CJSJ

Alzheimer's Disease and A Prion-Like Protein: A Toxic Relationship

Krithika Karthik

Abstract— **The role of TDP-43 and its prion-like features in Alzheimer's disease (AD) represents a new avenue of research concerning the pathogenesis of the disorder. Research has focused on identifying proteins involved in inducing aggregation/toxicity of the illness, with the Tau and ß-amyloid proteins being primarily responsible. The TDP-43 protein was first discovered in 1995 and has attracted considerable interest in recent years. This review details the structural and functional characteristics of TDP-43. Special emphasis is given to the post-translational modifications and mutations that accompany neurotoxicity and protein aggregates found in the brain tissue of AD patients. The interface of TDP-43 with other proteins involved in AD progression is also elucidated based on studies in this regard. Investigations using animal models with the intent to identify potential therapeutic strategies to combat the disease have also been outlined in this work.**

I. INTRODUCTION

Alzheimer's disease (AD) is a progressive neurodegenerative disease identified by the death of brain cells, initially observed in the frontotemporal lobes of the brain, characterized by symptoms such as impaired neuronal transmission, brain atrophy and consequent shrinkage [1], [2]. External manifestations of AD include dementia, involving memory loss and declining cognitive and social skills. Of the 50 million cases of dementia worldwide, 60-70% have been diagnosed as Alzheimer's, as indicated by WHO statistics [2].

In 1906, Alois Alzheimer, a clinical psychiatrist and neuropathologist at Frankfurt Psychiatric Hospital, provided the first description of Alzheimer's disease as a 'peculiar severe disease process of the cerebral cortex' [3], and this disease continues to remain a mystery in some major aspects. Brain scans of AD patients have revealed extracellular parenchymal and intraneuronal aggregates of proteins, primarily the beta-amyloid and Tau proteins, leading to the formation of amyloid-beta (Aß) plaques and neurofibrillary tangles (NFTs) respectively [4]. Authors have also hypothesized that the onset of the disease could be related to prions, or prion-like polymorphisms of proteins, as observed in the case of the Tau, Aß, or even the TDP-43 protein [5], [6].

Prions are misfolded proteins which have been identified as causative agents of disorders such as Creutzfeldt-Jakob disease. Also, prions multiply via the conformational conversion of normal cellular prion proteins (PrPc) to the disease-causing (PrPSc) isoforms [7], [8] rather than the conventional nucleic acid replication.

Aberrant processing during polypeptide synthesis due to mutations in the prion protein gene dictate the specific abnormality of the neurons in the disorder [9]. In the context of AD, versions of Tau and Aß proteins are observed to adopt prion-like properties, causing them to spread through the brain to induce neurotoxicity. Thus, authors have suggested that these proteins could be potential culprits for progression of the disease [10].

II. EFFECT OF MUTANT PROTEINS

Familial Alzheimer's disease (FAD) is an autosomal dominant disease. Mutations in the amyloid precursor protein (APP), presenilin 1 or 2 genes have been reported to result in FAD [9]. APP undergoes hydrolysis to form Aβ peptides that are responsible for the amyloid fibrils found in the plaques of AD brains [11]. APP is cleaved at residue 671 by β-secretase to form the Aβ (1-40) fragment and at residue 711 or 713 by γ-secretase to form the Aβ (1-42) fragment as depicted in Fig. 1 [9]. These Aβ (1-42) peptides can cause disruption in the central nervous system [9], [12] whereas the Aβ (1-40) fragment is mainly present in the amyloid fibrils of the plasma and cerebrospinal fluid [12]. Plaque formation is reported due to Aβ peptides that are influenced by specific mutations (APP695, APP751, APP770) of the truncated APP gene, which are commonly located at cleavage sites of β-secretase and γ-secretase [13].

