1-19-2023

# Susceptibility to Neurodegenerative Disorders: Insights from Paleogenomic Data

Draga Toncheva
*Medical Faculty, Department of Medical Genetics, Medical University of Sofia, Sofia, Bulgaria, Bulgarian Academy of Sciences, Sofia, Bulgaria*

Sena Karachanak-Yankova
*Medical Faculty, Department of Medical Genetics, Medical University of Sofia, Sofia, Bulgaria, Department of Genetics, Faculty of Biology, Sofia University, Sofia, Bulgaria*

Maria Marinova
*Department of Computer Systems and Technologies, Faculty of Electronics and Automation, Technical University of Sofia, Branch Plovdiv, Sofia, Bulgaria*

Plamenka Borovska
*Department of Informatics, Faculty of Applied Mathematics and Informatics, Technical University of Sofia, Sofia, Bulgaria*

Dimitar Serbezov
*Medical Faculty, Department of Medical Genetics, Medical University of Sofia, Sofia, Bulgaria*

# Susceptibility to Neurodegenerative Disorders: Insights from Paleogenomic Data

Draga Toncheva[1,2*], Sena Karachanak-Yankova [1,3], Maria Marinova [4], Plamenka Borovska[5], and Dimitar Serbezov[1]

[1]Medical Faculty, Department of Medical Genetics, Medical University of Sofia, Sofia, Bulgaria

[2]Bulgarian Academy of Sciences, Sofia, Bulgaria

[3]Department of Genetics, Faculty of Biology, Sofia University, Sofia, Bulgaria

[4]Department of Computer Systems and Technologies, Faculty of Electronics and Automation, Technical University of Sofia, Branch Plovdiv, Sofia, Bulgaria

[5]Department of Informatics, Faculty of Applied Mathematics and Informatics, Technical University of Sofia, Sofia, Bulgaria

*Correspondence to: Draga Toncheva, Department of Medical Genetics, Medical University of Sofia, Zdrave str 2 1431, Sofia, Bulgaria. Email: dragatoncheva@gmail.com

## Abstract

Ancient human genome data that has accumulated in recent years can be employed to establish the spatiotemporal trajectories of genetic variants associated with human diseases. Such knowledge might illuminate if and how past adaptations impact contemporary human health and medicine. Scarcely any studies have yet been attempted to evaluate the genetic susceptibility to neurodegenerative disorders in ancient human communities. Using publicly available ancient human genome-wide data the present study evaluates the molecular predisposition to neurodegenerative disorders in ancient human communities. To this end we screened the ancient genome-wide data for the presence of variants unequivocally associated with neurodegenerative disorders in modern populations, and their historical and geographic prevalence was assessed. These variants are two rare variants in the *LRRK2* gene associated with Mendelian Parkinson's disease, a pathogenic variant in the *CRH* gene, associated with autosomal dominant nocturnal frontal lobe epilepsy (ADNFLE), and a rare variant in the *TREM2* gene, a possible risk modifier associated with Alzheimer's disease. Our assessment of the historical and geographic prevalence indicates differing spatiotemporal frequency dynamics for these clinically significant variants. Neurodegenerative disorders are often with poorly understood pathogenesis that might be elucidated by studying the interaction of past genetic variability with ecological and evolutionary factors such as adverse environmental conditions, specific selective pressures, periods of population isolation and admixture processes. Data on molecular predisposition to neurodegenerative disorders in ancient genomes is instructive to modern medical diagnostic and therapeutic practices.

# Introduction

Neurodegeneration, the progressive loss of structure or function of neurons, is a feature of many debilitating, incurable diseases. Research on these diseases is aimed at elucidating the molecular mechanisms of the damaging processes, and also at finding new methods for diagnosis and therapy. Recent studies, for example, suggest that changes of the brain proteome are a key molecular mechanism at the onset of neurodegenerative processes (Schrötter et al. 2016). Comparative genome studies of different neurodegenerative diseases are therefore critical for getting insight into their etiology.

