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AN OVERVIEW OF SPHINGOLIPIDS AS A PROGNOSTIC BIOMARKER FOR THE PREDICTION OF DEGENERATIVE DISORDERS

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ABSTRACT

Sphingolipids are lipids that have a sphingoid base and contain a group of aliphatic amino alcohols that contain sphingosine. Sphingolipids are essential in treatment of neurodegenerative, neuroinflammatory, and neuropsychiatric diseases. Sphingolipid imbalances can cause neurological and psychological problems. The emergence of diseases such as AD, PD, HD are linked to abnormal Sphingolipid metabolism. Sphingolipids play a role in neuronal demyelination, which can lead to diseases including Multiple Sclerosis.

In clinical trials, prognostic biomarkers are used to investigate diseases and assess medication and treatment response. Prognostic biomarkers provide knowledge about the progression of the disease. In the case of ductal carcinoma of the breast, sphingosine kinase 1 (SPHK1) has prognostic significance. Sphingolipid metabolism is being studied as a possible prognostic biomarker for breast cancer. Sphingolipid changes have been identified in Melanoma, and Sphingomyelin synthase 1 (SMS1) has been linked to melanoma prognosis. Glycosphingolipids have also been identified as possible biomarkers for prostate cancer. Sphingolipidomics has advanced as a prognostic method for SLE an autoimmune disease. Sphingolipids and their metabolism are the focus of this study. S1P is also used to predict the outcome of CAP.

In this present review, we give an overview of how sphingolipids plays a role as prognostic biomarker in neurological disorders and will provide knowledge about recent advances in investigational techniques.

Keywords: Sphingolipids, Prognostic biomarkers, Methods of lipid investigation, neurological diseases, cancer

ABBREVIATIONS:
AD Alzheimer's disease
BDNF brain derived neurotrophic factor
CerS ceramide synthase
EI-MS electrospray ionization mass spectrometry
GlcCer glucosylceramide
GalCergalactosylceramide
GSLs glycosphingolipid
GCS glucosylceramide synthase
HD Huntington's disease
HTT huntingtin gene
LC-MS liquid chromatography based mass spectrometry
MALDI matrix assisted laser desorption ionization
MS Multiple sclerosis
MSI mass spectroscopy imageing
PD Parkinson's disease
RRMS relapsing remitting multiple sclerosis
SPHK sphingosine kinase
SPT serine palmitoyl transferase
SM sphingomyelin
S1P sphingosine-1-phosphate

1. INTRODUCTION

Sphingolipids are the class of lipids generally found in brain. More than a century ago Johann Ludwig Wilhelm Thudichum discovered Sphingolipids in brain **[1]**. Sphingolipids shows presence of backbone of sphingoid base or long chain which are represented as Sphingosine. Sphingoid base consist of amide linked to very long chain fatty acid which will form ceramide a simple type of sphingolipid, in which addition of head groups may lead to formation of more complex sphingolipids like SM, GlcCer, GalCer **[2]**. Bioactive sphingolipids play important role in transmission of signalling in the brain **[3]**.

Ceramide synthesis takes place by mechanism of de novo synthesis **(Figure 1)**. In animal cell, ceramide synthesis occurs in ER and mitochondria. Ceramide is de novo synthesized by serine and palmitoyl co-A condensation to give 3-ketosphinganine, with the help of enzyme SPT. 3-ketosphinganine is reduced to sphinganine catalyzed by NADPH dependent 3-dehydrosphinganine reductase or ketosphingosine reductase **[4-5]**. The sphingoid base is desaturated to sphingosine and then by acylation it forms ceramide with the help of dihydroceramide desaturase **[6]**.

GSLs get degrade into ceramide, inside lysosomes due to action of certain acid exohydrolase **[7-8]**. In lysosomes acid ceramidase (N-acylsphingosinedeacylase) which is a lipid hydrolase, causing degradation of ceramide to produce sphingosine and free fatty acids **(Figure 1) [9]**. Phosphorylation of this sphingosine gives two different types of kinases such as SK1 and SK2, which will form S1P **[10]**. FTY720 (fingolimod) which an analog of sphingosine was the first compound that binds to S1P receptors **[11-12]**.