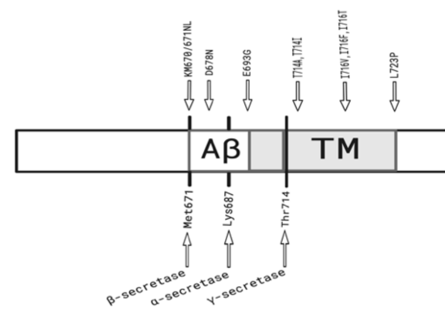

Figure 1. The C-terminus of the APP protein. The secretase cleavage sites and some mutations in the protein are depicted. Aβ, amyloid-beta; TM, transmembrane domain (Reproduced from [13])

Apart from the two key proteins Tau and Aβ, TDP-43 has also been implicated in the neurodegenerative processes occurring in AD [11]. For these reasons, studies conducted on TDP-43 have been reviewed in order to explore the likelihood of this protein as a potential therapeutic target.

III. TDP-43 PROTEIN

Structure

The TAR DNA binding protein (TDP-43) of 43 kDa is encoded by the TARDBP gene of 6 exons at the 1p36.22 locus [14]. This heterogenous ribonucleoprotein (hnRNP) of 414 amino acids [15] is localized primarily in the nucleus [16]. It consists of an N-terminal domain (NTD) and C-terminal domain (CTD), which is characterized as a prion-like domain [16]. The protein also has 2 RNA recognition motifs (RRM), and a nuclear localization signal (NLS) domain. The structure is depicted in Fig. 2.

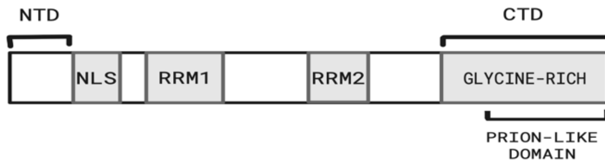

Figure 2. The TDP-43 protein structure and important domains (Reproduced from [17])

Function

The overall function of TDP-43 under nontoxic conditions includes the RNA regulation during transcription, mRNA stabilization, and alternative splicing [16]. The specific function of each domain is detailed in Table 1. The CTD is a low complexity sequence that can transition between alpha-helix and beta-sheet structures, similar to how prions trigger the onset of a disease [1], [18]. This, along with the prevalence of pathogenic TDP-43 in 20-50% of AD cases [11], has led researchers to suspect that TDP-43 could also be a possible contributor to the toxicity observed in AD brains [16].

TABLE I. STRUCTURAL AND FUNCTIONAL ANALYSIS OF THE TDP-43 PROTEIN DOMAINS [19]

Domain	Residual Position	Function under Non-toxic Conditions
NTD	1-77	Protein dimerization and oligomerization [16], [20]
NLS	78-100	Translocation of TDP-43 from nucleus to cytoplasm for cytoplasmic accumulation of proteins [4], [15]
RRM1	106-177	Specific RNA binding [4], [15]
RRM2	192-259	Specific RNA binding [4], [15]
CTD	260-414	Phase separation, aggregation, solubility, and protein homeostasis [16], [20]

Proteinopathies involving TDP-43

Studies have shown strong evidence for TDP-43 as a pathological hallmark for frontotemporal lobar degeneration (FTLD) and amyotrophic lateral sclerosis (ALS) [21]. TDP-43 localization in AD brains has also received attention in recent times. TDP-43 deposits as inclusion bodies in neuronal and glial cells of the central nervous system and spreads through the brain in 6 stages. The accumulation begins in the amygdala (stage 1), followed by the subiculum entorhinal cortex (stage 2), the hippocampus and occipitotemporal cortex (stage 3), the ventral striatum, insular and temporal cortices (stage 4), the brainstem (stage 5) and finally the basal ganglia and midfrontal cortex (stage 6) as reported by [6], [11].

The TARDBP gene was found to have mutations in genetic cases of ALS and FTLD [16], [22], but few researchers have assessed this gene in relation to AD [1]. A clinical trial consisting 181 AD patients and 130 controls in a Japanese population with 8 TARDBP gene polymorphisms observed no significant relationship [1]. Yet, it is still likely that the TDP-43 and Aβ/tau protein are related. Results show that Aβ deposits cause can cause abnormal TDP-43 aggregation and that TDP-43 is involved in tau aggregation [11]. Thus, TARDBP gene mutations could be responsible for the abnormal, mutated behavior of TDP-43, indicative of the need for more research on TDP-43 mutations pertaining to AD.

