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RESEARCH ARTICLE

Interaction between *APOE4* and herpes simplex virus type 1 in Alzheimer's disease

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Abstract

Introduction: Numerous results suggest the implication of infectious agents in the onset of Alzheimer's disease (AD).

Methods: In the Bordeaux-3C prospective cohort, we assessed the impact of herpes simplex virus type 1 (HSV-1) infection on the incidence of AD according to apolipoprotein E (*APOE*) status, a genetic susceptibility factor. Cox models were performed to estimate the 10-year risk of AD associated with anti-HSV antibodies in 1037 participants according to *APOE4* status.

Results: Among *APOE4* carriers, subjects for whom the frequency of HSV-1 reactivation is supposed to be high, that is, immunoglobulin M (IgM) positive or elevated levels of IgG, had an increased risk of AD with adjusted hazard ratios (HRs) of 3.68 (1.08–12.55) and 3.28 (1.19–9.03), respectively. No significant association was found in *APOE4*-negative subjects.

Discussion: These results, in accordance with a solid pathophysiological rationale, suggest a role for HSV-1 in AD development among subjects with a genetic susceptibility factor, the APOE4 allele.

KEYWORDS

Alzheimer's disease, APOE4, dementia, genetic susceptibility, herpes virus, prevention

1 | BACKGROUND

Although hallmarks of Alzheimer's disease (AD) such as β-amyloid plaques, neurofibrillary tangles, and neuroinflammation are well known,[1] the triggers of these hallmarks are still under investigation. Several results suggest an implication of infectious agents in the onset of AD. Indeed, the production of Aβ may be a physiological antimicrobial response.[2–4] Moreover, several genetic risk factors of AD have been identified to regulate the immune or antiviral response.[5–7]

Among the infectious agents suspected, herpes simplex virus type 1 (HSV-1) is an interesting candidate.[8] After a first infection occurring mainly at a young age, HSV-1 remains in a latent form within a sensory ganglion, the trigeminal ganglion. Thereafter, reactivation of the virus is periodically triggered by various stimuli, possibly leading to severe symptoms such as encephalitis in immunocompromised persons. In immunocompetent persons, reactivation causes minor symptoms, such as cold sores or, most of the time, asymptomatic shedding of the virus, highlighting a relatively good control of the infection by the immune system. Nevertheless, in elderly people, this control can be weakened by immunosenescence, which could explain the late development of AD.[9,10] Taking advantage of this decrease in immune defenses, HSV-1 could then migrate to the temporal cortex (the area usually affected in HSV-1 encephalitis) and as a neurotropic virus, could travel from neuron to neuron to other regions of the brain. Once in the brain, the direct action of the virus and/or the inflammation against it could be responsible for the main hallmarks of AD.[11] In fact, numerous postmortem studies have proven the presence of HSV-1 DNA in the brains of aged persons, specifically in areas affected by AD (reviewed in Steel[12]) and in amyloid plaques.[13] In vitro studies have also demonstrated that inoculation with HSV-1 could lead to neuroinflammation, amyloid and phosphorylated tau accumulations, impaired autophagy, and mitochondrial alterations (reviewed in Harris et al.[14]).

Epidemiological studies suggest an increased risk of AD in infected individuals.[15,16] As ≈80% of aged persons are infected by HSV-1[17] and not all of them develop AD, the existence of environmental, viral, or genetic susceptibility factors seems essential for this theory to be valid. Such a factor could explain why some infected persons remain "healthy carriers" as observed in numerous infectious diseases.[18] From this perspective, the ε4 allele of the *APOE* gene seems to be of particular interest regarding its frequency in the general population[19] and previous results suggesting an interaction with HSV-1 infection.[20]

The main goal of this study was to assess the impact of HSV-1 infection on the incidence of AD according to a genetic susceptibility factor, the *APOE* gene, which could modify the effect of the virus.