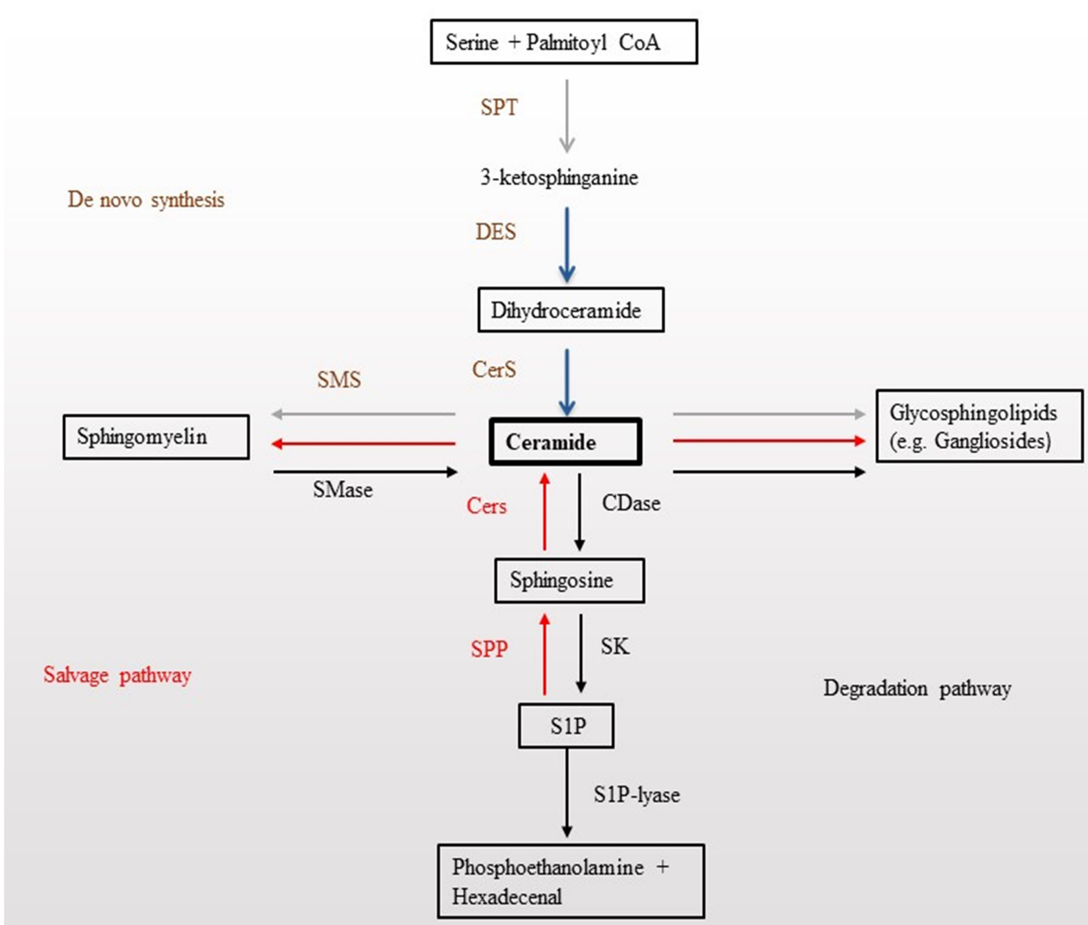

Figure 1: Representation of metabolism of Sphingolipids. De novo synthesis of ceramide occurs by combination of serine and palmitoyl co-A (shown by blue arrows) and recycling of S1P also gives ceramide (shown by red arrows). Ceramide is a precursor for glycosphingolipids and sphingomyelins. Hydrolytic degradation of ceramide gives fatty acids and sphingosine (shown by black arrows) which on phosphorylation gives S1P

2. METHODS OF LIPID INVESTIGATION:-

Lipidomic investigation is mainly a mass spectroscopy based analysis. Recent advances in lipidomic investigation by mass spectrometry gives detailed characterization of lipids which will includes sphingolipid classes and other categories of lipids. While doing MS analysis, lipid solution could be analyzed by shotgun method or by LC-MS **[13] (Figure 2)**.

2.1. Shotgun method: Shotgun method is also called as directly infusion based lipidomic. In this shotgun method unique chemical or physical properties of lipids and its subclasses are extracted from biological sample and is injected to mass spectrometer without purification. Lipid analysis after direct infusion is demonstrated by EI-MS **[14-16]**.