III. POST-TRANSLATIONAL MODIFICATIONS (PTMS) OF TDP-43

The TDP-43 protein can undergo multiple PTMs including truncation, ubiquitination, phosphorylation, and acetylation. As a target for multiple PTMs TDP-43 may contribute to AD progression in multiple ways. It should be noted that although there is evidence suggesting TDP-43 PTMs can stimulate aggregation [19], some studies also report PTMs occurring post-aggregation (refer to the Phosphorylation section) [23]. Consequently, whether these PTMs are the cause or the effect of aggregations is still unclear.

Truncation

The cleavage of TDP-43 leads to the formation of N terminal fragments (NTFs) or C terminal fragments (CTFs), depending on the cleavage site. The NTFs retain their function and break down in the nucleus, whereas CTFs translocate to the cytoplasm and aggregate [24]. The truncation of the RRMs affects the ability of the protein to carry out RNA regulation/binding and dimerization. Furthermore, the removal of part of/the whole NLS domain triggers further cytoplasmic protein aggregation [19]. Proteolytic cleavage at the 89/90 position and at position 169/170 or 174/175 produces TDP-35 and TDP-25, which are other commonly occurring fragments [19]. As studied by Li et al., western blot analyses of such TDP-43 fragments show higher levels of insolubility and therefore propensity to aggregate [23].

Ubiquitination

Ubiquitination involves the binding of the ubiquitin protein to TDP-43 [19]. This PTM regulates the activation/inactivation, localization and interactions of proteins. Common ubiquitination sites are lysine residues,

particularly at positions 160, 181, and 263. François-Moutal et al. found that the K181 residue could disrupt the secondary/tertiary structure of TDP-43 due to the changes in the interactions between the RRM domains or between the RRMs and NTD. It is also observed that the K263 residue can be easily ubiquitinated to decrease levels of RNA binding and aid in aggregative tendencies [19].

Phosphorylation

Phosphorylation involves the addition of phosphate groups to amino acids in the TDP-43 sequence. Abnormal or hyper-phosphorylation is also known to indicate neurotoxicity [19]. In fact, hyperphosphorylation of the tau protein in AD triggers the formation of NFTs [11], [25]. Serine residues, and less commonly threonine residues, are common phosphorylation sites. Interestingly, the CTD contains such serine and threonine residues [18]. This suggests that TDP-43 phosphorylation promotes aggregation and neurodegeneration [24]. However, the effect of this PTM is debatable, with certain studies suggesting the PTM serves a cytoprotective function, easing proteotoxic stress caused by accumulation of misfolded proteins and aggregates [24]. Either way, phosphorylation appears to play a key role in the progression of TDP-43 proteinopathies in a disease like AD.

Acetylation

Acetylation involves the addition of acetyl groups to amino acids in the TDP-43 sequence. The common target for acetylation are lysine residues [19]. This process can occur via various mechanisms such as RNA processing, cytoskeleton association, and cellular signaling [19]. The specific position undergoing acetylation could be Lys82 and 192 [19]. Cohen et al. have reported the occurrence of inclusion bodies in the cytoplasm reminiscent of TDP-43 proteinopathy aggregates [21].

IV. STUDIES INVOLVING ANIMAL MODELS OVEREXPRESSING PROTEINS IMPLICATED IN AD

Transgenic Mice Models: Advantages and Limitations

Mice are commonly used animals in studies due to their genetic similarity to humans. Wild type mice were found to exhibit 97% and 88% sequence homology to the human version of the APP and Tau proteins respectively [4]. When these mice overexpress APP, their pathology is reminiscent of AD pathogenesis. The Aß plaques are composed of the Aß (42) peptide, ubiquitin, and α-synuclein, among other constituents. α-synuclein is of special note as it has been characterized as “prion-like” following studies using animal models [12]. The mice, like human patients, were also observed to develop cognitive impairments which were not directly proportional to Aß plaque formation [26].

However, although mice can produce Aß, the plaque formation and development of AD-like characteristics are dependent on the APP mutations which resemble FAD, but not sporadic AD cases. The development of NFTs is also absent in these models. Nonetheless, if mutant APP and several other proteins are present, age-dependent development of both Aß plaques and NFTs can form in mice [26], [4].