RESEARCH IN CONTEXT

1. Systematic review: The biomedical database PubMed was used to search for previous literature. Previous reviews on the link between herpes simplex virus type 1 (HSV-1) and Alzheimer's disease (AD), cited in this article, highlight an increasingly solid pathophysiological rationale in favor of the infectious hypothesis in AD.
2. Interpretation: HSV-1 is a risk factor for AD, but its effect depends on genetic susceptibility. Among carriers of a genetic susceptibility factor, the *APOE4* allele, subjects with frequent reactivations of HSV-1 had a threefold increased risk of AD, whereas no association was found in APOE4 non-carriers.
3. Future directions: Because a large proportion of the population is infected with HSV-1, the identification of environmental, viral, or genetic susceptibility factors is essential to explain why only some infected individuals will develop AD. This information could help to identify participants for future preventive trials on AD.

Highlights

- Herpes simplex virus type 1 (HSV-1) is a risk factor for Alzheimer's disease (AD), but its effect depends on genetic susceptibility.
- Among *APOE4* carriers, those with frequent HSV reactivation are at higher risk of AD.
- Among *APOE4* non-carriers, no association was found between HSV-1 and AD.
- Susceptibility factors could help to design future preventive trials.

2 | METHODS

2.1 | Design, participants, and available data

The Three-City prospective cohort study (3C), the primary goal of which was to assess the effect of vascular factors on the incidence of dementia and cognitive impairment, began in 1999–2001 in three French areas (Bordeaux, Dijon, and Montpellier). A total of 9294 participants who were 65 years or older, non-institutionalized, and enrolled from electoral lists were included; due to financial constraints for the realization of serologies, only those from the Bordeaux center (n = 2104 at baseline) are considered in this ancillary study on HSV. Trained psychologists conducted home visits at 2, 4, 7, 10, 12, and 14 years after inclusion to collect sociodemographic, lifestyle, and health-related information using a standardized questionnaire. Cognitive functions and their impact on activities of daily living were assessed with a large battery of tests (including the Mini-Mental State Examination [MMSE]), and a neurologist examined participants with cognitive impairments to confirm the diagnosis of dementia according to Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSM-IV) criteria.[21] A group of experts reviewed all of these cases to confirm the dementia diagnosis and determine its subtype. Details of the study protocol have been fully described previously.[22] The ethical committee of the University Hospital of Kremlin-Bicêtre approved the study protocol, and each participant provided signed informed consent.

At the 4-year follow-up visit (T4) (2003–2005), the remaining 1568 participants from Bordeaux (171 participants were deceased and 365 lost to follow-up before T4) were asked to provide a blood sample that was stored at −80°C. Among the 1258 subjects with an available serum sample (257 participants did not provide a blood sample and 53 no longer had a blood sample available in the biobank), the presence of anti-HSV antibodies was assessed using LIAISON IgG HSV-1/2 and LIAISON IgM HSV-1/2 tests (Diasorin–Italy–chemiluminescence immunoassay technology). The presence of IgG and IgM were determined by an index value ≥1.10 and their absence by an index value <0.9 (as recommended by the manufacturer). Samples with values between 0.9 and 1.10 were tested a second time, and if their status remained inconclusive, they were excluded from our study sample (n = 33). We also excluded subjects with a status of IgG− IgM+ (n = 2) which could reflect either a primary infection (unlikely given the advanced age of the participants), or IgG false negative or IgM false positive. Finally, subjects with prevalent dementia at T4 (n = 60) and subjects without any follow-up after T4 (n = 126) were excluded for carrying out survival analyses on dementia incidence. Our study sample thus consisted of 1037 participants (Flow chart in Figure 1).

2.2 | Statistical analysis

Characteristics of the participants, HSV status, and missing data were described for the study sample. Because the rate of missing values was lower than 1.5% for each of the variables, we did not impute missing data. Different markers of HSV status were considered: presence of IgG anti-HSV, presence of IgM anti-HSV in IgG-positive subjects, and

