An Alzheimer-associated TREM2 variant occurs at the ADAM cleavage site and affects shedding and phagocytic function

Kai Schlepckow, Gernot Kleinberger, Akio Fukumori, Regina Feederle, Stefan F Lichtenthaler, Harald Steiner & Christian Haass

Abstract

Sequence variations occurring in the gene encoding the triggering receptor expressed on myeloid cells 2 (TREM2) support an essential function of microglia and innate immunity in the pathogenesis of Alzheimer’s disease (AD) and other neurodegenerative disorders. TREM2 matures within the secretory pathway, and its ectodomain is shed on the plasma membrane. Missense mutations in the immunoglobulin (Ig)-like domain such as p.T66M and p.Y38C retain TREM2 within the endoplasmic reticulum and reduce shedding as well as TREM2-dependent phagocytosis. Using mass spectrometry, we have now determined the cleavage site of TREM2. TREM2 is shed by proteases of the ADAM (a disintegrin and metalloproteinase domain containing protein) family C-terminal to histidine 157, a position where an AD-associated coding variant has been discovered (p.H157Y) in the Han Chinese population. Opposite to the characterized mutations within the Ig-like domain, such as p.T66M and p.Y38C, the p.H157Y variant within the stalk region leads to enhanced shedding of TREM2. Elevated ectodomain shedding reduces cell surface full-length TREM2 and lowers TREM2-dependent phagocytosis. Therefore, two seemingly opposite cellular effects of TREM2 variants, namely reduced versus enhanced shedding, result in similar phenotypic outcomes by reducing cell surface TREM2.

Keywords: Alzheimer’s disease; neurodegeneration; phagocytosis; regulated intramembrane proteolysis; TREM2

Introduction

Inflammation and activation of brain-resident immune cells are common hallmarks of numerous neurological disorders. A pivotal role of microgliosis has been recognized since a long time specifically in neurodegenerative disorders (Lyman et al, 2014; Villegas-Llerena et al, 2016). A central role of microglial function in disease pathogenesis is now further supported by the identification of sequence variants and mutations in the triggering receptor expressed on myeloid cells 2 (TREM2) that are associated with an increased risk for several neurodegenerative disorders such as Alzheimer’s disease (AD), frontotemporal lobar degeneration (FTLD), Parkinson’s disease, and FTLD-like syndrome (Guerrero & Hardy, 2013; Guerrero et al, 2013; Jonsson & Stefansson, 2013; Rayaprolu et al, 2013; Borroni et al, 2014; Cady et al, 2014; Cuyvers et al, 2014) and in a homozygous state cause Nasu–Hakola disease (Klunemann et al, 2005). In the brain, TREM2 is preferentially expressed in microglia and is functionally required for migration, cytokine release, phagocytosis, lipid sensing, ApoE binding, shielding of amyloid plaques, and microglia proliferation (Kleinberger et al, 2014; Atagi et al, 2015; Bailey et al, 2015; Colonna & Wang, 2016; Ulrich & Holtzman, 2016; Yeh et al, 2016; Yuan et al, 2016). TREM2 is a type-1 transmembrane protein that shuttles to the plasma membrane where it exerts its cell autonomous biological functions. TREM2 undergoes regulated intramembrane proteolysis (RIP) (Lichtenthaler et al, 2011; Wunderlich et al, 2013) (Fig 1A), which is initiated on the cell surface via shedding of full-length TREM2 by ADAM10 (a disintegrin and metalloproteinase domain containing protein) (Kleinberger et al, 2014). Shedding by ADAM10 results in the liberation of soluble TREM2 (sTREM2), which can be detected in human cerebrospinal fluid (CSF) (Kleinberger et al, 2014; Heslegrave et al, 2016; Piccio et al, 2016; Suarez-Calvet et al, 2016a,b). The membrane-retained C-terminal fragment (CTF) is...
Figure 1.

