Stimulation of TREM2 with agonistic antibodies - an emerging therapeutic option for Alzheimer's disease

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SUMMARY

Neurodegenerative disorders including Alzheimer's disease (AD) are associated with microgliosis. Microglia have long been considered to play detrimental roles in promoting the disease. However, functional analyses of genes encoding risk factors, which are linked to late onset AD and enriched or even exclusively expressed in microglia, revealed unexpected protective functions. One of the major risk genes is *TREM2*. *TREM2* risk variants turned out to be loss-of-function mutations affecting chemotaxis, phagocytosis, lipid and energy metabolism, as well as survival and proliferation. Agonistic anti-TREM2 antibodies have been developed to boost these protective functions in patients with intact *TREM2* alleles. Several anti-TREM2 antibodies are in early clinical trials, and current efforts aim to achieve more efficient transport across the blood brain barrier. TSPO-PET and FDG-PET may be used to monitor target engagement. Data from animal models and biomarker studies in patients further support a rationale for boosting TREM2 functions during the pre-symptomatic phase.

INTRODUCTION

Genome wide association studies have identified risk variants for late-onset AD (LOAD) in a number of genes, which are selectively expressed in microglia but not in other brain cells including neurons [1]. Among these genes, functional analyses of LOAD associated variants of TREM2 have allowed researchers to gain unexpected insights into microglia function and dysfunction [2,3].

TREM2 is predominantly expressed as a type-1 transmembrane protein, and it is targeted to the plasma membrane together with its co-receptor DAP12. The protein has been shown to bind to APOE4, which is well known as the major risk factor for developing LOAD [4]. However, the significance of this interaction for the increased risk of *APOE4* carriers to develop LOAD is currently unclear.

In this Review, we discuss evidence for TREM2 agonism as a mechanism to therapeutically stimulate protective TREM2 signaling. We describe the mechanisms whereby agonistic antibodies modify disease development in animal models, address potentially problematic interference with vital functions in peripheral myeloid cells, and present data from early clinical trials and biomarker studies. Finally, based on the proposal that microglia are integral components of the amyloid cascade [5], we will discuss combinatorial disease modifying strategies using amyloid lowering antibodies together with microglial modulating agonists. Other therapeutic approaches to modulating microglial function in AD have been extensively reviewed elsewhere [2].

TREM2 agonism for disease modification

TREM2 is a key player in regulation of microglial responses to pathological challenges (Panel 1; Figure 1). Mouse models strongly suggest a protective role for microglial Trem2 in the context of amyloid, as well as amyloid-induced tau pathology (Panel 2). Emerging data in humans also support protective functions of TREM2. Importantly, identification of a protective AD variant in the *PLCG2* gene, which activates the TREM2 signaling pathway in microglia, clearly argued in favor of TREM2 agonism in the context of AD[31]. These insights thus stimulated many researchers to explore therapeutic approaches centered around activating TREM2 with the goal of boosting beneficial microglial functions in AD patients with intact

TREM2 alleles or loss of function associated AD risk variants. In particular, monoclonal agonistic anti-Trem2 antibodies (Table 1) have been developed to enhance protective Trem2 signaling in mouse microglia [2]. Antibody 4D9 was the first antibody that successfully showed a reduction of the halo of amyloid plaques in a mouse model for amyloidosis [11]. This antibody was developed based on the finding that shedding of TREM2 terminates cell-autonomous signaling [40] (Figure 2A). Antibody 4D9 was shown to bind full-length Trem2 on the cell surface with high affinity and to inhibit its proteolytic cleavage in a dose-dependent manner. The antibody binds to an epitope 12 amino acids N-terminal of the cleavage site, activates downstream Syk signaling, and strongly increases survival of murine macrophages (Figure 3A; Table 1). Importantly, a monovalent Fab version of 4D9 failed to elicit Syk signaling suggesting that Trem2 receptor cross-linking is required for activation [11]. In line with that, independent studies employing AL002c and AL002a, human and mouse specific versions of an anti-TREM2 agonistic antibody (Table 1), confirmed that TREM2 activation leads to amelioration of Aβ pathology *in vivo* [32,33]. These studies, therefore, provided early support for further pursuing this innovative translational approach. Other studies further confirmed a reduction of the amyloid burden [34,36]. Furthermore, antibody treatment led to enhanced clustering of microglia around amyloid plaques [33,36,38]. Strikingly, antibody treatment was also consistently shown to lead to cognitive improvement in mouse models [32-34,36]. Altogether, there is now strong evidence from several laboratories that collectively suggest encouraging therapeutic effects of boosting TREM2 signaling in preclinical models. In apparent contrast to these promising reports, one study demonstrated that engineering a less cleavable Trem2 mutant, which enhances levels of cell surface Trem2 and should therefore constitutively activate downstream signaling leads to enhanced amyloid deposition at an early disease stage. This is particularly surprising, since, similar to the agonistic antibodies described above, less cleavable Trem2 stimulated survival of macrophages as well as clustering of microglia around amyloid plaques [42]. Importantly, the less cleavable Trem2 model implies constitutively enhanced Trem2 signaling throughout life while Trem2 activation via monoclonal antibodies occurs in a pulsatile fashion during the phase of amyloid deposition making these two different approaches only partially comparable. These findings suggest, however, that antibody-mediated Trem2 agonism may have to be finely tuned to achieve the desired outcome.

The beneficial effects of therapeutic anti-Trem2 antibodies in mouse models of amyloidosis raise the question whether the antibody modulates microglial cell states. Several scRNA-seq analyses in amyloid mouse models show that a single dose of a TREM2 agonist most prominently upregulates microglial gene modules related to metabolism and proliferation[32,35,39]. This is in line with previous findings showing that agonistic antibodies collectively increase microglial proliferation [2] (Table 1) and glucose uptake [39]. Further studies are required to understand how microglial transcriptomic signatures change upon chronic anti-Trem2 antibody administration, since this scenario will more closely model therapeutic treatment in a clinical setting. Altogether, these emerging data indicate a promising therapeutic potential for TREM2 modulation via monoclonal antibodies, providing a clear rationale for further exploration in the clinic.