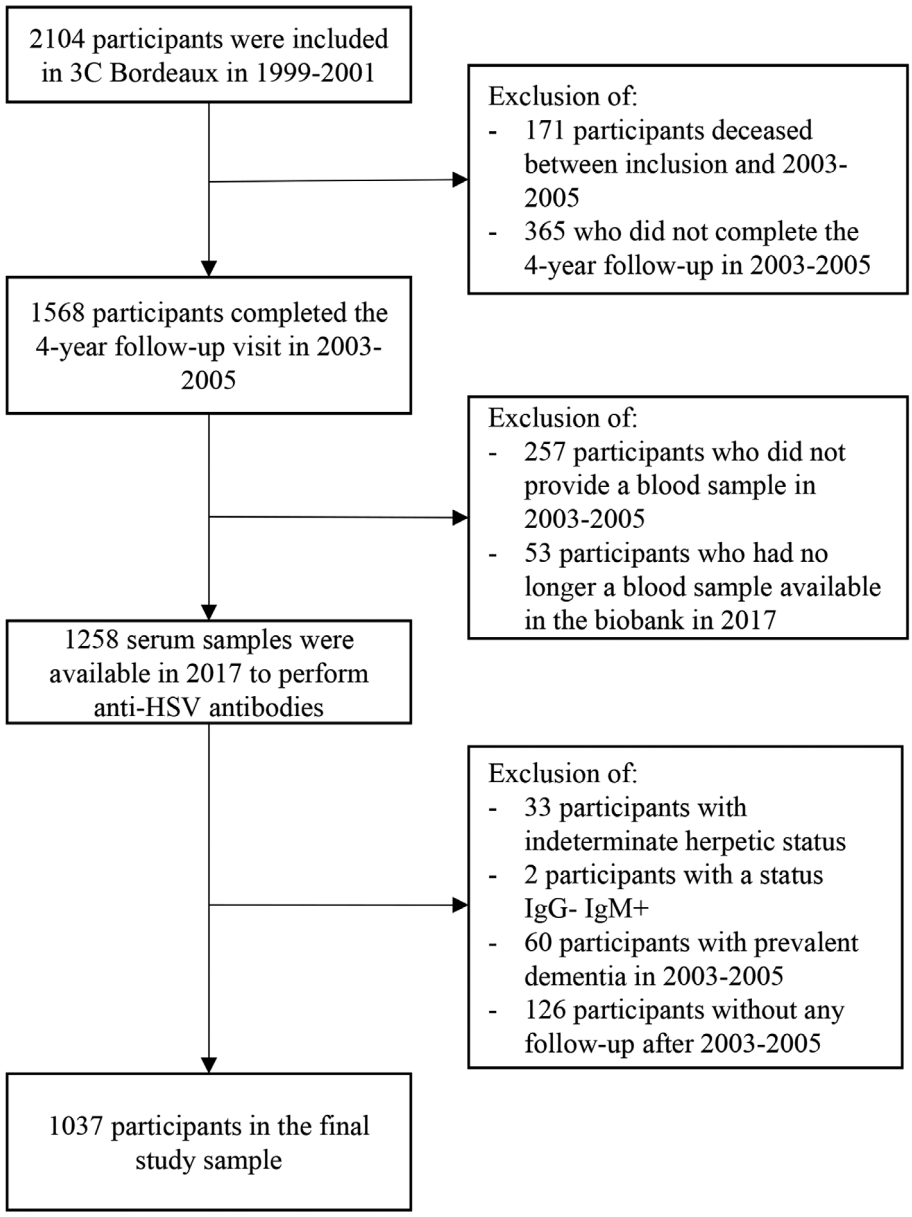

FIGURE 1 Flow chart of participants in the Three-City cohort of Bordeaux. Abbreviations: 3C, Three-City cohort; HSV, herpes simplex virus