Non-human Primate Models: Advantages and Limitations

Nonhuman primates are preferable models to study disease development in that their “behavioral complexity,” brain size, and genetics are highly similar to that of humans. In fact, their Aß and tau protein have 100% and 99.5-100% sequence homology with the human forms respectively, and protein accumulation occurs naturally. New world monkeys like squirrel monkeys experience neurotoxicity resembling that of humans, with possession of Aß (1-40) and Aß (1-42) of particular interest. These peptides accumulate and aggregate to form Aß plaques [4]. Thus, these models are favorable to study the propagation of the Aß protein and plaque formation.

However, primates suffer from the limitation of long lifespans and delayed neuropathology. Furthermore, cognitive symptoms are not evident and Aß and tau are found in smaller amounts than as seen in humans. Baboons, for example, only rarely have Aß accumulation and squirrel monkeys have no NFTs present [4].

Few studies have been done on TDP-43 using mammalian models. Mutant TDP-43 isoforms mainly remain in the nucleus or thinly distributed near the nucleus/in the cytoplasm in mice, but shuttle to the cytoplasm in primates [27], [24]. This distinction in the distribution of TDP-43 between mice and monkeys is to be noted, as it calls the reliability of the models into question.

V. THERAPEUTIC APPROACHES TARGETING PROTEINS IMPLICATED IN AD

Drugs can be used to target various aspects of AD, including preventing APP or TDP-43 fragmentation, reducing TDP-43 expression, or inhibiting PTMs of tau and TDP-43. In this review, the drugs targeting various aspects of TDP-43 proteinopathies are detailed in Table 2 [15].

TABLE II. DRUGS TARGETING TDP-43

Drug	Description	Model System Used	Effects Observed (Interactions with TDP-43)
Berberine	Medicinal herb that can be orally ingested [28]	Cell culture model Mouse model	Reduction of accumulation and aggregation of TDP-43 fragments [28] Decrease in levels of Aβ and phosphorylated tau, leading to improvements in cognitive symptoms (learning/motor skills and spatial

			memory) [29], [28]
N-Acetyl-cysteine (NAC)	Compound with antioxidative properties [15]	-	Modification of abnormal cytoplasmic accumulation of TDP-43 in neuron-like cells and reduction of toxic effects due to arsenite-induced insolubility and ubiquitination [15]
Dexamethasone	Synthetically produced steroid hormone (glucocorticoid) [15]	Transgenic mice model	Increase in TDP-25 solubility, improving cognitive symptoms [15]
Epigallocatechin gallate (EGCG)	Polyphenolic plant compound found [30]	-	Conversion of Aß protein, synuclein protein (eg- α-synuclein) and yeast prion (PSI), which tend to form amyloid deposits, into harmless oligomers [15]
Curcumin Dimethoxy	A compound, specifically monocarbonyl dimethoxy [15]	-	Reduce toxicity induced by mutant TDP-43 and halt aggregation due to pathological TDP-25 [15]
QBP1 (PolyQ peptide binding 1)	Peptide sequence targeting polyglutamine sequences reminiscent of amyloid fibrils [31]	*In vitro* models	Binding and inhibition of amyloid fibril production by targeting the Q/N rich CTD of TDP-43 [15]
Rolipram	Drug capable of inhibiting phosphodiesterase-4 enzyme	Transgenic mice models	Decrease in levels and aggregation of TDP-43 in specific neurons [15]
Riluzole	Drug clinically approved to treat ALS [15]	-	Decrease in TDP-43 interactions in a dose-dependent fashion [32]

VI. DISCUSSION

Studies investigating the role of TDP-43 in AD is still relatively minimal. Research has mainly focused upon ALS and FTLD, with relatively few studies directly extending this research to TDP-43 proteinopathies occurring in AD. Further studies are required to determine any reliable correlations between AD and TDP-43. The regulation of the Aβ/tau-protein by TDP-43 is unclear, so the interactions of TDP-43 requires further exploration [11]. This different perspective, of investigating the prion-like properties of the TDP-43 protein, should be seriously accounted for as it could help hasten the development of safer, more effective treatment methods for AD.