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subsequently cleared via an intramembranous cleavage by γ-secretase (Fig 1A) (Wunderlich et al., 2013; Glebov et al., 2016). So far several mutations have been functionally investigated. Mutations within the immunoglobulin (Ig)-like domain such as p.T66M and p.Y38C apparently result in misfolding of TREM2 and retention of the immature protein within the endoplasmic reticulum (Kleinberger et al., 2014; Park et al., 2015; Song et al., 2017) although upon strong transient overexpression mutant proteins may escape retention (Kober et al., 2016). As a consequence of misfolding, reduced cell surface levels of TREM2 are observed and shedding is dramatically lowered leading to reduced sTREM2 and TREM2 CTF levels (Kleinberger et al., 2014). Consistent with that, a patient with a homozygous TREM2 p.T66M mutation had extremely low or even no detectable sTREM2 in the CSF (Kleinberger et al., 2014; Piccio et al., 2016). Lowered cell surface TREM2 results in reduced phagocytic activity (Kleinberger et al., 2014). Although initially discrepant results regarding the effects of a loss of TREM2 function on amyloid plaque pathology were reported (Jay et al., 2015; Wang et al., 2015), TREM2 loss of function may lead to the accumulation of fuzzy amyloid plaques suggesting a lack of phagocytic clearance of the plaque halo or reduced prevention of amyloid plaque growth (Wang et al., 2016; Yuan et al., 2016). In support of reduced phagocytic plaque degradation, we showed recently that immunotherapeutic clearance of amyloid plaques via phagocytosis is reduced in the absence of TREM2 (Xiang et al., 2016).

Most of the functionally investigated mutations are located within the Ig-like domain of TREM2. Misfolding of this domain, retention, and consequently reduced shedding appear to be a common readout of at least some of these variants. Although many sequence variants were found within the Ig-like domain, genetic studies also identified sequence variants within the stalk region and such mutants are unlikely to affect folding of the Ig-like domain. Since mutations in the stalk region may affect the efficacy and precision of ADAM-mediated shedding, we first determined the TREM2 cleavage site. Strikingly, ADAM-mediated cleavage within the stalk region occurs C-terminal to histidine 157 exactly where the AD-associated variant p.H157Y is located (Guerreiro et al., 2013; Ma et al., 2014; Jiang et al., 2016; Song et al., 2017). Analysis of proteolytic processing of the mutant variant revealed that higher levels of sTREM2 are generated, a finding opposite to the reduced shedding observed for mutations within the Ig-like domain such as p.T66M and p.Y38C (Kleinberger et al., 2014). However, enhanced shedding of TREM2 p.H157Y leads to reduced cell surface full-length TREM2 and hence to reduced phagocytic activity. Thus, mutations located within the Ig-like domain or the stalk region reduce surface expression of TREM2 and probably its signaling activity via completely different cellular mechanisms.

Results and Discussion

TREM2 is cleaved by ADAM proteases between histidine 157 and serine 158

To determine the cleavage site of TREM2, we followed two independent approaches. First, we determined the N-terminus of the membrane retained CTF remaining after shedding of the full-length precursor (Kleinberger et al., 2014). To do so, we expressed C-terminally Flag-tagged TREM2 (TREM2-CFlag) in human kidney 293 cells (HEK 293) (strategy 1; Fig 1B). We enriched for the ADAM generated TREM2 CTF by inhibiting its γ-secretase-mediated intramembrane cleavage with DAPT (Dovey et al., 2001). Consistent with previous results (Wunderlich et al., 2013), Western blot analysis revealed a massive accumulation of the CTF upon γ-secretase inhibition (Fig 1C). Immunoprecipitation followed by mass spectrometry of the DAPT enriched CTF revealed one major peak corresponding to a molecular mass of 8,841.48 Da (Fig 1D; Table 1). This corresponds to a CTF with an N-terminus at serine 158 (see also Fig 1J). Additional very minor peaks may be due to proteolytic degradation of the CTF most likely within lysosomes. This is
supported by the almost complete absence of such minor peaks in the analysis of the cleavage site of sTREM2 (see below).

So far ADAM10 has been described as the sole sheddase of TREM2 (Kleinberger et al., 2014). However, shedding is well known to be stimulated after protein kinase C activation. Under these conditions, shedding is predominantly performed by ADAM17 (Black, 2002; Saltig & Lichtenhalter, 2015). To prove whether TREM2 shedding can be stimulated similarly and whether cleavage still occurs at the same site, we treated HEK293 cells stably expressing TREM2-CFlag with phorbol 12-myristate 13-acetate (PMA). This resulted in a robust increase in TREM2 shedding (Fig 1E). Similarly, and in line with previous results (Nitsch et al., 1992), shedding of the amyloid precursor protein (APP) was also stimulated (Fig 1E; lower panel). To determine the cleavage site of TREM2 after PMA-stimulated shedding, we again enriched for the CTF by γ-secretase inhibition (Fig 1C) followed by immunoprecipitation with an anti-Flag antibody. Mass spectrometry of the CTF revealed one major peak corresponding to a molecular mass of 8,837.09 Da (Fig 1F; Table 1). Thus, after PMA-stimulated TREM2 shedding, the proteolytic cleavage also occurs between histidine 157 and serine 158 (see also Fig 1J).

