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A Review: Metabolism of Lipid Via TREM-2A

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Abstract

This review investigated the metabolism of lipid via TREM-2. Lipid metabolism is the synthesis and degradation of lipids in cells, involving the breakdown and storage of fats for energy and the synthesis of structural and functional lipids, such as those involved in the construction of cell membranes. Lipid metabolism is often considered as the digestion and absorption process of dietary fat; however, there are two sources of fats that organisms can use to obtain energy: from consumed dietary fats and from stored fat. Triggering receptor expressed on myeloid cells 2 (TREM-2) is a membrane receptor on myeloid cells and plays an important role in the body's immune defense. Recently, TREM-2 has received extensive attention from researchers, and its activity has been found in Alzheimer's disease, neuroinflammation, and traumatic brain injury. The appearance of TREM-2 is usually accompanied by changes in apolipoprotein E (ApoE). Apolipoprotein E (ApoE) is a protein playing a pivotal role in lipid homeostasis since it regulates cholesterol, triglyceride and phospholipid metabolism in the blood and the brain. APOE gene regulates the expression of this protein and has three different alleles: ε2, ε3 and ε4. Carrying an APOE4 allele is recognized as a genetic risk factor of late-onset Alzheimer's disease (LOAD) and coronary heart disease (CHD). A

major function of apoE is to mediate the binding of lipoproteins or lipid complexes in the plasma or interstitial fluids to specific cell-surface receptors. These receptors internalize apoE-containing lipoprotein particles; thus, apoE participates in the distribution/redistribution of lipids among various tissues and cells of the body. It is likely that apoE, with its multiple cellular origins and multiple structural and biophysical properties, is involved widely in processes of lipid metabolism and neurobiology, possibly encompassing a variety of disorders of neuronal repair, remodeling, and degeneration by interacting with different factors through various pathways.

Keywords: Metabolism, Myeloid, Alleles, Neuroinflammation, Apolipoprotein E, Homeostasis, Phospholipid, Alzheimer, Coronary, Remodeling

Introduction

Lipid metabolism is the process by which lipids are synthesized and broken down in cells, It involves producing structural and functional lipids, such as those required to construct cell membranes, as well as digesting and storing fats for energy. Animal livers manufacture these fats, which are obtained through diet. Lipogenesis is the process by which these fats are produced. (Freifelder, 1987; Chemistry Encyclopedia, 2016). The majority of triglycerides in the circulatory system are composed of cholesterol and triglycerides. produces through food consumption (Baynes, 2014). Ophardt (2013) notes that hydrolysis, which is aided by several digestive system enzymes, is frequently the first step in lipid metabolism (Chemistry Encyclopedia, 2016).

The human TREM-2 gene codes for the Triggering receptor expressed on myeloid cells 2 (TREM-2) protein (Bouchon *et al*., 2000; Paloneva *et al*., 2002). According to Rodríguez-Gómez et al. (2020), TREM-2 is expressed in immune cells in the central nervous system, including macrophages, immature monocyte-derived dendritic cells, osteoclasts, and microglia (Masuda *et ai*., 2020). Numerous cell types in the liver, including damage-reactive macrophages, express TREM-2 (Sun *et al*., 2020). TREM-2 is expressed in the intestine by macrophages and dendritic cells generated from myeloid cells (Genua *et al*., 2014). TREM-2 has anti-inflammatory properties and is overexpressed in a variety of tumor forms. As a result, it could make a useful therapeutic target. Even while lipid metabolism is widely acknowledged to be important, there are still gaps in our knowledge of the complex regulatory systems governing these activities. To preserve homeostasis, cellular reactions to

lipid availability, storage, and consumption are strictly regulated. Numerous illnesses, such as metabolic disorders, cardiovascular diseases, and neurodegenerative ailments, are linked to dysregulation of lipid metabolism. Therefore, deciphering the intricacies of lipid metabolism and creating focused therapies for related illnesses require a thorough understanding of the regulatory systems.

Lipid Metabolism

According to Brownsey *et al.* (2006), The pancreas and small intestine secrete lipase, which hydrolyzes fat's fatty acids to create free fatty acids and monoglycerides. Lipid metabolism is the term for this process. Bile is the primary fluid that emulsifies most of the fat the body consumes into tiny particles. Fatty acids and cholesterol are produced by fully hydrolyzing a small quantity of fatty acids. These microscopic molecules, which include triglycerides and both short- and medium-chain lipids, are hydrolyzed in the small intestine and then absorbed into the bloodstream.

During the process of lipid metabolism, fatty acids undergo combustion to generate new lipids from smaller component molecules or to provide energy. Lipid metabolism and carbohydrate metabolism are related because acetyl CoA and other metabolites of glucose can be converted into lipids.

Metabolism Processes

Lipid digestion, absorption, transport, storage, catabolism, and biosynthesis are examples of metabolic processes. The process of beta-oxidation, which occurs in the peroxisome and mitochondrial cell organelles, is responsible for lipid breakdown.

Lipid Digestion

The first stage of lipid metabolism is called digestion, and it involves the use of carbohydrates to smaller monoglyceride molecules using lipase enzymes. Lingual lipase is the first enzyme involved in the chemical breakdown of fats in the mouth. The lipases do not degrade ingested cholesterol; instead, they remain undiminished until it reaches the small intestine's epithelial cells. After that, lipids proceed to the stomach, where gastric lipase continues the chemical breakdown of the food and peristalsis, or mechanical digestion, starts. Once the fats enter the small intestine, however, most of the digestion and absorption of lipids takes place there.

