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No Increased Detection of Nucleic Acids of CNS-related Viruses in the Brains of Patients with Schizophrenia, Bipolar Disorder, and Autism Spectrum Disorder

Shishi Min[1–4], Michael J. Gandal[5], Richard F. Kopp[3], ChunyuLiu[2,3,6,], and Chao Chen[*,2,7,,8,]

[1]Department of Neurology, Xiangya Hospital, Central South University, Changsha, Hunan, China; [2]Center for Medical Genetics & Hunan Key Laboratory of Medical Genetics, School of Life Sciences, and Department of Psychiatry, The Second Xiangya Hospital, Central South University, Changsha, Hunan, China; [3]Department of Psychiatry, SUNY Upstate Medical University, Syracuse, NY, USA; [4]National Clinical Research Center for Geriatric Disorders, Xiangya Hospital, Central South University, Changsha, Hunan, China; [5]Lifespan Brain Institute at Penn Medicine and The Children's Hospital of Philadelphia, Department of Psychiatry, University of Pennsylvania, Philadelphia, PA, USA; [6]School of Psychology, Shaanxi Normal University, Xi'an, Shaanxi, China; [7]Hunan Key Laboratory of Animal Models for Human Diseases, Central South University, Changsha, Hunan, China; [8]National Clinical Research Center on Mental Disorders, The Second Xiangya Hospital, Central South University, Changsha, Hunan, China

[*]To whom correspondence should be addressed; Center for Medical Genetics & Hunan Key Laboratory of Medical Genetics, School of Life Sciences, and Department of Psychiatry, The Second Xiangya Hospital, Central South University, 410008 Changsha, Hunan, China; tel: +86 18874114280; e-mail: chenchao@sklmg.edu.cn

Background and Hypothesis **Viral infections are increasingly recognized in the etiology of psychiatric disorders based on epidemiological and serological studies. Few studies have analyzed viruses directly within the brain and no comprehensive investigation of viral infection within diseased brains has been completed. This study aims to determine whether viral infection in brain tissues is a risk factor for 3 major psychiatric disorders, including schizophrenia, bipolar disorder, and autism spectrum disorder.** ***Study Design*** **This study directly evaluated the presence of viral DNA or RNA in 1569 brains of patients and controls using whole-genome sequencing and RNA sequencing data with 4 independent cohorts. The PathSeq tool was used to identify known human viruses in the genome and transcriptome of patients and controls.** ***Study Results*** **A variety of DNA and RNA viruses related to the central nervous system were detected in the brains of patients with major psychiatric disorders, including viruses belonging to Herpesviridae, Polyomaviridae, Retroviridae, Flaviviridae, Parvoviridae, and Adenoviridae. However, no consistent significant differences were found between patients and controls in terms of types and amount of virus detected at both DNA and RNA levels.** ***Conclusions*** **The findings of this study do not suggest an association between viral infection in postmortem brains and major psychiatric disorders.**

Key words: viral infection/brain tissue/major psychiatric disorders/whole-genome sequencing/RNA sequencing

Introduction

Genetic and environmental factors both play important roles in the etiology of major psychiatric disorders, such as schizophrenia (SCZ), bipolar disorder (BD), and autism spectrum disorder (ASD). Viral infection, as one of the environmental risk factors, has been found to be related to psychiatric disorders.[1–3] Most of the evidence for an association between viral infection and psychiatric disease comes from epidemiological and serological studies using blood samples.[4–9] For example, the levels of antibodies to Epstein–Barr virus (EBV) virions were elevated in the blood of patients with SCZ as compared to the controls.[4] A meta-analysis of SCZ data revealed association with human herpesvirus 2 (HHV-2) and Borna disease virus (BDV).[5] The cytomegalovirus (CMV)-positive immunoglobulin (Ig) G status was reported to be significantly elevated in BD patients.[6] Human immunodeficiency virus (HIV)-exposed uninfected children had a more than 2-fold increase in odds for ASD.[7] The positive findings of these epidemiological and serological studies supported the association between viral infection and mental illness, suggesting that viral infection may be involved in the pathogenesis of major psychiatric disorders.