Alzheimer's disease (AD) is one of the most common neurodegenerative brain disorders and is responsible for 60–70% for all dementia cases (WHO 2021). The prevalence of AD in people over 65 years in Europe is 4.4% (Lobo et al. 2000), and several meta-analyses and nationwide surveys have yielded roughly similar age-specific prevalence of AD across regions (Lobo et al. 2000, Dong et al. 2007, Plassman et al. 2007). These figures are however bound to increase as population aging has become a worldwide phenomenon. AD is a multifactorial disease that is caused by the interaction of genetic and environmental factors. The genetic basis of AD is better understood in familial cases with early onset (before the age 50) and autosomal dominant type of inheritance. These however represent only 1% of AD patients and are caused by rare variants in genes that play role in the formation of amyloid plaques: *APP*, *PSEN1*, *PSEN2* and *TREM2* (Khani et al. 2022). The genetic etiology of late sporadic forms of Alzheimer's disease is far less known. A small number of rare variants and a large number of common variants with small impact have been identified, among which the strongest effect is attributed to the ε4 allele of the *APOE* gene, long considered to be the leading genetic risk factor for late-onset AD (Corder et al. 1993, Huang and Mucke 2012).

Parkinson's disease (PD) is the second most common neurodegenerative disease after AD, the annual incidence of PD being estimated to be 4.5-21 cases per 100000 (Kasten et al. 2007). About 10–15% of the patients have a genetic cause of the disease (Verstraeten et al. 2015).

Epilepsy is a neurological disorder in which brain activity becomes abnormal, with an estimated annual cumulative incidence rate of around 68 per 100000 (Fiest et al. 2017). Tai et al., 2016 provide evidence that taupathy is associated with cognitive decline in temporal lobe epilepsy, the most prevalent form of chronic focal epilepsy (Tai et al. 2016). Taupathy is also recognized as the key driver of disease progression in Alzheimer's disease and other neurodegenerative disorders (Chang et al. 2018). Genetic defects can determine the pathogenesis of certain epilepsies with familial occurrence and with very early manifestations (Steinlein 2008). So far, however, the genetic basis of the brain disorders is based only on studies of modern populations.

Neurology as a modern science dates back to the 17th century with the publication of Thomas Willis, *Anatomy of the Brain* (Grand 1999). Before that Alcmaeon of Croton (6th B.C.) was the first of the Greek physicians to ascertain that the brain is the organ of mental faculties and that epilepsy is a brain's disease, and not "sacred disease" induced by gods, as was believed

in the Homeric Era (Baloyannis 2013). Deliberations on neurological concepts can also be found in the treatises of Hippocrates (5th century BC) (Tsoucalas et al. 2017). Data on neurological disorders in ancient populations were obtained indirectly by inspecting human busts from the Roman Republic and the early Roman Empire ages before 3rd century AD (Engmann 2013). The description of facial features of one bust suggests eyelid ptosis, probably due to the mitochondrial disorder Kearns-Sayre syndrome (Oculocraniosomatic disorder), or to progressive supranuclear palsy. In another bust, the pupils diverge, which is interpreted as probable left-sided paralysis of the oculomotor nerve with additional mild ptosis of the left eyelid, or possibly right-sided paralysis of the oculomotor nerve. Homeric Epics written down in the late 8th or 7th century BC contain stories with archaic descriptions of the clinical consequences of trauma to the head and neck, spine, and peripheral nerve injuries (Walshe III 2016). Trepanation is apparently the oldest practiced surgical intervention in antiquity for the treatment of cranial trauma, neurological diseases, tumors, or for religious reasons, e.g. to ward off evil spirits that caused mental illness, epilepsy or migraine symptoms. Evidence has been found that successful trepanations had been performed in the Neolithic Age in Anatolia (Erdal 2010), in Early Iron Age cemeteries on the Silk Road in Xinjiang, China (Zhang et al. 2018) in Ancient Egypt (Collado-Vázquez and Carrillo 2014), in the pre-Columbian Inca empire in South America and in other regions of the world (Kushner et al. 2018).

Data on the emergence and evolution of mutations associated with neurodegenerative diseases from ancient DNA samples is currently scarce (Simonti et al. 2016). For the present study, we examine human genome-wide data obtained from ancient DNA samples for the incidence of mutations in genes associated with neurodegenerative disorders.

# Materials and methods

We examined 2729 publicly available genome-wide data obtained from ancient human DNA samples from different geographical regions and dated 100-15000 BP (Allen Ancient DNA Resource, 2022). The age distribution of the samples analyzed is given on Fig. 1.

