Nat. Chem. Biol* 10, 443–449. [PubMed: 24747528]
- Merrifield CJ, Perrais D, Zenisek D, 2005. Coupling between clathrin-coated-pit invagination, cortactin recruitment, and membrane scission observed in live cells. *Cell* 121, 593–606. [PubMed: 15907472]
- Meyerholz A, Hinrichsen L, Groos S, Esk PC, Brandes G, Ungewickell EJ, 2005. Effect of clathrin assembly lymphoid myeloid leukemia protein depletion on clathrin coat formation. *Traffic* 6, 1225–1234. [PubMed: 16262731]
- Miller SE, Mathiasen S, Bright NA, Pierre F, Kelly BT, Kladt N, Schauss A, Merrifield CJ, Stamou D, Honing S, et al. , 2015. CALM regulates clathrin-coated vesicle size and maturation by directly sensing and driving membrane curvature. *Dev. Cell* 33, 163–175. [PubMed: 25898166]
- Miyagawa T, Ebinuma I, Morohashi Y, Hori Y, Young Chang M, Hattori H, Maehara T, Yokoshima S, Fukuyama T, Tsuji S, et al. , 2016. BIN1 regulates BACE1 intracellular trafficking and amyloid-beta production. *Hum. Mol. Genet* 25, 2948–2958. [PubMed: 27179792]
- Miyashita A, Koike A, Jun G, Wang LS, Takahashi S, Matsubara E, Kawarabayashi T, Shoji M, Tomita N, Arai H, et al. , 2013. SORL1 is genetically associated with late-onset Alzheimer's disease in Japanese, Koreans and Caucasians. *PLoS One* 8, e58618. [PubMed: 23565137]
- Morena F, Argentati C, Trotta R, Crispoltoni L, Stabile A, Pistilli A, di Baldassarre A, Calafiore R, Montanucci P, Basta G, et al. , 2017. A comparison of lysosomal enzymes expression levels in peripheral blood of mild- and severe-Alzheimer's disease and MCI patients: implications for regenerative medicine approaches. *Int. J. Mol. Sci* 18.
- Mungenast AE, Siegert S, Tsai LH, 2016. Modeling Alzheimer's disease with human induced pluripotent stem (iPS) cells. *Mol. Cell. Neurosci* 73, 13–31. [PubMed: 26657644]
- Nakatogawa H, 2020. Mechanisms governing autophagosome biogenesis. *Nat. Rev. Mol. Cell Biol* 21, 439–458. [PubMed: 32372019]
- Narayan P, Sienski G, Bonner JM, Lin YT, Seo J, Baru V, Haque A, Milo B, Akay LA, Graziosi A, et al. , 2020. PICALM rescues endocytic defects caused by the Alzheimer's disease risk factor APOE4. *Cell Rep.* 33, 108224. [PubMed: 33027662]
- Nixon RA, 2017. Amyloid precursor protein and endosomal-lysosomal dysfunction in Alzheimer's disease: inseparable partners in a multifactorial disease. *FASEB J.* 31, 2729–2743. [PubMed: 28663518]
- Nixon RA, Cataldo AM, 1995. The endosomal-lysosomal system of neurons: new roles. *Trends Neurosci.* 18, 489–496. [PubMed: 8592758]
- Novikova G, Kapoor M, Tew J, Abud EM, Efthymiou AG, Chen SX, Cheng H, Fullard JF, Bendl J, Liu Y, et al. , 2021. Integration of Alzheimer's disease genetics and myeloid genomics identifies disease risk regulatory elements and genes. *Nat. Commun* 12, 1610. [PubMed: 33712570]

- Pasqualato S, Senic-Matuglia F, Renault L, Goud B, Salamero J, Cherfils J, 2004. The structural GDP/GTP cycle of Rab11 reveals a novel interface involved in the dynamics of recycling endosomes. *J. Biol. Chem* 279, 11480–11488. [PubMed: 14699104]