In this study, known neurotropic viruses[10] or other viruses that are not considered neurotropic but can spread into the central nervous system (CNS) were defined as CNS-related viruses. Neurotropic viruses are those that specifically target the CNS.[10] In addition, some viruses do not infect specific cells in the CNS and are thus not considered neurotropic strictly speaking, but may enter the CNS as a result of high plasma viremia (such as a non-neurotropic strain of influenza,[11] or parvovirus B19[12]) or via infected peripheral blood mononuclear cells (such as

HHV6[13] or EBV[14]). Viruses can invade and access the well-protected CNS (reviewed by Koyuncu et al. 2013).[15] For example, most alpha-herpesviruses, including herpes simplex type-1 (HSV-1), HSV-2, and varicella-zoster virus (VZV), enter the CNS by the invasion of sensory nerve endings.[16] Some virus particles, such as Hepatitis C virus (HCV),[17] John Cunningham virus (JCV),[18] and EBV,[14] in the circulatory system can reach and infect brain microvascular endothelial cells and leading to the disruption of blood-brain barrier (BBB) integrity and finally infiltration into CNS. HSV-1 and HSV-2 are active in the CNS and have been linked with a variety of CNS diseases, including multiple sclerosis, epilepsy, and encephalitis.[19–21] Two studies by Readhead et al.[22] and Eimer et al.[23] have reported the important role of HSV-1 and HHV-6 in the pathogenesis of Alzheimer's disease, respectively. However, the association between viral infection in CNS and major psychiatric disorders is unclear.

The presence of viruses in genomes and transcriptomes has been explored in small studies of postmortem brains with inconsistent results. Early studies used polymerase chain reaction (PCR) techniques to directly detect viral genomes in brains. Most of these studies failed to find viral sequences in patients with major psychiatric disorders,[24–32] with a few exceptions.[33–35] Moreover, the presence of viruses did not always link viruses to disease. For example, a study by Hobbs et al. investigated the presence of the 2 parvoviruses (B19, AAV2) in the postmortem dorsolateral prefrontal cortex (DLPFC) by utilizing nested PCR and DNA sequencing with 35 patients with SCZ, 34 patients with BD, and 35 unaffected controls.[35] They found no statistical differences in the presence of B19 or AAV2 among groups. However, Gandal et al. identified up-regulated interferon (IFN) response in postmortem brains of patients with ASD and SCZ using large transcriptome data.[36] Type I interferons have been reported to be very important for the host's defense against viruses.[37] The up-regulated IFN response hints at the potential association between viral perturbation and major psychiatric disorders. Altered immune response in SCZ and ASD makes it reasonable to hypothesize that infectious agents trigger or aggravate the disease. The findings of these studies suggest that further investigation is needed, particularly with large brain samples, to explore the role of viral infections in mental illness.

The emergence of high-throughput next generation sequencing (NGS) technology makes it possible to detect viruses in large samples.[38,39] Known or annotated viruses can be detected in NGS data by sequence alignment. Moreover, NGS data makes it possible to simultaneously detect multiple types of viruses, a big advantage over PCR-based methods. In the present study, the hypothesis was that CNS-related viruses have excessive presence in the brains of patients with major psychiatric disorders and could be detected by NGS. Genomic and transcriptomic data from postmortem brains were used to systematically search for evidence of CNS-related viruses in SCZ, BD, ASD, and controls. The combination of genomic and transcriptomic data enables the comprehensive discovery of potential viruses, including DNA and RNA viruses. DNA and RNA viruses were closely examined separately. Whole genome sequencing (WGS) data from the BrainGVEX study and RNA sequencing (RNA-seq) datasets from the BrainGVEX, BipSeq, UCLA-ASD, and CommonMind studies were screened for DNA and RNA viruses, using the computational viral detection pipeline called PathSeq.[40] The detection of viral sequences in the brains of major psychiatric disorders is critical to determine whether the presence of the virus in the brain samples is associated with major psychiatric disorders and understand the role of viral infection.