level of IgG anti-HSV in terciles. Incidence rates of dementia were estimated, and survival curves were generated using the Kaplan-Meier method. Cox proportional hazard regression models with delayed entry were used to estimate cause-specific hazard ratios (HRs) and 95% confidence intervals (95% CIs) for all types of dementia and for AD or mixed dementia. The baseline time was the 4-year follow-up visit (ie, the follow-up visit when blood samples were collected). The age at onset of dementia was defined as the age between the last visit without dementia and the diagnosis visit. Participants without dementia at the end of the follow-up, those lost to follow-up, or those who passed away before the onset of dementia were censored at the age of the last visit without dementia. In preliminary analyses, Cox models were also used to assess the association between HSV status and age of death; as there was no association, we chose not to implement an illness-death model. In adjusted Cox models, sex, level of education (elementary school without diploma, short secondary school, and higher levels), marital status, presence of at least one allele of *APOEε4*, hypertension (defined as systolic blood pressure higher than 140 and/or diastolic blood pressure higher than 90 and/or use of an antihypertensive treatment), diabetes (defined as fasting blood sugar ≥7 mmol/L or blood sugar ≥11 mmol/L and/or intake of diabetes therapy), hypercholesterolemia (defined as cholesterol ≥6.2 mmol/L and/or intake of cholesterol-lowering drug), and tobacco consumption (defined as non-smoker, former smoker, or current smoker) were considered potential confounding factors. We adjusted our analyses for factors that may be related to either the clinical diagnosis of AD or to HSV status to limit a potential confusion bias. In fact, marital status and socio-economic status have been associated with HSV infection in previous literature[23,24] and such associations were also found in our study sample (results not showed). Moreover, the clinical diagnosis of AD reflects most of the time a

TABLE 1 Characteristics of the participants in the study sample 3C-Bordeaux, 2003–2005, n = 1037

		Total (N = 1037)
Age	Minimum/maximum	69.7/92.2
	Mean (standard deviation)	77.7 (4.7)
Sex	Men	373 (36.0)
Level of education	Elementary school without diploma	97 (9.4)
	Short secondary school	519 (50.0)
	Higher levels	421 (40.6)
Marital status[a]	Married	533 (51.5)
	Divorced or separate	81 (7.8)
	Widowed	350 (33.8)
	Single	71 (6.9)
APOE4[a]	At least one allele	178 (17.4)
Hypertension[a]	Yes	775 (74.8)
Diabetes[a]	Yes	126 (12.2)
Hypercholesterolemia	Yes	605 (58.3)
Tobacco consumption	Non-smoker	673 (64.9)
	Former smoker	312 (30.1)
	Smoker	52 (5.0)
Mini-Mental Status Examination[a]	Minimum/maximum	18.0/30.0
	Mean (standard deviation)	27.7 (1.9)

Values are numbers and percentages unless otherwise indicated.
[a]Number of missing data: Two for marital status, 14 for *APOE4*, one for hypertension, eight for diabetes, and two for Mini-Mental Status Examination.

multifactorial process, especially in older people, depending certainly on the presence of specific hallmarks of AD but also on other types of alterations (vascular pathology being frequently associated with AD), on the level of cognitive reserve (linked to the level of education) and the level of social support (linked to marital status).[25] Interactions with the presence of at least one allele $\varepsilon 4$ of the *APOE* gene were tested and considered significant if <0.10. Cox models were then performed according to *APOE4* status. In these models, adjustment variables were restricted to sex and level of education due to the smaller sample size. We verified the proportional hazards assumption with Schoenfeld residuals and by testing interaction with time. All statistical tests were two-tailed, and the threshold for statistical significance was 5%. Analyses were performed with the statistical software SAS (version 9.4; SAS Institute).

3 | RESULTS

3.1 | Characteristics of the participants

The main characteristics of the participants are available in Table 1. The skew toward female participants reflects mainly the French elderly population, with longer life expectancy for women and thus higher proportion of women among elderly, and could also partly be explained by a volunteer bias. The mean follow-up time was 7.4 years (standard deviation 2.9, min-max 1.1–10.9). IgG and IgM anti-HSV were present in 81.4% (844/1037) and 4.5% (47/1037) of the participants, respectively. In IgG-positive participants, the percentage of positive IgM was 5.6% (47/844).

3.2 | Association between HSV status and incidence of dementia

During the follow-up, 204 participants (19.7%) were diagnosed with dementia (including 87 probable AD, 47 possible AD, and 22 mixed dementia). The incidence of dementia was 26.7 cases per 1000 person-years.

