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FYN, SARS-CoV-2, and IFITM3 in the neurobiology of Alzheimer's disease

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ABSTRACT

Introduction: (IFITM3) is an innate immune protein that has been identified as a novel γ-secretase (γs) modulator. FYN is a kinase that stabilizes IFITM3 on the membrane, primes APP for amyloidogenic γs processing and mediates tau oligomerization. The purpose of this study is to explore the role of FYN and IFITM3 in AD and COVID-19, expanding on previous research from our group.
Methods: A 520 gene signature containing FYN and IFITM3 (termed Ia) was extracted from a previously published meta-analysis of Alzheimer's disease (AD) bulk- and single nuclei sequencing data. Exploratory analyses involved meta-analysis of bulk and single cell RNA data for IFITM3 and FYN differential expression per CNS site and cellular type. Confirmatory analyses, gene set enrichment analysis (GSEA) on I_a was performed to detect overlapping enriched biological networks between COVID-19 with AD.
Results: Bulk RNA data analysis revealed that IFITM3 and FYN were overexpressed in two CNS regions in AD vs. Controls: the temporal cortex Wilcoxon p-value=1.3e-6) and the parahippocampal cortex Wilcoxon p-value=0.012). Correspondingly, single cell RNA analysis of IFITM3 and FYN revealed that it was differentially expressed in neurons, glial and endothelial cells donated b AD patients, when compared to controls.
Discussion: IFITM3 and FYN were found as interactors within biological networks overlapping between AD and SARS-CoV-2 infection. Within the context of SARS-CoV-2 induced tau aggregation and interactions between tau and Ab1–42, the FYN – IFITM3 regulome may outline an important innate immunity element responsive to viral infection and IFN-I signaling in both AD and COVID-19.

1. Introduction

Interferon-induced transmembrane protein 3 (IFITM3) belongs to a family of proteins that act as a second line of defense against enveloped viruses, including SARS-CoV-2. IFITM3's role in intercepting and shuttling viral particles to the lysosomes[1] was recently complemented by its discovery as a novel γ-secretase modulator that promotes Aβ production[2]. Considering the accumulating evidence on common pathways between COVID-19 and Alzheimer's disease (AD)[3], we aimed to examine whether FYN, a kinase regulating IFITM3's membrane localization [4,5] is accordingly perturbed in both AD and COVID-19 transcriptomes. Current state of the art transcriptomic evidence suggest that FYN interacts with SARS-CoV-2 during the course of infection[6], and was found to be upregulated in a recent meta-analysis of SARS-CoV-2 expression datasets[7]. Expanding on our previous research on IFITM3 networks in AD[8] and their overlap with COVID-19 [9], we propose a comprehensive model of AD pathogenesis where viral induction of the IFITM3/FYN endocytosis signal could account for increased Aβ oligomerization via γ-secretase activation[2]. Furthermore, SARS-CoV-2 induced FYN dysregulation / overactivation [6,7] would concomitantly and independently promote Tau fibrilization[10], abrogate autophagy[4], and prepare APP[11] for processing by the previously $IFITM3^2$-activated γ-secretase complex.

In order to explore FYN and IFITM3's expression patterns and networks in AD vs. COVID-19, we applied a composite systems biology approach[12]. Gene expression data from both bulk tissue and single cell RNA sequencing studies were used in order to explore FYN and IFITM3's expression patterns in CNS cites and cells beyond those examined by previous research from our group [8,9]. Subsequently, we aimed to investigate the overlap between FYN/IFITM3's biological

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networks and SARS-CoV-2 infectomics. Finally, we provide data on FYN/IFITM3 networks that arose in our previous study and integrated them in a comprehensive model of AD and AD-like manifestations of NeuroCOVID-19′s pathogenesis.

2. Methods

2.1. Data acquisition

For this study, we utilized consensus gene module and differential gene expression data on IFITM3 and FYN from a previously published [13] of AD brain transcriptomes. These datasets included data generated by the Accelerating Medicines Partnership Alzheimer's Disease Project (AMP-AD)[14] as well as publicly available datasets [15-17]; The AMP-AD datasets included RNA-seq provided by the Mayo Clinic Brain Bank[18], the Religious Orders Study and Memory and Aging Project (ROSMAP) [19] and the Mount Sinai School of Medicine (MSSM)[20] studies.

Consensus gene expression modules represent gene co-expression networks detected via consensus Weighted Gene Co-expression Network Analysis (cWGCNA)[21]. These networks contain highly correlated genes that are conserved across studies, and are furthermore associated with specific sample traits, such as cell type and diagnosis, and combined with other sources of data, such as single nuclei RNA-seq experiments and genome wide association studies (GWAS) . For both genes of interest, module membership and the eigengene-based connectivity ($k_{ME;}$ range of values: −1 to 1) are reported according to primary data.

