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REGULAR ARTICLE

Paediatric Acute onset Neuropsychiatric Syndrome: Exploratory study finds no evidence of HLA class II association but high rate of autoimmunity in first-degree relatives

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Abstract

Aim: Paediatric acute-onset neuropsychiatric syndrome (PANS) is defined by an acute onset of obsessive-compulsive disorder and/or eating restrictions and at least two other severe neuropsychiatric symptoms. The condition is suspected to have an immune-mediated pathophysiology, but reliable biomarkers have not been identified.

Methods: We hypothesised that PANS, like narcolepsy, might have a human leucocyte antigen (HLA) association, as found in 95% of children developing narcolepsy after H1N1 immunisation. Low resolution genotyping of the MHC class II antigens HLA-DRB1 and HLA-DQB1 was performed using two different PCR-based methods. In addition, parents were interviewed regarding a detailed family history of autoimmune diseases in first-degree relatives. A total of 18 children, aged 5–14 (mean 8.2) years at onset of PANS met symptom criteria.

Results: No evident association between PANS and the specific HLA alleles examined was observed. In first-degree relatives of 10 of the 18 children, an autoimmune disease had been diagnosed, and three of the 18 children themselves had an autoimmune disease.

Conclusion: No HLA allele association such as seen in children with narcolepsy after H1N1 immunisation could be confirmed in this group of children with PANS. However, more than half the group had a first-degree relative with a diagnosed autoimmune disease.

KEYWORDS
children, human leucocyte antigen, obsessive-compulsive disorder, paediatric acute-onset neuropsychiatric syndrome

1 | INTRODUCTION

The condition behind the acronym PANS (paediatric acute-onset neuropsychiatric syndrome) is characterised by an abrupt onset of obsessive-compulsive symptoms/obsessive-compulsive disorder (OCD) and/or eating restrictions, combined with at least two other severe neuropsychiatric symptoms. These symptoms may be separation anxiety, sensory symptoms, emotional lability, choreiform

Abbreviations: HLA, Human leucocyte antigen; OCD, Obsessive-compulsive disorder; PANS, Paediatric acute-onset neuropsychiatric syndrome.

movements and hallucinations. PANS is also a diagnosis of exclusion, which means that other neurological or medical disorders must be ruled out.[1,2]

In contrast to PANS, which is purely symptom-based and specifies no environmental or immune-related trigger, the acronym PANDAS (paediatric autoimmune neuropsychiatric disorder associated with streptococcal infections), implicates a temporal association with group A streptococcal infection.[3] An immune-mediated reaction may follow a streptococcus infection, and antistreptococcal antibodies can cross-react with basal ganglia neurons. Such autoimmune reactions may disrupt the basal ganglia-thalamocortical circuit and generate obsessive-compulsive symptoms.[4] Another example of such possible reactions is Sydenham's chorea, first described in the 17th century, a hyperkinetic movement disorder, temporally associated with a group A beta-haemolytic streptococcus infection and well known to be associated with OCD and other neuropsychiatric disorders.[5]

With regard to underlying aetiologies of PANS, a variety of disease mechanisms such as neurological, endocrine and metabolic disorders as well as postinfectious, autoimmune and neuroinflammatory disorders have been proposed.[6-9] In a recent Swedish study, a cohort of 45 children, autoimmune diseases or inflammatory disorder in first-, second-, and third-degree relatives were reported in 76% of the patients with PANS.[9]

Treatment of PANS currently involves several approaches, dependent on presumed aetiology, antibiotics and anti-inflammatory and immune modulating therapies. In addition, psychopharmacological agents and cognitive behavioural therapy can provide symptomatic relief.[7] However, to date, while some biological studies have suggested that PANS/PANDAS has an underlying immune basis, more data are needed to support or refute this association. Concerns have been raised against the assumption that inflammation has a prominent role in cases with severe, disabling psychiatric-only presentations,[10] and some researchers have pointed out that biological studies have failed to show a clear immune basis in PANS/PANDAS cases.[11]