Methods

Whole Genome Sequencing Data

WGS data (synapse ID: syn21578890) used in this study were obtained from the BrainGVEX study of the PsychENCODE project.[36] The data were generated from the frontal cortex (Brodmann's area (BA) 46/9) of the postmortem brains of 269 individuals, including 45 patients with SCZ, 26 patients with BD, and 198 non-psychiatric controls (table 1). The average depth of WGS data (104 million 150 bp paired end reads on average) was 4.83 per individual, with a mean of 48.5% of the genome with at least 5× coverage.

RNA Sequencing Datasets

RNA-seq datasets from 1300 brain samples used in this study were available from the BrainGVEX, BipSeq, UCLA-ASD, and CommonMind studies of the PsychENCODE project. The full details of these studies and ethical approval have been described in the original articles.[36,41–43] RNA integrity of all postmortem samples was assessed and samples with RNA Integrity Number lower than 5.5 were excluded.[36,41–43] For the BrainGVEX study, RNA-seq data (synapse ID: syn3270015) was generated from postmortem prefrontal cortex samples (BA46) with 419 individuals, including 93 patients with SCZ, 72 patients with BD, and 254 non-psychiatric controls. Total RNA was isolated using the miRNeasy Kit (Qiagen) according to the manufacturer's instructions. RNA integrity of samples was assessed with Agilent Technologies RNA 600 nano kit.[36] The paired end 100 bp library was sequenced on an Illumina HiSeq2000 with average read depth >70 million reads. For the BipSeq study, RNA-seq data (synapse ID: syn8403872) was generated from DLPFC approximating BA46/9, including 69 patients with BD. Total RNA was extracted from ~100 mg of tissue using the RNeasy kit (Qiagen) according to the manufacturer's protocol.[41] The library was sequenced using an Illumina HiSeq2000 with paired end 2 × 100bp reads with an average of 85 million reads. For the UCLA-ASD study, RNA-seq data (synapse ID: syn4587615) was generated from 96 unique

Table 1. Sample Demographics

Data	Cohort	Diagnosis	Brain Regions	N	Age at Death: Mean (SD)	PMI (h) Mean (SD)	Male (Female)
RNA-seq	BrainGVEX	SCZ	PFC	93	42.5 (10.4)	40.4 (26.7)	66 (27)
	(*n* = 419)	BD	PFC	72	43.9 (11.6)	37.0 (18.8)	37 (35)
		Control	PFC	254	69.6 (18.3)	10.1 (13.6)	163 (91)
	BipSeq	BD	DLPFC	69	46.0 (14.3)	31.4 (17.8)	38 (31)
	(*n* = 69)						
	CMC	SCZ	DLPFC	261	68.8 (16.6)	20.8 (13.5)	166 (95)
	(*n* = 589)	BD	DLPFC	46	50.8 (15.3)	19.1 (7.7)	25 (21)
		Control	DLPFC	282	65.5 (19.3)	13.6 (7.8)	160 (122)
	UCLA-ASD[a]	ASD	CB, PFC, TC	47	25.3 (17.3)	22.3 (9.7)	38 (9)
	(*n* = 96)	Control	CB, PFC, TC	49	26.1 (16.6)	19.4 (7.6)	40 (9)
WGS	BrainGVEX	SCZ	PFC	45	42.6 (11.4)	47.5 (33.3)	30 (15)
	(*n* = 269)	BD	PFC	26	44.2 (12.0)	40.4 (22.1)	13 (13)
		Control	PFC	198	76.2 (14.2)	6.1 (10.4)	124 (74)

Note: PMI, postmortem interval; SCZ, schizophrenia; BD, bipolar disorder; ASD, autism spectrum disorder; PFC, prefrontal cortex; DLPFC, dorsolateral prefrontal cortex; CB, cerebellum; TC, temporal cortex.
[a]Up to 3 brain regions per individual from the UCLA-ASD study were used, the total number of samples for ASD and non-ASD controls equal to 223.