Immunotherapy appears to be a promising path [9], [12]. For example, in one study [9], [26], the administration of antibodies decreased Aβ plaque levels and improved the cognitive symptoms. However, any immunotherapeutic treatment comes with a high risk factor. One example of a problem observed is vasogenic edema or encephalitis, which in some cases is also accompanied by hemorrhage [4]. Some drugs are promising, however, especially those that can slow the 6-stage transgression of TDP-43 through the brain. Such a drug could have important implications as the majority of AD diagnoses occur late into the disease—80% in stage 3 and 85% in stage 4 or 5 [6]—and slowing movement of TDP-43 can allow for earlier detection of proteinopathies. It should also be noted that certain drug molecules described in this review, such as NAC, are nutritional supplements and their potential as preventative/protective drugs should be explored in greater detail.

Caenorhabditis elegans models have been used extensively to study AD, contributing to major advances in the repertoire of this disease [22], [33]. *Drosophila melanogaster* is another common model organism. The *C. elegans* worms and *D. melanogaster* flies can be rapidly multiplied at low costs [33], [34], such that they are a convenient choice for *in vivo* studies, although phenotypic features may not resemble that of humans. For mammalian models, numerous ethical guidelines must be considered. Therefore, there is no ideal model organism to study AD, so the model should be selected cognizant of what aspect needs to be studied.

Mutations and PTMs play vital roles on the behavior of TDP-43 and its consequent effect on AD cases. Most cases of AD have been associated with mutations in the APP, PSEN1, PSEN2 genes among others. These mutations should be studied further, including their interaction with the TARDBP gene. The interaction of different PTMs could also be explored regarding effects on pathogenesis. as ubiquitination and truncation appear to drive further neurotoxicity together [18]. Overall, the PTMs of TDP-43 seem to be interrelated and codependent in certain situations, and hence they pave the way for the use of interesting study designs in order to further understand the role of TDP-43 in AD.

VII. CONCLUSION

The gravity of this situation, in which both the diagnosis and treatment/prevention of AD is unclear, is not to be understated. Different mechanisms by which drugs can target TDP-43 in AD brain tissues must be explored further. The relationship between TDP-43 and AD is not definite but has strong supporting evidence, indicating that more research is required to directly associate both. Although TDP-43 is not prevalent in all AD cases, it presents a novel approach due to

its prion-like characteristics. The TDP-43 protein could therefore pioneer breakthroughs in studies directed towards a more comprehensive knowledge of AD with the objective to develop possible prevention/therapeutic strategies.

VIII. ACKNOWLEDGEMENT

I would like to extend my gratitude to my mentor Dr. Mrinalini Menon for her valuable guidance and feedback in researching for and writing my article. I would also like to thank Ms. Neha Kumar for providing this opportunity and guiding me through formatting and referencing in my article. Finally, I would like to thank Ms. Paulomi Choudhury for her inputs on writing and organizing a review article.