Pelley (2012) claims that to help with the breakdown of triglycerides, the pancreas secretes two substances into the small intestine: bile salt-dependent lipase and the pancreatic lipase family. The triglycerides are subsequently further physically broken down into individual fatty acid units so that the small intestine's epithelial cells may absorb them (Voet *et al.*, 2013). Pancreatic lipase produces the signal for the decomposition of triglyceride to separate components of free fatty acid and glycerol.

Absorption of Lipids

The second stage in the chemical breakdown of lipids is fat absorption. Although the small intestine is the primary site of fat absorption, the gastrointestinal tract can also absorb short-chain fatty acids. Once triglyceride molecules are broken down into individual lipids such as fatty acids, glycerols, and cholesterol, they will assemble to form structures called micelles. Fatty acids and monoglycerides break free from the micelles, traverse the membrane, and enter the intestinal epithelial cells. In the cytoplasm of epithelial cells, fatty acids and monoglycerides are recombined back into triglycerides. In the cytoplasm of epithelial cells, triglycerides, and cholesterol are encased in bigger particles known as chylomicrons, which are amphipathic structures that contain digested lipids. Chylomicrons enter the body's adipose and other tissues through the bloodstream (Chemistry Encyclopedia, 2016).

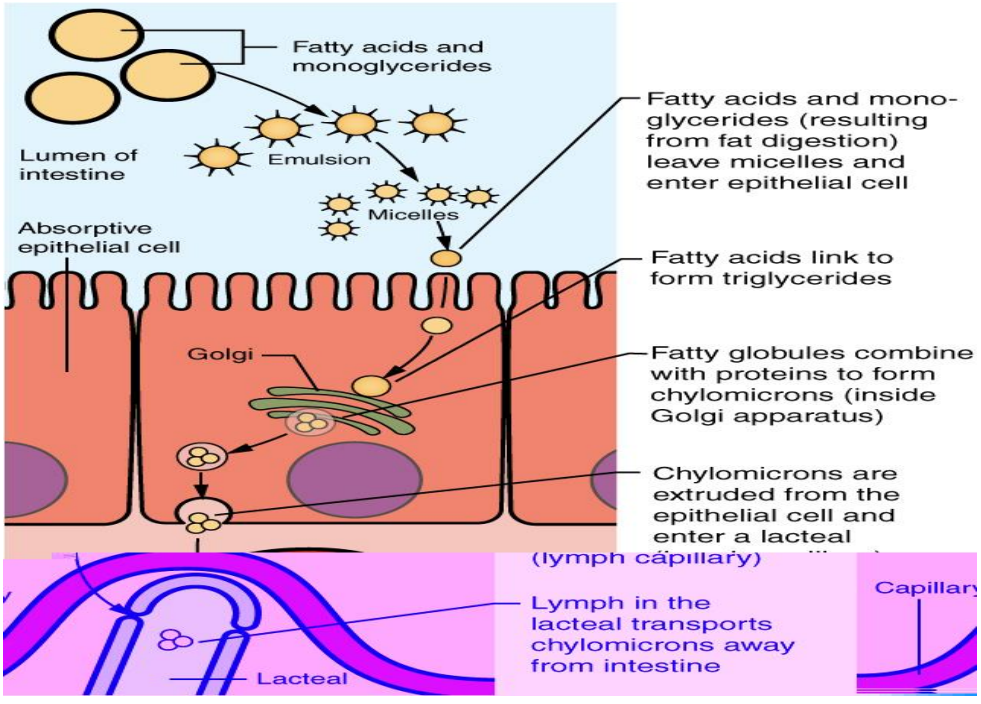

Figure 1: Chart showing the lipid absorption process

Lipid transportation

Triglycerides, cholesterol, and other membrane lipids are hydrophobic, necessitating the use of unique transport proteins called lipoproteins. The blood can carry cholesterol and triglycerides due to the lipoproteins' amphipathic nature. One subclass of lipoproteins that transports Chylomicrons are digested lipids that are transported from the small intestine to the remainder of the body. The various lipid types that lipoproteins transport is reflected in their varying densities (Harris, 2009).

Lipid storage

Triglycerides, a type of lipid, are kept in white adipose tissue. The mass of accumulated triglycerides in a lean young adult human is approximately 10-20 kilos. Glycerol with three fatty acids forms the backbone of triglycerides. The triglyceride droplet is ultimately reached via the esterification of free fatty acids once they are converted to acyl-CoA. Lipoprotein lipase has a crucial function.

Lipid Catabolism

When cytoplasmic (or simply any lipoproteins) travel through tissues, triglycerides are liberated and subsequently hydrolyzed on the luminal surface of capillary endothelial cells by lipoprotein lipase (Feingold and Grunfeld, 2000). Triglycerides are changed into glycerol as well as fatty acids before entering cells; any remaining cholesterol is then transported back via the circulatory system to the liver.

In the cytoplasm of the cell (such as a muscle cell), glycerol is converted into glyceraldehyde 3-phosphate, an intermediary in glycolysis, where it is further oxidized and yields energy. However, the majority of the breakdown of fatty acids occurs in the mitochondria (Scheffler, 2008). Fatty acids with long chains (those having over 14 carbons) need to be converted into fatty acyl-CoA to pass through the mitochondrial membrane. (Lehninger *et al.*, 2000). Fatty acid breakdown begins in the cytoplasm of cells when acyl-CoA synthetase uses energy derived from ATP cleavage to catalyze the incorporation of coenzyme A into the fatty acid (Lehninger *et al.*, 2000).

Lipid biosynthesis

Apart from fats found in food, one of the primary energy sources for living things is fat deposited in adipose cells. The organisms can synthesize cholesterol, lipid membranes, and triacylglycerols by a variety of processes.