Controlling for sex, level of education, marital status, presence of at least one *APOE4* allele, hypertension, diabetes, hypercholesterolemia, and tobacco consumption, no significant association was found between HSV status and incidence of dementia, regardless of the marker of HSV status considered (see Table 2). Nevertheless, there were several significant interactions with *APOE4* status, and global analyses were thus incorrect. Cox models were then performed separately according to *APOE4* status.

In individuals with at least one *APOE4* allele (Table 3), the association between IgG status and incidence of dementia (all types) was close to significance ($P = 0.07$, adjusted hazard ratio [aHR] = 2.63, 95% CI [0.92–7.50]), whereas among IgG-positive individuals, being IgM+ or having a high level of IgG was associated with a threefold higher risk of developing AD or mixed dementia (aHR = 3.68 [1.08–12.55] for IgM status and aHR = 3.28 [1.19–9.03] for IgG level in the highest tercile). As expected, HRs were generally higher for analysis of AD or mixed dementia than for analyses concerning all types of dementia. No significant association was found between HSV status and incidence of dementia in *APOE4*-negative subjects with aHR close to 1 in most of the analyses (Table 4).

4 | DISCUSSION

4.1 | Main results

Our results highlight an increased risk of AD in aged individuals infected with HSV-1 and carriers of a genetic susceptibility factor, the $\varepsilon 4$ allele of the *APOE* gene. Among individuals for whom the frequency of HSV-1 reactivation is supposed to be high, that is, IgM positive at blood sampling or with an elevated level of IgG, the risk of AD was increased threefold with aHRs of 3.68 and 3.28, respectively. No significant association was found in *APOE4*-negative persons.

4.2 | Strengths and limits

The main strengths of our study are its prospective design, its long follow-up, and the methods used for the diagnosis of dementia. The

TABLE 2 Association between HSV status and incidence of dementia—Cox models 3C Bordeaux 2003–2014

	Unadjusted model (n = 1037 or 844)[a]				Adjusted model[b] (n = 1012 or 824)[a]			
	Cases	HR	95% CI	*P*-value	aHR	95% CI	*P*-value	Interaction with APOE4
IgG anti-HSV positive (vs negative)								
All dementia	204	1.25	0.85–1.84	0.25	1.37	0.91–2.06	0.14	0.08
Alzheimer's disease or mixed dementia	156	1.24	0.80–1.91	0.34	1.27	0.81–2.01	0.30	0.13
In IgG+ subjects, IgM anti-HSV positive (vs negative)								
All dementia	173	0.73	0.36–1.49	0.39	0.84	0.41–1.73	0.63	0.03
Alzheimer's disease or mixed dementia	132	0.83	0.39–1.79	0.64	1.00	0.46–2.17	0.99	0.02
In IgG+ subjects, IgG level in terciles[c]								
All dementia	173							0.29
Second tercile (vs first tercile)		1.26	0.84–1.90	0.26	1.24	0.82–1.89	0.31	
Third tercile (vs first tercile)		1.16	0.80–1.67	0.43	1.14	0.78–1.68	0.48	
Alzheimer's disease or mixed dementia	132							0.09
Second tercile (vs first tercile)		1.26	0.79–1.99	0.34	1.23	0.76–1.97	0.40	
Third tercile (vs first tercile)		1.12	0.74–1.70	0.59	1.10	0.71–1.69	0.68	

Abbreviations: 95% CI, 95% confidence interval; aHR, adjusted hazard ratio; HR, hazard ratio.
[a]Number of subjects for analysis in the whole sample or in the subsample of IgG+ subjects, respectively.
[b]Adjustment for sex, level of education, marital status, presence of at least one allele of *APOE4*, hypertension, diabetes, hypercholesterolemia, and tobacco consumption.
[c]IgG level in the first tercile was <19.2, in the second tercile, ≥19.2 and <30.01, and in the third tercile, ≥30.01.