IX. REFERENCES

[1] A. C. Wilson, B. N. Dugger, D. W. Dickson, and D.-S. Wang, "TDP-43 in aging and Alzheimer's disease - a review," *Int. J. Clin. Exp. Pathol.*, vol. 4, no. 2, pp. 147–155, Jan. 2011.
[2] "Dementia." https://www.who.int/news-room/fact-sheets/detail/dementia
[3] H. Hippius and G. Neundörfer, "The discovery of Alzheimer's disease," *Dialogues Clin. Neurosci.*, vol. 5, no. 1, pp. 101–108, Mar. 2003, doi: 10.31887/DCNS.2003.5.1/hhippius.
[4] E. Drummond and T. Wisniewski, "Alzheimer's disease: experimental models and reality," *Acta Neuropathol. (Berl.)*, vol. 133, no. 2, pp. 155–175, Feb. 2017, doi: 10.1007/s00401-016-1662-x.
[5] K. A. Josephs *et al.*, "Staging TDP-43 pathology in Alzheimer's disease," *Acta Neuropathol. (Berl.)*, vol. 127, no. 3, pp. 441–450, Mar. 2014, doi: 10.1007/s00401-013-1211-9.
[6] S. Nag, L. Yu, P. A. Boyle, S. E. Leurgans, D. A. Bennett, and J. A. Schneider, "TDP-43 pathology in anterior temporal pole cortex in aging and Alzheimer's disease," *Acta Neuropathol. Commun.*, vol. 6, no. 1, p. 33, May 2018, doi: 10.1186/s40478-018-0531-3.
[7] H. Liu *et al.*, "Solution Structure of Syrian Hamster Prion Protein rPrP(90−231)," *Biochemistry*, vol. 38, no. 17, pp. 5362–5377, Apr. 1999, doi: 10.1021/bi982878x.
[8] R. Riek, S. Hornemann, G. Wider, M. Billeter, R. Glockshuber, and K. Wüthrich, "NMR structure of the mouse prion protein domain PrP(121–231)," *Nature*, vol. 382, no. 6587, pp. 180–182, Jul. 1996, doi: 10.1038/382180a0.
[9] S. B. Prusiner, "Neurodegenerative Diseases and Prions," *N. Engl. J. Med.*, vol. 344, no. 20, pp. 1516–1526, May 2001, doi: 10.1056/NEJM200105173442006.
[10] "Alzheimer's Disease is a 'Double-Prion Disorder,' Study Shows." https://www.ucsf.edu/news/2019/05/414326/alzheimers-disease-double-prion-disorder-study-shows
[11] W. Huang *et al.*, "TDP-43: From Alzheimer's Disease to Limbic-Predominant Age-Related TDP-43 Encephalopathy," *Front. Mol. Neurosci.*, vol. 13, p. 26, 2020, doi: 10.3389/fnmol.2020.00026.
[12] F. Ciccocioppo *et al.*, "Neurodegenerative diseases as proteinopathies-driven immune disorders," *Neural Regen. Res.*, vol. 15, no. 5, pp. 850–856, May 2020, doi: 10.4103/1673-5374.268971.
[13] L. M. Bekris, C.-E. Yu, T. D. Bird, and D. W. Tsuang, "Genetics of Alzheimer disease," *J. Geriatr. Psychiatry Neurol.*, vol. 23, no. 4, pp. 213–227, Dec. 2010, doi: 10.1177/0891988710383571.
[14] H.-Y. Wang, I.-F. Wang, J. Bose, and C.-K. J. Shen, "Structural diversity and functional implications of the eukaryotic TDP gene family," *Genomics*, vol. 83, no. 1, pp. 130–139, Jan. 2004, doi: 10.1016/S0888-7543(03)00214-3.
[15] E. Buratti, "Targeting TDP-43 proteinopathy with drugs and drug-like small molecules," *Br. J. Pharmacol.*, vol. 178, no. 6, pp. 1298–1315, Mar. 2021, doi: 10.1111/bph.15148.
[16] M. Jo, S. Lee, Y.-M. Jeon, S. Kim, Y. Kwon, and H.-J. Kim, "The role of TDP-43 propagation in neurodegenerative diseases: integrating insights from clinical and experimental studies," *Exp. Mol. Med.*, vol. 52, no. 10, pp. 1652–1662, Oct. 2020, doi: 10.1038/s12276-020-00513-7.
[17] S. Farina, F. Esposito, M. Battistoni, G. Biamonti, and S. Francia, "Post-Translational Modifications Modulate Proteinopathies of TDP-43, FUS and hnRNP-A/B in Amyotrophic Lateral Sclerosis," *Front. Mol. Biosci.*, vol. 8, p. 693325, Jul. 2021, doi: 10.3389/fmolb.2021.693325.
[18] R. C. Hergesheimer *et al.*, "The debated toxic role of aggregated TDP-43 in amyotrophic lateral sclerosis: a resolution in sight?," *Brain J. Neurol.*, vol. 142, no. 5, pp. 1176–1194, May 2019, doi: 10.1093/brain/awz078.