2.2. Liquid chromatography basedmass spectrometry (LC-MS): LC-MS in normal phase is used for the identification of glycerophospholipids. LC-MS in reversed phase is used for acyl chain separation which is associated with the study of various neurodegenerative diseases. Lipids are investigated by using MSI which is associated with MALDI **[17]**.

3. ROLE OF SPHINGOLIPIDS IN NEURODEGENERATIVE DISEASES:

3.1. Alzheimer's disease (AD): Alzheimer's disease is a neurodegenerative disorder, associated with dementia and decline of cognitive disability. Pathology of AD is characterized by accumulation of extracellular amyloid, chronic neuroinflammation and neurofibrillary tangles or tau protein and causes neuronal death.

Aggregation of β-amyloid (Aβ) associated with accumulation of extracellular senile plaque. β-amyloidare monomers released in external environment of cell by amyloidogenic pathway, aggregation forms oligomers converted to plaques showing toxic effect for neurons. Elevated level of ceramide in CSF and brain white matter is seen in AD dementia hence, early AD stage shows alteration in plasma ceramide level **[18]**.

While most of the studies found association with β-amyloid and sphingolipids, some recent studies found link between sphingolipids and tau protein phosphorylation. Ceramide modulates the activity of PP2A enzyme hence, as the ceramide level increases tau protein leads to hyperphosphorylation which will leads to neurodegeneration with deposition of amyloid.Hence alteration in sphingolipid mechanism increases the risk of AD development **[19]**. Dose response study by using fingolimod was carried out on transgenic AD mice where 1mg/kg/day dose is given and it shows that fingolimod decreases β-amyloid level in brain and improves memory. Hence, fingolimod may have potential therapeutic benefit in AD **[20]**. In animal model female FVB-Tg mice which express human AβPP mutation is studied and FTY720 in 0.9% NaCl is administered, it shows that FTY720 blocks neuronal death occurring due to aggregation of Aβ and it restore production of neurotrophin **[21]**.

3.2. Parkinson's Disease (PD): Parkinson's disease is a neurological disease, affecting movement of body which often include tremors, associated with loss of dopaminergic neurons which leads to reduction of dopamine level in brain causing neurodegeneration and chronic neuroinflammation.

Glycosphingolipid a subclass of lipid regulates inflammatory process of brain functions. Hence, deregulation in metabolism of glycosphingolipid is seen in Parkinson's disease. Various proinflammatory cytokine like TGF-β can be considered as therapeutic targets or biomarkers in neurodegenerative disorders **[22-23]**. Glucocerebrosidase (GBA) which is an enzyme that breakdown GlcCerinto glucose and ceramide is involved PD and mutation in gene coding of this enzyme is genetic risk factor in PD **[24]**. Recently study has been carried out on A53T transgenic mouse model which shows fingolimod (FTY720) elevates BDNF and lowers aggregation of α-synuclein. Solubilization of α-synuclein decreases level of ganglioside in PD by which fingolimod gives neuroprotective effect **[25-26]**.

3.3. Huntington's Disease (HD): Huntington's disease (HD) is an inherited neurodegenerative disease caused by faulty CAG trinucleotide expansion of HTT gene. Disturbances in motor function, loss of thinking ability is seen in HD **[27]**. Study showed alteration in upregulation of SGPL1 and SK1 in HD **(Table 1)**. Impaired ganglioside metabolism was shown in HD, ganglioside GM1 reduced in fibroblast of HD patient. Administration of GM1 decreases mutation of HTT gene hence, GM1treatment decreases the rate of degeneration of neurons, atrophy of white matter and body weight loss **[28]**.

4. ROLE OF SPHINGOLIPIDS IN NEUROINFLAMMATORY DISEASES:

4.1. Multiple sclerosis (MS): Multiple sclerosis is an autoimmune, chronic neuroinflammatory disease, associated with demyelination of neurons results into loss of vision, pain and impaired coordination **[29-30]**.