Lipid biosynthesis in membranes: Membrane lipids fall into two main categories: sphingolipids and glycerophospholipids. Our bodies generate a wide variety of membrane lipids, but the mechanisms all follow the same pattern. Sphingosine or glycerol, the backbone, is first produced. Next, phosphatidic acid is created by adding fatty acids to the backbone. Additional alteration occurs when different hydrophilic head groups are added to the phosphatidic acid backbone. The synthesis of membrane lipids occurs in the endoplasmic reticulum membrane (Gault *et al*., 2010).

Triglyceride biosynthesis: Another crucial step in the process is the phosphatidic acid. Phosphatase catalyzes the transformation of the phosphatidic acid to diacylglyceride, which acyltransferase subsequently converts to triglycerides. The cytosol is where triglyceride biosynthesis takes place.

TREM-2

The immunoglobulin superfamily of cell surface receptors, which include the one that triggers receptor expressed on myeloid cell lines 2 (TREM-2) (Allcock *et al*., 2003; Klesney-Tait *et al*., 2006), is encoded by a gene cluster on chromosome 6p21. Microglia, osteoclasts, dendritic cells, and tissue macrophages are some different myeloid-derived varieties of cells that express it (Colonna, 2003b; Ibach *et al*., 2021). The immunoglobulin superfamily of cell surface receptors, comprising the triggering receptor expressed on myeloid cells 2 (TREM-2) is encoded by a gene cluster on chromosome 6p21 (Allcock *et al*., 2003; Klesney-Tait *et al*., 2006). Amongst the myeloid-derived cell categories that express it are dendritic cell osteoclasts, macrophages from the tissues, and microglia (Zhao *et al.,* 2018; Zhong *et al.,* 2018). The short cytoplasmic domain of TREM-2 has no known signaling function. On the other hand, TREM-2 can connect to its adaptor protein DAP12 (DNAX activation protein of 12 kDa, TYROBP) through charged residue linkages inside the transmembrane regions of both enzymes. The short cytoplasmic domain of TREM-2 has no known signaling function. On the other hand, TREM-2 can connect to its adaptor protein DAP12 (DNAX activation protein of 12 kDa, TYROBP) through charged residue linkages inside the transmembrane regions of both enzymes (Ibach *et al*., 2021). The short cytoplasmic domain of TREM-2 has no known signaling function. On the other hand, TREM-2 can connect to its adaptor protein DAP12 (DNAX activation protein of 12 kDa, TYROBP) through charged residue linkages inside the transmembrane regions of both enzymes (Paloneva *et al*., 2002). A distinctive immunoreceptor tyrosine-based activation motif found in the

cytoplasmic domain of DAP12 is phosphorylated upon ligand-binding to TREM-2, thereby regulating multiple intracellular signaling pathways that govern processes such as calcium mobilization, cytokine production, phagocytosis, cell proliferation and differentiation, survival, and cytoskeletal remodeling (Jay *et al.*, 2017).

The primary intracellular locations of TREM-2 are exocytic vesicles and the trans-Golgi network (Prada *et al.*, 2006). TREM-2 can be proteolytically cleaved between amino acids H157 and S158 by members of the disintegrin and metalloproteinase (ADAM) family (Feuerbach *et al.*, 2017; Schlepckow *et al.*, 2017; Thornton *et al.*, 2017). This cleavage results in the release of a soluble TREM-2 ectodomain (sTREM-2) and a membrane-tethered C-terminal fragment (CTF) into extracellular fluids. (Kleinberger *et al.*, 2014; Wunderlich *et al.*, 2013). The TREM-2 CTF, which is also expressed in microglia (Nadler *et al.*, 2008; Farfara *et al.*, 2011; Kemmerling *et al.*, 2017; Walter *et al.*, 2017), serves as a substrate for intramembrane proteolysis by γ-secretase (Wunderlich *et al.*, 2013) TREM-2 can be proteolytically cleaved between amino acids H157 and S158 by members of the disintegrin and metalloproteinase (ADAM) family (Feuerbach *et al.*, 2017; Schlepckow *et al.*, 2017; Thornton *et al.*, 2017). This cleavage results in the release of a soluble TREM-2 ectodomain (sTREM-2) and a membrane-tethered C-terminal fragment (CTF) into extracellular fluids (Frank *et al.*, 2008).

Expression Patterns

Myeloid Cells: TREM-2 is mostly expressed in macrophages, dendritic cells, and microglia are examples of myeloid lineage cells. These cells play a major role in innate immune responses (Kiialainen *et al.*, 2005). The resident immune cells of the central nervous system (CNS) are called microglia, and these cells express TREM-2 in particularly high concentrations. The brain's immunological responses are influenced by microglial TREM-2 (Kiialainen *et al.*, 2005). TREM-2 Upregulation in Neurodegenerative diseases: Studies have demonstrated an upregulation of TREM-2 expression in response to neurodegenerative diseases, indicating a possible function for TREM-2 in the central nervous system's defense against injury or disease (Jay *et al.*, 2017).

Roles in Immune Regulation

Phagocytosis and Clearance: TREM-2 regulates phagocytosis, which aids in the removal of apoptotic cells and other biological detritus. For tissue homeostasis to be maintained, this function is essential (Takahashi *et al.*, 2005).

Inflammatory Response Modulation: TREM-2 has been linked to the regulation of inflammatory reactions. According to Turnbull *et al.* (2006), when it is engaged, myeloid cells' pro-inflammatory reactions can be counteracted by anti-inflammatory signals.