{~?~IM: insert Fig 1 here.}
**Figure 1**. Age distribution of the evaluated ancient DNA samples (100 BP–15000 BP).
As a starting point for our analyses, we consider variants listed in the freely available DisGeNet database of disease associated variants (Piñero et al. 2020), and we establish 32644 of these

variants in the ancient genome-wide data. From these we select mutations in genes that have been determined to be associated with neurodegenerative diseases in contemporary patients. Considered were only functionally significant variants which are localized in coding sequences and are either missense, frameshift, splice, start-gain, stop-loss or stop-gain mutations as these types of mutations entail changes in the amino acid sequence. Also, we selected variants with contemporary population frequency of < 0.05, as low population frequencies might be indicative of negative selection resulting from their pathogenic effect. The clinical significance as monogenic defects or risk factors for neurodegenerative diseases was further determined using the VarSome platform (Kopanos et al. 2019), whose inferences are used in American College of Medical Genetics (ACMG) guidelines, the ClinVar database (Landrum et al. 2018) as well as in relevant forums such as the Alzheimer's disease forum (ALZFORUM, 2022) and LRRK2 Cohort Consortium (2022). VarSome utilizes a wide range of data collected from multiple resources, including the variant's coding effect for different transcripts, its genomic location, the genes it affects, its population frequency, the function of the produced protein, associated phenotypes, relevant literature, clinical studies and pathogenicity.

# Results

Our selection procedure to single out monogenic defects or risk factors for neurodegenerative diseases in the analyzed 2729 ancient DNA samples yielded four variants in three different genes: two variants in the *LRRK2* gene (rs34637584 G>A and rs34778348 G>A) associated with Parkinson's disease type 8 (PARK8), one variant in the *CRH* gene, rs12721510 C>A), associated with autosomal dominant nocturnal frontal lobe epilepsy (ADNFLE) and one variant in *TREM2*, rs2234255 C>T, predisposing to Alzheimer's Disease. These four variants were established 88 times in the analyzed 2729 ancient DNA samples (Table 1).

{~?~IM: insert Table 1 here.}

| Population | Ancient samples | | | Contemporary samples | | | χ2-test |
|---|---|---|---|---|---|---|---|
| | Allele Count | Total Count | af | Allele Count | Total Count | af | |

| LRRK2 rs34637584 G>A, p.G2019S | | | | | | | |
|---|---|---|---|---|---|---|---|
| African | 0 | 92 | 0 | 2 | 16252 | 0.0001 | p = 1 |
| Ashkenazi | - | - | - | 86 | 10072 | 0.0085 | |
| East Asian | 3 | 422 | 0.014 | 0 | 18394 | 0 | p < 2.2e-16 |
| European-Finnish | - | - | - | 0 | 21644 | 0 | |
| European Non-Finnish | 9 | 2762 | 0.007 | 29 | 113486 | 0.0003 | p = 6.53e-16 |
| Latino | - | - | - | 9 | 34558 | 0.0003 | |
| South Asian | 1 | 358 | 0.006 | 0 | 30616 | 0 | p = 5.04e-06 |
| Other | 4 | 1824 | 0.004 | 6 | 6132 | 0.0010 | p = 0.36 |
| **Total** | **17** | **5458** | **0.003** | **132** | **251154** | **0.0005** | **p = 4.12e-14** |
| LRRK2 rs34778348 G>A, p.Gly2385Arg | | | | | | | |
| African | 0 | 92 | 0 | 0 | 16232 | 0 | |
| Ashkenazi | - | - | - | 0 | 10060 | 0 | |
| East Asian | 1 | 422 | 0.005 | 401 | 18382 | 0.022 | p = 0.012 |
| European-Finnish | - | - | - | 1 | 21612 | 0.0001 | |
| European Non-Finnish | 1 | 2762 | 0.0007 | 5 | 113372 | 0.00004 | p = 0.34 |
| Latino | - | - | - | 0 | 34390 | 0 | |
| South Asian | 0 | 358 | 0 | 17 | 30594 | 0.0006 | p = 1 |
| Other | 1 | 1824 | 0,001 | 5 | 6092 | 0.0008 | p = 0.49 |
| **Total** | **3** | **5458** | **0.001** | **429** | **250734** | **0.002** | **p = 0.057** |
| CRH rs12721510 C>A | | | | | | | |
| African | 0 | 92 | 0 | 445 | 41428 | 0.011 | p = 0.63 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Ashkenazi | - | - | - | 320 | 3472 | 0.092 | |
| East Asian | 1 | 422 | 0.005 | 1 | 5192 | 0.0002 | p = 0.35 |
| European-Finnish | - | - | - | 637 | 10594 | 0.060 | |
| European Non-Finnish | 32 | 2762 | 0.023 | 4250 | 68010 | 0.063 | p < 2.2e-16 |
| Latino | - | - | - | 529 | 15272 | 0.035 | |
| South Asian | 0 | 358 | 0 | 61 | 4826 | 0.013 | p = 0.06 |
| Other | 10 | 1824 | 0,011 | 88 | 2090 | 0.042 | p = 1.90e-12 |
| **Total** | **43** | **5458** | **0.016** | **6376** | **152110** | **0.042** | **p < 2.2e-16** |
| *TREM2* rs2234255 C>T, p. His157Tyr | | | | | | | |
| African | 0 | 92 | 0 | 9 | 16252 | 0.0006 | p = 1 |
| Ashkenazi | - | - | - | 7 | 10 076 | 0.0007 | |
| East Asian | 3 | 422 | 0.0142 | 35 | 18390 | 0.002 | p = 0.07 |
| European-Finnish | - | - | - | 1 | 21 636 | 0.00004 | |
| European Non-Finnish | 0 | 2762 | 0 | 34 | 113714 | 0.0003 | p = 0.73 |
| Latino | - | - | - | 1112 | 34576 | 0.032 | |
| South Asian | 0 | 358 | 0 | 22 | 30612 | 0.0007 | p = 1 |
| Other | 22 | 1824 | 0,024 | 13 | 6140 | 0.002 | p = 6.78e-08 |
| **Total** | **25** | **5458** | **0.009** | **1233** | **251396** | **0.005** | **p = 0.81** |