Several studies suggested that sphingolipid is associated with Multiple sclerosis. Sphingolipid involve in demyelination of neurons during pathogenesis of MS. Level of sphingolipid elevated in MS brain due to stimulation of inflammatory cytokines leading to death of oligodendrocytes **[31]**. Conversely S1Preceptor modulators act as a modifier in MS, it decreases demyelination of neurons and increases the course of remyelination. More specifically, sphingosine-1-phosphate receptor 2 (S1PR2) reduces demyelination **[32]**. Hence, S1PRs are considered as therapeutic targets for inflammatory diseases like Multiple sclerosis **[33]**. Fingolimod (FTY720) which is S1P receptor agonist was first drug approved by US-FDA and is considered as safe and effective therapy for relapsing of MS. Fingolimod protects astrocytes from neuroinflammation **(Figure 3)** **[34]**. Three drugs are clinically approved for treatment of RRMS such as fingolimod, teriflunomide, dimethyl fumarate **[35]**. Siponimod (BAF312) is another novel therapy which gives anti-inflammatory and neuroprotective mode of action. Siponimod modulates glial cell function and attenuates demyelination **[36]**. Study shows that Siponimod administration gives beneficial effects for experimental autoimmune encephalomyelitis (EAE) in MS rodent model by altering glutamatergic transmission and gives neuroprotective effects in CNS of EAE mice **[37]**.

5. ROLE OF SPHINGOLIPIDS IN NEUROPSYCHIATRIC DISEASES:

Pathological processes of various neuropsychiatric diseases include metabolism of sphingolipids. Chronic stress animal model was studied for advancements of understanding depression and anxiety. Imbalance in various types of sphingolipids such as ceramide, sphingomyelin and GalCers involve in psychiatric disease development. Interestingly, it was shown that anxiety was decreased and depressive symptoms was improved by fingolimod in MS patients. Study showed that fingolimod gives protective role for chronic stress induced anxiety like behavior. Sphingolipids acts as surrogate markers for treatment of neuropsychiatric diseases. Study was carried out on animal model where tricyclic antidepressant such as amitriptyline was administered, results into reduction in ceramide concentration and improvement in behavior **[1]**.

6. ROLE OF SPHINGOLIPIDS IN CANCER:

6.1. Breast cancer : Breast cancer is common type of cancer among females. Breast cancer metastasis is determined by genetic changes that result in cancer cells surviving, proliferating, migrating away from primary site and gaining or losing functions. SPHK (sphingosine kinase) act as key enzyme in pathophysiology of cancer **(Figure 3)**. S1P forms by phosphorylation of sphingosine by SPHK1, to prevent apoptosis and stimulate cellular proliferation and angiogenesis. Study suggested that SPHK1 expression involve in development and progression of breast cancer and act as potential prognostic biomarker in breast cancer **[38]**. Study has investigated gene expression could be correlation coefficient for diagnosis and prognosis of cancer **[39]**. Mass spectrometry (LC-HRMS) identifies various types of sphingolipids in breast cancer. Upregulation in SPHK1 expression observed in tumor cells. Conversely, ceramide causes apoptosis of cancerous cells and inhibit growth of cancerous cells and hence, ceramide phosphate is potential diagnostic and clinical relevance of breast cancer **[40]**. Recent translational studies gives evidence of clinical importance of sphingolipids as biomarker in cancer diagnosis and prognosis. Bioactive sphingolipids demonstrated correlation between patient survival and treatment response in different types of tumors **[41]**.

6.2. Melanoma: Melanoma is frequently reported, most dangerous and deadliest type of skin cancer. Study shows progression in melanoma due GCS enzyme in mice. Downregulation of sphingomyelin synthase 1 (SMS1) occurs frequently in melanoma, associated with reprogramming of sphingolipids **[42]**.

6.3. Prostate cancer: Prostate cancer is common cancer in male, accounting over 350,000 death worldwide in 2018. Detection of Prostate-specific antigen (PSA) became standard test for prostate cancer in 1990. Study found glycosphingolipids are associated with prostate cancer. Hence, glycosphingolipids could be prognostic biomarker for prostate cancer **[43-44]**.

7. ROLE OF SPHINGOLIPIDS IN OTHER DISEASES:

7.1. Systemic Lupus Erythematosus (SLE): SLEis an autoimmune disease involving multiple organ damage including kidney, lungs, cardiovascular and neuropsychiatric systems. Recent study shows elevation in level of sphingolipid in serum sample of SLE patient. Sphingolipids such as sphingomyelin, ceramide shows elevation in SLE. Hence, sphingolipidomic could be potential prognostic tool for SLE **[45]**.