2.2. Reconstruction of the FYN – IFITM3 interactome and comparative transcriptomics with COVID-19 datasets

Consensus gene modules containing FYN and IFITM3 were considered as candidate interactomes, visualized via STRING (available from: https://string-db.org)[22] and used for comparative gene set enrichment analyses (GSEA) via Enrichr[23] (Available from: (Available from: https://maayanlab.cloud/Enrichr/). Enrichment analyses aimed both to (a) determine the biological functions and pathways associated with FYN/IFITM3 signatures and (b) determine overlap with gene signatures extracted from COVID-19 datasets. For GSEA, adjusted p-values <0.05 were considered statistically significant.

2.3. Determination of IFITM3-FYN's cell- and CNS site- associations

Consensus gene modules represent gene co-expression networks that are conserved across datasets, CNS regions and enriched for specific states (i.e. upregulated in AD) and cell types[24]. In order to determine whether FYN and IFITM3's differential expression was region specific, we inquired each gene's comparative expression per study and CNS region (accessible via: http://swaruplab.bio.uci.edu:3838/bulkRNA/) [13]. Expression data were reported as reported as log2 transformed hub gene expression, with unpaired Wilcoxon test p-values<0.05 determining differential gene expression between AD vs. Controls.

While data on asymptomatic AD are also provided, we did not consider them in the context of determining FYN / IFITM3 associations with CNS sites in AD. Furthermore, is should be noted that differential gene expression were complimentary to the extraction of the FYN / IFITM3 interactome, as detailed previously.

2.4. Determination of FYN and IFITM3's cell- and disease- associations in AD via single cell RNA transcriptomics

For single-cell expression studies, the scREAD database (Available from: https://bmbls.bmi.osumc.edu/scread/) was interrogated, to further characterize FYN and IFITM3's expression in AD-donated cells and associated CNS regions [10,11]. The scREAD platform is a unique database compiling single cell RNA sequencing (scRNA-Seq) data from AD studies along with matched control atlases. Furthermore, it provides differential gene expression data that account for gender, brain region, and age. Herein, we screened gene expression data from human datasets, and examined all cross-dataset, disease – control comparisons. For all comparisons, a gene was considered as differentially expressed when the independent samples Wilcoxon p-values <0.05 (Bonferroni adjusted) and the absolute value of the logFC in the single cell resolution was >0.25[25].

2.5. Data availability statement

Primary data are available from their respective sources, as cited herein. All data meta-data generated by the analyses in this manuscript are available online via Mendeley Data (Available from: https://data.mendeley.com/datasets/5bypp2h5kj/1) and as supplementary files.

3. Results

3.1. Reconstruction of the FYN – IFITM3 interactome, its associations with AD and cell types

Both FYN (k_{ME} = 0.87) and IFITM3 (k_{ME}= 0.77) were detected in a consensus module (CM9; n_{genes}=520) that was positively correlated with AD diagnosis, significantly enriched in astrocytes and endothelial cells, and methylation quantitative trait loci (mQTL)[13]. This FYN/IFITM3 containing module was used as a candidate interactome (henceforth dubbed I_a; Fig. 1) and was subsequently used in GSEA).

Analysis of bulk RNA data revealed that IFITM3 was overexpressed in two regions in the discovery datasets: the temporal cortex (TC; Mayo Clinic Study, AD vs. Controls, Wilcoxon test p-value=$1.3e^{-6}$) and the parahippocampal gyrus (PHCG; MSSM study, AD vs. controls, Wilcoxon test p-value=0.012); (Supplementary Fig.1a). In the validation datasets, IFITM3 was differentially expressed in Zhang et al's study [9] (AD vs. Controls, Wilcoxon test p-value<$2.2e^{-6}$; Supplementary Fig. 1b). FYN was differentially expressed in the temporal cortex (Mayo Clinic Study, AD vs. Controls, adj. p-value=$2.9e^{-5}$), the prefrontal cortex (PFC; ROSMAP Study, AD vs. Controls, Wilcoxon test p-value=0.004) and the parahippocampal gyrus (MSSM study, AD vs. controls, Wilcoxon test p-value=$2.9e^{-5}$); Supplementary Fig. 1c. In the validation datasets, FYN was differentially expressed in two datasets, including Zhang et al's study [13] (AD vs. Controls, Wilcoxon test p-value<$2.2e^{-16}$; Supplementary Fig 1d). Taken together, these results indicate that the overexpression of FYN and IFITM3 can be localized to the PFC, the TC and the PHCG (Fig. 1 and Supplementary Materials $1_{a\text{-}d}$).