**Table 1**. Number of variant alleles, number of instances and estimated overall frequencies (af) from ancient and contemporary (gnomAD exomes, Version: 2.1.1) samples in different populations.

The ratio of the estimated population frequency of all disease associated variants established in the ancient samples and their contemporary frequencies is shown on Figure 2. The four variants that we focus on in this study are all among these with largest overall relative drop between

ancient and contemporary frequencies indicating that these specific mutations might have been subject to negative selection.

{~?~IM: insert Fig 2 here.}

**Figure 2.** Ancient/contemporary ratio of estimated population frequency of disease associated variants listed in DiGeNet database. The variants are ranged from those with lowest ratio (left side of the figure) to those with highest ratio (right side of the figure). The four pathogenic neurodegenerative disease associated variants considered in this study are marked with arrows. Plotted are 26131 variants for which are available contemporary population frequency estimates, out of the 32644 DisGeNet variants found in the ancient genome-wide data.

# Variant rs34637584 G>A in the *LRRK2* gene, G2019S

The variant rs34637584 G>A in the *LRRK2* gene was found in 17 ancient samples, with overall ancient population frequency estimate of 0.003. The highest frequency is estimated in East Asia (0.014), and is lower in South Asia (0.006) and Europe (0.007) (Table 1). The oldest samples are dated to 8000–8500 BP from today`s Latvia and 7000-7500 BC from today`s Serbia (Table 2). The frequency of this variant is significantly lower in contemporary populations ($\chi^2$-test, p = 4.12e-14), and in contrast to its ancient distribution, it seems to be absent in contemporary South Asian and East Asian populations. This variant has been established as *Pathogenic* by VarSome and ClinVar, and as associated with Parkinson's Disease by DisGeNet, with a variant-disease association (VDA score) of 0.9.

{~?~IM: insert Table 2 here.}

| Age pool | nr. of samples | LRRK2 | | | | CRH | | TREM2 | |
|---|---|---|---|---|---|---|---|---|---|
| | | rs34637584 | | rs34778348 | | rs12721510 | | rs2234255 | |
| | | nr. | af | nr. | af | nr. | af | nr. | af |
| 0-1000 | 267 | 1 | 0.004 | 1 | 0.004 | 0 | 0 | 16 | 0.06 |
| 1000-2000 | 272 | 3 | 0.011 | 0 | 0 | 2 | 0.007 | 3 | 0.011 |
| 2000-3000 | 272 | 1 | 0.004 | 0 | 0 | 1 | 0.004 | 0 | 0 |
| 3000-4000 | 549 | 5 | 0.009 | 0 | 0 | 9 | 0.016 | 1 | 0.002 |
| 4000-5000 | 626 | 3 | 0.005 | 0 | 0 | 10 | 0.016 | 2 | 0.003 |
| 5000-6000 | 241 | 0 | 0 | 1 | 0.004 | 10 | 0.041 | 0 | 0 |
| 6000-7000 | 127 | 1 | 0.008 | 1 | 0.008 | 1 | 0.008 | 1 | 0.008 |
| 7000-8000 | 214 | 1 | 0.005 | 0 | 0 | 8 | 0.037 | 0 | 0 |
| 8000-9000 | 75 | 2 | 0.027 | 0 | 0 | 2 | 0.027 | 1 | 0.013 |
| 9000-10000 | 34 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0.029 |
| 10000-52000 | 52 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

| Total | 2729 | 17 | 0.006 | 3 | 0.001 | 43 | 0.016 | 25 | 0.009 |
|---|---|---|---|---|---|---|---|---|---|

**Table 2.** Number and population frequency of variants associated with neurological and neurodegenerative disorders in ancient human genome-wide data, allocated into 1000 -year age pools.