Effect on Immune Activation: TREM-2 is thought to control the activation and differentiation of immune cells. Research indicates that TREM-2 signaling could affect macrophage polarization toward anti-inflammatory phenotypes (Colonna, 2007).

Association with CNS Disorders:

TREM-2 has been discovered to be soluble in MS patients' cerebral fluid, suggesting that it may be involved in CNS inflammatory illnesses (Piccio *et al.*, 2008). TREM-2 depletion has been shown to impact the progression of Alzheimer's disease (AD) in a mouse model, highlighting its importance in neurodegenerative illnesses (Jay *et al.*, 2017).

Immune-Metabolic Crosstalk

The fundamental activity that underlies almost all biological processes is metabolism. Immunometabolism, the fusion of immunity and metabolism, is leading the way in immunology research and is revolutionizing the discipline. Research conducted in the past ten years has demonstrated that the immunity system function in both health and illness depends fundamentally on metabolism. The significant connection between metabolism and immunity is highlighted by the finding that cellular metabolism affects immune cell state and fate and contributes to infectious disease, inflammation, and cancer (Chapman and Chi, 2022). Moreover, organismal homeostasis is mediated by interactions between the immunological and metabolic systems (Lercher *et al.*, 2020).

Immunometabolism has seen the emergence of several new concepts recently. Immune receptors and metabolism-related environmental signals (such as nutrition) interact extensively, cooperating to facilitate both adaptive and innate immune responses. Bidirectional metabolic signaling, a mechanism that involves signal transduction and metabolic reprogramming, further influences the course of immune responses (Chapman *et al.*, 2020). Recent research on a variety of cells, including macrophages, T cells, B cells, and dendritic cells (DCs), has shown that metabolic signaling influences both tissue immunity and the state and fate of immune cells. The immunometabolism, which is characterized by extensive interaction between the immune system, adipose tissue, and diet, is finally gaining recognition of its intracellular and intercellular networks. This is because metabolism functions as highly complex networks for cell signaling as well as interactions between cells

and the environment (Trim and Lynch, 2021). All things considered, our understanding of immunometabolism across the signaling, cells, and systems stages has enhanced our understanding of immunity and metabolism and created novel therapeutic opportunities for immune-mediated illnesses, cancer, and metabolic disorders (Fig. 1). This special edition of Cellular and Molecular Immunology has ten thorough review articles written by leading authorities on immunometabolism. They provide an overview of these novel ideas and discuss how an integrative knowledge of immunometabolism could result in the identification of novel biological pathways and medical interventions.

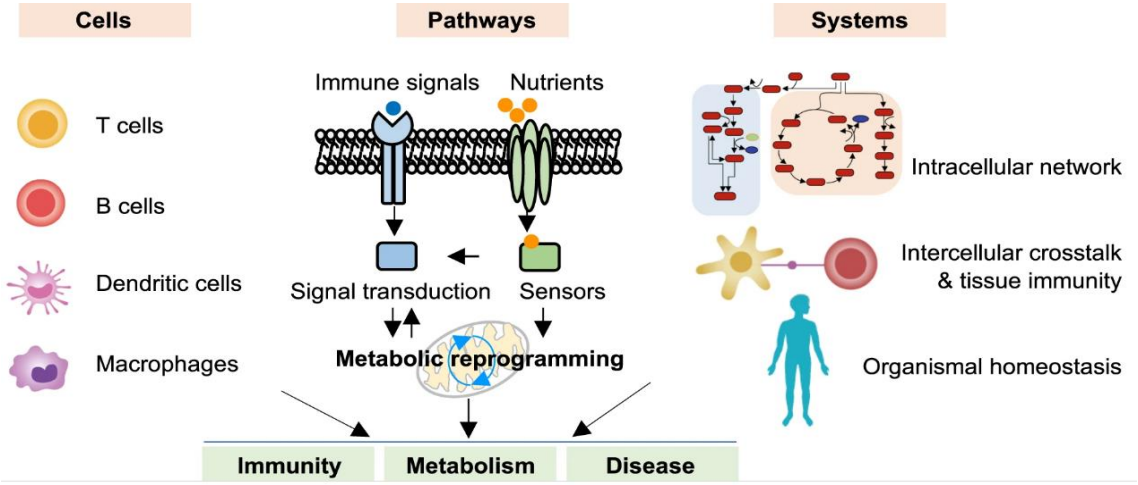

Figure 2: An integrative perspective on immunometabolism that connects systemic metabolism, immune cell biology, including metabolic pathways.

TREM-2

Numerous enzymes, cellular transporters, lipid carrier and transfer proteins, and cell surface receptors work together to facilitate lipid transport throughout the circulatory system, extracellular space, and plasma membrane. It most likely arose as an ancient evolutionary process because of the distribution of essential dietary or endogenously generated lipids and hormones, as well as lipid-modified signaling proteins and other related macromolecules, between progressively metabolically specialized tissues. One of the more intricate biochemical systems' first parts is the lipoprotein receptor. These receptors on the cell's surface fall into two primary groups: endocytic receptors, which bind their cargo in the form of lipid-carrying lipoproteins and mediate their internalization and eventual lysosomal delivery, and those that facilitate lipid exchange at the plasma membrane without cellular uptake of the particle's protein component. While well-known

members of the first group include the scavenger type A receptors (SRAs) and the low-density lipoprotein (LDL) receptor and LDL receptor-related proteins, the latter group includes, for example, the scavenger type B receptors SR-B1, SR-B2, and CD36. Some of these proteins have been identified in recent years for purposes that are often unrelated to their specific roles as mediators of cellular lipid uptake. These roles include that of signal modulators and transducers in cells. We will confine our analysis to a single, incredibly adaptable subgroup: the proteins bound to LDL receptors. We will briefly examine the evolutionary history of the family before discussing the more conventional and restricted roles that the LDL receptor gene family plays in the metabolism of other macromolecules and in "classic" lipoprotein transport. Subsequently, our focus will mainly be on the more extensive and quickly expanding functions of the LDL receptor gene family, including tyrosine kinase activation and modulation, integration and regulation of basic cellular signaling pathways in the central nervous system and during development, and its involvement in cellular