individuals, including 47 patients with ASD and 49 age-matched non-ASD controls, with up to 3 different regions, across the prefrontal cortex (BA9/46), temporal cortex (BA41/42/22), and cerebellum (vermis). The total number of specimens for ASD and non-ASD controls was 223. Approximately 50–100 mg of tissue across the cortical region of interest was isolated from each sample using the miRNeasy kit with no modifications (Qiagen). For each RNA sample, RNA quality was quantified using the RIN on an Agilent Bioanalyzer.[42] Libraries were sequenced on an Illumina HiSeq2500 instrument using high output mode with standard chemistry and protocols for 50 bp paired end reads to achieve a target depth of 70 million reads. Sequence files downloaded from the BrainGVEX, BipSeq, and UCLA-ASD studies were read in fastq format. For the CommonMind study, RNA-seq data (synapse ID: syn2759798) from human postmortem DLPFC brain samples were obtained from 589 subjects, including 261 patients with SCZ, 46 patients with BD, and 282 controls. Total RNA was extracted from 50 mg of homogenized dorsolateral prefrontal cortex tissue using the RNeasy kit. The remaining samples had a mean RIN of 7.7.[43] The RNA-Seq library was sequenced using an Illumina HiSeq 2500 with 100 bp paired-end reads with an average of 85 million reads. Sequence files downloaded from the CommonMind study were read in bam format, including mapped and unmapped files. Bam files were merged and converted to fastq format using samtool.[44]

Screening for CNS-related Viruses using WGS and RNA-seq Datasets

The GATK PathSeq tool[40] of Broad Institute was used to detect viral sequences in WGS and RNA-seq data. FastQC (v0.10.1) was used to check the quality of the raw reads in WGS and RNA-seq data first. Before running the "PathSeq" pipeline on these data, the fastq files of RNA-Seq data and WGS data were converted to unmapped BAM (uBAM) format files using the "FastqToSam" command in Picard tool (v1.119) (https://broadinstitute.github.io/picard/). Afterward, the uBAM format files were passed into the "PathSeqPipelineSpark" command supported in the PathSeq tool. Processing sequencing data with the PathSeq tool involves a series of steps including preprocessing and quality control, filtering host sequences, and identifying viruses. The remaining non-host reads were aligned to reference of a microbial genome using the BWA-MEM algorithm and classified. The pre-built reference (https://console.cloud.google.com/storage/browser/gcp-public-data--broad-references) of PathSeq was downloaded and used. The total number of microorganisms in the reference is approximately 25 000, including 118 human viruses. To capture as much of viruses as possible while maintaining the accuracy and stringency of virus alignment, a cutoff value of 70% alignment identity score was used for the classification of viral reads. Each detected virus was scored. The PathSeq Score based on the number of reads aligned with viral references indicates the relative abundance of the virus present in the sample. These scores are defined by the developer to quantify microbe abundance: "Alignments with sufficient identity score (70% of the read length in this study) estimate the read counts and relative abundance of microbes at each level of the taxonomic tree (eg, strain, species, genus, family, etc.) in the sample. Reads with the best alignment will add a score of 1 to the species or strain. For reads maps to more than one organism, a score of 1/(number of the mapped organism) will be added to each organism. Scores are totaled for

Table 2. Statistical Analysis of Viral Detection Between SCZ, BD, and Controls Based on DNA-seq

	Individuals (N)	Adenoviridae	Herpesviridae				Polyomaviridae		Parvoviridae			Papillomaviridae
		HAdV-C	EBV	CMV	HHV-6A	HHV-6B	JCV	HPyV6	AAV	B19V	V9	HPV
SCZ	45	1	0	0	3	4	0	0	0	0	0	0
BD	26	1	0	0	1	0	0	0	1	1	0	0
Controls	198	0	1	1	11	12	1	1	1	0	2	1
Fisher test SCZ vs. Controls (*P*-value)		.185	1	1	.728	.507	1	1	1	—	1	1
Fisher test BD vs. Controls (*P*-value)		.116	1	1	1	.369	1	1	.219	.116	1	1

Note: HAdV-C, human adenovirus species C; EBV, Epstein-Barr virus; CMV, cytomegalovirus; HHV-6A, human beta herpesvirus 6A; HHV-6B, human beta herpesvirus 6B; JCV, John Cunningham virus; HPyV6, human polyomavirus 6; AAV, adeno associated virus; B19V, human parvovirus B19; V9, human erythrovirus V9; HPV, human papillomavirus.

each taxon by summing the scores across all reads and the scores of any descendent taxa."[40] Scores of detected viruses from the PathSeq tool were then imported into R (https://cran.r-project.org/) for visualization. The available sources of all datasets and software included and used in this study are described in table S1.