## Variant rs34778348 G>A in the *LRRK2* gene, Gly2385Arg

The second dominant mutation in the *LRRK2* gene, rs34778348 G>A (Gly2385Arg), was established in only three ancient samples (Table 1 & Table 2), one from Greece (dated 6000–6500 BP); one from Mongolia (1000–1500 BP) and one from the Levant (5500–6000 BC). The estimated population frequency of this variant from ancient samples is very low, as it is in contemporary populations, 0.001 and 0.002, respectively. The variant has been designated in modern populations as *Likely Pathogenic* by Varsome, and with *Conflicting interpretations of pathogenicity* by ClinVar. This variant is autosomal dominant and denoted as associated with Parkinson's Disease by DisGeNet with VDA score of 0.9.

## Variant rs12721510 C>A in the *CRH* gene

The *CRH* gene encodes the corticotropin-releasing hormone. The rs12721510 C>A variant was established in 43 ancient genomes, with overall population frequency of 0.016. The majority of samples containing the mutation (n=32) were from Europe (af = 0.023) (Table 1). The oldest instances this variant was identified is in two samples from Anatolia dated to 8000-8500 BC, followed by two samples from the Balkan region from the Neolithic period dated to 7500-8000 BP. The contemporary population frequency is higher compared to that in ancient populations (0.016 vs 0.042). The highest contemporary frequency is determined in Ashkenazi Jews (0.092) and in European populations (Non-Finnish European, 0.063 and European-Finnish, 0.060). Compared to the limited geographic distribution in ancient communities, this variant is widespread in modern populations and is found in, along Ashkenazi and European, in African, South Asian, and Latino populations. Although designated as *benign* in VarSome, the variant is determined *pathogenic* by ClinVar and as associated with autosomal dominant nocturnal frontal lobe epilepsy (ADNFLE), with VDA score of 0.7.

## Variant rs2234255 C> T in the *TREM2* gene, His157Tyr

The variant rs2234255 was identified in 25 ancient genomes with an overall allele frequency of 0.009. Three instances are in samples from East Asia (AF = 0.014), one from West Asia, thirteen from the Caribbean region, one from North America and seven from South America. The oldest samples are dated to 9500–10000 BP from Brazil and to 8000–8500 BP from Anatolia. The overall frequency of the variant in modern populations is 0.005. The variant allele is globally distributed in contemporary populations. It is classified as *benign* by VarSome, *benign/likely benign* by ClinVar, and is reported as a possible risk modifier for Alzheimer's disease in the Alzheimer Forum. The variant has been found to increase the risk of Alzheimer's disease in the Han Chinese cohort (odds ratio: 11.01, p = 0.02) (Jiang et al. 2016). No association with AD has been demonstrated in Caucasian, Japanese, and African-American populations, but meta-analyzes of cohorts from the same populations have shown an increased risk for AD (odds ratio 3.65, p = 0.002), in accordance with results of The Alzheimer's Disease Sequencing Project (odds ratio: 4.7, p = 0.01).

# Discussion

The present study investigates the historical and geographic prevalence of four variants in genes associated with neurodegenerative disorders in modern populations. This is attained using ancient human genome-wide data and variant frequency data from contemporary populations. These variants are two rare variants in the *LRRK2* gene (the pathogenic rs34637584 and the possibly pathogenic rs34778348) associated with Mendelian Parkinson's disease), the variant rs12721510 in the *CRH* gene, associated with autosomal dominant nocturnal frontal lobe epilepsy, and a rare variant in the *TREM2* gene, the possible risk modifier rs2234255, associated with Alzheimer's disease.