A longstanding challenge in neurologic diseases, however, has been that systemic antibody administration requires very high doses to achieve therapeutically relevant antibody concentrations in the brain. Typically, only about 0.1% of an administered antibody crosses the blood brain barrier (BBB) and enters the brain parenchyma [43]. The vast majority of administered antibodies consequently remains in the periphery which in the case of TREM2 might be relevant since it is now well documented that TREM2 is expressed in a number of peripheral tissues where agonistic effects of anti-TREM2 antibodies might have deleterious consequences (see below). Therefore, efforts are on the way to enhance antibody transcytosis across the blood-brain-barrier via a brain shuttle system [43-46] involving engineered antibodies containing a transferrin receptor (TfR) binding site (Figure 3B). Upon binding to TfR the modified antibodies are internalized by endothelial cells, likely released in the acidic environment of the endosomal pathway, and secreted on the other side of the BBB into the brain parenchyma. The engineered antibodies thus not only allow to increase brain penetrance, but also allow to substantially lower antibody doses to be administered. Independent studies have shown that either coupling the Fc part of the antibody with a monovalent TfR-binding scFv [36,37] or adding a TfR binding site to the Fc part of the antibody (ATV:TREM2 (DNL919); ATV: antibody transport vehicle; Figure 3B) dramatically enhances antibody uptake by an order of magnitude [36,37,39,45]. As a consequence, enhanced amelioration of amyloid pathology in mice treated with TfR-bound antibodies compared to those treated with conventional antibodies has been reported [36]. Furthermore, a single dose of ATV:TREM2 longitudinally increased microglial activation and glucose uptake as measured by TSPO- and

FDG-PET imaging [39]. This also suggests a possibility of TSPO- and FDG-PET imaging to be used as a biomarker for target engagement in clinical trials (summarized below). The biodistribution of ATV:TREM2 depends on tissue expression of TfR rather than TREM2 expression. High levels of antibody binding were accordingly detected in spleen, bone marrow, plasma, lung, and brain. Nevertheless, no histopathological abnormalities were observed, suggesting that the modified antibodies are well tolerated, although they do not only accumulate within the brain [39].

A crucial aspect in further exploring TREM2 agonism in the context of AD will be to evaluate its effect on amyloid-induced tau pathology (see Panel 2). Surprisingly, although various studies examining *Trem2* deficiency in mouse models for amyloid and tau pathology suggest that Trem2 limits seeding and spreading of amyloid-induced tau pathology [26-28], it was reported that chronic AL002a treatment enhances seeding and spreading of phospho-tau as well as neuritic dystrophy suggesting it may even worsen cognitive performance [38]. This indicates a need for further studies to address the complexity of different stages of AD-related pathology, particularly in regard to the timing of therapeutic anti-TREM2 antibody treatment [47].

A close comparison of currently published therapeutic anti-Trem2 antibodies suggests that they act via similar mechanisms, namely by prevention of shedding, crosslinking of cell-surface Trem2, and stimulating similar protective functions via Syk signaling (Table 1) [11,34-37,39,48]. Antibody-mediated cross-linking of Trem2 on the cell surface is essential for activation of Syk signaling as shown by both absence of activation by monovalent antibody versions [11,35,39] and enhanced signaling when employing tetravalent antibodies [36,37]. In addition, several of these antibodies share a similar epitope [11,39], which is located N-terminal in the immediate vicinity of the cleavage site [49-51] (Figure 3A), indicating that these agonistic antibodies promote TREM2 signaling via steric hindrance of ADAM10/17 cleavage of cell surface located signaling competent TREM2. Similar binding sites of independently screened therapeutic antibodies and their similar mechanisms of action suggest that indeed most if not all currently known agonistic antibodies utilize the same signaling pathway to promote protective activities.

However, the binding site N-terminal of the cleavage site also implies that agonistic antibodies not only bind to cell surface full-length TREM2, but also to sTREM2. The functional consequences of antibody binding to sTREM2 are not yet entirely clear but there is accumulating evidence that sTREM2 inhibits amyloid fibrillization *in vitro* [52-54]. In addition,

sTREM2 has been reported to ameliorate amyloid pathology in an animal model, albeit at very high non-physiological concentrations warranting independent confirmation [55]. Furthermore, it has been reported that sTREM2 binds to neurons indicating that sTREM2 may have a non-cell autonomous function [56]. In future studies, it will therefore be important to address the roles of sTREM2 in health and disease in much more detail since this has direct implications for therapeutic TREM2 modulation as we outline below.

Concerns were also raised that agonistic antibodies may over-activate microglia and drive them into a dead end of hyperactivation with no return to a homeostatic state. However, microglia can apparently switch dynamically between homeostatic and disease associated states [57]. These findings may therefore indicate a wider therapeutic window of anti-TREM2 antibodies, than originally anticipated.

Importantly, one also has to consider the consequences of TREM2 stimulation in the periphery. In that regard, the well documented immunosuppressive role of TREM2 expressing tumor associated macrophages (TAMs) in the tumor microenvironment (TME) of various types of cancer may be concerning . In line with these findings, treatment with an antagonistic anti-Trem2 antibody [58,59] or Trem2 ablation [59,60] both lead to disease amelioration in certain mouse models of cancer. Further animal studies are therefore required to prove safety of agonistic anti-TREM2 antibodies.

Finally, abnormal synaptic pruning upon chronic treatment with TREM2 agonists must be investigated, since this mechanism has been shown to affect memory function in mouse models and AD patients (see Panel 1) [61].

Protective TREM2 functions in humans: from genetics to biomarker-based clinical studies

The above-described findings clearly support protective effects of TREM2 agonism in model systems. But what about protective functions in humans? TREM2 loss-of-function variants have been related to atypical AD phenotypes, such as non-amnestic syndromes and faster disease progression.[62-65] In line with that, AD cases carrying TREM2 risk variants also showed a different plaque morphology with less APOE and a higher burden of tau tangle deposition with an atypical relative preservation of hippocampal regions compared to neocortical regions[62,66-68] supporting protective TREM2 functions on AD pathology. Predicted risk nonsynonymous single nucleotide polymorphisms (nsSNPs) within the V-type Ig-like ectodomain of TREM2 (Figure 2A) have been found to cause dysfunction, supporting a

relevant role of ligand binding for the protective functions attributed to TREM2-dependent activated microglia.[69] Additionally, mRNA transcripts of at least four TREM2 isoforms (ENST00000373113, TREM2Δe2, ENST00000338469, and ENST00000373122), the last three coming from alternative splicing, have been described specifically in human brain[70-75]. The most abundant isoform ENST00000373113 is the canonical full-length TREM2 protein (Figure 2A). Isoforms ENST00000338469 and ENST00000373122 are proposed to be directly secreted soluble forms (i.e. alternative sTREM2 forms), potentially detectable in biological fluids,[71] while isoform TREM2Δe2 lacks exon 2, possibly producing a non-functional TREM2 receptor [72,73,75] (Figure 2B). The last three isoforms are proposed to be possible modulators of full-length TREM2 activation pathways.[71-74] The impact of alternative splicing on TREM2 functions has also been pointed out by another study that predicts 10 low-frequency AD risk variants by a splicing-guided aggregation approach to affect splicing regulatory elements and potentially facilitate exon 2 skipping.[75] Importantly, agonistic TREM2 antibodies may bind to these four described isoforms of TREM2 (Figure 2 and Figure 3A) and thus affect their potential biological activities. This may indeed be relevant as both sTREM2 as generated by cleavage of full-length TREM2 and the two alternative soluble TREM2 isoforms have been shown to reduce synaptic plasticity in mouse brain slices[76]. Since in mice only one naturally occurring alternative isoform has been described it will only be possible to rigorously evaluate such effects in humans.