- Pasternak SH, Bagshaw RD, Guiral M, Zhang S, Ackerley CA, Pak BJ, Callahan JW, Mahuran DJ, 2003. Presenilin-1, nicastrin, amyloid precursor protein, and gamma-secretase activity are co-localized in the lysosomal membrane. *J. Biol. Chem* 278, 26687–26694. [PubMed: 12736250]
- Patel D, Zhang X, Farrell JJ, Chung J, Stein TD, Lunetta KL, Farrer LA, 2021. Cell-type-specific expression quantitative trait loci associated with Alzheimer disease in blood and brain tissue. *Transl. Psychiatry* 11, 250. [PubMed: 33907181]
- Pearse BM, 1976. Clathrin: a unique protein associated with intracellular transfer of membrane by coated vesicles. *Proc. Natl. Acad. Sci. U. S. A* 73, 1255–1259. [PubMed: 1063406]
- Penney J, Ralvenius WT, Tsai LH, 2020. Modeling Alzheimer’s disease with iPSC-derived brain cells. *Mol. Psychiatry* 25, 148–167. [PubMed: 31391546]
- Pimenova AA, Raj T, Goate AM, 2018. Untangling genetic risk for Alzheimer’s disease. *Biol. Psychiatry* 83, 300–310. [PubMed: 28666525]
- Pitulescu ME, Adams RH, 2010. Eph/ephrin molecules-a hub for signaling and endocytosis. *Genes Dev.* 24, 2480–2492. [PubMed: 21078817]
- Poteryaev D, Datta S, Ackema K, Zerial M, Spang A, 2010. Identification of the switch in early-to-late endosome transition. *Cell* 141, 497–508. [PubMed: 20434987]
- Pottier C, Hannequin D, Coutant S, Rovelet-Lecrux A, Wallon D, Rousseau S, Legallic S, Paquet C, Bombois S, Pariente J, et al. , 2012. High frequency of potentially pathogenic SORL1 mutations in autosomal dominant early-onset Alzheimer disease. *Mol. Psychiatry* 17, 875–879. [PubMed: 22472873]
- Prokic I, Cowling BS, Laporte J, 2014. Amphiphysin 2 (BIN1) in physiology and diseases. *J. Mol. Med. (Berl)* 92, 453–463. [PubMed: 24590001]
- Puri C, Vicinanza M, Rubinsztein DC, 2018. Phagophores evolve from recycling endosomes. *Autophagy* 14, 1475–1477. [PubMed: 29940791]
- Qian L, Tew J, 2021. Human iPSC-based Modeling of central nerve system disorders for drug discovery. *Int. J. Mol. Sci* 22. [PubMed: 35008458]
- Querfurth HW, LaFerla FM, 2010. Alzheimer’s disease. *N. Engl. J. Med* 362, 329–344. [PubMed: 20107219]
- Raghavan NS, Brickman AM, Andrews H, Manly JJ, Schupf N, Lantigua R, Wolock CJ, Kamalakaran S, Petrovski S, Tosto G, et al. , 2018. Whole-exome sequencing in 20,197 persons for rare variants in Alzheimer’s disease. *Ann. Clin. Transl. Neurol* 5, 832–842. [PubMed: 30009200]
- Raman S, Brookhouser N, Braffnan DA, 2020. Using human induced pluripotent stem cells (hiPSCs) to investigate the mechanisms by which apolipoprotein E (APOE) contributes to Alzheimer’s disease (AD) risk. *Neurobiol. Dis* 138, 104788. [PubMed: 32032733]
- Reddy K, Cusack CL, Nnah IC, Khayati K, Saqena C, Huynh TB, Noggle SA, Ballabio A, Dobrowolski R, 2016. Dysregulation of nutrient sensing and CLEARance in Presenilin deficiency. *Cell Rep.* 14, 2166–2179. [PubMed: 26923592]
- Research in Medical Education, 1992. Proceedings of the 31st Annual Conference. New Orleans, Louisiana, November 1992. *Acad. Med* 67. S1–88.