3.2. FYN and IFITM3's cell- and disease- associations in AD via single cell RNA transcriptomics

Cross-dataset comparisons of the scREAD database revealed that IFITM3 and FYN were differentially expressed in AD when compared to controls (adj. p-value<0.05) in a cell-type and CNS site-specific manner (Figs. 2 and 3)

Specifically, that IFITM3 was overexpressed (a) in astrocytes, microglia and endothelial cells in the entorhinal cortex datasets (b) underexpressed astrocytes, microglia and endothelial cells and oligodendrocyte precursor cells in the prefrontal cortex datasets(d) overexpressed in microglia the superior parietal lobe datasets AD01203, AD01204 and underexpressed in microglia from the superior parietal lobe dataset AD01205 (Supplementary Materials 2a,b).

FYN was differentially expressed in astrocytes, microglia, oligodendrocytes, oligodendrocyte precursors, endothelial cells, excitatory and inhibitory neurons in the entorhinal cortex, prefrontal cortex and the superior frontal gyrus. No clear pattern of differential expression (ubiquitously up or down) could be established between cell types or CNS regions, indicating significant cell-to-cell variability[25]

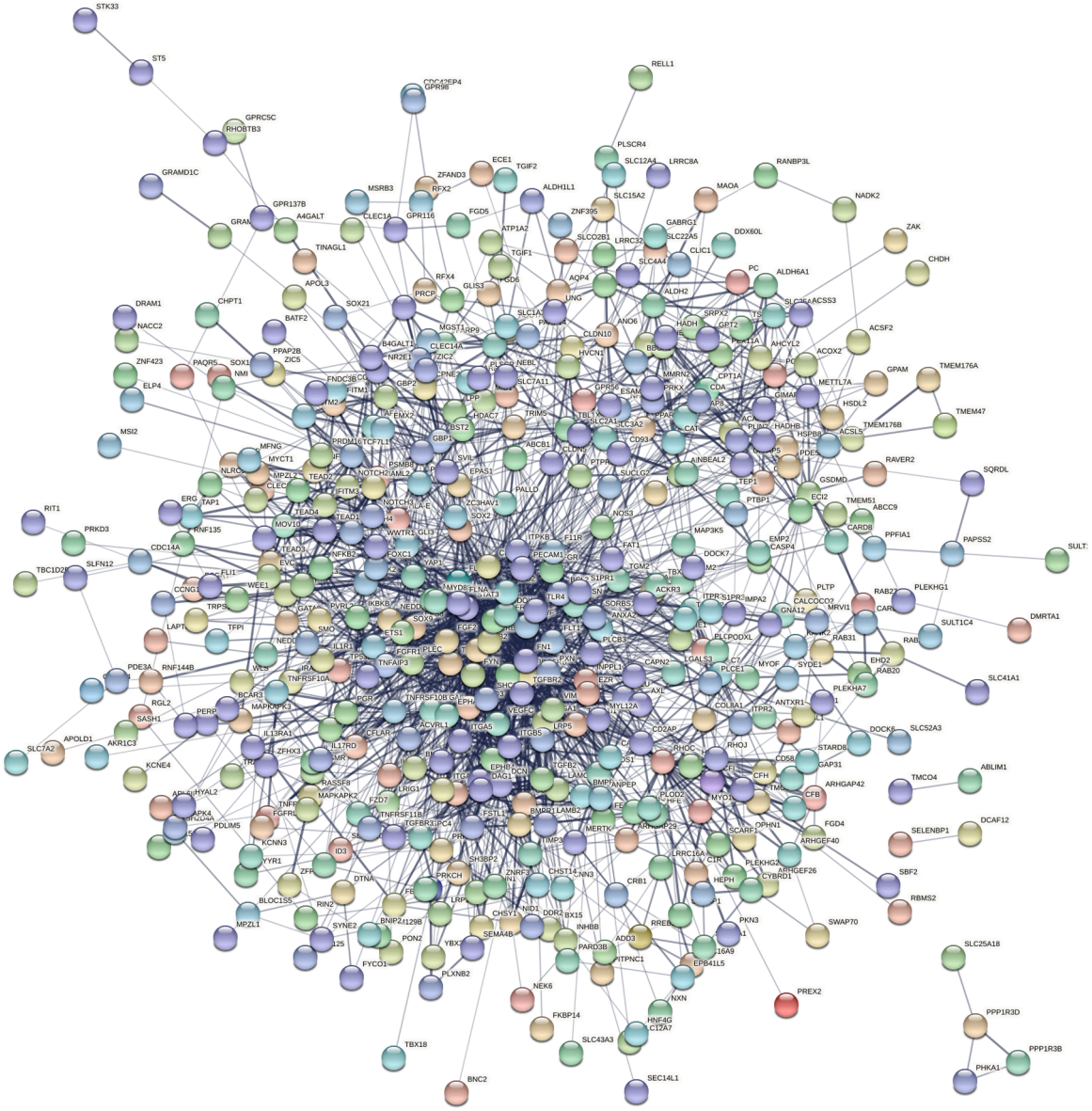

Fig. 1. Representation of the FYN-IFITM3 interactome via STRING.

(Supplementary Materials 2a,b).

3.3. Biological pathways and functions associated with FYN / IFITM3 and comparative transcriptomics with COVID-19 datasets

GSEA revealed several distinct, significantly enriched pathways and ontologies for FYN and IFITM3 (Supplementary Materials 3). Both FYN and IFITM3 were detected in the following significantly enriched pathways / ontologies:

"Cytokine Signaling in Immune system Homo sapiens" (Reactome Identifier: R-HSA-1280215; adjusted p-value=$5.98e^{-04}$), "Immune System Homo Sapiens" (Reactome Identifier: R-HSA-168256; adjusted p-value=0.00349), "Cytokine-mediated signaling pathway" (Gene Ontology Identifier:0019221; adjusted p-value=$2.94\ e^{-06}$).