CSF sTREM2 has been widely investigated as a biomarker for TREM2 function and microglia activity in human cohorts. CSF sTREM2 levels represent mostly the soluble fragment released after cleavage of full-length TREM2 on the cell surface, with a possible small contribution of the other soluble isoforms (Figure 2).[71] Until now, only two studies specifically reported levels of sTREM2 derived from cleavage using a neo-epitope specific antibody (1H3) that does not detect other soluble isoforms.[77,78] Thus, this assay exclusively detects sTREM2 derived from shedding of cell surface located, signaling competent TREM2. Regarding the AD continuum, most cross-sectional studies have shown increased levels of sTREM2 in early AD stages compared to healthy individuals, starting in the preclinical phase, with a maximum in the prodromal AD group and smaller differences in later clinical stages.[74,79] In sporadic late onset AD, two cross-sectional studies based on independent cohorts showed decreased levels of sTREM2 compared to controls in cognitively normal participants with CSF biomarkers suggestive of amyloid deposition, but normal tau-related markers.[80,81] However, the proposed early decrease in sporadic AD has not been found in autosomal dominant AD, where an

extremely early increase of sTREM2 up to 21 years before expected symptom onset has been reported in carriers of pathogenic mutations compared to non-carriers.[77] This elevation was observed immediately after the first changes in biomarkers representing amyloid accumulation and deposition and before changes in CSF tau-related markers, in line with a very early response of microglia to Aβ pathology, already reported in mouse models, where TREM2 related protective activities of microglia restrict amyloid seeding.[77,66,82] This finding also has direct consequences for the timing of therapeutic approaches using TREM2 agonists, as it indicates that they must be initiated as early as possible (Figure 4).

Despite the increased levels of sTREM2 found early in AD development, sTREM2 is not a valid diagnostic marker for AD since there is a large inter-individual variation of baseline sTREM2 levels which generates a wide range of sTREM2 values within the same diagnostic group and, therefore, a considerable overlap regarding sTREM2 levels between these groups. Nevertheless, sTREM2 can be used to study TREM2 related protective functions during the development of AD. Regarding AD pathology, there is a well-established correlation between sTREM2 and CSF tau-related markers in cross-sectional studies.[74,83] In autosomal-dominant AD, lower levels of $Aβ_{1-42}$ and $Aβ_{1-42}/Aβ_{1-40}$ in CSF at baseline were the most important predictor of sTREM2 longitudinal elevation in CSF in carriers of pathogenic mutations, while tau-related markers showed no relationship with longitudinal changes in sTREM2.[77] Further supporting the relationship between sTREM2 and Aβ biomarkers, a later study reported cross-sectional associations between CSF levels of sTREM2 and different Aβ species in CSF (truncated $Aβ_{x-40}$, $Aβ_{x-42}$, $Aβ_{1-40}$ and $Aβ_{1-38)}$ in two independent sporadic LOAD cohorts. The same study also showed that $Aβ_{x-40}$ levels independently explained 21.2% of sTREM2 variance in CSF.[84] Regarding the influence of TREM2 on the development of amyloid pathology in AD, higher longitudinal increases of sTREM2 levels in CSF have been related to a slower decrease in CSF $Aβ_{1-42}$ (indicating Aβ deposition within the brain) in presymptomatic autosomal dominant AD cases and slower increase in the PiB-PET signal in the symptomatic phase suggesting a possible biphasic function of TREM2 limiting amyloid pathology[77], in line with findings in mouse models [66,85]. Two independent studies focused on sporadic late-onset AD also showed that higher baseline levels of CSF sTREM2 were associated with lower rates of increase in amyloid-PET signal[86,87] further supporting TREM2-dependent protective functions on amyloid pathology.

Beyond the well-established correlation between sTREM2 and tau-related markers in CSF, the biological relationship between TREM2 and tau pathology in AD, extracted from biomarker-based studies, remains controversial, mirroring findings in disease models. Faster hippocampal atrophy has been associated with higher sTREM2 CSF levels at baseline in cognitively normal adults with abnormal tau-related markers independently of Aβ-related markers, pointing to a relationship between TREM2 dependent microglial activation and a worse neuroimaging outcome related to Aβ-independent tau pathology.[88] Network analysis based on tau ([18F]MK-6240) and microglial activation ([11C]PBR28 -TSPO-) PET imaging studies from aging and AD-continuum participants reported a hierarchical correlation between tau- and TSPO-PET signals along Braak-like stages, supporting a role of activated microglia in the spreading of tau-pathology.[89] These findings contrast with the higher burden of tau pathology and its atypical distribution found in neuropathology in AD cases carrying loss-of-function *TREM2* genetic variants.[62,67,68] Additionally, in autosomal-dominant AD, higher longitudinal increases in sTREM2 were found to ameliorate the increase in CSF p-tau related to the increase of amyloid-PET signal in presymptomatic participants.[77] Higher baseline levels of sTREM2 were also related to a slower tau deposition as measured by tau-PET in pre-demented individuals with biomarker evidence of amyloid and tau deposition.[87] Both these studies further support the protective effect of TREM2 activation on Aβ-dependent tau pathology in AD.[77,87]

Longitudinal studies in sporadic late-onset AD have related higher levels of sTREM2 at baseline to a slower hippocampal atrophy and better cognitive progression during early clinical phases, together with a lower rate of MCI-to-dementia conversion.[90-92] During the presymptomatic phase of autosomal-dominant AD, higher rates of sTREM2 increase in CSF strongly correlated with a slower cognitive decline together with a moderately slower cortical shrinkage in the precuneus.[77] Additionally, variants in the MS4A gene cluster related to higher sTREM2 levels in the CSF were also associated with a lower AD risk and a later symptom onset in cases with cognitive decline.[93] Nevertheless, one study reported differential effects of sTREM2 levels in CSF at baseline on the neuroimaging outcome (grey and white matter volume changes and mean diffusivity variation) of cognitively normal adults (57 to 78 years old) depending on age, with a beneficial effect of higher sTREM2 in younger participants and a detrimental effect in older participants.[94] Some studies based on cross-sectional data also argue for a detrimental effect of TREM2 activation in AD based on correlations between sTREM2 levels in CSF and different markers of disease progression, such as MMSE scores, white matter hyperintensities

or cortical thickness.[89,95] However, these cross-sectional associations might only represent the increase of sTREM2 as a marker of microglial response to pathology along AD development.

Taken together, studies based on human cohorts show a rather consistent beneficial effect of high sTREM2 levels in CSF on the development of Aβ pathology (Figure 4). Most studies also indicate a benefit of high levels of sTREM2 on neuroimaging biomarkers and cognition in early AD phases, while studies focused on the relationship between sTREM2 and tau-related pathology in AD are still controversial. This suggests that the possible protective TREM2 functions in AD may depend on disease stage. During the initial stages, when amyloid deposition is the main pathological event, boosting TREM2 functions may have the highest beneficial effect on disease progression (Figure 4). Then, in later stages, when tau-related pathology is more prominent, the protective role of TREM2 would be less evident or even absent.