- Riemens RJM, Kenis G, van den Beucken T, 2020. Human-induced pluripotent stem cells as a model for studying sporadic Alzheimer’s disease. *Neurobiol. Learn. Mem* 175, 107318. [PubMed: 32977028]
- Rink J, Ghigo E, Kalaidzidis Y, Zerial M, 2005. Rab conversion as a mechanism of progression from early to late endosomes. *Cell* 122, 735–749. [PubMed: 16143105]
- Rogaeva E, Meng Y, Lee JH, Gu Y, Kawarai T, Zou F, Katayama T, Baldwin CT, Cheng R, Hasegawa H, et al. , 2007. The neuronal sortilin-related receptor SORL1 is genetically associated with Alzheimer disease. *Nat. Genet* 39, 168–177. [PubMed: 17220890]
- Rohe M, Haiti D, Fjorback AN, Klose J, Willnow TE, 2013. SORLA-mediated trafficking of TrkB enhances the response of neurons to BDNF. *PLoS One* 8, e72164. [PubMed: 23977241]
- Roher A, Gray EG, Paula-Barbosa M, 1988. Alzheimer’s disease: coated vesicles, coated pits and the amyloid-related cell. *Proc. R. Soc. Lond. B Biol. Sci* 232, 367–373. [PubMed: 2895476]

- Rojas R, van Vlijmen T, Mardones GA, Prabhu Y, Rojas AL, Mohammed S, Heck AJ, Raposo G, van der Sluijs P, Bonifacino JS, 2008. Regulation of retromer recruitment to endosomes by sequential action of Rab5 and Rab7. *J. Cell Biol* 183, 513–526. [PubMed: 18981234]
- Sakamuro D, Elliott KJ, Wechsler-Reya R, Prendergast GC, 1996. BIN1 is a novel MYC-interacting protein with features of a tumour suppressor. *Nat. Genet* 14, 69–77. [PubMed: 8782822]
- Sandvig K, Kavaliauskiene S, Skodand T, 2018. Clathrin-independent endocytosis: an increasing degree of complexity. *Histochem. Cell Biol* 150, 107–118. [PubMed: 29774430]
- Sannerud R, Esselens C, Ejsmont P, Mattera R, Rochin L, Tharkeshwar AK, De Baets G, De Wever V, Habets R, Baert V, et al. , 2016. Restricted location of PSEN2/gamma-secretase determines substrate specificity and generates an intracellular Abeta Pool. *Cell* 166, 193–208. [PubMed: 27293189]
- Scheltens P, Blennow K, Breteler MM, de Strooper B, Frisoni GB, Salloway S, Van der Flier WM, 2016. Alzheimer's disease. *Lancet* 388, 505–517. [PubMed: 26921134]
- Scheltens P, De Strooper B, Kivipelto M, Holstege H, Chetelat G, Teunissen CE, Cummings J, van der Flier WM, 2021. Alzheimer's disease. *Lancet* 397, 1577–1590. [PubMed: 33667416]
- Scherzer CR, Offe K, Gearing M, Rees HD, Fang G, Heilman CJ, Schaller C, Bujo H, Levey AI, Lah JJ, 2004. Loss of apolipoprotein E receptor LR11 in Alzheimer disease. *Arch. Neurol* 61, 1200–1205. [PubMed: 15313836]
- Schwagerl AL, Mohan PS, Cataldo AM, Vonsattel JP, Kowall NW, Nixon RA, 1995. Elevated levels of the endosomal-lysosomal proteinase cathepsin D in cerebrospinal fluid in Alzheimer disease. *J. Neurochem* 64, 443–446. [PubMed: 7798944]
- Schwartzentruber J, Cooper S, Liu JZ, Barrio-Hernandez I, Bello E, Kumasaka N, Young AMH, Franklin RJM, Johnson T, Estrada K, et al. , 2021. Genome-wide meta-analysis, fine-mapping and integrative prioritization implicate new Alzheimer's disease risk genes. *Nat. Genet* 53, 392–402. [PubMed: 33589840]