Confirmatory GSEA of the I_a interactome indicated that FYN and IFITM3 biological networks were significantly enriched in several COVID-19 datasets containing SARS-CoV-2 upregulated genes (Supplementary Materials 3; adjusted p-value<0.05). Table 1 presents the 10 first (by order of adjusted p-value) entries of significantly enriched human COVID-19 datasets, out of a total of 447.

Finally, we include significantly enriched FYN signatures from entorhinal cortex neurons containing neurofibrillary tangles and hippocampal neurons from our previous study[5] for comparisons (Supplementary Materials $4_{a\text{-}d}$). Notably, the "cytokine-mediated signalling pathway" GO term, containing both FYN and IFITM3,was significantly enriched in both I_a and our previous study (FDR<0.05).

4. Discussion

In our study, we explored FYN and IFITM3's expression patterns and networks in AD vs. COVID-19 using a meta-analytical approach. We extracted FYN/IFITM3's putative interactome from a published consensus gene module in AD. GSEA revealed that common networks involving both FYN and IFITM3 mediate cytokine signaling, and immune processes as significantly enriched biological pathways and ontologies. Furthermore, FYN/IFITM3's interactome was significantly enriched in several human ex vivo and in vitro COVID-19 datasets. Differential gene expression analysis of bulk RNA-seq data indicated that IFITM3 was differentially expressed in non-neuronal cells (glia and endothelial cells) in the temporal cortex, the prefrontal cortex, the

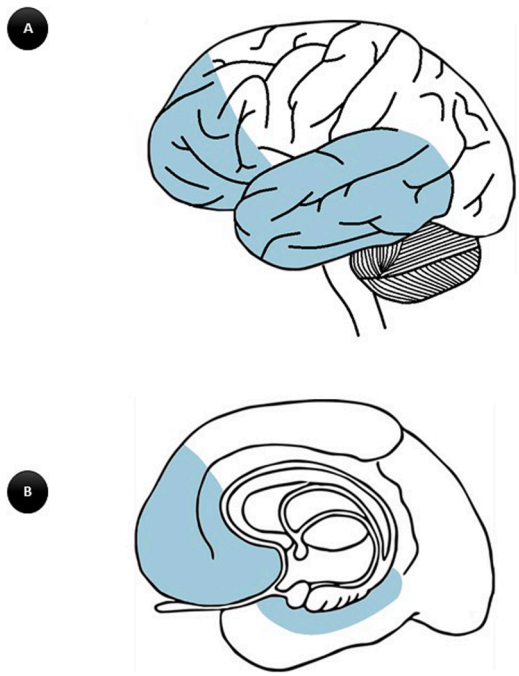

Fig. 2. Schematic representation of central nervous sites associated with the overexpression of FYN and IFITM3, i.e. the prefrontal cortex, the temporal cortex and the parahippocampal gyrus.

superior parietal lobe and the parahippocampal gyrus. FYN was differentially expressed in glial, endothelial cells and neurons in the entorhinal cortex, the prefrontal cortex and the superior frontal cortex, and its expression was characterized by high cell-to-cell variability.