The human leucocyte antigens (HLA) on chromosome 6 encode molecules involved in antigen presentation and inflammation regulation, and they are therefore of importance in immune-mediated autoimmune and infectious diseases.[12] A gene-environment interaction and a possible role of the immune system in the aetiology of OCD have been suggested. In the study by Rodriguez et al.,[13] the allele variability in HLA class II genes (*HLA-DRB1*, *HLA-DQB1*) was analysed in a sample of early-onset OCD and a reference sample. Pooling the different alleles that comprise HLA-DR4 (including *DRB1*04:01*, *DRB1*04:04* and *DRB1*04:05* alleles) revealed a significantly higher frequency of these alleles in the early-onset OCD sample than in a reference population. The role of HLA class II genes in the central nervous system and results supporting a role of the immune system in the pathophysiological model of OCD motivated us to analyse these genes in our current cohort of children with PANS. In a previous study,[8] we found that 48% of the children had a first-degree relative with an autoimmune disease.

Key notes

- Paediatric acute-onset neuropsychiatric syndrome (PANS) is an obsessive-compulsive disorder of unknown pathophysiology, with a presumed immune-mediated trigger.
- The possibility of a specific HLA allele association—as found in 95% of children developing narcolepsy after H1N1 immunisation—and the frequency of autoimmune diseases in first-degree relatives was investigated in 18 children with PANS.
- No HLA allele association was confirmed. Ten children (55%) had a first-degree relative with a diagnosed autoimmune disease.

In 2010, a significant increase in childhood narcolepsy was reported after H1N1 influenza vaccination in Finland and in Sweden.[14,15] A combination of genetic and environmental factors, triggering an autoimmune process leading to hypothalamic destruction with loss of hypocretin-1-containing cells, was found to be the pathogenic mechanism behind this type of narcolepsy.[16] A strong association of narcolepsy with the allele HLA-DQB1*0602 was documented, with more than 95% of the patients with narcolepsy after H1N1 vaccination carrying this allele, compared to about 25% in the general population. This finding, and the fact that high rates of neuropsychiatric comorbidity have also been observed in patients with narcolepsy,[17] further motivated our decision to analyse HLA allele types in our current PANS cohort in Gothenburg, Sweden. In addition, we investigated autoimmune diseases in first-degree relatives in more detail to address possible involvement of the adaptive immune responses in this patient group.

2 | METHODS

2.1 | Study participants

Eighteen children with PANS were included in the present study. Thirteen of these came from our previously published PANS cohort that included 23 children.[8] These 13 children and their parents had given consent to have blood tests taken. The children were 5 to 14 years old at the time of clinical assessment and blood sampling and had been comprehensively examined at the Child Neuropsychiatry Clinic in collaboration with the Gillberg Neuropsychiatry Centre (GNC) in Gothenburg after referral between 2015 and 2017. Another five children with PANS, in the same age range referred in 2018, were also included after a

similar clinical assessment and providing consent for participation in the study. Most children were from the southwest of Sweden and were referred by their local paediatrician or child psychiatrist, or, in a few cases, by the parents directly. Paediatric/neuropaediatric assessments to identify other possible medical disorders underlying the symptoms were carried out. All 18 children (11 boys) met diagnostic criteria for PANS defined according to Swedo et al.[1] with sudden onset of OCD combined with at least two other neuropsychiatric symptoms. The mean age was 8.2 years (SD = 2.1 years). Sixteen of the children were of Swedish, one of the Swedish-Southern Europeans, and one of the Swedish-Asian origins. All patients were assessed by two of the authors (EF and MJ), that is, two experienced senior consultants in paediatrics/paediatric neurology, at a clinical visit, as reported in our previous observational cohort study.[8] In all cases, the onset of PANS symptoms was preceded by a bacterial or viral infection. Four of the 18 children had previously diagnosed developmental disorder/s, ADHD and/or autism. No child had an intellectual disability.

2.2 | HLA genotyping

HLA genotyping of HLA-DRB1 and HLA-DQB1 was performed with low resolution (first field) using two different PCR-based methods. Typing for HLA-DRB1 was performed with a Luminex-based reverse PCR-SSO (LABType® SSO from OneLambda; Thermofischer). HLA-DQB1 was typed with PCR-SSP (Olerup-SSP; CareDx).

2.3 | Parental interview

In connection with the clinical assessment, parents of the 18 children were interviewed regarding a detailed family history of autoimmune diseases in first-degree relatives.