- Seshadri S, Fitzpatrick AL, Ikram MA, DeStefano AL, Gudnason V, Boada M, Bis JC, Smith AV, Carassquillo MM, Lambert JC, et al. , 2010. Genome-wide analysis of genetic loci associated with Alzheimer disease. *JAMA* 303, 1832–1840. [PubMed: 20460622]
- Shafaq-Zadah M, Dransart E, Johannes L, 2020. Clathrin-independent endocytosis, retrograde trafficking, and cell polarity. *Curr. Opin. Cell Biol* 65, 112–121. [PubMed: 32688213]
- Simon AM, de Maturana RL, Ricobaraza A, Escribano L, Schiapparelli L, Cuadrado-Tejedor M, Perez-Mediavilla A, Avila J, Del Rio J, Frechilla D, 2009. Early changes in hippocampal Eph receptors precede the onset of memory decline in mouse models of Alzheimer's disease. *J. Alzheimers Dis* 17, 773–786. [PubMed: 19542617]
- Sjodin S, Brinkmalm G, Ohrfelt A, Parnetti L, Paciotti S, Hansson O, Hardy J, Blennow K, Zetterberg H, Brinkmalm A, 2019. Endo-lysosomal proteins and ubiquitin CSF concentrations in Alzheimer's and Parkinson's disease. *Alzheimers Res. Ther* 11, 82. [PubMed: 31521194]
- Small SA, Petsko GA, 2020. Endosomal recycling reconciles the Alzheimer's disease paradox. *Sci. Transl. Med* 12.
- Sonnichsen B, De Renzis S, Nielsen E, Rietdorf J, Zerial M, 2000. Distinct membrane domains on endosomes in the recycling pathway visualized by multicolor imaging of Rab4, Rab5, and Rab11. *J. Cell Biol* 149, 901–914. [PubMed: 10811830]
- Sottejeau Y, Bretteville A, Cantrelle FX, Malmanche N, Demiaute F, Mendes T, Delay C, Alves Dos Alves H, Flaig A, Davies P, et al. , 2015. Tau phosphorylation regulates the interaction between BIN1's SH3 domain and Tau's proline-rich domain. *Acta Neuropathol. Commun* 3, 58.
- Srivatsan S, Swiecki M, Otero K, Celia M, Shaw AS, 2013. CD2-associated protein regulates plasmacytoid dendritic cell migration, but is dispensable for their development and cytokine production. *J. Immunol* 191, 5933–5940. [PubMed: 24218450]
- Takei K, Slepnev VI, Haucke V, De Camilli P, 1999. Functional partnership between amphiphysin and dynamin in clathrin-mediated endocytosis. *Nat. Cell Biol* 1, 33–39. [PubMed: 10559861]
- Tao QQ, Chen YC, Wu ZY, 2019. The role of CD2AP in the pathogenesis of Alzheimer's disease. *Aging Dis.* 10, 901–907. [PubMed: 31440393]



- Tebar F, Bohlander SK, Sorkin A, 1999. Clathrin assembly lymphoid myeloid leukemia (CALM) protein: localization in endocytic-coated pits, interactions with clathrin, and the impact of overexpression on clathrin-mediated traffic. *Mol. Biol. Cell* 10, 2687–2702. [PubMed: 10436022]
- Thinakaran G, Koo EH, 2008. Amyloid precursor protein trafficking, processing, and function. *J. Biol. Chem* 283, 29615–29619. [PubMed: 18650430]
- Ubelmann F, Burrinha T, Salavessa L, Gomes R, Ferreira C, Moreno N, Guimas Almeida C, 2017. Binl and CD2AP polarise the endocytic generation of beta-amyloid. *EMBO Rep.* 18, 102–122. [PubMed: 27895104]