Statistical Analysis of PathSeq Results

The Fisher's exact test was performed using R to determine whether any significant differences exist in the frequency of viral detection between major psychiatric disorders and controls.

Results

No Significant Difference in the Low Frequency of CNS-related DNA Viruses Between SCZ (nor BD) and Controls in Genomic DNA Data

Eleven CNS-related DNA viruses representing 5 viral families were detected in brain samples of patients with SCZ, BD, and controls from the BrainGVEX cohort (table 2). The detection frequency of viruses ranged from 0.5% (1/198) to 8.9% (4/45) in the 3 groups. No significant differences were found in the detection frequencies of any DNA virus between SCZ, or BD and controls based on DNA-seq (table 2). The PathSeq score represents the amount evidence of the presence of a virus. PathSeq scores of those viruses detected in cases and controls ranged from 0.5 to 2079 (figure S1). Most scores (86%, 37/43) were low, falling between 0 and 4. The PathSeq Score of greater than 0 indicates positive, and a higher score indicates stronger evidence of a virus is present in the individuals based on the number of reads that aligned to viral references. Viruses with scores equal to or greater than 20, including HHV-6A, HHV-6B, CMV, JCV, were only detected in the control group.

No Consistent Difference in the Detection of Viruses Between SCZ (nor BD) and Controls in Multiple Transcriptome Data Sets

To further explore the association between viral infection and major psychiatric disorders, RNA-seq data available from the BrainGVEX and BipSeq studies (table 1) were combined and used to detect viruses based on RNA expression. Eight CNS-related viruses (6 DNA viruses and 2 RNA viruses) representing 4 viral families were detected in the brains of patients and controls (table 3). Similar to DNA-seq level, PathSeq scores were low. PathSeq scores for these viruses ranged from 0.67 to 56, with most scores (94.4%, 51/54) falling between 0 and 10 (figure S2). The frequency of detection for viruses ranged from 0.4% (1/254) to 11% (28/254) in 3 groups. Except for Human adenovirus species C (HAdV-C) which was significantly increased in control brains, there were no significant

differences in the detection frequencies for other viruses between cases, and controls (table 3).

RNA-seq data from the CommonMind study were used as a replication cohort. The dataset included 589 individuals with SCZ, BD, and controls (table 1). Similar to the results of the BrainGEVX and BipSeq cohort, PathSeq analysis demonstrated a low frequency of CNS-related virus detection, ranging from 0.35% (1/282) to 2.48% (7/282), with PathSeq scores varying from 0.5 to 53 (figure S3). Most (95.9%, 47/49) of the scores were between 0 and 10 (table S2; figure S3). Also, the detection frequency of HAdV-C did not increase in the brains of controls in replicated cohorts compared with cases.

No Abnormal Levels of CNS-related Viruses in ASD Based on RNA-seq from Multiple Brain Regions

RNA-seq data from the UCLA-ASD study were available from multiple brain regions for 223 total samples (table 1). Eleven CNS-related viruses (8 DNA and 3 RNA viruses) representing 8 virus families were detected in 3 brain regions from patients with ASD and controls (table 4). PathSeq scores for these viruses ranged from 0.1 to 28.5. Most scores (90%, 54/60) were low and between 0 and 10 (figure S4). The detection frequency of viruses ranged from 2.4% (1/41) to 23.3% (7/30). For each of the 3 brain regions, the detection frequency of any of the viruses was not significantly different between patients and controls.

Discussion

In this study, WGS data and RNA-seq data were screened for CNS-related viruses in 1569 brains of patients with 3 major psychiatric disorders (SCZ, BD, ASD) and controls. CNS-related DNA and RNA viruses were detected in the brain tissues of patients and controls, including viruses belonging to Herpesviridae (EBV, CMV, HHV-6A, HHV-6B, HHV7), Polyomaviridae (JCV, HPyV6), Retroviridae (HIV), Flaviviridae (GBV-C, HCV), Parvoviridae (B19V, V9, AAV), and Adenoviridae (HAdV-C). However, no significant increases in the detection frequency of viruses in brains of patients were detected compared with brains of controls based on both DNA-seq and RNA-seq data. Although previous epidemiological and serological studies using blood samples have reported possible associations between viral infection and psychiatric disorders,[4–8,45–47] our study does not support an association between viral infection in postmortem brains and major psychiatric disorders.