Clinical trials using TREM2 agonists and clinical perspectives

Until today, data on TREM2 boosting therapies derived from clinical studies are very limited. Results from only one phase 1 clinical trial have been published so far, showing that the TREM2 agonist antibody AL002 is safe and well-tolerated in 69 participants comprising healthy adults and AD patients.[32] A second phase 2 study assessing the efficacy of the same antibody in MCI and mild dementia due to AD is currently ongoing (ClinicalTrials.gov identifier: NCT04592874). The independently developed human agonistic anti-TREM2 antibody DNL919 [39] is currently being tested for safety and tolerability in healthy adults (Table 2), and results are expected in the next two years.

In this context, considering the results from clinical studies focused on CSF sTREM2 will be crucial for an appropriate design of future phase 2 and 3 clinical trials. Biomarker-based studies suggest that the beneficial role of TREM2 is most likely disease stage-dependent, therefore, the outcome obtained after treatment initiated in different phases of the disease might differ substantially. Several studies have shown that a higher increase of sTREM2, as a biomarker of TREM2 activity, is associated with better clinical outcome specifically during early AD phases.[77,91] Consequently, therapies aimed to enhance TREM2 functions should be tested first in early AD phases, including the presymptomatic stage where seeding of amyloid

is initiated. In fact, accounting for the suggested close relationship with Aβ pathology, boosting TREM2 activation during the presymptomatic phase of AD could be the most effective approach to obtain the highest clinical benefit, which is line with TREM2 dependent clearance of seeds in mouse models [66,82]. TREM2 boosting therapies should be considered to be initiated in at least three clinical groups reflecting different stages of AD: in cognitively normal individuals with abnormal Aβ markers, but normal tau-related markers (A^+/T^-); in cognitively normal individuals with abnormality of both Aβ and tau-related markers (A^+/T^+), and in individuals with MCI and biomarker evidence of underlying AD pathology (Figure 4). Considering the conflicting results regarding the relationship between TREM2 activation and tau-related pathology both in biomarker-based clinical studies and animal models [38,89,96], initiation of TREM2 agonist treatments in later stages may not be favored.

In that regard, sTREM2 could also be useful as a stratification marker for TREM2 boosting therapies. Based on the above-described findings that increased sTREM2 levels associate with beneficial outcomes, we propose that specifically patients with low sTREM2 levels at baseline should be treated with TREM2 agonists.

Unfortunately, sTREM2 levels cannot be used as a biomarker for target engagement. Although several agonistic antibodies reportedly reduce TREM2 shedding, these antibodies all bind to an epitope N-terminal of the cleavage site and may therefore stabilize the remaining sTREM2 [11,39] Furthermore, detection of CSF sTREM2 derived from alternative splicing could make interpretations difficult [71,74]. Although TSPO-PET does not reflect selectively TREM2-activated microglia, some studies have found a correlation between sTREM2 levels and the TSPO signal in neurodegenerative diseases[39,78,89], which suggests TSPO-PET as a possible tool to assess target engagement during TREM2 boosting therapies. Studies in both mouse models and human cohorts have reported a correlation between microglial activation, as measured by sTREM2 levels and TSPO-PET, and FDG-PET tracer uptake, pointing to a possible use of FDG-PET as an additional biomarker for microglial activation,[21,95] with the advantage of being a functional neuroimaging tool broadly used in the clinical practice.

The recently approved anti-amyloid drug Lecanemab[97] and similar antibodies, which are currently tested, additionally opens new perspectives regarding the use of sTREM2 as a biomarker of TREM2 activated microglia in the clinical practice. First, baseline levels of sTREM2 in CSF could be useful to stratify patients in those with a better intrinsic response towards amyloid deposition and those prone to a faster Aβ accumulation[77,98], which could

help to adjust treatment dosage. Levels of sTREM2 could additionally serve to predict which patients will have a better cognitive outcome and adjust the expected effect of anti-amyloid treatment by sTREM2 levels at baseline. If TREM2-boosting therapies demonstrate a beneficial effect in AD, combined treatments using anti-amyloid drugs and TREM2 agonistic antibodies will be a potentially synergistic therapeutic option, given the suggested beneficial role of TREM2 functions on Aβ pathology. In that case, future studies will be required to assess the time point and the order (sequential or concomitant treatments) of these proposed disease-modifying therapies.

Conclusions and future directions

TREM2 agonism may indeed be a novel approach for treatment of AD. Although it is becoming increasingly clear that TREM2 agonism stimulates protective microglia functions in the brain, a number of questions remain. Most importantly, long-term studies are required, which not only address amelioration of AD phenotypes but also take potentially deleterious effects such as increased risk for tumorigenesis into account. We also need a much better understanding of sTREM2 function, since basically all therapeutic antibodies investigated so far not only bind to full-length TREM2 but also to sTREM2. In addition, microglial activity state markers would be required to monitor target engagement of agonistic anti-TREM2 antibodies in patients. We also want to emphasize that it is very important to consider TREM2 agonism not as an alternative treatment approach to the conventional anti-amyloid treatments currently tested. Based on the assumption that microglia are integral parts of the amyloid cascade [5], we rather propose that TREM2 agonists could further boost the ability of anti-Aβ antibodies, such as Lecanemab [97] to efficiently lower amyloid accumulation and enhance the chance to slow disease progression. Combinatorial treatment paradigms may therefore be considered in the future. Finally, boosting protective activities of microglia may not only decrease the risk for AD, but probably also for other neurological disorders. Indeed, there is evidence that agonistic anti-TREM2 antibodies may ameliorate pathological phenotypes in de-myelination disorders such as multiple sclerosis [99,100].

Figure Legends

Figure 1 Microglial responses to pathological challenges

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(**Upper left**) TREM2 is essential for controlling lipid homeostasis in microglia. The protein mediates microglial recognition of damage-associated lipids, including sensing and metabolizing of lipids during injury. (**Lower left**) TREM2 recognises externalized phosphatidylserine on synapses for targeted engulfment during development, raising the question of whether there is a protective role for TREM2-mediated synapse engulfment in Alzheimer's disease. (**Upper right**) On stress or pathological challenge, microglia turn on mTOR in a TREM2-dependent manner to meet increased demands for energy and protein synthesis. (**Lower right**) A defensive response to deposition of amyloid plaques is to form a protective barrier around plaques and prevent further release of toxic peptides.