- Ungewickell EJ, Hinrichsen L, 2007. Endocytosis: clathrin-mediated membrane budding. *Curr. Opin. Cell Biol* 19, 417–425. [PubMed: 17631994]
- Ungewickell A, Ward ME, Ungewickell E, Majerus PW, 2004. The inositol polyphosphate 5-phosphatase Ocr1 associates with endosomes that are partially coated with clathrin. *Proc. Natl. Acad. Sci. U. S. A* 101, 13501–13506. [PubMed: 15353600]
- Van Acker ZP, Bretou M, Annaert W, 2019. Endo-lysosomal dysregulations and late-onset Alzheimer's disease: impact of genetic risk factors. *Mol. Neurodegener* 14, 20. [PubMed: 31159836]
- van der Blik AM, Redelmeier TE, Damke H, Tisdale EJ, Meyerowitz EM, Schmid SL, 1993. Mutations in human dynamin block an intermediate stage in coated vesicle formation. *J. Cell Biol* 122, 553–563. [PubMed: 8101525]
- van Niel G, D'Angelo G, Raposo G, 2018. Shedding light on the cell biology of extracellular vesicles. *Nat. Rev. Mol. Cell Biol* 19, 213–228. [PubMed: 29339798]
- Vargas LM, Cerpa W, Munoz FJ, Zanlungo S, Alvarez AR, 2018. Amyloid-beta oligomers synaptotoxicity: the emerging role of EphA4/c-Abl signaling in Alzheimer's disease. *Biochim. Biophys. Acta Mol. basis Dis* 1864, 1148–1159. [PubMed: 29378302]
- Wigge P, Kohler K, Vallis Y, Doyle CA, Owen D, Hunt SP, McMahon HT, 1997. Amphiphysin heterodimers: potential role in clathrin-mediated endocytosis. *Mol. Biol. Cell* 8, 2003–2015. [PubMed: 9348539]
- Willnow TE, Petersen CM, Nykjaer A, 2008. VPS10P-domain receptors - regulators of neuronal viability and function. *Nat. Rev. Neurosci* 9, 899–909. [PubMed: 19002190]
- Winckler B, Faundez V, Maday S, Cai Q, Guimas Almeida C, Zhang H, 2018. The Endolysosomal system and Proteostasis: from development to degeneration. *J. Neurosci.* 38, 9364–9374. [PubMed: 30381428]
- Winterstein C, Trotter J, Kramer-Albers EM, 2008. Distinct endocytic recycling of myelin proteins promotes oligodendroglial membrane remodeling. *J. Cell Sci* 121, 834–842. [PubMed: 18303048]
- Yamazaki H, Bujo H, Kusunoki J, Seimiya K, Kanaki T, Morisaki N, Schneider WJ, Saito Y, 1996. Elements of neural adhesion molecules and a yeast vacuolar protein sorting receptor are present in a novel mammalian low density lipoprotein receptor family member. *J. Biol. Chem* 271, 24761–24768. [PubMed: 8798746]
- Yamazaki T, Masuda J, Omori T, Usui R, Akiyama H, Mara Y, 2009. EphA1 interacts with integrin-linked kinase and regulates cell morphology and motility. *J. Cell Sci* 122, 243–255. [PubMed: 19118217]
- Yang A, Vest Ryan T., Fabian Kern, Lee Davis P., Maat Christina A., Losada Patricia M., Chen Michelle B., Maayan Agam, Nicholas Schaum, Nathalie Khoury, Kruti Calcuttawala, Róbert Pálovics, Andrew Shin, Wang Elizabeth Y., Jian Luo, David Gate, Siegenthaler Julie A., McNerney M. Windy, Andreas Keller, Tony Wyss-Coray, 2021. A human brain vascular atlas reveals diverse cell mediators of Alzheimer's disease risk. In: *Biondv bioRxiv* 2021.04.26.441262 10.1101/2021.04.26.441262.