Table 1. Summary of identified peptides and comparison of observed masses to calculated masses. [M + H]+ indicates a singly charged peptide.

<table>
<thead>
<tr>
<th>Peptide</th>
<th>Cleavage</th>
<th>Sequence</th>
<th>Mass [M + H]+ (Da)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TREM2-CFlag WT (HEK)</td>
<td>N-terminal of serine 158 (P1')</td>
<td>SISRSLEEIEIP…DYKDDDDK</td>
<td>8,840.08 8,841.48</td>
</tr>
<tr>
<td>TREM2-CFlag WT (PMA, HEK)</td>
<td>N-terminal of serine 158 (P1')</td>
<td>SISRSLEEIEIP…DYKDDDDK</td>
<td>8,840.08 8,837.09</td>
</tr>
<tr>
<td>TREM2-CFlag p.H157Y (HEK)</td>
<td>N-terminal of serine 158 (P1')</td>
<td>SISRSLEEIEIP…DYKDDDDK</td>
<td>8,840.08 8,832.76</td>
</tr>
<tr>
<td>TREM2-TEVFlag (HEK)</td>
<td>C-terminal of histidine 157 (P1)</td>
<td>GDYKDDDDKLDHRDAGLWFGESESFEDAHVEH</td>
<td>3,948.01 3,947.78</td>
</tr>
<tr>
<td>TREM2-TEVFlag (THP-1)</td>
<td>C-terminal of histidine 157 (P1)</td>
<td>GDYKDDDDKLDHRDAGLWFGESESFEDAHVEH</td>
<td>3,948.01 3,943.80</td>
</tr>
</tbody>
</table>

The cleavage site of TREM2 after histidine 157 coincides with the p.H157Y late-onset AD-associated mutation recently described in the Han Chinese population (Ma et al., 2014; Jiang et al., 2016) (Fig 2A). Since it is well known that an AD-related mutation at the site of β-secretase-mediated shedding of APP pathologically affects APP proteolysis (Citron et al., 1992; Cai et al., 1993; Haass et al., 1995b; Thinakaran et al., 1996), we investigated proteolytic processing of TREM2 p.H157Y. Surprisingly, and opposite to the p.T66M and p.Y38C mutations, we found enhanced shedding of the p.H157Y mutant by Western blotting (Fig 2B). This was independently confirmed by anti-HA ELISA-mediated quantitation (Fig 2C). Since shedding of p.H157Y is significantly increased and the amino acid sequence is changed at the P1 site, we next asked whether the cleavage still occurs after amino acid 157. As described above, we enriched for the CTF produced from TREM2 p.H157Y via inhibition of γ-secretase by DAPT (see also Fig 2H). Mass spectrometry of the enriched CTF revealed one major peak corresponding to a molecular mass of 8,832.76 Da, which is consistent with a predominant cleavage after amino acid 157 (Fig 2D and E; Table 1). Thus, TREM2 p.H157Y is cleaved at the very same position as wild-type (wt) TREM2. Furthermore, TREM2 p.H157Y is also shed by members of the ADAM family, since the protease inhibitors GM6001 and GI254023X significantly reduce shedding (Fig 2F). Enhanced shedding of the p.H157Y variant is accompanied by reduced levels of mature fully glycosylated TREM2 (Fig 2G). Despite increased shedding and reduced mature TREM2, we surprisingly observed less CTFs suggesting enhanced degradation (Fig 2G). In line with that, the CTF derived from TREM2 p.H157Y is recovered after inhibition of γ-secretase cleavage (Fig 2H). Furthermore, additional expression of DAP12, which forms a tight complex with TREM2 (Paradowska-Gorycka & Jurkowska, 2013), prevents γ-secretase-dependent degradation of the p.H157Y TREM2 CTF (Fig 2I). Enhanced shedding
Figure 2.
suggests reduced levels of TREM2 on the plasma membrane. Levels of cell surface mutant and wt TREM2 were determined by cell surface biotinylation. In line with our previous findings (Kleinberger et al., 2014), wt TREM2 was readily observed on the plasma membrane (Fig 2j). In contrast, p.H157Y could not be biotinylated on the cell surface similar to the previously investigated p.T66M mutant (Kleinberger et al., 2014) (Fig 2i). Thus, the histidine-to-tyrosine exchange at amino acid 157 increases shedding of mutant TREM2 and, as a consequence, reduces cell surface levels of the fully mature protein.