Based on our findings, we will review the state-of-the art regarding the role of FYN and IFITM3 in the pathobiology of AD. Furthermore, we will discuss the role of immune processes in tau and Aβ processing, and its potential perturbations. Finally, we will synthesize our findings and the literature in a quasinfectious hypothesis on AD pathogenesis that may be applicable to the newly emerging COVID-19 associated dyscognitive disorder.

4.1. FYN / IFITM3 pathways in Alzheimer's disease and innate immunity

The recent identification of IFITM3, an established antiviral factor as a gamma secretase modulator[2] has provided deeper foundations to the antimicrobial protection hypothesis of AD pathogenesis[26]. Our focus on FYN is founded on its regulatory role in stabilizing IFITM3 in the membrane via phosphorylation of a critical motif[5], enabling its potential interaction with the gamma secretase complex. Notably, FYN has been shown to phosphorylate APP in AD neurons, a process that enhances its amyloidogenic processing[11]. Notably, oligomeric Aβ can upregulate FYN by interacting with PrP^{c}-mGluR5[27] inducing FYN-mediated phosphorylation of NMDA receptors[28]. Taken together, these studies indicate that FYN may regulate IFITM3 – gamma secretase – APP amyloidogenic processing via phosphorylation, with Aβ-PrP^{c}-mGluR5 cascades providing a feed-forward loop at the autocrine and paracrine milieu.

Aside from Aβ oligomerization, FYN has been shown to mediate tau aggregation in vivo[10], with process that involve both direct phosphorylation and activation of other kinases such as GSK-3β via NMDAR activation[27]. Notably, GSK-3β activation can lead to both FYN's activation and altered subcellular localization, either (a) in the plasma membrane, constituting a feedback loop GSK3-β-FYN—NMDAR-GSK-3β feedback loop) or in other subcellular compartments, including the nucleus [29,30].

Within the context of the antimicrobial protection hypothesis, both IFITM3 and FYN offer unique insight in the interactions between viral modulations of the transcriptome, innate immunity and AD pathology. We have previously shown that IFITM3 gene signatures enriched for IFN-I signaling represent overlapping pathways between AD and COVID-19[9]. IFITM3's role in SARS-CoV-2 has been shown to be structure dependent. Specifically, mutational alterations of its YxxΦ motif, FYN's phosphorylation target, may maintain the balance between SARS-CoV-2′s restriction or permission[31]. To our knowledge, we are the first to outline this functional relationship in the literature. Furthermore, mutational ablation of this site indicates that SARS-CoV-2-IFITM3 mediated entry may be mediated by hijacking of IFITM3's endocytic signal[32], a concept that has recently been corroborated[33].

FYN's role in cytokine signaling is upstream compared to IFITM3's, positively regulating (i.e. proinflammatory) signal transduction following immune receptor activation[34]. This modus operandi has been observed in microglia in animal models of Parkinson's disease[35]. Aside from its homeostatic role, FYN may be recruited in viral processes that require phosphorylation[36] and subversion of specific compartments such as autophagosomes[37]. In SARS-CoV-2, FYN has been reported as a differentially expressed gene via multiple omics approaches [7,6] albeit the consequences of its recruitment or perturbation have not been explored.

4.2. SARS-CoV-2-related neurocognitive deficits and their importance in AD pathobiology: from phenotypes to genes

COVID-19 has been lately recognized as a spectrum, one that includes both phenotypic and genomic overlap with neurodegenerative disease including Alzheimer's disease[38]. Among the more albeit easily underdiagnosed manifestations are neurocognitive symptoms, including memory defects, even among those patients recovered from mild disease [39].

A study reporting on a 3-month follow-up of patients recovering from COVID-19 uncovered microstructural alterations in the entorhinal cortex, associated with hyposmia, whereas memory loss was associated with hippocampal cortex remodeling[40]. As a previous hypothesis from our group[41], this concept has been subsequently validated by neuropathological studies that have detected SARS-CoV-2 in the olfactory cortex and the hippocampi [42,43]. Notably, a primate model of SARS-CoV-2 neurotropism has provided further corroboration to the our previously stated hypothesis[44].

Aside from the phenotypical overlap, genetic and epigenetic mechanisms have been shown to overlap between Alzheimer's disease and COVID-19. Independent meta-analyses of gene expression data have corroborated IFITM3 as a commonly perturbed gene in both conditions [45,46]. In the setting of SARS-CoV-2 infection, single nuclei RNA sequencing has identified FYN and IFITM3 perturbations spatially linked them to blood-barrier endothelial and glial cells[47]. A link between SARS-CoV-2 and the secretory phenotype of senescent endothelial cells has recently identified IFITM3 downregulation as a result of paracrine cellular communication[48]. Fisetin, a senolytic that has been shown to improve cognition and soluble Aβ burden[49], was used in the previous study to upregulate IFITM3 and reduce senescent cell burden[48].