TREM-2 controls the brain's lipid metabolism

Most recent studies on TREM-2 have focused on how the brain uses it. TREM-2 expresses itself in the brain in a very specific way. TREM-2 exhibits specific cellular expression in microglia, and significant anatomical manifestation occurs in the corpus callosum and base of the ganglia (Nugent *et al.*, 2020). It has been determined that the lipid receptor TREM-2 regulates the metabolism of cholesterol and phospholipids in the central nervous system (Andreone *et al.*, 2020; Nugent *et al.*, 2020).

TREM-2 regulates brain cholesterol and myelin metabolism

TREM-2's role in the central nervous system has been the main focus of recent research on the protein. TREM-2 exhibits a highly recognizable pattern of expression in the brain. TREM-2 exhibits specific expression in microglia at the cellular level, although at the anatomical level, it is significantly expressed in the corpus callosum and basal ganglia (Nugent *et al.*, 2020). According to research (Andreon *et al.*, 2020; Nugent *et al.*, 2020), TREM-2 is a lipid receptor that regulates the metabolism of cholesterol and phospholipids in the central nervous system. Moreover, TREM-2 increases the microglial transition to DAM, which influences AD pathogenesis and interacts with brain lipid metabolism, according to Park *et al.* (2018). Interestingly, the current study shows that TREM-2 binds to ApoE, an essential lipid transporter in the central nervous system. The ε2, ε3, and ε4 alleles

of the APOE gene encode the three major variations of the human ApoE protein, which are ApoE2, ApoE3, and ApoE4. All three isoforms can bind to TREM-2. As to the findings of Atagi *et al.* (2015) and Fernandez *et al.* (2019), APOE ε2 protects against AD, while APOE ε4 is the primary genetic risk factor for late-onset AD. The mouse ApoE protein and the most common form of ApoE protein, human ApoE3, are the most comparable to one another. ApoE, which mediates endocytosis and the outflow of lipids and cholesterol, is necessary for the conversion to DAM. (Krasemann *et al.*, 2017).

Consequently, ApoE may be connected to TREM-2's effects on these lipid metabolism-related pathways. In addition, we analyze the TREM-2-affected lipid metabolism pathways in the CNS and their interaction with ApoE, and we address the idea that the disruption of lipid metabolism in the CNS contributes to the pathogenesis of AD.

TREM-2 regulates brain cholesterol and myelin metabolism

Growing research has connected TREM-2 function and microglia's lipid metabolism (Poliani *et al.*, 2015). Nevertheless, previous research has primarily focused on lipids, which are hypothesized TREM-2 ligands in the form of signals or lipoprotein particles that are exposed to the cell surface (Sudom *et al.*, 2018).

TREM-2 signaling is essential for controlling the metabolism of cholesterol in microglia, according to fascinating new research. In a cuprizone (CPZ)-treated model, which is used to simulate demyelination and the effect of demyelination-induced lipid overload, Nugent et al. discovered that microglia in Trem-2-/- mice had a more than tenfold increase in cholesterol esters (CEs) and oxidized Ces compared with Trem-2 +/+ mice (Nugent *et al.*, 2020). To further understand the mechanisms causing CE formation, these researchers blocked the endoplasmic reticulum (ER) enzyme acetyl-CoA acetyltransferase 1 and boosted the levels of the cholesterol transporters ABCA1 and ABCG1 to reduce the generation of Ces from free cholesterol. Nugent *et al.* (2020) report that both treatments reduced the accumulation of CE in Trem-2-/- mouse models, suggesting a connection between intracellular cholesterol deposited as CE or aberrant microglial cholesterol export and Trem-2 deficiency. A lipidomic analysis of microglia (IMG) derived from induced pluripotent stem cells (iPSCs) confirmed and extended the function of TREM-2 in the control of lipids. The intracellular enzyme PLCγ2, expressed by the AD-linked gene PLCG2, cleaves the membrane phospholipid phosphatidylinositol-4,5-bisphosphate (PIP2)

into inositol-1,4,5-trisphosphate (IP3) and diacylglycerol (DAG) (Kadamur and Ross, 2013).

Using lipidomic analysis of cell extracts, Andreone *et al.* evaluated the quantities of 100 lipids in TREM-2 KO IMG and phospholipase C γ2 (PLCG2) KO IMG after myelin exposure. In contrast to cells of the wild type (WT) exposed Numerous lipids, such as free cholesterol and CE, were confirmed in further independent PLCG2 KO IMG clones and TREM-2 KO IMG clones made using various techniques (Andreone *et al.*, 2020). TREM-2 may control the transport of cholesterol in human microglia in a PLCγ2-dependent way, given that intracellular cholesterol is typically deposited as CE and that PLCγ2 signals downstream of the TREM-2-DAP12 Andreone *et al.* (2020).

Brain cholesterol is produced locally because the BBB prevents lipoproteins rich in cholesterol from entering the central nervous system. About eighty percent of the free cholesterol in the brain is contained in the myelin sheath that oligodendrocytes produce to shield axons. Myelin is therefore a crucial and reliable marker of the brain's cholesterol metabolism (Martin *et al.*, 2014). Studies have shown that TREM-2 participates in the microglial response to myelin damage, which affects remyelination (Canton *et al.*, 2015). A recent study found that in response to myelin injury, Trem-2 microglia are unable to multiply genes associated with activation, lipid catabolism, and phagocytosis. As a result, there is a decrease in oligodendrocyte production, axonal dystrophy, chronic demyelination, and myelin debris clearing.