Large sample sizes can help detect rare viruses and determine if there is indeed a difference in viral infection between patients and controls. The cohorts combined in this study contained the largest brain samples assembled to date to evaluate the association between viral infection and psychiatric disorders. A study of virus discovery on postmortem frontal cortex of SCZ and controls using NGS data by Tomasik et al. only detected one CNS-related virus, Hepatitis C.[48] Their sample size was relatively small with 15 patients and 15 controls, which may have provided limited power to detect viruses. In our study, a large sample of patients with SCZ was used, enabling detection of multiple CNS-related viruses that were not detected in a previous study,[48] eg, HHV-6A, HHV-6B, HHV-7, CMV, EBV, HCV. Although no statistical difference was found in the frequency of viruses between patients and controls in the current study, these results provide a summary of the types of latent CNS-related viruses present in brains of patients with these psychiatric disorders. One limitation of this study is that the number of patients with SCZ and BD was lower than that of healthy controls. The sample size of ASD and patients with WGS data is also relatively small. Therefore, the results of this study need to be replicated in a larger and independent cohort in the future.

The difference in detection frequency of HHV-6 in BD appears to be related to difference in brain regions analyzed. In the current study, all instances of HHV-6 detection occurred in BD from only 1 patient at the DNA level (out of 26) (table 2), and 1 patient at the RNA level (out of 141) (table 3) both with low Pathseq scores of 1.0. No HHV-6 was detected in the replicate cohort. In contrast, a study by Prusty et al. using postmortem posterior

Table 3. Statistical Analysis of viral Detection Between SCZ, BD, and Controls Based on RNA-seq

		Adenoviridae	Herpesviridae				Polyomaviridae	Flaviviridae	
	Individuals (N)	HAdV-C[1]	EBV[a]	CMV[a]	HHV-6A[a]	`HHV-6B[a]	JCV[a]	GBV-C[b]	HCV[b]
SCZ	93	0	1	0	0	1	0	0	2
BD	141	4	3	0	0	1	0	1	0
Controls	254	28	5	2	1	1	1	3	0
Fisher test SCZ vs. Controls (*P*-value)		.0001	1	1	1	.468	1	0.567	.071
Fisher test BD vs. Controls (*P*-value)		.0023	1	1	1	1	1	1	—

[a]DNA viruses: Abbreviations are explained in the first footnote to table 2.
[b]RNA viruses: GBV-C, Hepatitis G virus; HCV; Hepatitis C virus; HIV, Human immunodeficiency virus; HERV-K113, Human endogenous retrovirus K113.

Table 4. Statistical Analysis of Viral RNA Detection Between ASD and Controls in 3 Brain Regions

	Individuals (N)	Adenoviridae	Herpesviridae			Polyomaviridae	Parvoviridae	Papilloma-viridae	Hepadna-viridae	Retroviridae		Flavi-viridae
		HAdV-C[a]	EBV[a]	HHV-6B[a]	HHV-7[a]	HPyV6[a]	B19V[a]	HPV[a]	HBV[a]	HIV[b]	HERV-K113[b]	GBV-C[b]
ASD_FC	39	0	1	—	—	—	2	—	—	6	1	0
Control_FC	38	1	2	—	—	—	0	—	—	4	2	2
ASD_TC	38	0	3	—	—	—	0	1	1	4	1	0
Control_TC	30	1	3	—	—	—	1	0	0	7	0	2
ASD_CB	37	—	0	1	0	1	—	—	1	2	0	1
Control_CB	41	—	1	0	1	0	—	—	0	3	3	1
Fisher test ASD_FC vs. Controls_FC (*P*-value=)		.493	1	—	—	—	.493	—	—	.737	.615	.24
Fisher test ASD_TC vs. Controls_TC (*P*-value=)		.441	1	—	—	—	.441	1	1	.194	1	.191
Fisher test ASD_CB vs. Controls_CB (*P*-value=)		—	1	.474	1	.474	—	—	.474	1	.242	1