Taken together, these studies outline known and interdependent connections of FYN and IFITM3 with innate immunity, viral lifecycles and the pathophysiological processes underlying AD.

4.3. Fitting SARS-CoV-2 in the antimicrobial protection hypothesis of Alzheimer's disease: a demi-infectious hypothesis

Several recent studies have bolstered the concepts underlying the antimicrobial protection hypothesis[26]. Soluble Aβ has been shown to function as an opsonin-like molecule, forming complexes with nucleic acids (both endogenous and viral) and enhancing their recognition by glial, while upregulating IFITM3-containing IFN-I signatures[50]. This

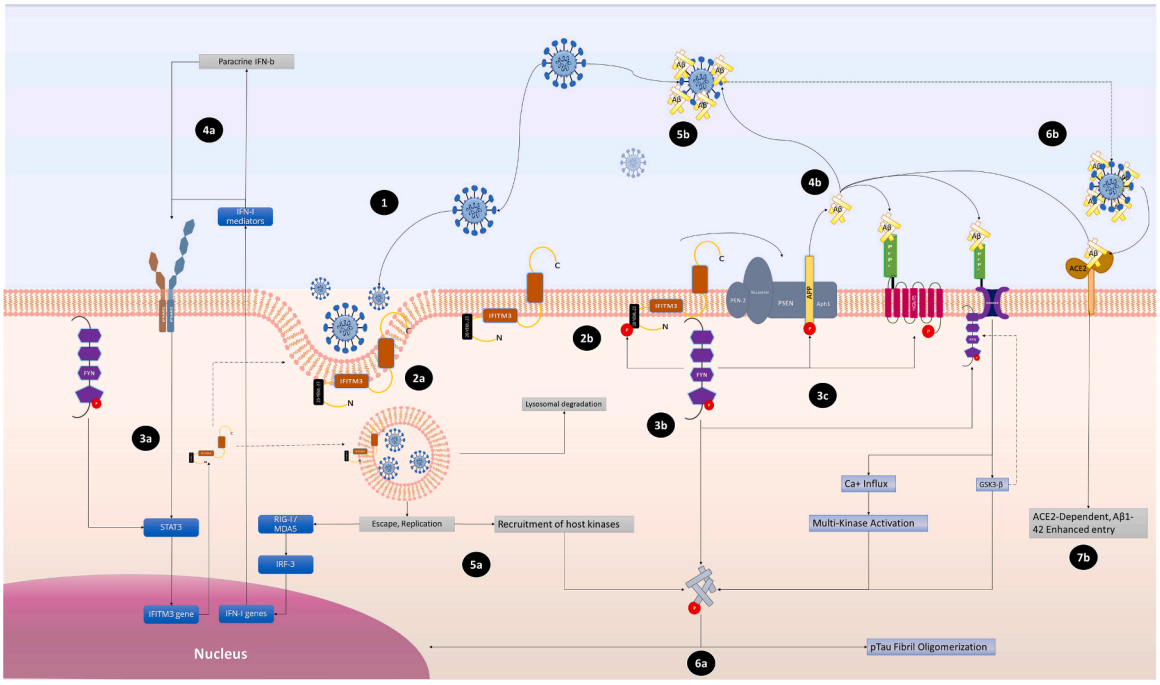

Fig. 3. IFITM3 and FYN in the setting of a deminfectious hypothesis of Alzheimer's disease pathogenesis, with SARS-CoV-2 as the pathogen. Lipid-raft mediated SARS-CoV-2 endocytosis in lipid rafts leads to a SARS-CoV-2 – IFITM3 interaction and endosomal sequestration (1). Depending on the phosphorylation status of IFITM3, the virus is either sequestered in IFITM3-enriched endosomes (2a) or retained in the surface (2b). Successful clearance of an invading virus via endosome – lysosome fusion leads to the recognition of viral fragments as DAMPs by PPRs such as RIG-I / MDA-5, which mediate upregulate IFN-I genes. IFN-I responses can function in an autocrine manner via their receptors (4a) and via STAT3 signaling, provide positive feedback of IFITM3's expression in IFN-I primed cells, or induce its expression in naïve cells. Notably, FYN has been shown to promote STAT3 signaling as part of proinflammatory priming of peripheral immune cells and microglia. IFITM3 mediates IRF3 autosomal degradation (not shown here), introducing a negative feedback loop in DAMP recognition. Viruses that escape endosomal sequestration or capitalize mutant IFITM3 isoforms, as has been shown in SARS-CoV-2, may escape and replicate productively (5a), recruiting host kinases. Tau hyperphosphorylations may occur as a result of viral processes, culminating in toxicity and oligomerization (6a). In the alternative scenario of IFITM3 phosphorylation, the latter becomes stabilized on the membrane, priming gamma secretase (2b). Concomitant phosphorylation of APP by FYN completes amyloidogenic priming (3b), and Aβ oligomer release in the paracrine milieu (4b). Isoform-specific interactions between Aβ oligomers and SARS-CoV-2's S1 have been reported, and may alter its infectivity; $A\beta_{1-42}$ in particular has been shown to promote rather than abrogate S1-ACE2 interactions (5b), and facilitate canonical SARS-CoV-2 entry (6b). Autocrine Aβ-PrP^{c} interactions provide further feedback to FYN via phosphorylation targets such as mGluR5 (3c). Physical interactions between FYN and NMDAR link Aβ and tau pathology via the activation of multiple kinases (indirectly, following calcium influx), the direct activation of GSK-3b, or FYN-mediated tau oligomerization.

non-specific mechanism of recognizing danger-associated molecular patterns (DAMPs) represents a sentinel aspect of innate immunity that is fundamentally perturbed in the setting of AD[51]. DAMP dysregulation in AD furthermore extends to other pattern-recognition receptors (PRR), such retinoic acid-inducible gene-I (RIG-1), and feeds back to Aβ production[52].