Figure 2 Effect of shedding of TREM2 on cell autonomous signalling

(**A**) TREM2 is predominantly expressed as a type-1 transmembrane protein, which is transported to the plasma membrane together with its co-receptor DAP12 (occurring as a disulfide-bonded dimer). On the cell surface, TREM2 can bind to a variety of ligands and stimulate cell-autonomous signaling via DAP12 and spleen tyrosine kinase (SYK) phosphorylation [41]. Cell autonomous TREM2 signaling is terminated by shedding of its ectodomain (ie cleavage after His157 by ADAM10 or ADAM17), which releases soluble TREM2 (sTREM2) [40] and terminates signaling. (**B**) TREM2 isoforms generated by alternative splicing. The two soluble isoforms 1 and 2 (ENST00000338469 and ENST00000373122, respectively), which differ from the canonical isoform shown in panel (A) (ENST00000373113) from residue 162 (C termini colored in light green and yellow, respectively), have no cell autonomous signaling capacity. Whether the TREM2Δe2 isoform bound to DAP12 has signaling capacity is currently unknown. CTF: C-terminal fragment; ITAM: immunoreceptor tyrosine-based activation motif; Ig: immunoglobulin.

Figure 3 Binding of agonistic anti-TREM2 antibodies and mechanism of transport across the BBB

(**A**) 4D9 and DNL919 (i.e. ATV:TREM2) bind to overlapping epitopes in the TREM2 stalk region close to the ADAM10/17 cleavage site (colored in yellow and orange, respectively). The human TREM2 sequence is shown. (**B**) ATV:TREM2 binds to the transferrin receptor on the luminal side of the BBB via its modified Fc domain. Upon internalization, ATV:TREM2 is likely released

in the acidic environment of the endosomal pathway and secreted on the parenchymal side of the BBB for engagement of microglial TREM2. ATV: antibody transport vehicle.

Figure 4 CSF and neuroimaging biomarker trajectories along the AD continuum and proposed therapeutic window for TREM2 agonists

Each line in the upper part illustrates the proposed and approximated trajectory of biomarkers representing Aβ pathology (Aβ-PET and CSF $Aβ_{42}$), tau-related pathology (tau-PET and CSF p-tau), CSF sTREM2 and neuronal dysfunction and death (FDG-PET and atrophy in the MRI), from normality to abnormality and according to literature, along the AD development (represented in the lower bar from normality to the clinical impairment). The green box represents the interval in which a biomarker would be considered as normal. The lower bars represent 1) the clinical evolution towards the symptomatic phase (red); 2) the proposed development of Aβ pathology according to the Aβ pathology biomarker trajectories along the AD continuum (orange); 3) the postulated TREM2-dependent microglial responses to pathology along the AD continuum (violet); 4) the proposed development of tau-related pathology according to the tau-related pathology biomarker trajectories along the AD continuum (blue); 5) the proposed sequence for neuronal dysfunction and death according to the corresponding biomarkers. Grey boxes illustrate the three different proposed windows for TREM2 agonist treatment according to the clinical and biomarker stage. CSF = cerebrospinal fluid. A^+ = biomarkers suggestive of underlying Aβ pathology (abnormality in Aβ-PET and/or CSF $Aβ_{42)}$. T^- = normal tau-related pathology biomarkers (tau-PET and CSF p-tau). T^+ = Biomarkers suggestive of underlying tau-related pathology (abnormality in tau-PET and/or CSF p-tau). p-tau = phosphorylated tau at threonine 181. FDG = 18F-fluorodeoxyglucose. PET = Positron Emission Tomography. MCI = Mild cognitive impairment. MG = Microglia. MRI = Magnetic Resonance Imaging.

Table 1 Agonistic anti-TREM2 antibodies with protective TREM2-dependent microglial functions both *in vitro* and *in vivo*.

Table 2 Clinical trials testing the therapeutic potential of anti-TREM2 antibodies in Alzheimer's disease.

Search Strategy and selection criteria

We performed our search in PubMed databases for articles that were peer-reviewed and published in English with a specific focus on those papers published since 2018 and until 15 May 2023. We used the search terms "TREM2" AND "dementia", "TREM2" AND "Alzheimer", "TREM2" AND "neurodegeneration", "TREM2" AND "synapse", "TREM2" AND "microglia", and "TREM2" AND "macrophages". We searched for clinical trials using TREM2 agonists on the ClinicalTrials.gov webpage (https://clinicaltrials.gov) with the terms "TREM2", "TREM-2" and by the name of each individual human antibody developed until now.

Author contributions

KS and CH wrote the Summary, Introduction, TREM2 agonism for disease modification, Role of TREM2 in models of amyloid and tau pathology, and Conclusions and future directions. EM-R wrote Protective TREM2 functions in humans: from genetics to biomarker-based clinical studies and Clinical trials using TREM2 agonists and clinical perspectives. SH wrote Microglial responses to pathological challenges.

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Literature

1.	Romero-Molina C, Garretti F, Andrews SJ, Marcora E, Goate AM. Microglial efferocytosis: Diving into the Alzheimer's disease gene pool. *Neuron* 2022; **110**(21): 3513-33.
2.	Lewcock JW, Schlepckow K, Di Paolo G, Tahirovic S, Monroe KM, Haass C. Emerging Microglia Biology Defines Novel Therapeutic Approaches for Alzheimer's Disease. *Neuron* 2020; **108**(5): 801-21.
3.	Deczkowska A, Weiner A, Amit I. The Physiology, Pathology, and Potential Therapeutic Applications of the TREM2 Signaling Pathway. *Cell* 2020; **181**(6): 1207-17.
4.	Hou J, Chen Y, Grajales-Reyes G, Colonna M. TREM2 dependent and independent functions of microglia in Alzheimer's disease. *Mol Neurodegener* 2022; **17**(1): 84.
5.	Haass C, Selkoe D. If amyloid drives Alzheimer disease, why have anti-amyloid therapies not yet slowed cognitive decline? *PLoS Biol* 2022; **20**(7): e3001694.
6.	Keren-Shaul H, Spinrad A, Weiner A, et al. A Unique Microglia Type Associated with Restricting Development of Alzheimer's Disease. *Cell* 2017; **169**(7): 1276-90 e17.
7.	Krasemann S, Madore C, Cialic R, et al. The TREM2-APOE Pathway Drives the Transcriptional Phenotype of Dysfunctional Microglia in Neurodegenerative Diseases. *Immunity* 2017; **47**(3): 566-81 e9.
8.	Hong S, Dissing-Olesen L, Stevens B. New insights on the role of microglia in synaptic pruning in health and disease. *Curr Opin Neurobiol* 2016; **36**: 128-34.
9.	Ulland TK, Song WM, Huang SC, et al. TREM2 Maintains Microglial Metabolic Fitness in Alzheimer's Disease. *Cell* 2017; **170**(4): 649-63 e13.
10.	Stern AM, Yang Y, Jin S, et al. Abundant Abeta fibrils in ultracentrifugal supernatants of aqueous extracts from Alzheimer's disease brains. *Neuron* 2023.
11.	Schlepckow K, Monroe KM, Kleinberger G, et al. Enhancing protective microglial activities with a dual function TREM2 antibody to the stalk region. *EMBO Mol Med* 2020; **12**(4): e11227.
12.	Klunemann HH, Ridha BH, Magy L, et al. The genetic causes of basal ganglia calcification, dementia, and bone cysts: DAP12 and TREM2. *Neurology* 2005; **64**(9): 1502-7.
13.	Paolicelli RC, Bolasco G, Pagani F, et al. Synaptic pruning by microglia is necessary for normal brain development. *Science* 2011; **333**(6048): 1456-8.
14.	Hong S, Beja-Glasser VF, Nfonoyim BM, et al. Complement and microglia mediate early synapse loss in Alzheimer mouse models. *Science* 2016; **352**(6286): 712-6.
15.	Shi Q, Chowdhury S, Ma R, et al. Complement C3 deficiency protects against neurodegeneration in aged plaque-rich APP/PS1 mice. *Sci Transl Med* 2017; **9**(392).
16.	Vukojicic A, Delestree N, Fletcher EV, et al. The Classical Complement Pathway Mediates Microglia-Dependent Remodeling of Spinal Motor Circuits during Development and in SMA. *Cell Rep* 2019; **29**(10): 3087-100 e7.
17.	Werneburg S, Jung J, Kunjamma RB, et al. Targeted Complement Inhibition at Synapses Prevents Microglial Synaptic Engulfment and Synapse Loss in Demyelinating Disease. *Immunity* 2020; **52**(1): 167-82 e7.