7.2. Community Acquired Pneumonia (CAP): CAP is an acute respiratory system infection. S1P plays integral part in immune defense and maintenance of endothelial barriers. S1P protect lungs from lung injury and pulmonary leak. Hence, S1P acts as potential biomarker for pneumonia **[46]**.

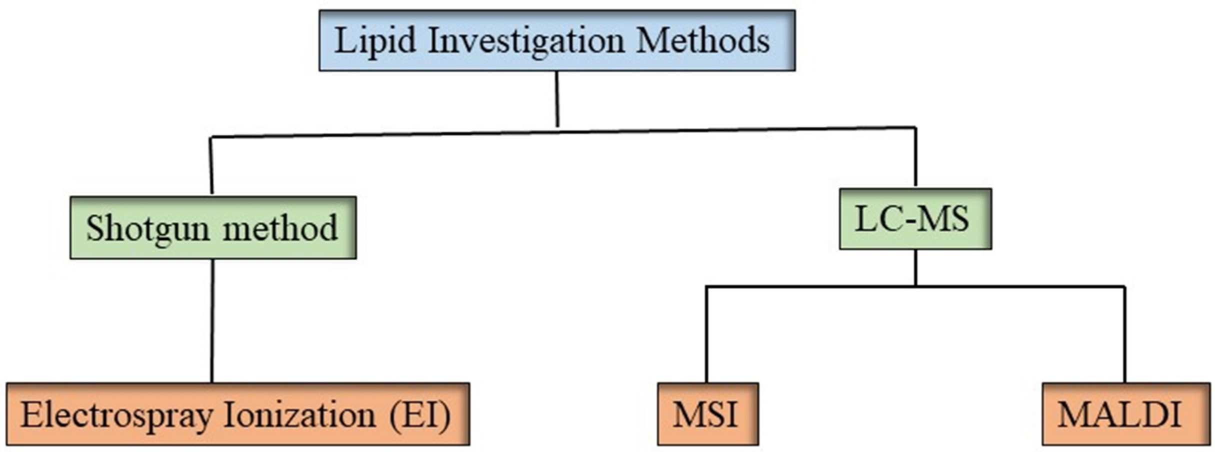

Figure 2: Classification of Methods of lipid investigation

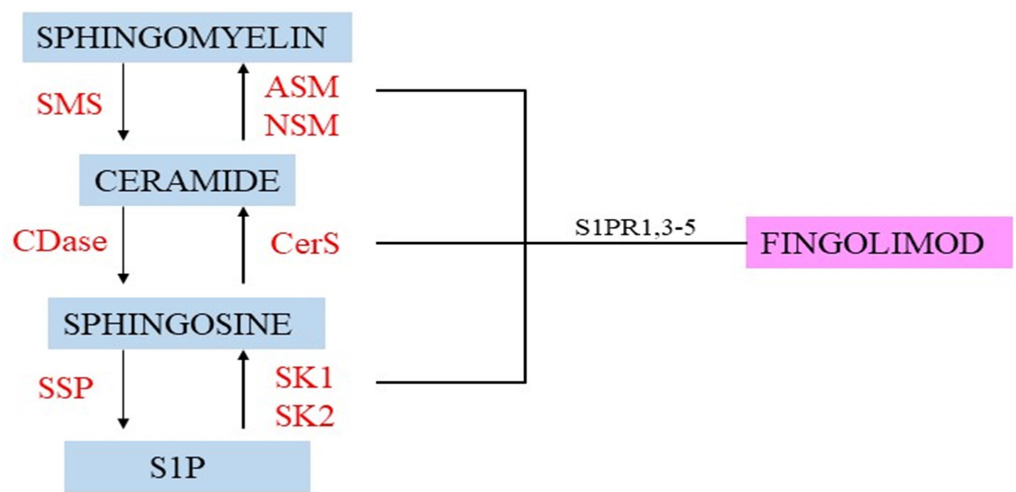

Figure 3: Representation of effect of fingolimod (FTY720) on hyperactive sphingomyelin-ceramide-sphingosine-S1P pathway in MS

Table 1: Representation of Downregulation and Upregulation of sphingolipids in AD, PD and HD