These results could affect the actual process of remyelination, specifically the altered removal of myelin debris (a required step for proper remyelination) and the decrease in oligodendrocytes (Poliani *et al.*, 2015). Park *et al.* (2018) discovered that TREM-2 promotes the microglial transition to DAM, which affects AD pathogenesis and interacts with brain lipid metabolism. Interestingly, the current study shows that TREM-2 binds to ApoE, an essential lipid transporter in the central nervous system. The ε2, ε3, and ε4 alleles of the APOE gene encode the three major variations of the human ApoE protein, which are ApoE2, ApoE3, and ApoE4. All three isoforms can bind to TREM-2. As to the findings of Atagi *et al.* (2015) and Fernandez *et al.* (2019), APOE ε2 protects against AD, while APOE ε4 is the primary genetic risk factor for late-onset AD. The mouse ApoE protein and the human ApoE3, the most prevalent form of ApoE protein, are most similar to one another.

ApoE, which mediates endocytosis and the outflow of lipids and cholesterol, is necessary for the conversion to DAM (Krasemann *et al*., 2017).

Consequently, ApoE may be connected to TREM-2's effects on these lipid metabolism-related pathways. In addition, we analyze the TREM-2-affected lipid metabolism pathways in the CNS and their interaction with ApoE, and we address the idea that the disruption of lipid metabolism in the CNS contributes to the pathogenesis of AD growth regulation and cancer.

Conclusion

Since the disease-associated variants studied here cause a partial or almost complete loss-of-function, activation of TREM-2-DAP12 signaling could potentially represent a therapeutic approach to normalize regulation of microglial activity. Indeed, agonistic antibodies have been used to activate TREM-2 signaling and even restore some cellular deficits observed in TREM-2 R47H expressing cells. The monoclonal anti TREM-2 antibody 4B2A3 described here also had agonistic effects on TREM-2-DAP12 signaling. This antibody does not bind to the ligand binding domain, however, it still stimulated TREM-2 signaling through binding to the stalk region of TREM-2 within amino acids 131-148 and activated TREM-2 through receptor cross-linking. This could also be confirmed with iPSdMiG expressing endogenous TREM-2, albeit at much lower levels as compared to overexpressed TREM-2 and DAP12 in the HEK 293 reporter line.

Notably, beside the T66M variant, stimulation by the 4B2A3 antibody was observed for the other disease-associated variants, even though to a lower extent as compared to the stimulation of the common TREM-2 variant in both, HEK 293 reporter cells and iPSdMiG. Taken together, the reporter cell model described here represents a suitable system for studying cellular transport and metabolism of TREM-2 and DAP12 and effects of disease associated TREM-2 mutations. This system also allows the identification and characterization of compounds that modulate TREM-2 activity for the development of future therapeutic strategies for AD and other neurodegenerative diseases. As the 4B2A3 anti-TREM-2 specific antibody showed specific activation of TREM-2 signaling in AD-associated risk variants, it might also represent a promising candidate for therapeutic approaches.

References

Allcock, R. J., Barrow, A. D., Forbes, S., Beck, S. and Trowsdale, J. (2003). The human TREM gene cluster at 6p21.1 encodes both activating and inhibitory single IgV domain receptors and includes NKp44. *European journal of immunology*, *33*(2):567-577.

Andreone, B. J., Przybyla, L., Llapashtica, C., Rana, A., Davis, S. S., van Lengerich, B., ... and Lewcock, J. W. (2020). Alzheimer's-associated PLCγ2 is a signaling node required for both TREM2 function and the inflammatory response in human microglia. *Nature Neuroscience*, *23*(8):927-938.

Atagi, Y., Liu, C. C., Painter, M. M., Chen, X. F., Verbeeck, C., Zheng, H., ... and Bu, G. (2015). Apolipoprotein E is a ligand for triggering receptor expressed on myeloid cells 2 (TREM2). *Journal of Biological Chemistry*, *290*(43):6043-26050.

Baynes, D. (2014). Medical Biochemistry. Saunders, Elsevier Ltd. pp. 121-122.

Bouchon, A., Dietrich, J. and Colonna, M. (2000). Cutting edge: inflammatory responses can be triggered by TREM-1, a novel receptor expressed on neutrophils and monocytes. *The Journal of Immunology*, *164*(10):4991-4995.

Brownsey, R. W., Boone, A. N. and Elliott, J. E. (2006). Regulation of Acetyl-CoA Carboxylase. *Biochemical Society Transactions*, 34(2):223-227.

Cantoni, C., Bollman, B., Licastro, D., Xie, M., Mikesell, R., Schmidt, R., ... and Piccio, L. (2015). TREM2 regulates microglial cell activation in response to demyelination in vivo. *Acta neuropathologica*, *129*: 429-447.

Chapman, N. M. and Chi, H. (2022). Metabolic adaptation of lymphocytes in immunity and disease. *Immunity*, *55*(1): 14-30.

Chapman, N. M., Boothby, M. R., and Chi, H. (2020). Metabolic coordination of T cell quiescence and activation. *Nature reviews immunology*, *20*(1): 55-70.

Chemistry Encyclopedia "Hydrolysis structure, reaction, water, proteins, examples, salt, molecule". chemistryexplained.com. Retrieved 2016-11-01.