2.4 | Statistical analysis

We conducted one-sided proportion tests (prop. test in R 3.5.3) with Yates continuity correction to test whether the proportions of patients with HLA-DRB1*04 and HLA-DQB1*06 in our sample were higher than the proportions found in a sample of 966 adults in the general Swedish population.

2.5 | Ethics approval

The study was approved by the regional ethical review board in Gothenburg, Sweden (registration number 173–16). All parents and participants provided informed consent/assent after receiving oral and written information.

TABLE 1 HLA-DRB1 and HLA-DQB1 genotyping in 18 children with PANS

HLA-DRB1 PCR-SSO	HLA-DRB1 PCR-SSO	HLA-DQB1 PCR-SSP
1. DRB1*04:ANDCG	DRB1*13:ASVTF	03,06
2. DRB1*04:AJERH	DRB1*15:ASVGE	03,06
3. DRB1*04:ANEDJ	DRB1*15:EUGZ	03,06
4. DRB1*03:ASPEU	DRB1*14:ANCVP	02,05
5. DRB1*01:ATCPK	DRB1*01:03	05
6. DRB1*13:AYDDK	DRB1*13:AYDDM	06
7. DRB1*12:AUSBS	DRB1*15:AYRAZ	03,06
8. DRB1*04:AYMTR	DRB1*07:AXZBC	02,03
9. DRB1*04:AYJAV	DRB1*10:AYGBP	03,05
10. DRB1*03:AYFJM	DRB1*13:AYFJP	02,06
11. DRB1*03:BCUHG	DRB1*08:AUZYC	02,06
12. DRB1*04:AYDEF	DRB1*13:AYKVR	02,06
13. DRB1*13:AYDDK	DRB1*13:AYDDM	06
14. DRB1*13:AYJAU	DRB1*15:AYJBF	06
15. DRB1*03:BEVRA	DRB1*04:AYJCJ	02,03
16. DRB1*07:BHMVA	DRB1*14:WNUA	03
17. DRB1*01:BECUH	DRB1*07:BHMVA	02,05
18. DRB1*03:BPHRB	DRB1*15:BPHRD	02,03

3 | RESULTS

3.1 | HLA analyses

There was no association between PANS and the specific HLA-DRB1 and HLA-DQB1 alleles. Seven of the 18 children (39%) carried the HLA-DRB1*04 allele compared to 32% in the general Swedish population,[18] and 10 (55%) carried the HLA-DQB1*06 allele compared to 48% in the same Swedish population.[18] One-sided proportion tests with Yates continuity correction revealed that the proportions in our sample, did not differ significantly from the general Swedish population (HLA-DRB1*04: 95% CI of mean difference: −18.7%-32.3%, p =0.361 and HLA-DQB1*06, 95% CI of mean difference −18.5%, 33.5%, p =0.347).

Details of the HLA-DRB1 and HLA-DQB1 genotyping are found in Table 1.

3.2 | Autoimmune diseases in first-degree relatives of children with PANS

In 10 of the 18 children, parents reported that there was a first-degree family member with a diagnosed autoimmune disease: coeliac disease (n = 2; a boy with a sister and a boy with a father with the disorder), psoriasis (n = 1; a boy with a father with the disorder), ulcerative colitis (n = 1; a boy with a father and a brother

with the disorder), diabetes mellitus type 1 (n = 1; a boy with a sister with the disorder), rheumatoid arthritis (n = 1; a girl with a sister with rheumatoid arthritis), SLE (n = 1; a boy with a mother with the disorder), Guillain-Barré syndrome (n = 1; a girl with a mother with the disorder) and hypothyroidism (n = 2; a girl and a boy with a mother with the disorder). In addition, one child had a first-degree relative with a thrombocyte disorder, but not of certain autoimmune origin. Three of the 18 children (17%) themselves had a diagnosed autoimmune disease: a boy with ulcerative colitis, a boy with coeliac disease and a boy with diabetes mellitus type 1.