	Alzheimer's Disease	Parkinson's Disease	Huntington's Disease
Sphingolipids			
Ceramide	Upregulated	Downregulated	Upregulated
Sphingomyelin	Downregulated	Downregulated	Not Available
Sphingosine	Upregulated	Not Available	Not Available
Sphingosine-1-Phosphate (S1P)	Downregulated	Downregulated	Downregulated
Ganglioside GM1	Upregulated	Downregulated	Downregulated
Ganglioside GM3	Upregulated	Not Available	Not Available
Enzymes			
Sphingosine kinase 1 (SK1)	Downregulated	Downregulated	Downregulated
Sphingosine kinase 2 (SK2)	Downregulated	Downregulated	Not Available
Acid Sphigomyelinase (ASM)	Upregulated	Downregulated	Not Available
Acid Ceramidase (AC)	Upregulated	Not Available	Not Available
Ceramide synthase (CerS)	CerS1 Upregulated, CerS2 and CerS6 Downregulated	Upregulated	CerS1 Downregulated

8. CONCLUSION

The study has shown that, lipid plays vital role in neurodegenerative, psychiatric and neuroinflammatory diseases as well as in cancer because the abnormal level of sphingolipids may lead to diseased condition. Sphingolipids are used as prognostic biomarkers as it makes easier and beneficial determination of therapy and treatment in clinical trials. This study has given overview of sphingolipids in neurological disorders, cancer and advances in investigational methods. Imbalance in level of Sphingolipids like ceramide and sphingomyelin results in various diseases like AD, PD, HD, MS and many psychiatric disorders such as anxiety, depression and cancer like breast cancer, melanoma and prostate cancer. Prognostic biomarkers give information about how the disease has been developed. Sphingosine kinase 1 (SPHK1) gives prognostic value in case of ductal carcinoma of breast. Metabolic pathway of Sphingolipid is considered as a potential prognostic biomarker for Breast cancer. Alteration in Sphingolipids have been reported in Melanoma as Sphingomyelin synthase 1 (SMS1) is associated with prognosis of melanoma. Similarly, identification of Glycosphingolipids can be used as potential biomarkers for Prostate cancer. Sphingolipidomic could be potential prognostic tool in Systemic Lupus Erythematosus (SLE). Similarly, sphingosine-1-phosphate is used in prognosis of Community Acquired Pneumonia (CAP).

CONFLICTS OF INTEREST: The authors declare no conflict of interest.

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REFERENCES

[1] Daan van Kruining *et al.* Sphingolipids as prognostic biomarkers of neurodegeneration, neuroinflammation and psychiatric diseases and their emerging role in lipidomic investigation methods. Advanced Drug Delivery Reviews. 2020; 232-244.

[2] Alfred H. Merrill, Jr. Sphingolipid and Glycosphingolipid metabolic pathways in the era of sphingolipidomics. Chemical reviews. 2011; 111: 6387-6422.

[3] Kinga Czubowicz, Henryk Jesko, Przemyslaw Wencel, Walter J. Lukiw, Robert P. Strosznajder. The role of ceramide and sphingosine-1-phosphate in Alzheimer's disease and other neurodegenerative disorders. Molecular Neurobiology. 2019; 56: 5436-5455.

[4] Chiara Mencarelli, Pilar Martinez-Martinez. Ceramide function in the brain: when a slight tilt is enough. Cellular and Molecular Life Sciences. 2012; 70: 181-203.

[5] Christoph Michel, Gerhild van Echten-Deckert. Conversion of dihydroceramide to ceramide occurs at cytosolic face of the endoplasmic reticulum. FEBS Letters. 1997; 153-155.

[6] Christoph Michel *et al.* Characterization of ceramide synthesis: A dihydroceramide desaturase introduces the 4,5-trans double bond of sphingosine at the level of dihydroceramide. The Journal of Biological Chemistry. 1997; 22432-22437.

[7] Tomoko Funakoshi, Satoshi Yasuda, Masayoshi Fukasawa, Masahiro Nishijima, Kentaro Hanada. Reconstitution of ATP- and Cytosol-dependent Transport of de Novo Synthesized Ceramide to the Site of Sphingomyelin Synthesis in Semi-intact Cells. The Journal of Biological Chemistry. 2000; 29938-29945.