## Variant rs34637584 G>A in the *LRRK2* gene, G2019S mutation

Functional interpretations illustrate the pathogenic effect of rs34637584 in heterozygous state in modern populations (Landrum et al. 2018, Kopanos et al. 2019). The variant is associated with an autosomal dominant type of monogenic Parkinson's disease type 8. This is the most common monogenic mutation in the early and late forms of the disease, and in familial and sporadic cases

(Bouhouche et al. 2017). The *LRRK2* gene encodes the enzyme Leucine-rich repeat kinase 2 from the leucine-rich repeat kinase family. The international *LRRK2* consortium determined the presence of this mutation in 1% of sporadic and 4% of familial patients with PARK8 (Healy et al. 2008). The onset is usually after the age of 50 years, but there are also early forms (in the 20s) and late forms (in the 90s). Mutations in *LRRK2* increase the activity of the LRRK2 protein. Published data from phenotype-genotype correlation studies demonstrate that heterozygous carriers have similar clinical characteristics to homozygotes and lack gene dosage effect (Ishihara et al. 2006). Homozygous mutant mice with the *LRRK2* G2019S point mutation appear normal, but have behavioral and electrophysiological changes, as well as lack of behavioral plasticity that correlates with abnormal synaptic plasticity in the striatum (Matikainen-Ankney et al. 2016). The estimated frequency of the mutation is significantly higher in ancient than in modern populations, i.e. 12.5 times higher (0.0062 vs 0.0005, cf. Table 1). Monogenic mutations associated with AD are clinically manifested later than the age usually reached by individuals in ancient populations, which may weaken the effect of natural selection. The significant reduction in the incidence of pathogenic mutations in modern populations may be further due to unknown environmental factors affecting the pathogenesis of complex diseases. Small population sizes, limited founder number and a lack of naturally occurring gene flow could have led to higher levels of inbreeding, as often might have been the case in ancient human communities (Ceballos et al. 2021). Overall, our results suggest consistently low prevalence of this variant through time (Fig. 3).

{~?~IM: insert Fig 3 here.}

**Figure 3.** Frequency of rs34637584 (*LRRK2* gene), rs12721510 (*CRH* gene) and rs2234255 (*TREM2* gene) variants associated with neurodegenerative disorders in different periods BP. The frequency trajectories are plotted using bins of 3000 years and sliding windows of 500 years. Uncertainty of the frequency estimation is indicated by a gray colored area, representing the normal approximation of the 95% binomial proportion CI.

## Variant rs34778348 G>A in the *LRRK2* gene, Gly2385Arg

The LRRK2 protein has two domains with kinase and GTPase activities that are disrupted by dominant mutations (Nguyen and Moore 2017). The variant Gly2385Arg is localized in the C-terminal WD40 domain of the LRRK2 protein. It causes partial or complete loss of kinase function of the protein and has been assessed to be a risk factor for Parkinson's disease (PD) in some populations, e.g. ethnic Chinese, Japanese, Taiwanese, Singaporean and Korean and

Malaysian (Gopalai et al. 2014). A meta-analysis of 61 published case-control studies also demonstrates that the G2385R mutation is a risk factor for PD (Wu et al. 2012).

## Variant rs12721510 C>A in the *CRH* gene

The *CRH* gene encodes corticotropin-releasing hormone, which is a 48 amino acid peptide extensively present in the CNS. As a neurotransmitter and neuromodulator in extrahypothalamic circuits it integrates a multisystem response to stress, and controls numerous behaviors such as locomotor activity, anxiety, food intake, sexual behavior, sleep, arousal, and learning. The rs12721510 variant is located in the promoter region of the *CRH* gene and it alters the expression of the protein, and is associated with familial autosomal dominant nocturnal frontal lobe epilepsy (ADNFLE) (Combi et al. 2005). The estimated overall population frequency of the variant in ancient samples is significantly lower than in modern populations (cf. Table 1). The temporal dynamic of its frequency (based mostly on European samples) indicates gradual but significant decrease in frequency throughout the time period analyzed (Fig. 2). As the disease manifests in childhood, a substantial demographic subgroup of ancient communities, negative selection has likely had the opportunity to reduce its frequency. Medical advances, e.g. diagnostic and therapeutic developments, might have weakened the strength of negative selection in recent times and explain the high frequency of this variant in modern populations.

## Variant rs2234255 C> T in the *TREM2* gene, His157Tyr

The *TREM2* gene encodes a transmembrane immune receptor in the microglial cells. It acts as a modulator of the inflammatory response in the CNS by regulating the number of myeloid cells and enhancing phagocytosis. Disrupted functions of the *TREM2* gene provokes pathogenesis of neurodegenerative diseases and has been shown to increase the risk of Alzheimer's disease in certain populations (Gratuze et al. 2018). The population frequency of this rare risk variant in the *TREM2* gene in ancient populations has been estimated to be significantly higher than that in modern populations (cf. Table 1). The temporal frequency dynamics curve indicates a peak at around 10000 BP, but also a recent upsurge from about 1000 BP. The high number of variant alleles established in during the ceramic era in the Caribbean region from the Ceramic Era (Fernandes et al. 2021) cause this apparent recent frequency upsurge.