- Young JE, Boulanger-Weill J, Williams DA, Woodruff G, Buen F, Revilla AC, Herrera C, Israel MA, Yuan SH, Edland SD, et al. , 2015. Elucidating molecular phenotypes caused by the SORL1 Alzheimer's disease genetic risk factor using human induced pluripotent stem cells. *Cell Stem Cell* 16, 373–385. [PubMed: 25772071]
- Yuyama K, Igarashi Y, 2016. Physiological and pathological roles of exosomes in the nervous system. *Biomol Concepts* 7, 53–68. [PubMed: 26812803]



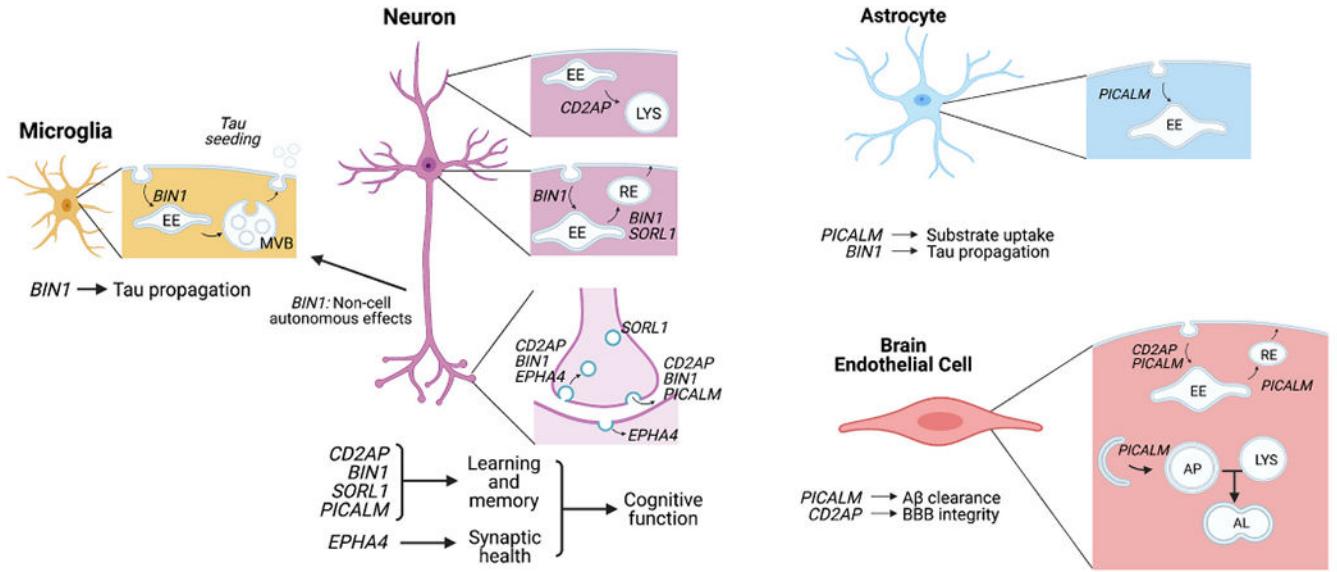
- Zerial M, McBride H, 2001. Rab proteins as membrane organizers. *Nat. Rev. Mol. Cell Biol* 2, 107–117. [PubMed: 11252952]
- Zhang M, Chen L, Wang S, Wang T, 2009. Rab7: roles in membrane trafficking and disease. *Biosci. Rep* 29, 193–209. [PubMed: 19392663]
- Zhang X, Garbett K, Veeraraghavalu K, Wilburn B, Gilmore R, Mimics K, Sisodia SS, 2012. A role for presenilins in autophagy revisited: normal acidification of lysosomes in cells lacking PSEN1 and PSEN2. *J. Neurosci* 32, 8633–8648. [PubMed: 22723704]

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**Fig. 1.** AD risk genes in the ELN function in diverse cell types in the brain. In microglia, *BIN1* regulates *Tau* propagation via its role in mediating extracellular vesicle release and may also be involved in extracellular cross-talk between neurons and microglia. *CD2AP* regulates APP transport from early endosomes to lysosomes and may contribute to APP degradation in neurons and other cell types as well as functioning in growth-factor mediated endocytosis in neurons which may stimulate pathways involved in learning and memory. *SORL1* can mediate trafficking from the early endosome through various arms of the ELN and, in conjunction with *EPHA4*, play an important role in synaptic structure and function. *PICALM* is a clathrin-binding protein that mediates trafficking between early endosomes and the TGN. Furthermore, *PICALM* may interact with *APOE* in astrocytes to control internalization of cell surface ligands. In brain endothelial cells, multiple ELN associated genes function in roles of *Aβ* clearance, maintenance of BBB integrity and cerebral blood flow through regulation of early endosomes, recycling endosomes, and the autophagy pathway. (Image created with [BioRender.com](https://www.biorender.com)).

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