[8] Thomas Kolter, Konrad Sandhoff. Lysosomal degradation of membrane lipids. FEBS Letters. 2009; 1700–1712.

[9] Jae-Ho Park, Edward H. Schuchman. Acid ceramidase and human disease. Biochimica et Biophysica Acta. 2006; 2133-2138.

[10] Michael Maceyka *et al*. SphK1 and SphK2, Sphingosine Kinase Isoenzymes with Opposing Functions in Sphingolipid Metabolism. Journal of Biological Chemistry. 2005; 37118-37129.

[11] Arielle M. Bryan and Maurizio Del Poeta. Sphingosine-1-phosphate receptors and innate immunity. Cell Microbiol. 2018; 1-18.

[12] Andreas V. Thuy, Christina-Maria Reimann, Nasr Y. A. Hemdan, Markus H. Gräler. Sphingosine 1-Phosphate in Blood: Function, Metabolism, and Fate. Cell PhysiolBiochem. 2014; 34: 158-171.

[13] Kui Yang and Xianlin Han. Lipidomics: Techniques, applications, and outcomes related to biomedical sciences. Trends Biochem Sci. 2016; 41(11): 954–969.

[14] Changfeng Hu, Qiao Duan, Xianlin Han. Strategies to improve/eliminate the limitations in shotgun lipidomics. Proteomics. 2020; 20(11): 1-23.

[15] Changfeng Hu, Chunyan Wang, Lijiao He, Xianlin Han. Novel strategies for enhancing shotgun lipidomics for comprehensive analysis of cellular lipidomes. Trends Analyt Chem. 2019; 120: 1-25.

[16] Thomas Züllig, Martin Trötzmüller and Harald C. Köfeler. Lipidomics from sample preparation to data analysis: a primer. Analytical and Bioanalytical Chemistry. 2020; 412: 2191–2209.

[17] Alexander Triebl *et al*. Shared reference materials harmonize lipidomics across MS-based detection platforms and laboratories. Journal of Lipid Research. 2020; 105-115.

[18] Nienke M. de Wit, Kevin Mol, Sabela Rodrıguez-Lorenzo, Helga E. de Vriesand Gijs Kooij. The Role of Sphingolipids and Specialized Pro-Resolving Mediators in Alzheimer's Disease. Frontier in immunology. 2021; 1-14.

[19] Michelle M Mielke and Norman J Haughey. Could plasma sphingolipids be diagnostic orprognostic biomarkers for Alzheimer's disease?. Clinical Lipidology. 2012; 7(5): 525-536.

[20] Isabel Carreras *et al.* Dual dose-dependent efects of fingolimod in a mouse model of Alzheimer's disease. Scientific report. 2019; 1-11.

[21] Henryk Jesko *et al.* Fingolimod Affects Transcription of Genes Encoding Enzymesof Ceramide

Metabolism in Animal Model of Alzheimer's Disease. Molecular Neurobiology. 2020; 57: 2799–2811.

[22] Patrick Oeckland Markus Otto. A Review on MS-Based Blood Biomarkersfor Alzheimer's Disease. Neurol Ther. 2019; 113–127.

[23] Karim Belarbi *et al.* Glycosphingolipids and neuro-inflammationin Parkinson's disease. Molecular Neurodegeneration. 2020; 1-16.

[24] Yumiko V. Taguchi *et al.* Glucosylsphingosine Promotes - Synuclein Pathology inMutant GBA-Associated Parkinson's Disease. The Journal of Neuroscience. 2017; 37(40): 9617-9631.

[25] Silvia Paciotti, Elisabetta Albi, Lucilla Parnetti and Tommaso Beccari. Lysosomal Ceramide Metabolism Disorders: Implications in Parkinson's Disease. Journal of Clinical Medicine. 2020; 1-20.

[26] Guadalupe Vidal-Martínez *et al.* FTY720/Fingolimod Reduces Synucleinopathy and Improves Gut Motility in A53T Mice. The Journal of Biological Chemistry. 2016; 39: 20811–20821.

[27] P. McColgana and S. J. Tabrizia. Huntington's disease: a clinical review. Europeon Journal of Neurology. 2017; 24-34.

[28] Melanie Alpaugh *et al.* Disease-modifying effects of ganglioside GM1 inHuntington's disease models. EMBO Molecular Medicine. 2017; 9: 1537–1557.