# Conclusion

To our knowledge no studies have yet attempted to evaluate the genetic susceptibility to neurodegenerative disorders in ancient human communities. The present study evaluates the molecular predisposition to disease using ancient human genome-wide data, but with absence of information on any probable clinical presentations. Data on molecular predisposition to neurodegenerative disorders in ancient genomes might however be instructive for modern medical diagnostic and therapeutic methods. Many of these diseases are with poorly understood pathogenesis, where along with the genetic, other factors might have influence such as adverse environmental effects, specific selective pressures, migration processes. Various environmental factors combined with differential population histories might have led to the diverse spatiotemporal frequency dynamics of clinically significant variants.

# Disclosure statement

No potential conflict of interest was reported by the authors.

# Data availability statement

Data available on request from the authors.

# References

ALZFORUM. 2022. https://www.alzforum.org/ (accessed May 2022).

Baloyannis, S. J. 2013. Epilepsy: A way from Herodotus to Hippocrates. *Epilepsy Behav.* 28:303.

Bouhouche, A., H. Tibar, R. Ben El Haj et al. 2017. LRRK2 G2019S mutation: Prevalence and clinical features in Moroccans with Parkinson's disease. *Parkinsons Dis*. 2017:2412486.

Ceballos, F. C., K. Gürün, N. E. Altınışık et al. 2021. Human inbreeding has decreased in time through the Holocene. *Curr. Biol*. 31:3925–3934.e8.

Chang, H.-Y., T.-K. Sang, and A.-S. Chiang. 2018. Untangling the Tauopathy for Alzheimer's disease and parkinsonism. *J. Biomed. Sci.* 25:54.

Collado-Vázquez, S. and J. M. Carrillo. 2014. Cranial trepanation in The Egyptian. *Neurologia* 29:433–440.

Combi, R., L. Dalprà, L. Ferini-Strambi et al. 2005. Frontal lobe epilepsy and mutations of the corticotropin-releasing hormone gene. *Ann. Neurol.* 58:899–904.

Corder, E. H., A. M. Saunders, W. J. Strittmatter et al. 1993. Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. *Science* 261:921–923.

David Reich Lab. 2022. Allen Ancient DNA Resource. https://reich.hms.harvard.edu/allen-ancient-dna-resource-aadr-downloadable-genotypes-present-day-and-ancient-dna-data. (accessed May 2022).

Dong, M.-j., B. Peng, X.-t. Lin et al. 2007. The prevalence of dementia in the People's Republic of China: A systematic analysis of 1980–2004 studies. *Age Ageing* 36:619–624.

Engmann, B. 2013. Neurologic diseases in ancient Roman sculpture busts. *Neurol. Clin. Pract.* 3:539–541.

Erdal, Y. S. 2010. A retrospective study on trepanation in Anatolia. *Proc. 6th Int. Congress Archaeol. Ancient Near East.*

Fernandes, D. M., K. A. Sirak, H. Ringbauer et al. 2021. A genetic history of the pre-contact Caribbean. *Nature* 590:103–110.

Fiest, K. M., K. M. Sauro, S. Wiebe et al. 2017. Prevalence and incidence of epilepsy: A systematic review and meta-analysis of international studies. *Neurology* 88:296–303.

Gopalai, A. A., S. Y. Lim, J. Y. Chua et al. 2014. LRRK2 G2385R and R1628P mutations are associated with an increased risk of Parkinson's disease in the Malaysian population. *Biomed. Res. Int.* 2014:867321.

Grand, W. 1999. The Anatomy of the Brain, by Thomas Willis. *Neurosurgery* 45:1234–1237.

Gratuze, M., C. E. G. Leyns, and D. M. Holtzman. 2018. New insights into the role of TREM2 in Alzheimer's disease. *Mol. Neurodegener.* 13:66.

Healy, D. G., M. Falchi, S. S. O'Sullivan et al. 2008. Phenotype, genotype, and worldwide genetic

penetrance of LRRK2-associated Parkinson's disease: A case-control study. *Lancet Neurol.* 7:583–590.

Huang, Y., and L. Mucke. 2012. Alzheimer mechanisms and therapeutic strategies. *Cell* 148:1204–1222.

Ishihara, L., L. Warren, R. Gibson et al. 2006. Clinical features of Parkinson disease patients with homozygous leucine-rich repeat kinase 2 G2019S mutations. *Arch. Neurol.* 63:1250–1254.

Jiang, T., J. K. Hou, Q. Gao et al. 2016. TREM2 p.H157Y variant and the risk of Alzheimer's Disease: A aeta-Analysis involving 14,510 subjects. *Curr. Neurovasc. Res.* 13:318–320.