4 | DISCUSSION

In this exploratory study, we investigated if, in children meeting the criteria for PANS, there would be a similar strong HLA association as that reported in children with narcolepsy after H1N1 immunisation, where about 95% of the children having the allele, DQB1*0602. To the best of our knowledge, there are no published studies reporting findings from analyses of HLA specific alleles in children meeting criteria for PANS. In this sample of 18 children with PANS, the frequency of HLA specific alleles was not significantly different from the population. Studies of larger or pooled PANS cohorts, with controls matched for ethnic origin and comparison to population HLA allele frequencies, are needed to verify whether there may be an involvement of HLA in the aetiology of PANS, as these HLA alleles have been found to impact other autoimmune diseases and narcolepsy.[19,20]

An increased rate of immune-related symptoms and syndromes has been demonstrated among patients with obsessive-compulsive disorder (OCD) in comparison with other psychiatric groups.[4] Obsessive-compulsive disorder or obsessive-compulsive symptoms are core symptoms in PANS. In OCD in general, a possible role of the immune system and a gene-environment interaction model have been suggested as aetiological mechanisms. In the previously cited study from Spain,[13] a significant enrichment of HLA-DRB1*04 alleles was present in the OCD sample in comparison with the reference sample.

Based on the hypothesis that not only genetic, but also environmental factors, interactively account for susceptibility to OCD, an examination for genome-wide DNA methylation was performed on blood samples from 65 patients with OCD and 96 healthy controls in a study by Yue et al.[21] A total of 2,190 unique genes were found to be differentially methylated between OCD and healthy control subjects. These genes included *BCYRN1*, *BCOR*, *FGF13*, *HLA-DRB1* and *ARX*, which have previously been reported to be associated with OCD.

Autoimmune diseases of different types were found in the PANS children's first-degree relatives at a high rate: 55% of the children had a sibling and/or a parent with a diagnosed autoimmune disease. A similar high rate (48%) was demonstrated in our previous study.[8] The prevalence of autoimmune diseases has been estimated to be between 3%[22] and up to 9% in US populations,[23] and first-degree relatives of patients with autoimmune diseases have an increased risk of exhibiting autoimmune diseases.[23] Among the children in our study group, the rate of autoimmune diseases was 17% (3/18). Whether or not PANS represents an autoimmune disease has not been demonstrated,[11] but our findings may indicate that individuals with PANS may be prone to such diseases.

Murphy et al[24] studied history of autoimmune diseases in mothers of children exhibiting tics and/or OCD (one subgroup defined as 'likely PANDAS' and one subgroup as 'unlikely PANDAS') and reported that 17.8% of study mothers had autoimmune disorders. This prevalence rate was significantly greater than the general prevalence among women in the United States. In addition, study mothers were more likely to report having an autoimmune disease whether their children were considered 'likely PANDAS' cases versus 'unlikely PANDAS' cases.

Our results, based on children with PANS and thus with a main symptom of OCD, are in accord with findings from a nationwide Swedish study, demonstrating that the risk of any autoimmune disease was higher in first-degree relatives of individuals with OCD and/or Tourette's syndrome/chronic tic disorder—40%, 31% and 17% in mothers, fathers and siblings, respectively—than among second- and third-degree relatives of probands with OCD and these tic disorders.[25]

A limitation of our study was the small cohort that was examined with regard to HLA alleles and autoimmune heredity in first-degree relatives. Nevertheless, if this patient group had a common HLA allele type, a more homogeneous HLA allele association would have been expected. Another limitation of our study is that no HLA class I alleles were analysed as also HLA class I can be associated to autoimmune disorders. A strength of the study was the clinically well-characterised study group, where every patient was assessed by the same physicians at the same child neuropsychiatric clinic.

5 | CONCLUSION

In conclusion, this exploratory pilot study of children with PANS could not confirm any HLA class II allele association. The results, however, indicated a high rate of autoimmune diseases in first-degree relatives. Further studies of larger samples are needed to verify or disprove our results. Whether or not symptoms consistent with PANS, in the absence of an identified streptococcus infection, can be shown to represent a postinfectious, autoimmune and immune-mediated disruption of a basal ganglia-thalamocortical circuit generating OCD, will be decisive when it comes to treatment alternatives with immunomodulatory interventions.

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CONFLICT OF INTEREST

The authors have no conflicts of interest to report.

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