[19] L. François-Moutal, S. Perez-Miller, D. D. Scott, V. G. Miranda, N. Mollasalehi, and M. Khanna, "Structural Insights Into TDP-43 and Effects of Post-translational Modifications," *Front. Mol. Neurosci.*, vol. 12, pp. 301–301, Dec. 2019, doi: 10.3389/fnmol.2019.00301.
[20] B. Borroni, A. Alberici, and E. Buratti, "Review: Molecular pathology of frontotemporal lobar degenerations," *Neuropathol. Appl. Neurobiol.*, vol. 45, no. 1, pp. 41–57, Feb. 2019, doi: 10.1111/nan.12534.
[21] T. J. Cohen, A. W. Hwang, C. R. Restrepo, C.-X. Yuan, J. Q. Trojanowski, and V. M. Y. Lee, "An acetylation switch controls TDP-43 function and aggregation propensity," *Nat. Commun.*, vol. 6, no. 1, p. 5845, Jan. 2015, doi: 10.1038/ncomms6845.
[22] N. F. Liachko, C. R. Guthrie, and B. C. Kraemer, "Phosphorylation Promotes Neurotoxicity in a Caenorhabditis elegans Model of TDP-43 Proteinopathy," *J. Neurosci.*, vol. 30, no. 48, pp. 16208–16219, Dec. 2010, doi: 10.1523/JNEUROSCI.2911-10.2010.
[23] H.-Y. Li, P.-A. Yeh, H.-C. Chiu, C.-Y. Tang, and B. P. Tu, "Hyperphosphorylation as a Defense Mechanism to Reduce TDP-43 Aggregation," *PLOS ONE*, vol. 6, no. 8, p. e23075, Aug. 2011, doi: 10.1371/journal.pone.0023075.
[24] D. Chhangani, A. Martín-Peña, and D. E. Rincon-Limas, "Molecular, functional, and pathological aspects of TDP-43 fragmentation," *iScience*, vol. 24, no. 5, p. 102459, May 2021, doi: 10.1016/j.isci.2021.102459.
[25] M. A. DeTure and D. W. Dickson, "The neuropathological diagnosis of Alzheimer's disease," *Mol. Neurodegener.*, vol. 14, no. 1, p. 32, Dec. 2019, doi: 10.1186/s13024-019-0333-5.
[26] F. M. LaFerla and K. N. Green, "Animal Models of Alzheimer Disease," *Cold Spring Harb. Perspect. Med.*, vol. 2, no. 11, pp. a006320–a006320, Nov. 2012, doi: 10.1101/cshperspect.a006320.
[27] P. Yin *et al.*, "Caspase-4 mediates cytoplasmic accumulation of TDP-43 in the primate brains," *Acta Neuropathol. (Berl.)*, vol. 137, no. 6, pp. 919–937, Jun. 2019, doi: 10.1007/s00401-019-01979-0.
[28] C.-F. Chang *et al.*, "Therapeutic effect of berberine on TDP-43-related pathogenesis in FTLD and ALS," *J. Biomed. Sci.*, vol. 23, no. 1, p. 72, Oct. 2016, doi: 10.1186/s12929-016-0290-z.
[29] S. S. K. Durairajan *et al.*, "Berberine ameliorates β-amyloid pathology, gliosis, and cognitive impairment in an Alzheimer's disease transgenic mouse model," *Neurobiol. Aging*, vol. 33, no. 12, pp. 2903–2919, Dec. 2012, doi: 10.1016/j.neurobiolaging.2012.02.016.
[30] Q. Y. Eng, P. V. Thanikachalam, and S. Ramamurthy, "Molecular understanding of Epigallocatechin gallate (EGCG) in cardiovascular and metabolic diseases," *J. Ethnopharmacol.*, vol. 210, pp. 296–310, Jan. 2018, doi: 10.1016/j.jep.2017.08.035.
[31] S. Chen, V. Berthelier, J. B. Hamilton, B. O'Nuallai, and R. Wetzel, "Amyloid-like Features of Polyglutamine Aggregates and Their Assembly Kinetics," *Biochemistry*, vol. 41, no. 23, pp. 7391–7399, Jun. 2002, doi: 10.1021/bi011772q.
[32] M. Oberstadt *et al.*, "TDP-43 self-interaction is modulated by redox-active compounds Auranofin, Chelerythrine and Riluzole," *Sci. Rep.*, vol. 8, no. 1, p. 2248, Feb. 2018, doi: 10.1038/s41598-018-20565-0.
[33] T. Kaletta and M. O. Hengartner, "Finding function in novel targets: C. elegans as a model organism," *Nat. Rev. Drug Discov.*, vol. 5, no. 5, pp. 387–399, May 2006, doi: 10.1038/nrd2031.
[34] N. S. Tolwinski, "Introduction: Drosophila-A Model System for Developmental Biology," *J. Dev. Biol.*, vol. 5, no. 3, p. 9, Sep. 2017, doi: 10.3390/jdb5030009.