TABLE 3 Association between HSV status and incidence of dementia in APOE4-positive subjects—Cox models. 3C Bordeaux 2003–2014

	Unadjusted model (n = 178 or 140)[a]				Adjusted model[b] (n = 178 or 140)[a]		
	Cases	HR	95% CI	*P*-value	aHR	95% CI	*P*-value
IgG anti-HSV positive (vs negative)							
All dementia	43	3.06	1.09–8.57	0.03	2.63	0.92–7.50	0.07
Alzheimer's disease or mixed dementia	36	2.61	0.92–7.39	0.07	2.24	0.78–6.46	0.14
In IgG+ subjects, IgM anti-HSV positive (vs negative)							
All dementia	39	2.72	0.82–8.99	0.10	3.09	0.92–10.42	0.07
Alzheimer's disease or mixed dementia	32	3.21	0.96–10.69	0.06	3.68	1.08–12.55	0.04
In IgG+ subjects, IgG level in terciles[c]							
All dementia	39						
Second tercile (vs first tercile)		1.51	0.62–3.72	0.37	1.62	0.65–4.03	0.30
Third tercile (vs first tercile)		2.18	0.94–5.04	0.07	2.25	0.97–5.24	0.06
Alzheimer's disease or mixed dementia	32						
Second tercile (vs first tercile)		2.11	0.72–6.17	0.17	2.16	0.73–6.43	0.17
Third tercile (vs first tercile)		3.19	1.17–8.73	0.02	3.28	1.19–9.03	0.02

Abbreviations: 95% CI, 95% confidence interval; aHR, adjusted hazard ratio; HR, hazard ratio.
[a]Number of subjects for analysis in the whole sample or in the subsample of IgG+ subjects, respectively.
[b]Adjustment for sex and level of education.
[c]IgG level in the first tercile was <19.2, in the second tercile, ≥19.2 and <30.01, and in the third tercile, ≥30.01.

repeated evaluations of various neurocognitive functions by trained psychologists, the second examination by a neurologist or a geriatrician for subjects with suspicion of cognitive decline, and the validation and classification of dementia cases by a group of experts have allowed a high-quality diagnosis. Nevertheless, the risk of informative censorship, inherent to our prospective design, cannot be excluded and the small number of participants diagnosed with non-AD dementias (n = 48 among them seven APOE4 carriers) prevented us from carrying out analyses to further test the specificity of the association between HSV and AD. The absence of available amyloid or tau markers for AD diagnosis is due to the logistic difficulties in performing cerebrospinal fluid examinations or PET scans on such a large sample living in the community and to the absence of validated markers in plasma. Despite some known selection bias (volunteers, urban recruitment, duration of the follow-up, and acceptance of blood sampling), the study has the advantage of including a population cohort, rather than a hospital

TABLE 4 Association between HSV status and incidence of dementia in APOE4- negative subjects—Cox models. 3C Bordeaux 2003–2014

	Unadjusted model (n = 845 or 693)[a]				Adjusted model[b] (n = 845 or 693)[a]		
	Cases	HR	95% CI	*P*-value	aHR	95% CI	*P*-value
IgG anti-HSV positive (vs negative)							
All dementia	158	1.08	0.71–1.64	0.72	1.01	0.66–1.55	0.96
Alzheimer's disease or mixed dementia	117	1.09	0.66–1.78	0.75	0.99	0.60–1.64	0.98
In IgG+ subjects, IgM anti-HSV positive (vs negative)							
All dementia	132	0.53	0.22–1.31	0.17	0.58	0.24–1.44	0.24
Alzheimer's disease or mixed dementia	98	0.55	0.20–1.51	0.25	0.60	0.22–1.65	0.32
In IgG+ subjects, IgG level in terciles[c]							
All dementia	132						
Second tercile (vs first tercile)		1.21	0.76–1.93	0.41	1.16	0.73–1.85	0.53
Third tercile (vs first tercile)		1.04	0.69–1.57	0.87	0.98	0.64–1.49	0.92
Alzheimer's disease or mixed dementia	98						
Second tercile (vs first tercile)		1.14	0.67–1.93	0.63	1.09	0.64–1.85	0.75
Third tercile (vs first tercile)		0.89	0.55–1.43	0.63	0.83	0.51–1.34	0.44

Abbreviations: 95% CI, 95% confidence interval; aHR, adjusted hazard ratio; HR, hazard ratio.