Recognition of invading viruses, including SARS-CoV-2, by RIG-I/MDA5 is followed by an IRF3 mediated induction of IFN-β. Autocrine and paracrine IFN-β induction would upregulate IFITM3 and enhance its trafficking in endosomes and the membrane[53]. While this is physiologically this represents an antiviral mechanism, SARS-CoV-2′s ORF3a has been shown to block lysosomal fusion and may thus escape into the cytosol [54,55]. furthermore, FYN-mediated enhancement of STAT3 cascades would enhance this feedback loop and maintain proinflammatory signaling[56]. Up to this point, viral invasion, global and cell-level IFN-I signaling represents a common niche between AD and COVID-19 [50,57]. FYN dysregulation at this point may contribute to global IFN-I perturbations via proinflammatory signaling[34] in peripheral immune cells [58,59]. Alternatively, FYN has been shown to enhance neuroinflammation in AD, Parkinson's disease dementia (PDD) and Dementia with Lewy bodies (DLB) by maintaining proinflammatory signaling microglia, associated with increased Aβ and pTau and dysproteostasis[60]. This paradigm of peripheral and CNS immune perturbations in AD has been recently outlined in an extended IFITM3 gene signature including OAS1, and overlapping with transcriptome perturbations in COVID-19[61]. Beyond SARS-CoV-2, interferon-related genes such as OAS1[62] and IFITM3[2] and downstream signal modulators such as FYN[34] are key players in both AD and susceptibility to wide variety of such as HIV-1 [63], WNV[64] and DENV[65].

IFITM3's recent addition as a gamma secretase modulator[2] further raises the question of context, in its interaction with a HSV-1[66] and HSV-1′s rapid seeding by Aβ as a potentially protective mechanism[67]. Notably, in silico analyses have indicated that the previously mentioned model can be realized by SARS-CoV-2; it's S1 has high affinity for soluble Aβ and tau [68]. This novel interaction has been shown to be isoform specific, with $A\beta_{1-42}$ enhancing rather abrogating infectivity by modulating ACE2-S1 interactions[69].

Within this context, FYN dysregulation can link Aβ accumulation, Tau hyperphosphorylation and innate immunity in the setting of COVID-19. SARS-CoV-2 infection experiments with brain organoids have recently revealed that neuroinvasion is followed by altered neuronal distribution of hyperphosphorylated Tau[70]. Furthermore, data from SARS-CoV-2 infectomics have consistently outlined the induction of the tau kinase pathway and impaired autophagy following SARS-CoV-2 neuroinvasion[71]. Via both direct and indirect mechanisms, FYN perturbations could account for SARS-CoV-2 -induced tau aggregation[10]. FYN-mediated stabilization of IFITM3 on the membrane would serve to prime its interactions with gamma secretase[2] along with amyloidogenic APP phosphorylation[11], and abort viral entry via endosomal escape[31]. FYN-enhanced Aβ-autocrine signaling would promote tau

Table 1
Significantly enriched COVID-19 datasets.