Kasten, M., A. Chade, and C. M. Tanner. 2007. Epidemiology of Parkinson's disease. *Handb. Clin. Neurol.* 83:129–151.

Khani, M., E. Gibbons, J. Bras et al. 2022. Challenge accepted: Uncovering the role of rare genetic variants in Alzheimer's disease. *Mol. Neurodegener.* 17:3.

Kopanos, C., V. Tsiolkas, A. Kouris et al. 2019. VarSome: the human genomic variant search engine. *Bioinformatics* 35:1978–1980.

Kushner, D. S., J. W. Verano, and A. R. Titelbaum. 2018. Trepanation procedures/outcomes: Comparison of prehistoric peru with other ancient, medieval, and american civil war cranial surgery. *World Neurosurg.* 114:245–251.

Landrum, M. J., J. M. Lee, M. Benson et al. 2018. ClinVar: Improving access to variant interpretations and supporting evidence. *Nucleic Acids Res.* 46:D1062-D1067.

LRRK2 Cohort Consortium. 2022. https://www.neurodegenerationresearch.eu/cohort/lrrk2-cohort-consortium/ (accessed May 2022).

Lobo, A., L. J. Launer, L. Fratiglioni et al. 2000. Prevalence of dementia and major subtypes in Europe: A collaborative study of population-based cohorts. Neurologic diseases in the Elderly Research Group. *Neurology* 54:S4–9.

Matikainen-Ankney, B. A., N. Kezunovic, R. E. Mesias et al. 2016. Altered development of synapse structure and function in striatum caused by Parkinson's disease-linked LRRK2-G2019S mutation. *J. Neurosci.* 36:7128–7141.

Nguyen, A. P. T., and D. J. Moore. 2017. Understanding the GTPase activity of LRRK2: regulation, function, and neurotoxicity. *Adv. Neurobiol.* 14:71–88.

Piñero, J., J. M. Ramírez-Anguita, J. Saüch-Pitarch et al. 2020. The DisGeNET knowledge platform for disease genomics: 2019 update. *Nucleic Acids Res.* 48:D845–D855.

Plassman, B. L., K. M. Langa, G. G. Fisher et al. 2007. Prevalence of dementia in the United States: The aging, demographics, and memory study. *Neuroepidemiology* 29:125–132.

Schrötter, A., Y. M. Park, K. Marcus et al. 2016. Key players in neurodegenerative disorders in focus-New insights into the proteomic profile of Alzheimer's disease, schizophrenia, ALS, and multiple

sclerosis-24th HUPO BPP Workshop: September 29, 2015, Vancouver, Canada. *Proteomics* 16:1047–1050.

Simonti, C. N., B. Vernot, L. Bastarache et al. 2016. The phenotypic legacy of admixture between modern humans and Neandertals. *Science* 351:737–741.

Steinlein, O. K. 2008. Genetics and epilepsy. *Dialogues Clin. Neurosci.* 10:29–38.

Tai, X. Y., M. Koepp, J. S. Duncan et al. 2016. Hyperphosphorylated tau in patients with refractory epilepsy correlates with cognitive decline: A study of temporal lobe resections. *Brain* 139:2441–2455.

Tsoucalas, G., K. Spengos, G. Panayiotakopoulos et al. 2017. Epilepsy, theories and treatment inside Corpus Hippocraticum. *Curr. Pharm. Des.* 23:6369–6372.

Verstraeten, A., J. Theuns, and C. Van Broeckhoven. 2015. Progress in unraveling the genetic etiology of Parkinson disease in a genomic era. *Trends Genet.* 31:140–149.

Walshe III, T. M. 2016. *Neurological Concepts in Ancient Greek Medicine.* Oxford, UK: Oxford University Press.

WHO. 2021. Dementia. https://www.who.int/news-room/fact-sheets/detail/dementia#:~:text=Alzheimer's%20disease%20is%20the%20most,dependency%20among%20older%20people%20globally (accessed 2021).

Wu, X., K. F. Tang, Y. Li et al. 2012. Quantitative assessment of the effect of LRRK2 exonic variants on the risk of Parkinson's disease: A meta-analysis. *Parkinsonism Relat. Disord.* 18:722–730.

Zhang, Q., Q. Wang, B. Kong et al. 2018. A scientific analysis of cranial trepanation from an Early Iron Age cemetery on the ancient Silk Road in Xinjiang, China. *Archaeol. Anthropol. Sci.* 10:1319–1327.