The disease-associated mutation reduces phagocytic activity

Both the p.T66M and the p.H157Y mutations lead to reduced cell surface TREM2, albeit via opposite cellular mechanisms. Since the amount of cell surface TREM2 correlates with its cell autonomous function (Kleinberger et al., 2014; Song et al., 2017), we hypothesized that both mutations may result in a similar loss of function. To investigate TREM2-mediated function, we analyzed TREM2-dependent phagocytosis. In line with our previous findings (Kleinberger et al., 2014), wt TREM2 readily promoted uptake of Escherichia coli conjugated to pHrodo (Fig 3). Uptake was specific since treatment with cytochalasin D blocked engulfment of E. coli. Strikingly, cells expressing TREM2 p.H157Y exhibited a significantly reduced phagocytic capacity (Fig 3), demonstrating that this mutation at least affects phagocytic uptake of bacteria.

Sequence variations of TREM2 have been found in all domains of the protein. However, most of the mutations occur within the Ig-like domain. At least some of the mutant proteins, such as p.T66M and p.V38C, are likely to be misfolded and retained within the endoplasmic reticulum (Kleinberger et al., 2014; Park et al., 2015; Song et al., 2017). Therefore, these types of mutations reduce cell surface TREM2 and consequently the release of sTREM2, as shedding predominantly occurs on the plasma membrane. Consistent with reduced cell surface TREM2, biological functions of TREM2 such as lipid sensing, ApoE binding, and phagocytosis are all decreased by such mutations (Kleinberger et al., 2014; Atagi et al., 2015; Bailey et al., 2015; Yeh et al., 2016). Thus, for this class of mutations reduced release of sTREM2 may serve as a surrogate marker for
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Materials and Methods

cDNA constructs

cDNA constructs that were used in this study have been previously described (Kleinberger et al., 2014). Briefly, TREM2-CFlag as denoted in the main text is full-length TREM2 with N- and C-terminal HA and Flag tags, respectively. TREM2-DAP12 fusion constructs contained the ectodomain of TREM2 including aa169 (Proline169) fused to DAP12 (aa28–113). Furthermore, an amino acid change in the transmembrane domain of DAP12 from aspartic acid to alanine (p.D50A) was included (Hamerman et al., 2006). Additionally, the TREM2-DAP12 fusion constructs included a HA-tag and a linker sequence (SGGGGGL) after the endogenous TREM2 signal peptide. The TREM2 missense mutation p.H157Y (CAC>CAC) was introduced into the respective plasmid by site-directed mutagenesis (Stratagene). The TREM2-TEVFlag construct was generated according to a previously reported strategy (Fukumori et al., 2010). The TEVFlag sequence (ENLYFQGDYKDDDDK) was introduced after amino acid 132. All constructs were verified by DNA sequencing (GATC Biotech).

Cell culture

Generation and maintenance of stable HEK293 Flp-In cells lines were performed as previously described (Kleinberger et al., 2014). Human monocytic THP-1 cells were cultured in RPMI1640 medium supplemented with 10% FCS (Sigma), 1% penicillin/streptomycin, and 50 μM β-mercaptoethanol. THP-1 cells were kept in culture at densities between 2 × 10^5 and 1 × 10^6 cells per ml of cell culture medium, and transfections were performed at densities between 5 × 10^5 and 1 × 10^6 cells per ml of cell culture medium. Transfections were carried out with 1 μg of TREM2-TEVFlag DNA per 1 × 10^6 of THP-1 cells using a nucleofector device and the 4G Cell Line 4D-Nucleofector® X Kit (both Lonza). GFP cDNA was transfected in parallel to check for transfection efficiency. Transient transfection of cells stably expressing wt or mutant TREM2 with human DAP12 was performed in a 6-well plate using 2 μg DNA/well and Lipofectamine 2000. If not stated otherwise, cell culture reagents were purchased from Thermo Fisher Scientific.