Colonna, M. (2003b). TREMs in the immune system and beyond. *Nature Reviews Immunology*, 3(6):445–453.

Colonna, M. (2007). TREMs in the immune system and beyond. *Nature Reviews Immunology*, 7(6): 464–475.

Farfara, D., Trudler, D., Segev-Amzaleg, N., Galron, R., Stein, R., and Frenkel, D. (2011). γ-Secretase component presenilin is important for microglia β-amyloid clearance. Annals of Neurology, 69(1):170–180.

Feingold, K. R. and Grunfeld, C. (2000). "Introduction to Lipids and Lipoproteins". In De Groot L. J., Chrousos, G., Dungan, K., Feingold, K. R., Grossman, A., Hershman, J. M., Koch, C., Korbonits, M and McLachlan R. (eds.). *Endotext*. South Dartmouth (MA): MDText.com, Inc.

Fernandez, C. G., Hamby, M. E., McReynolds, M. L. and Ray, W. J. (2019). The role of APOE4 in disrupting the homeostatic functions of astrocytes and microglia in aging and Alzheimer's disease. *Frontiers in aging neuroscience*, *11*: 14.

Feuerbach, D., Schindler, P., Barske, C., Joller, S., Beng-Louka, E., Worringer, K. A., … Neumann, U. (2017). ADAM17 is the main sheddase for the generation of human

triggering receptor expressed in myeloid cells (hTREM2) ectodomain and cleaves TREM2 after histidine 157. *Neuroscience Letters*, 660:109–114.

Frank, S., Burbach, G. J., Bonin, M., Walter, M., Streit, W., Bechmann, I. and Deller, T. (2008). TREM2 is upregulated in amyloid plaque-associated microglia in aged APP23 transgenic mice. Glia, 56(13):1438–1447.

Freifelder, D. (1987). Molecular biology (2nd ed.). Jones and Bartlett, Boston.

Gault, C. R., Obeid, L. M., and Hannun, Y. A. (2010). An overview of sphingolipid metabolism: from synthesis to breakdown. *Sphingolipids as signaling and regulatory molecules*, 1-23.

Genua, M., Rutella, S., Correale, C. and Danese, S. (2014). "The triggering receptor expressed on myeloid cells (TREM) in inflammatory bowel disease pathogenesis". *Journal of Translational Medicine,* 12: 293.

Harris, J. R. (Ed.). (2010). *Cholesterol Binding and Cholesterol Transport Proteins:: Structure and Function in Health and Disease* (Vol. 51). Springer Science and Business Media.

Jay, T. R., Hirsch, A. M., Broihier, M. L., Miller, C. M., Neilson, L. E., Ransohoff, R. M., ... and Landreth, G. E. (2017). Disease progression-dependent effects of TREM2 deficiency in a mouse model of Alzheimer's disease. *Journal of Neuroscience*, *37*(3), 637-647.

Jay, T. R., von Saucken, V. E. and Landreth, G. E. (2017). TREM2 in neurodegenerative diseases. Molecular Neurodegeneration, 12(1):56.

Kadamur, G., and Ross, E. M. (2013). Mammalian phospholipase C. *Annual review of physiology*, *75*(1): 127-154.

Kemmerling, N., Wunderlich, P., Theil, S., Linnartz-Gerlach, B., Hersch, N., Hoffmann, B., … Walter, J. (2017). Intramembranous processing by γ-secretase regulates reverse signaling of ephrin-B2 in migration of microglia. Glia, 65(7):1103–1118.

Kiialainen, A., Hovanes, K. and Paloneva, J. (2005). Dap12 and TREM-2, molecules involved in innate immunity and neurodegeneration, are co-expressed in the CNS. *Neurobiology of Disease*, 18(2):314-322.

Kleinberger, G., Yamanishi, Y., Suárez-Calvet, M., Czirr, E., Lohmann, E., Cuyvers, E., ... and Haass, C. (2014). TREM2 mutations implicated in neurodegeneration impair cell surface transport and phagocytosis. *Science translational medicine*, *6*(243), 243ra86-243ra86.

Klesney-Tait, J., Turnbull, I. R. and Colonna, M. (2006). The TREM receptor family and signal integration. *Nature Immunology*, 7(12):1266–1273.

Krasemann, S., Madore, C., Cialic, R., Baufeld, C., Calcagno, N and El Fatimy, R. (2017). The TREM-2-APOE pathway drives the transcriptional phenotype of dysfunctional microglia in neurodegenerative diseases. *Immunity*, 47(3):566-81 e9.

Lehninger, A. L., Nelson, D. L. and Cox, M. M. (2000). *Lehninger Principles of Biochemistry* (3rd ed.). Worth Publishers, New York.

Lercher, A., Baazim, H. and Bergthaler, A. (2020). Systemic immunometabolism: challenges and opportunities. *Immunity*, 53:496-509.

Masuda, T., Sankowski, R., Staszewski, O. and Prinz, M (2020). "Microglia Heterogeneity in the Single-Cell Era". *Cell Reports,* 30 (5):1271-1281.

Nadler, Y., Alexandrovich, A., Grigoriadis, N., Hartmann, T., Rao, K. S. J., Shohami, E. and Stein, R. (2008). Increased expression of the γ-secretase components presenilin-1 and nicastrin in activated astrocytes and microglia following traumatic brain injury. Glia, 56(5):552–567.

Nugent, A. A., Lin, K., Van Lengerich, B., Lianoglou, S., Przybyla, L., Davis, S. S., ... and Di Paolo, G. (2020). TREM2 regulates microglial cholesterol metabolism upon chronic phagocytic challenge. *Neuron*, *105*(5): 837-854.