Note: FC, frontal cortex; TC, temporal cortex; CB, cerebellum. HBV, Hepatitis B virus; HIV, Human immunodeficiency virus; HERV-K113, Human endogenous retrovirus K113.
[a]DNA viruses: Abbreviations are explained in the first footnote to table 2.
[b]RNA viruses: Abbreviations are explained in the first footnote to table 3.

cerebellum reported that HHV-6 was detected more frequently ($P < .001$) in BD relative to controls, and that such infection may be associated with BD.[49] The DNA and RNA used in the current study were collected from the DLPFC. However, combined with the results of no difference in virus infection among all 3 brain regions for ASD patients, the question of whether viral infections are localized to specific regions of the brain remains to be explored in the future. DNA and RNA sequencing based on bulk brain tissues is a limitation of this study. There may be only small fractions of cells that are infected by the virus. Single-nucleus DNA sequencing or single-nucleus RNA sequencing technologies are powerful tools for detecting virus infections in individual cells and identifying cell type specific viral infections.[50,51]

The frequency of infection and Pathseq scores of detected viruses in brain tissue was low for most viruses and there could be several reasons. Similar to the PathSeq score distribution reported in a study showing HHV-6 detection with low PathSeq scores (mostly between 0 and 10) in postmortem brains of Alzheimer's disease patients both in the Mount Sinai Brain Bank (MSBB) and the Religious Orders Study/Memory and Aging Project (ROSMAP) cohorts.[39] One of the reasons could be that postmortem brain tissue samples used in this study may have hindered the detection of early infection. Viral infections during early neurodevelopmental processes may not be present or detected in postmortem brains because of the clearance of viral infection from the CNS.[52] This may be especially relevant for acute viral infections, characterized by the rapid production of virus and followed by clearance and resolution within days.[53] Moreover, if the clearance of acute infectious viruses is incomplete, lifelong persistent infections and a mechanism for prevention of viral replication and reactivation will be established in CNS.[52,53] During persistent infection, the virus usually maintains a low replication rate and is therefore difficult to detect. To determine the role of viral infections in the pathology of major psychiatric disorders, a study focusing on the developmental brain should be highly interesting for future research. However, it is important to note that the developmental brain tissues at the early stage of the disease are almost impossible to get. Viral detection using cerebrospinal fluid collected from living psychiatric patients may be a feasible approach.

A second reason may be associated with the observation that type I IFN is important for controlling viral replication and is necessary for continuing to reduce viral RNA levels.[52,54] The effect of interferon on host infection response is not only limited to acute antiviral response but also includes chronic antiviral response.[37] Type I IFN affects immune cells to enhance immune response, respond more effectively to viral infection, and improve memory response production for future viral challenges.[37] This also provides a possible explanation for why Gandal et al. found that IFN response modules were enriched in major psychiatric disorders,[36] while only

a small number of viral nucleic acid sequences were detected in the current study using the same postmortem brain tissues. It is worth noting that some of the low PathSeq scores for viruses may also be caused by contamination in sample preparations. In this study, the frequency of HIV detection in the UCLA-ASD cohort is unusually high, ranging from 5.4% to 23.3% (table 4). Actually, only 2 control samples showed high HIV PathSeq scores (13–28.5) consistently in all brain regions where data were available (table S3). In contrast, HIV appears to be randomly detected with low PathSeq scores (1–6) in only a subset of data for the other individuals.

In conclusion, this study directly examined the presence of CNS-related viral sequences in the brains of patients with psychiatric disorders and controls using NGS datasets from multiple sources. A survey of viral DNA and RNA present in brains of patients and controls was reported. However, the frequencies of these viral infections in postmortem brain were not associated with disease status. Results of this study do not support an association between CNS-related viruses and psychiatric disorders, though the possibility of transient acute viral infection followed by a complete clearance and an associated inflammatory response leading to these disorders cannot be ruled out.

Supplementary Material

Supplementary material is available at https://academic.oup.com/schizophreniabulletin/.

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