18. Scott-Hewitt N, Perrucci F, Morini R, et al. Local externalization of phosphatidylserine mediates developmental synaptic pruning by microglia. *EMBO J* 2020; **39**(16): e105380.

19. Filipello F, Morini R, Corradini I, et al. The Microglial Innate Immune Receptor TREM2 Is Required for Synapse Elimination and Normal Brain Connectivity. *Immunity* 2018; **48**(5): 979-91 e8.

20. Bernier LP, York EM, Kamyabi A, Choi HB, Weilinger NL, MacVicar BA. Microglial metabolic flexibility supports immune surveillance of the brain parenchyma. *Nat Commun* 2020; **11**(1): 1559.

21. Xiang X, Wind K, Wiedemann T, et al. Microglial activation states drive glucose uptake and FDG-PET alterations in neurodegenerative diseases. *Sci Transl Med* 2021; **13**(615): eabe5640.

22. Nugent AA, Lin K, van Lengerich B, et al. TREM2 Regulates Microglial Cholesterol Metabolism upon Chronic Phagocytic Challenge. *Neuron* 2020; **105**(5): 837-54 e9.

23. Gouna G, Klose C, Bosch-Queralt M, et al. TREM2-dependent lipid droplet biogenesis in phagocytes is required for remyelination. *J Exp Med* 2021; **218**(10).

24. 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): 107-16.

25. Guerreiro R, Wojtas A, Bras J, et al. TREM2 variants in Alzheimer's disease. *N Engl J Med* 2013; **368**(2): 117-127.

26. Leyns CEG, Gratuze M, Narasimhan S, et al. TREM2 function impedes tau seeding in neuritic plaques. *Nat Neurosci* 2019; **22**(8): 1217-22.

27. Gratuze M, Chen Y, Parhizkar S, et al. Activated microglia mitigate Abeta-associated tau seeding and spreading. *J Exp Med* 2021; **218**(8).

28. Lee SH, Meilandt WJ, Xie L, et al. Trem2 restrains the enhancement of tau accumulation and neurodegeneration by beta-amyloid pathology. *Neuron* 2021; **109**(8): 1283-301 e6.

29. Bemiller SM, McCray TJ, Allan K, et al. TREM2 deficiency exacerbates tau pathology through dysregulated kinase signaling in a mouse model of tauopathy. *Mol Neurodegener* 2017; **12**(1): 74.

30. Leyns CEG, Ulrich JD, Finn MB, et al. TREM2 deficiency attenuates neuroinflammation and protects against neurodegeneration in a mouse model of tauopathy. *Proc Natl Acad Sci U S A* 2017; **114**(43): 11524-9.

31. Sims R, van der Lee SJ, Naj AC, et al. Rare coding variants in PLCG2, ABI3, and TREM2 implicate microglial-mediated innate immunity in Alzheimer's disease. *Nat Genet* 2017; **49**(9): 1373-84.

32. Wang S, Mustafa M, Yuede CM, et al. Anti-human TREM2 induces microglia proliferation and reduces pathology in an Alzheimer's disease model. *J Exp Med* 2020; **217**(9).

33. Price BR, Sudduth TL, Weekman EM, et al. Therapeutic Trem2 activation ameliorates amyloid-beta deposition and improves cognition in the 5XFAD model of amyloid deposition. *J Neuroinflammation* 2020; **17**(1): 238.

34. Fassler M, Rappaport MS, Cuno CB, George J. Engagement of TREM2 by a novel monoclonal antibody induces activation of microglia and improves cognitive function in Alzheimer's disease models. *J Neuroinflammation* 2021; **18**(1): 19.

35. Ellwanger DC, Wang S, Brioschi S, et al. Prior activation state shapes the microglia response to antihuman TREM2 in a mouse model of Alzheimer's disease. *Proc Natl Acad Sci U S A* 2021; **118**(3).

36.   Zhao P, Xu Y, Jiang L, et al. A tetravalent TREM2 agonistic antibody reduced amyloid pathology in a mouse model of Alzheimer's disease. *Sci Transl Med* 2022; **14**(661): eabq0095.

37.   Zhao P, Xu Y, Fan X, et al. Discovery and engineering of an anti-TREM2 antibody to promote amyloid plaque clearance by microglia in 5XFAD mice. *MAbs* 2022; **14**(1): 2107971.

38.   Jain N, Lewis CA, Ulrich JD, Holtzman DM. Chronic TREM2 activation exacerbates Abeta-associated tau seeding and spreading. *J Exp Med* 2023; **220**(1): e20220654.

39.   van Lengerich B, Zhan L, Xia D, et al. A TREM2-activating antibody with a blood-brain barrier transport vehicle enhances microglial metabolism in Alzheimer's disease models. *Nat Neurosci* 2023; **26**(3): 416-429.

40.   Kleinberger G, Yamanishi Y, Suarez-Calvet M, et al. TREM2 mutations implicated in neurodegeneration impair cell surface transport and phagocytosis. *Sci Transl Med* 2014; **6**(243): 243ra86.

41.   Ulland TK, Colonna M. TREM2 - a key player in microglial biology and Alzheimer disease. *Nat Rev Neurol* 2018; **14**(11): 667-75.

42.   Dhandapani R, Neri M, Bernhard M, et al. Sustained Trem2 stabilization accelerates microglia heterogeneity and Abeta pathology in a mouse model of Alzheimer's disease. *Cell Rep* 2022; **39**(9): 110883.