Antibodies

For immunoblot detection, the following antibodies were used: rat monoclonal anti-HA conjugated to HRP (3F10; 1:700 to 1:2,000; Roche), goat polyclonal anti-human TREM2 (AF1828; 1:100; R&D Systems), rat monoclonal antibody against a C-terminal peptide of human TREM2 (9D11; 1:20; C-HQKPGTHPPSELDCGHDGP), rat monoclonal antibody against sAPPα (2D8 (Shirotani et al., 2007); 1:100), and rabbit anti-calnexin (1:3,000, Enzo Life Sciences). The HRP-conjugated goat anti-rat (1:10,000; Santa Cruz Biotechnology), donkey anti-goat (1:10,000; Santa Cruz Biotechnology), and goat anti-rabbit IgG (1:10,000; Promega) were used as secondary antibodies.

Cell surface biotinylation

Surface biotinylations were carried out as described previously (Kleinberger et al., 2014).
Preparation of conditioned media, cell lysates, and immunoblotting

HEK293 Flp-In cells stably expressing respective cDNA constructs were seeded at a density of $1.5 \times 10^6$/cm$^2$, and medium was changed 48 h post-seeding. Inhibitors/activators used were GM 6001 (25 μM), GI 254023X (5 μM), DAPT (5 μM, all Sigma), and phorbol 12-myristate 13-acetate (PMA; 100 nM; Enzo Life Sciences). Conditioned medium was collected after 18–20 h except for PMA-treated cells where media were collected after 2 h. Membrane fractions were prepared as previously described (Kleinberger et al., 2014). Alternatively, total lysates were prepared using STE lysis buffer (150 mM NaCl, 50 mM Tris/HCl pH 7.6, 2 mM EDTA, 1% Triton X-100). Lysis was carried out on ice for 20 min, and lysates were obtained upon centrifugation at 15,871 g for 30 min at 4°C. To generate lysates upon transient transfection of THP-1 monocytes, cells were gently centrifuged (100 g for 10 min at 4°C) after overnight incubation at 37°C, washed once with ice-cold phosphate-buffered saline (PBS), centrifuged again (100 g for 10 min at 4°C), and subsequently lysed. Protein concentrations were measured using the BCA method, equal amounts of protein were mixed with Laemmli sample buffer supplemented with β-mercaptoethanol, separated by standard 15% SDS-PAGE, transferred onto polyvinylidene difluoride membranes (Hybond P; Amersharm Biosciences), processed with respective antibodies, and developed using enhanced chemiluminescence technique (Pierce).

Phagocytosis assay

Phagocytosis of fluorogenic E. coli particles (pHrodo Green, Molecular Probes) after 60- or 120-min incubation at 37°C was performed as described before (Kleinberger et al., 2014). As a negative control, phagocytosis was inhibited with cytochalasin D (10 μM; Sigma).

sTREM2 ELISA

sTREM2 levels from conditioned media were essentially determined as previously described (Kleinberger et al., 2014). Rat anti-HA antibody (3F10; 1:1,000; Roche) was used as detection antibody.

MALDI-TOF mass spectrometry analysis of ectodomain cleavage

For the identification of the N-terminus of the TREM2 CTF, HEK293 Flp-In cells stably expressing TREM2-CTFlag (both wt and p.H157Y) were harvested in ice-cold PBS upon overnight treatment using DAPT. Upon treatment with PMA, cells were harvested 5 h post-treatment. Cell pellets were frozen at −20°C until use. Cells were lysed in lysis buffer (4% n-dodecyl β-D-maltoside, 0.1% N-octylglycoside, 10 mM Tris–HCl, pH 8.0, 5 mM EDTA, and 140 mM NaCl) containing protease inhibitor mix (Sigma) for 20 min on ice. Following a clarifying spin at 21,000 g for 10 min, supernatants were subjected to a second clarifying spin by ultracentrifugation at 100,000 g for 1 h and incubated with anti-FLAG M2-agarose (Sigma) overnight by rotation at 4°C. Beads were washed four times with immunoprecipitation/mass spectrometry (IP/MS) buffer (0.1% N-octylglucoside, 10 mM Tris–HCl, pH 8.0, 5 mM EDTA, and 140 mM NaCl) and two times with water. Beads were stored at −20°C until MS analysis. Control samples were generated using HEK293 Flp-In cells stably transfected with an empty expression vector (pcDNA5/FRT/TO, Thermo Fisher Scientific).