[a]Number of subjects for analysis in the whole sample or in the subsample of IgG+ subjects, respectively.

[b]Adjustment for sex and level of education.

[c]IgG level in the first tercile was <19.2, in the second tercile, ≥19.2 and <30.01, and in the third tercile, ≥30.01.

cohort, and is, thus, more representative of the general population. The availability of HSV serologies across the entire 3C cohort would have increased the statistical power of our analyses and is indeed a limitation of our study. However, despite this reduced sample and thanks to the availability of genetic data, we were able to perform stratified analyses on APOE4 status, which led to statistically significant results but constrained us to limit the set of adjustment variables. Other strengths of our analysis are the low level of missing data, use of an appropriate statistical model, and low risk of reverse causality due to the prospective design. The HSV serology used in this study has a sensitivity of 99% and a specificity of 97% according to the manufacturer, but it does not distinguish between HSV-1 and HSV-2. In subjects over 50 years old, the lower prevalence of HSV-2 (≈20%) compared to HSV-1 (≈80%)[17] minimizes the importance of this limitation. Moreover, because HSV serology simultaneously reflects the intensity of HSV-1 infection and the intensity of the related immune response, its interpretation is difficult. We hypothesized that IgM-positive subjects at blood sampling were more likely to have a higher frequency of reactivation than others. We also supposed that in infected subjects, a higher level of IgG was a marker of a more important history of reactivations; the hypothesis was that each reactivation over time led to an increase in the number of B lymphocytes specialized for HSV-1 and, consequently, to a higher level of anti-HSV IgG.

4.3 | Interpretation in light of literature

4.3.1 | Previous studies

Epidemiological results concerning the association of HSV-1 infection and dementia are heterogeneous, as recently reviewed in a meta-analysis by Warren et al.[26] Several studies that investigated the association between IgG status and dementia have found either no association or, more rarely, an increased risk of dementia in infected subjects. Concerning the level of IgG, the results were in favor of either no association with dementia or an increased risk of dementia in subjects with high levels of IgG. Of interest, Mancuso et al. and Costa et al. showed a significantly higher level of IgG anti-HSV in AD versus controls and an intermediate level in subjects with mild cognitive impairment.[27,28] Fewer studies have investigated the impact of recent reactivations (ie, IgM+) on the risk of AD. The results from the French cohort PAQUID have highlighted an increased risk of incident AD in IgM-positive subjects (aHR = 2.55 [1.38–4.72]).[15] These results have been replicated in the Swedish cohort BETULA, where similar results were found (aHR = 1.693 [1.003–2.858]).[16] In this last cohort, a lower decline in episodic memory was paradoxically reported in infected subjects with a recent reactivation (IgG+ IgM+) versus infected subjects without a recent reactivation (IgG+ IgM–),[29] and no association between IgM status and dementia was highlighted in a nested case-control study[30] from the same group. This heterogeneity could partly be explained by methodological discrepancies. In older studies, small sample sizes and/or less sensitive methods to detect HSV antibodies could have led to non-significant results.[31–33] Two factors seem essential to consider for the hypothesis of an implication of HSV-1 in the onset of AD to be valid: age and presence of susceptibility factors.

4.3.2 | Delayed onset of Alzheimer's disease and immunosenescence

Although HSV-1 primary infection often occurs at a young age, the late onset of AD suggests a delayed effect of HSV-1 on cognitive functions.