43.   Kariolis MS, Wells RC, Getz JA, et al. Brain delivery of therapeutic proteins using an Fc fragment blood-brain barrier transport vehicle in mice and monkeys. *Sci Transl Med* 2020; **12**(545).

44.   Ullman JC, Arguello A, Getz JA, et al. Brain delivery and activity of a lysosomal enzyme using a blood-brain barrier transport vehicle in mice. *Sci Transl Med* 2020; **12**(545).

45.   Yu YJ, Zhang Y, Kenrick M, et al. Boosting brain uptake of a therapeutic antibody by reducing its affinity for a transcytosis target. *Sci Transl Med* 2011; **3**(84): 84ra44.

46.   Niewoehner J, Bohrmann B, Collin L, et al. Increased brain penetration and potency of a therapeutic antibody using a monovalent molecular shuttle. *Neuron* 2014; **81**(1): 49-60.

47.   Jay TR, Hirsch AM, Broihier ML, et al. Disease Progression-Dependent Effects of TREM2 Deficiency in a Mouse Model of Alzheimer's Disease. *J Neurosci* 2017; **37**(3): 637-47.

48.   Cheng Q, Danao J, Talreja S, et al. TREM2-activating antibodies abrogate the negative pleiotropic effects of the Alzheimer's disease variant Trem2(R47H) on murine myeloid cell function. *J Biol Chem* 2018; **293**(32): 12620-33.

49.   Schlepckow K, Kleinberger G, Fukumori A, et al. An Alzheimer-associated TREM2 variant occurs at the ADAM cleavage site and affects shedding and phagocytic function. *EMBO Mol Med* 2017; **9**(10): 1356-65.

50.   Thornton P, Sevalle J, Deery MJ, et al. TREM2 shedding by cleavage at the H157-S158 bond is accelerated for the Alzheimer's disease-associated H157Y variant. *EMBO Mol Med* 2017; **9**(10): 1366-78.

51.   Feuerbach D, Schindler P, Barske C, et al. ADAM17 is the main sheddase for the generation of human triggering receptor expressed in myeloid cells (hTREM2) ectodomain and cleaves TREM2 after Histidine 157. *Neurosci Lett* 2017; **660**: 109-14.

52.   Kober DL, Stuchell-Brereton MD, Kluender CE, et al. Functional insights from biophysical study of TREM2 interactions with apoE and Abeta(1-42). *Alzheimers Dement* 2020.

53.   Vilalta A, Zhou Y, Sevalle J, et al. Wild-type sTREM2 blocks Abeta aggregation and neurotoxicity, but the Alzheimer's R47H mutant increases Abeta aggregation. *J Biol Chem* 2021; **296**: 100631.

54.   Belsare KD, Wu H, Mondal D, et al. Soluble TREM2 inhibits secondary nucleation of Abeta fibrillization and enhances cellular uptake of fibrillar Abeta. *Proc Natl Acad Sci U S A* 2022; **119**(5).

55.   Zhong L, Xu Y, Zhuo R et al. Soluble TREM2 ameliorates pathological phenotypes by modulating microglial functions in an Alzheimer's disease model. *Nat Commun* 2019; **10**(1): 1365.

56.   Song WM, Joshita S, Zhou Y, Ulland TK, Gilfillan S, Colonna M. Humanized TREM2 mice reveal microglia-intrinsic and -extrinsic effects of R47H polymorphism. *J Exp Med* 2018; **215**(3): 745-60.

57.   Reifschneider A, Robinson S, van Lengerich B, et al. Loss of TREM2 rescues hyperactivation of microglia, but not lysosomal deficits and neurotoxicity in models of progranulin deficiency. *EMBO J* 2022; **41**(4): e109108.

58.   Molgora M, Esaulova E, Vermi W, et al. TREM2 Modulation Remodels the Tumor Myeloid Landscape Enhancing Anti-PD-1 Immunotherapy. *Cell* 2020; **182**(4): 886-900 e17.

59.   Sun R, Han R, McCornack C, et al. TREM2 inhibition triggers antitumor cell activity of myeloid cells in glioblastoma. *Sci Adv* 2023; **9**(19): eade3559.

60.   Katzenelenbogen Y, Sheban F, Yalin A, et al. Coupled scRNA-Seq and Intracellular Protein Activity Reveal an Immunosuppressive Role of TREM2 in Cancer. *Cell* 2020; **182**(4): 872-85 e19.

61.   Bartels T, De Schepper S, Hong S. Microglia modulate neurodegeneration in Alzheimer's and Parkinson's diseases. *Science* 2020; **370**(6512): 66-9.

62.   Kim B, Suh E, Nguyen AT, et al. TREM2 risk variants are associated with atypical Alzheimer's disease. *Acta Neuropathol* 2022; **144**(6): 1085-102.

63.   Engelman CD, Darst BF, Bilgel M, et al. The effect of rare variants in TREM2 and PLD3 on longitudinal cognitive function in the Wisconsin Registry for Alzheimer's Prevention. *Neurobiol Aging* 2018; **66**: 177.e1-.e5.

64.   Olive C, Ibanez L, Farias FHG, et al. Examination of the Effect of Rare Variants in TREM2, ABI3, and PLCG2 in LOAD Through Multiple Phenotypes. *J Alzheimers Dis* 2020; **77**(4): 1469-82.

65.   Del-Aguila JL, Fernández MV, Schindler S, et al. Assessment of the Genetic Architecture of Alzheimer's Disease Risk in Rate of Memory Decline. *J Alzheimers Dis* 2018; **62**(2): 745-56.

66.   Parhizkar S, Arzberger T, Brendel M, et al. Loss of TREM2 function increases amyloid seeding but reduces plaque-associated ApoE. *Nat Neurosci* 2019; **22**(2): 191-204.

67.   Prokop S, Miller KR, Labra SR, et al. Impact of TREM2 risk variants on brain region-specific immune activation and plaque microenvironment in Alzheimer's disease patient brain samples. *Acta Neuropathol* 2019; **138**(4): 613-30.

68.   Joshi P, Riffel F, Kumar S, et al. TREM2 modulates differential deposition of modified and non-modified Aβ species in extracellular plaques and intraneuronal deposits. *Acta Neuropathol Commun* 2021; **9**(1): 168.

69.   Dash R, Munni YA, Mitra S, et al. Dynamic insights into the effects of nonsynonymous polymorphisms (nsSNPs) on loss of TREM2 function. *Sci Rep* 2022; **12**(1): 9378.

70.   Urdánoz-Casado A, de Gordoa JS, Robles M, et al. Profile of TREM2-Derived circRNA and mRNA Variants in the Entorhinal Cortex of Alzheimer's Disease Patients. *Int J Mol Sci* 2022; **23**(14).

71.   Del-Aguila JL, Benitez BA, Li Z, et al. TREM2 brain transcript-specific studies in AD and TREM2 mutation carriers. *Mol Neurodegener* 2019; **14**(1): 18.