For the identification of the C-terminus of the shed ectodomain, the TREM2-TEVFlag cDNA construct was transfected transiently into HEK293 Flp-In cells followed by addition of fresh medium 24 h post-transfection. After further 5 h, the supernatant was collected and cleared by centrifugation at 15,000 g for 30 min at 4°C. The pH of the supernatant was adjusted to pH 8.0 using 1 M Tris/HisCl (30 mM final concentration). 0.5 M EDTA pH 8.0 (3.5 mM final concentration) was added, and the supernatant was incubated with 40 μl anti-FLAG M2-agarose overnight by rotation at 4°C. Beads were washed four times with IP/MS buffer and two times with water. The TREM2 ectodomain was eluted from the beads with 40 μl 100 mM glycine pH 2.5 for 10 min by rotation at 4°C. Upon centrifugation (5 min at 1,200 g), the supernatant was neutralized by addition of 1/8 volume 1 M Tris–HCl pH 8.0. After addition of EDTA, DTT (final concentrations of 5 mM and 1 mM, respectively), and complete protease inhibitor (Roche), 10 units of AcTEV protease (Thermo Fisher Scientific) were added and digestion was carried out overnight at 4°C. Upon addition of 1 ml of IP/MS buffer, 10 μl anti-FLAG M2-agarose was added and immunoprecipitation was conducted for 1 h by rotation at 4°C. Beads were washed three times with IP/MS buffer and three times with water and stored at −20°C until MS analysis. In case of THP-1 monocytes, 30 × 10⁶ cells were transiently transfected with the TREM2-TEVFlag cDNA construct. Upon overnight incubation at 37°C, cells were spun down and media were processed as described above to give samples for mass spectrometry. Control samples were generated using either HEK293 Flp-In cells or THP-1 cells transiently transfected with an empty expression vector (EV; pcDNA5/FRT/TO).

MS analysis was performed using a 4800 MALDI TOF/TOF analyzer (Applied Biosystems) essentially as described previously (Fukumori et al., 2010). Immunoprecipitated peptides were eluted from the beads using 0.3% trifluoroacetic acid (TFA) in 50% acetonitrile and saturated with α-cyano-4-hydroxy cinnamic acid. The dissolved samples were dried on a stainless plate and subjected to MALDI-TOF MS analysis.

Statistics

The data were analyzed using GraphPad Prism 5 (GraphPad Software, Inc.) and in SPSS IBM, version 20.0. Group comparisons in the levels of sTREM2 (Fig 2C) or in phagocytosis assays (Fig 3) were analyzed by one-way ANOVA test followed by Dunnett's or Tukey's post hoc tests for pairwise comparisons, respectively. All tests were 2-tailed, and statistical significance was set to $P < 0.05$.

Expanded View for this article is available online.

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The paper explained

Problem
A central role of microglial function in disease pathogenesis is strongly supported by the identification of sequence variants in the triggering receptor expressed on myeloid cells 2 (TREM2) that are associated with an increased risk for several neurodegenerative disorders. Most of the functionally investigated mutations are located within the Ig-like domain of TREM2. Misfolding of this domain, retention, and consequently reduced shedding appear to be a common read-out of at least some of these variants. Although many sequence variants were found within the Ig-like domain, genetic studies also identified sequence variants within the stalk region and such mutants are unlikely to affect folding of the Ig-like domain. Since mutations in the stalk region may rather affect the efficacy and precision of ADAM-mediated shedding, we determined the TREM2 cleavage site. Moreover, modulation of TREM2 shedding, for example, by its selective inhibition, may be an innovative therapeutic approach, for which knowledge of the exact cleavage site is absolutely required.

Results
TREM2 is shed by proteases of the ADAM (a disintegrin and metallo-proteinase domain containing protein) family C-terminal to histidine 157, a position where an AD-associate coding variant has been discovered (p.H157Y) in the Han Chinese population. Opposite to the characterized mutations within the Ig-like domain, such as p.T66M and p.Y58C, the p.H157Y variant within the stalk region leads to enhanced shedding of TREM2. Elevated ectodomain shedding reduces cell surface full-length TREM2 and lowers TREM2-dependent phagocytosis.

Impact
Two seemingly opposite cellular effects of TREM2 variants, namely reduced versus enhanced shedding, result in similar phenotypic outcomes by reducing cell surface TREM2. Moreover, these findings may open the opportunity to therapeutically modulate TREM2 function by selectively blocking access of ADAM proteases to the cleavage site between histidine 157 and serine 158. In contrast to blocking ADAM 10/17 activity with conventional protease inhibitors, this approach would be selective and avoid cleavage inhibition of numerous ADAM10/17 substrates.

References
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