This effect could be either initiated or strengthened by a trigger occurring in aged subjects. Immunosenescence is supposed to be that trigger. In fact, it affects cells essential to the maintenance of HSV-1 latency,[34] the CD8+ T lymphocytes. Considering this information, research needs to assess the association of HSV-1 and dementia in subjects old enough to allow HSV-1 to express its deleterious impact. In a cohort including younger subjects, an interaction with age would need to be considered, looking for an increased effect of HSV-1 with age, as shown in a recent analysis from Lövheim et al.[29] Testing an interaction directly with immunosenescent markers could enhance the accuracy of future analyses, as age is only one of the determinants of immunosenescence. For example, the presence of chronic coinfections could prematurely exhaust the immune system, and a recent study has already highlighted an interaction between HSV-1 and a known contributor of immunosenescence, CMV infection, for the risk of AD development.[35]

4.3.3 | Presence of susceptibility factors

Because a high frequency of the population is infected by HSV-1, the presence of susceptibility factors (environmental, viral, or genetic) is needed to explain why some infected subjects develop AD but not all and may partially explain variations concerning the age at onset of AD. Viral factors (such as viral load, strain, neurovirulence, coinfections, and frequency of reactivation) have not yet been investigated and may help to identify subjects at higher risk of AD. Concerning genetic susceptibility factors, previous studies have mainly investigated the *APOE* gene, showing arguments in favor of an interaction between HSV-1 infection and *APOE* status. In murine studies, *APOE4* was associated with a higher HSV-1 load in mouse brains compared to *APOE3*.[36–38] *APOE4* carriers were also shown to be more at risk of cold sores.[39–41] At the same time, *APOE* status has been linked to either the frequency or extent of other viral infections such as HIV, hepatitis C virus, or varicella zoster virus (reviewed in Itzhaki et al.[42]) and to some related neurological complications such as dementia or peripheral neuropathy in HIV-infected subjects.[43] Among epidemiological studies, the results are heterogeneous. Letenneur et al. found no interaction between HSV status and *APOE4*,[15] whereas Lövheim et al. evidenced an increased risk of decline in episodic memory associated with HSV-1 seropositivity in *APOE4*-positive subjects.[29] Several mechanisms have been proposed to explain the interaction between apoE4 and HSV-1. apoE and HSV-1 use the same binding site to enter the cell, the heparan sulfate proteoglycans.[44] Thus, it was proposed that HSV-1 competes more effectively with apoE4 than apoE3, facilitating its entry into neurons and its spread from neuron to neuron. Other results suggest an impact of the APOE allele on the response to HSV-1 damage. Indeed, it was shown that the antioxidant activity of apoE4 was less important than that of apoE3 and apoE2.[45]

Numerous other genetic risk factors of AD have also been linked to the viral cycle of HSV-1 in the neuron and to the immune response.[5–7,46] The combination of all these genetic risk factors may explain the propensity of a subject to develop cognitive disorders due to HSV-1.

4.3.4 | CONCLUSION

Our results suggest an increased risk of AD in aged subjects infected with HSV-1 and carriers of a genetic susceptibility factor, the $\varepsilon 4$ allele of the *APOE* gene. This finding is in accordance with an increasingly solid pathophysiological rationale in favor of the implication of HSV-1 in the onset of AD. Our hypothesis provide arguments to some of the key questions regarding the origins of AD as proposed in a paper published in 2015.[47] Among them, we bring results in favor of an interaction between HSV-1 infection and *APOE* as well as elements for the possible underlying mechanism, which may explain at least partially the relation between *APOE4* and the onset of AD. Moreover, the decrease of immune defenses with age and its implication on HSV-1 reactivations may also explain the relationship between aging and AD. In addition, due to its affinity for the temporal cortex and its capacity to travel from neuron to neuron to progressively affect other areas of the brain, HSV-1 infection may also explain the patterns of progression of the disease. Furthermore, HSV-1 virus could interfere with the key hallmarks of AD pathology, via the possible role of aβ in the antimicrobial response and the possibility to provoke other hallmarks of AD, as shown in cells or animals models. Considering the optimistic results regarding the efficacy of treatment in in vitro or animal studies,[48–50] the infectious hypothesis deserves further investigation in regard to the possibilities of effective preventive treatments of AD. Two clinical trials evaluating the efficacy of valaciclovir in subjects with AD or mild cognitive impairment are currently underway in Sweden and in the United States (NCT02997982 and NCT03282916). A further understanding of susceptibility factors implicated in the relation between HSV-1 and AD could help to select participants for further preventive trials.

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CONFLICT OF INTEREST

The authors report no conflicts of interest related to this study.

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