72.   Shaw BC, Snider HC, Turner AK, Zajac DJ, Simpson JF, Estus S. An Alternatively Spliced TREM2 Isoform Lacking the Ligand Binding Domain is Expressed in Human Brain. *J Alzheimers Dis* 2022; **87**(4): 1647-57.

73.   Kiianitsa K, Kurtz I, Beeman N, et al. Novel TREM2 splicing isoform that lacks the V-set immunoglobulin domain is abundant in the human brain. *J Leukoc Biol* 2021; **110**(5): 829-37.

74.   Filipello F, Goldsbury C, You SF, Locca A, Karch CM, Piccio L. Soluble TREM2: Innocent bystander or active player in neurological diseases? *Neurobiol Dis* 2022; **165**: 105630.

75.   Han S, Na Y, Koh I, Nho K, Lee Y. Alternative Splicing Regulation of Low-Frequency Genetic Variants in Exon 2 of TREM2 in Alzheimer's Disease by Splicing-Based Aggregation. *Int J Mol Sci* 2021; **22**(18).

76.   Moutinho M, Coronel I, Tsai AP, et al. TREM2 splice isoforms generate soluble TREM2 species that disrupt long-term potentiation. *Genome Med* 2023; **15**(1): 11.

77.   Morenas-Rodriguez E, Li Y, Nuscher B, et al. Soluble TREM2 in CSF and its association with other biomarkers and cognition in autosomal-dominant Alzheimer's disease: a longitudinal observational study. *Lancet Neurol* 2022; **21**(4): 329-41.

78.   Palleis C, Sauerbeck J, Beyer L, et al. In Vivo Assessment of Neuroinflammation in 4-Repeat Tauopathies. *Mov Disord* 2021; **36**(4): 883-94.

79.   Liu D, Cao B, Zhao Y, et al. Soluble TREM2 changes during the clinical course of Alzheimer's disease: A meta-analysis. *Neurosci Lett* 2018; **686**: 10-6.

80.   Suarez-Calvet M, Morenas-Rodriguez E, Kleinberger G, et al. Early increase of CSF sTREM2 in Alzheimer's disease is associated with tau related-neurodegeneration but not with amyloid-beta pathology. *Mol Neurodegener* 2019; **14**(1): 1.

81.   Ma LZ, Tan L, Bi YL, et al. Dynamic changes of CSF sTREM2 in preclinical Alzheimer's disease: the CABLE study. *Mol Neurodegener* 2020; **15**(1): 25.

82.   Zhao N, Qiao W, Li F, et al. Elevating microglia TREM2 reduces amyloid seeding and suppresses disease-associated microglia. *J Exp Med* 2022; **219**(12).

83.   Carmona S, Zahs K, Wu E, Dakin K, Bras J, Guerreiro R. The role of TREM2 in Alzheimer's disease and other neurodegenerative disorders. *Lancet Neurol* 2018; **17**(8): 721-30.

84.   Winfree RL, Dumitrescu L, Blennow K, et al. Biological correlates of elevated soluble TREM2 in cerebrospinal fluid. *Neurobiol Aging* 2022; **118**: 88-98.

85.   Uhlmann RE, Rother C, Rasmussen J, et al. Acute targeting of pre-amyloid seeds in transgenic mice reduces Alzheimer-like pathology later in life. *Nat Neurosci* 2020; **23**(12): 1580-8.

86.   Ewers M, Biechele G, Suarez-Calvet M, et al. Higher CSF sTREM2 and microglia activation are associated with slower rates of beta-amyloid accumulation. *EMBO Mol Med* 2020; **12**(9): e12308.

87.   Pereira JB, Janelidze, S., Strandberg, O., Whelan, C.D., Zetterberg, H., Blennow, K., Plamqvist, S., Stomrud, E., Mattsson-Carlgren, N., Hansson, O. Microglial activation protects against accumulation of tau aggregates in nondemented individuals with underlying Alzheimer's disease pathology. *Nature Aging* 2022; **2**(12): 1138-1144.

88.   Araque Caballero M, Suárez-Calvet M, Duering M, et al. White matter diffusion alterations precede symptom onset in autosomal dominant Alzheimer's disease. *Brain* 2018; **141**(10): 3065-80.

89.   Pascoal TA, Benedet AL, Ashton NJ, et al. Microglial activation and tau propagate jointly across Braak stages. *Nat Med* 2021.

90.   Hu WT, Ozturk T, Kollhoff A, Wharton W, Christina Howell J. Higher CSF sTNFR1-related proteins associate with better prognosis in very early Alzheimer's disease. *Nat Commun* 2021; **12**(1): 4001.

91.   Ewers M, Franzmeier N, Suarez-Calvet M, et al. Increased soluble TREM2 in cerebrospinal fluid is associated with reduced cognitive and clinical decline in Alzheimer's disease. *Sci Transl Med* 2019; **11**(507).

92.   Zhao A, Jiao Y, Ye G, et al. Soluble TREM2 levels associate with conversion from mild cognitive impairment to Alzheimer's disease. *J Clin Invest* 2022; **132**(24).

93.   Deming Y, Filipello F, Cignarella F, et al. The MS4A gene cluster is a key modulator of soluble TREM2 and Alzheimer's disease risk. *Sci Transl Med* 2019; **11**(505).

94.   Falcon C, Monte-Rubio GC, Grau-Rivera O, et al. CSF glial biomarkers YKL40 and sTREM2 are associated with longitudinal volume and diffusivity changes in cognitively unimpaired individuals. *Neuroimage Clin* 2019; **23**: 101801.

95.   Choi H, Choi Y, Lee EJ, et al. Hippocampal glucose uptake as a surrogate of metabolic change of microglia in Alzheimer's disease. *J Neuroinflammation* 2021; **18**(1): 190.

96.   Araque Caballero MA, Suarez-Calvet M, Duering M, et al. White matter diffusion alterations precede symptom onset in autosomal dominant Alzheimer's disease. *Brain* 2018; **141**(10): 3065-80.

97.   van Dyck CH, Swanson CJ, Aisen P, et al. Lecanemab in Early Alzheimer's Disease. *N Engl J Med* 2023; **388**(1): 9-21.

98.   Ewers M, Biechele G, Suárez-Calvet M, et al. Higher CSF sTREM2 and microglia activation are associated with slower rates of beta-amyloid accumulation. *EMBO Mol Med* 2020; **12**(9): e12308.

99.   Cignarella F, Filipello F, Bollman B, et al. TREM2 activation on microglia promotes myelin debris clearance and remyelination in a model of multiple sclerosis. *Acta Neuropathol* 2020.

100.   Bosch-Queralt M, Cantuti-Castelvetri L, Damkou A, et al. Diet-dependent regulation of TGFbeta impairs reparative innate immune responses after demyelination. *Nat Metab* 2021; **3**(2): 211-27.