Index	Name	P-value	Adjusted p-value
1	500 genes up-regulated by SARS-CoV-2 in human Organoids cells from GSE154613	1.001e-19	4.474e-17
2	Top 500 upregulated genes for SARS-CoV-2 infection in human sclera from GSE164073	1.477e-18	3.301e-16
3	Top 500 up genes for SARS-CoV-2 infection 48 hpi in human alveolar organoids for GSE152586	9.499e-18	1.061e-15
4	500 genes up-regulated by SARS-CoV-2 in human Lung Organoid cells at 24 hpi from GSE148697	6.699e-17	4.991e-15
5	Top 500 upregulated genes for SARS-CoV-2 infection in human lung organoids from GSE148697	6.699e-17	4.991e-15
6	500 genes up-regulated by SARS-CoV-2 in human Calu3 cells at 24 h from GSE148729 s1 polyA	1.061e-15	5.926e-14
7	500 top upregulated genes from SARS-CoV-2 infection at 72 HPI from GSE157852	1.995e-14	9.189e-13
8	SARS-CoV perturbation; 402 Up Genes from GEN3VA; Human bronchial epithelial 2B4 cells; Accession: GSE17400 Platform: GPL570; Entry 6	2.469e-14	9.647e-13
9	500 genes up-regulated by SARS-CoV-2 in human pancreatic organoids from GSE151803	3.327e-13	7.435e-12
10	500 genes up-regulated by SARS-CoV-2 in human pancreatic organoids from GSE151803	3.327e-13	7.435e-12

toxicity [28,60], and prepare an Aβ-enriched antiviral milieu that would intercept incoming viruses via opsonin-like interactions [71] that prime microglia and provide feedback to IFN-I and IFITM3 specifically[50].

How would this local immune stimulation translate into a network-expanding neurodegenerative disease? Brain endothelial cells and glia cells could be the prime culprits in SARS-CoV-2, a concept supported by both our current findings and others[72]. Neuroinvasion and trans-olfactory spread of SARS-CoV-2 to the hippocampi, supported by the clinicoradiological course of COVID-19 dyscognitive syndromes[40] and neuropathology [42,44] would then represent the clinically evident stage. As we have previously proposed, at the neuroinvasion stage, a primary hub such as the olfactory cortex could inform its distal connectome of an invader and glia via the efflux of Aβ and Tau[9]. In the setting of the neuroimmune hypothesis, both these molecules would have to be reconsidered as a novel class of immune mediator that functions as a "blind" guardian in innate immunity signaling[71], following DAMP recognition by PPRs. A comprehensive model is presented in Figure 3.

4.4. Limitations and context

Our current work should be considered within its limitations and context. Currently, no study has evaluated the longitudinal development of pathologically proven AD following exposure to COVID-19, and such a study is required to verify both our findings and our hypotheses.

A study that combines Another important limitation is that other AD-related genes are also implicated in COVID-19 infection, such as APOE [73] and ACE2[74]. While this implication further bolsters SARS-CoV-2′s potential role in AD pathogenesis, genetic variability and gene interactions between these genes and the FYN/IFITM3 switch should be studied in detail, in order to elucidate their mechanistic effects. Cell-to-cell variability, as it arose in our scRNA-seq scrutiny of AD vs. Controls data, is another confounding factor in interpretating our results. This variability indicates the departure from the concept of tissues as homogenates, and the recognition of their heterogeneity. This paradigm has been acknowledge in microglia, along with is potential contribution to AD[75], and may furthermore indicate the need to account for senescence and the PASP phenotype in general in tissues[48].

Another limitation in our study is that the differential expression data from Morabito et al[13] only provide the p-values for pairwise comparisons and not raw data. As such, the direction of differential expression is derived from studying the plots, rather than the output. As both FYN and IFITM3 are a priori selected in a consensus gene module, this limitation does not affect their implication in AD or the validity of the interactome.

5. Conclusions

FYN represents a known modulator of IFITM3's stabilization in the membrane and its endocytic pathway. In this analysis, we determine the function, cell-, CNS-site specific context of their interaction within cytokine signaling in AD. Furthermore, we are the first to propose a deminfectious model of AD pathogenesis that can be initiated by SARS-CoV-2, building upon our previous research and contemporary knowledge. Future studies should aim to link clinical, radiological and neuropathological findings with genomics and basic science in order to fully characterize both the emerging entity of COVID-19 dyscognitive syndromes, and their implication in AD.

Author contributions

Conceptualization, analyses, first draft, final draft, revisions: GDV. Final draft, confirmation of analyses: TM, MB; Revisions, conceptualization: KAK. All authors have read and approved of the final draft. TM and MB contributed equally.

Declaration of Competing Interest

None declared.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.dscb.2021.100022.

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