Ophardt, C. E. (2013). Lipid Metabolism Summary. *Virtual Chembook. Elmhurst College*.

Paloneva, J., Manninen ,T., Christman, G., Hovanes, K., Mandelin, J. and Adolfsson, R. (2002). "Mutations in two genes encoding different subunits of a receptor signaling complex result in an identical disease phenotype". *American Journal of Human Genetics*. 71 (3): 656-662.

Paloneva, J., Manninen, T., Christman, G., Hovanes, K., Mandelin, J., Adolfsson, R., … Peltonen, L. (2002). Mutations in two genes encoding different subunits of a receptor signaling complex result in an identical disease phenotype. American Journal of Human Genetics, 71(3):656–662.

Park, J., Wetzel, I., Marriott, I., Dréau, D., D'Avanzo, C., Kim, D. Y., ... and Cho, H. (2018). A 3D human triculture system modeling neurodegeneration and neuroinflammation in Alzheimer's disease. *Nature neuroscience*, *21*(7):941-951.

Pelley, J. W. (2012). *Elsevier's Integrated Review Biochemistry* (2nd ed.). Philadelphia: Elsevier/Mosby.

Piccio, L., Buonsanti, C. and Cella, M. (2008). Identification of soluble TREM-2 in the cerebrospinal fluid and its association with multiple sclerosis and CNS inflammation. *Brain*, 131(11): 3081-3091.

Poliani, P. L., Wang, Y., Fontana, E., Robinette, M. L., Yamanishi, Y., Gilfillan, S. and Colonna, M. (2015). TREM2 sustains microglial expansion during aging and response to demyelination. *The Journal of clinical investigation*, *125*(5): 2161-2170.

Prada, I., Ongania, G. N., Buonsanti, C., Panina-Bordignon, P. and Meldolesi, J. (2006). Triggering receptor expressed in myeloid cells 2 (TREM2) trafficking in microglial cells: Continuous shuttling to and from the plasmamembrane regulated by cell stimulation. Neuroscience, 140(4):1139–1148.

Schlepckow, K., Kleinberger, G., Fukumori, A., Feederle, R., Lichtenthaler, S. F., Steiner, H. and Haass, C. (2017). An Alzheimer associated TREM2 variant occurs at the ADAM cleavage site and affects shedding and phagocytic function. EMBO Molecular Medicine, 9(10):1356–1365.

Shirotani, K., Hori, Y., Yoshizaki, R., Higuchi, E., Colonna, M., Saito, T., ... and Iwata, N. (2019). Aminophospholipids are signal-transducing TREM2 ligands on apoptotic cells. *Scientific Reports*, *9*(1): 7508.

Sudom, A., Talreja, S., Danao, J., Bragg, E., Kegel, R., Min, X., ... and Wang, Z. (2018). Molecular basis for the loss-of-function effects of the Alzheimer's disease–associated R47H variant of the immune receptor TREM2. *Journal of Biological Chemistry*, *293*(32), 12634-12646.

Sun, H., Feng, J. and Tang, L. (2020). "Function of TREM1 and TREM-2 in Liver-Related Diseases". *Cells*. 9 (12):2626.

Takahashi, K, Rochford, C. D. and Neumann, H. (2005). Clearance of apoptotic neurons without inflammation by microglial triggering receptor expressed on myeloid cells-2. *Journal of Experimental Medicine*, 201(4):647-657.

Thornton, P., Sevalle, J., Deery, M. J., Fraser, G., Zhou, Y., Ståhl, S., … Crowther, D. C. (2017). TREM2 shedding by cleavage at the H157-S158 bond is accelerated for the Alzheimer's disease-associated H157Y variant. EMBO Molecular Medicine, 9(10):1366–1378.

Trim, W. V. and Lynch, L. (2022). Immune and non-immune functions of adipose tissue leukocytes. *Nature Reviews Immunology*, *22*(6):371-386.

Turnbull, I. R., Gilfillan, S. and Cella, M. (2006). Cutting edge: TREM-2 attenuates macrophage activation. *Journal of Immunology*, 177(6):3520-3524.

Voet, D., Voet, J. G., and Pratt, C. W. (2013). *Fundamentals of biochemistry: life at the molecular level*. Hoboken, NJ: Wiley. pp. 581-620.

Walter, J., Kemmerling, N., Wunderlich, P. and Glebov, K. (2017). γ-Secretase in microglia - implications for neurodegeneration and neuroinflammation. *Journal of Neurochemistry*, 143(4):445–454.

Wculek, S. K., Dunphy, G., Heras-Murillo, I., Mastrangelo, A., and Sancho, D. (2022). Metabolism of tissue macrophages in homeostasis and pathology. *Cellular and molecular immunology*, *19*(3):384-408.

Wunderlich, P., Glebov, K., Kemmerling, N., Tien, N. T., Neumann, H. and Walter, J. (2013). Sequential proteolytic processing of the triggering receptor expressed on myeloid cells-2 (TREM2) protein by ectodomain shedding and γ-secretase-dependent intramembranous cleavage. *Journal of Biological Chemistry*, 288(46):33027–33036.

Zhao, Y., Wu, X., Li, X., Jiang, L.-L., Gui, X., Liu, Y., … Xu, H. (2018). TREM2 is a receptor for β-amyloid that mediates microglial function. *Neuron*, 97(5):1023–1031.

Zhong, L., Wang, Z., Wang, D., Wang, Z., Martens, Y. A., Wu, L., … Chen, X.-F. (2018). Amyloid-beta modulates microglial responses by binding to the triggering receptor expressed on myeloid cells 2 (TREM2). Molecular Neurodegeneration, 13(1):15.