[29] Laura Mangiarini *et al.* Exon 1 of the HD Gene with an Expanded CAG Repeat Is Sufficient to Cause a Progressive Neurological Phenotype in Transgenic Mice. Cell. 1996; 493–506.

[30] Clare Baecher-Allan, Belinda J. Kaskow, and Howard L. Weiner1. Multiple Sclerosis: Mechanisms and Immunotherapy. Neuron 97. 2018; 742-768.

[31] Somsankar Dasgupta, and Swapan K. Ray. Diverse Biological Functions of Sphingolipids in the CNS: Ceramide and Sphingosine Regulate Myelination in Developing Brain but Stimulate Demyelination during Pathogenesis of Multiple Sclerosis. J Neurol Psychol. 2017; 5: 1-17.

[32] Adnan M. Subei and Jeffrey A. Cohen. Sphingosine 1-Phosphate Receptor Modulators in Multiple Sclerosis. CNS Drugs. 2015; 29(7): 1-18.

[33] Veit Rothhammer *et al.* Sphingosine 1-phosphate receptor modulation suppresses pathogenic astrocyte activation and chronic progressive CNS inflammation. PNAS. 2017; 8: 1-5.

[34] Jerold Chun, Yasuyuki Kihara, Deepa Jonnalagadda, and Victoria A. Blaho. Fingolimod: Lessons Learned and New Opportunities for Treating Multiple Sclerosis and Other Disorders. Annu Rev Pharmacol Toxicol. 2019; 59: 149–170.

[35] Cristina Guarnera, Placido Bramanti and Emanuela Mazzon. Comparison of efficacy and safety of oral agents for the treatment of relapsing–remitting multiple sclerosis. Drug Design Development and Theory. 2017; 11: 2193–2207.

[36] Lesley J. Scott. Siponimod: A Review in Secondary Progressive Multiple Sclerosis. CNS Drugs. 2020; 34: 1191–1200.

[37] Antonietta Gentile *et al*. Siponimod (BAF312) prevents synaptic neuro-degeneration in experimental multiple sclerosis. Journal of Neuro-inflammation. 2016; 13(207): 1-13.

[38] Sung-Im Do *et al*. Predictive and prognostic value of sphingosine kinase 1 expression in patients with invasive ductal carcinoma of the breast. Am J Transl Res. 2017; 9(12): 5684-5695.

[39] Meena Kishore Sakharkar, Sarinder Kaur Dhillon, Saravana Babu Chidambaram, Musthafa Mohamed Essa and Jian Yang. Gene Pair Correlation Coefficients in Sphingolipid Metabolic Pathway as a Potential Prognostic Biomarker for Breast Cancer. Cancers. 2020; 1-8.

[40] Priyanka Bhadwal *et al*. LC-HRMS based approach to identify novel sphingolipid biomarkers in breast cancer patients. Scientific Reports. 2020; 10(4668): 1-15.

[41] Mirela Sedic, Petra Grbcic and Sandra Kraljevic Pavelic. Bioactive Sphingolipids as Biomarkers Predictive of Disease Severity and Treatment Response in Cancer: Current Status and Translational Challenges. Anti-cancer Research. 2019; 39: 41-56.

[42] Fatima Bilal *et al*. Sphingomyelin Synthase 1 (SMS1) Downregulation Is Associated With Sphingolipid Reprogramming and a Worse Prognosis in Melanoma. Frontiers in Pharmacology. 2019; 1-7.

[43] Ashley J. Snider *et al*. Identification of Plasma Glycosphingolipids as Potential Biomarkers for Prostate Cancer (PCa) Status. Biomolecules. 2020; 1-15.

[44] Peng Wang *et al*. Roles of sphingosine-1-phosphate signaling in cancer. Cancer Cell International. 2019; 19(295): 1-12.

[45] Olivia C. Harden and Samar M. Hammad. Sphingolipids and Diagnosis, Prognosis and Organ Damage in Systemic Lupus Erythematous. Frontiers in Immun-ology. 2020; 1-14.

[46] Shih-Chang Hsu *et al.* Circulating sphingosine-1-phosphate as aprog-nostic biomarker for community acquired pneumonia. Plos One. 2019; 1-13.