Children with Tourette's Syndrome May Suffer Immunoglobulin A Dysgammaglobulinemia: Preliminary Report

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Background: Postinfectious autoimmunity has been implicated in Tourette's syndrome and obsessive-compulsive disorder (TS/OCD), whereas increased frequency of upper respiratory tract infections (URTI) in TS/OCD patients suggests immune deficiency. We hypothesized that antineuronal antibodies may be elevated in patients (reflecting autoimmune processes), and levels of total immunoglobulins (Igs) may be decreased (reflecting immune deficiency).

Methods: We analyzed plasma of TS/OCD patients ($n = 24$) and healthy age- and sex-matched control subjects ($n = 22$) by enzyme-linked immunosorbent assay (ELISA) for the levels of total and specific IgG, IgM, and IgA against antigens previously identified in multiple sclerosis (myelin basic protein and myelin-associated glycoprotein) and Sydenham's chorea (ganglioside-GM1, lysoganglioside, and tubulin).

Results: Total IgA was decreased in TS/OCD patients (median 115 mg/100 mL) compared with control subjects (141 mg/100 mL; $p = .02$). Specific IgA against all antigens, except tubulin were also decreased in the patients (MPB 0 vs. 13 [ELISA units [EU]; myelin-associated glycoprotein 29 vs. 44 EU, $p = .04$; ganglioside GM1 21 vs. 35 EU, $p = .01$; lysoganglioside 44 vs. 56 EU, $p = .03$; tubulin 44 vs. 44 EU, $p = .8$). The levels of total IgA and anti-myelin basic protein (MBP) IgA were significantly lower in the subgroup of pediatric autoimmune neuropsychiatric disorder associated with *Streptococcus* (PANDAS) cases ($n = 10$) than in non-PANDAS cases ($n = 9$; total IgA 98 mg/100 mL vs. 133 mg/mL, $p = .03$; anti-MBP IgA 1 vs. 6 EU, $p = .03$) or healthy control subjects (total IgA 141 mg/100 mL, $p = .02$; anti-MBP IgA 13 EU, $p = .005$).

Conclusions: At least some TS/OCD patients may suffer IgA dysgammaglobulinemia, possibly rendering the children more prone to URTI.

Key Words: Autoimmunity, immune deficiency, immunoglobulins, Group A β-hemolytic streptococcus, PANDAS, Tourette's syndrome

Tourette's syndrome (TS) is a chronic, relapsing disorder characterized by involuntary motor and phonic tics, and obsessive-compulsive disorder (OCD) with unknown etiology.

Postinfectious autoimmunity was implied in TS/OCD because of its clinical similarity to Sydenham's chorea (SC), a component of autoimmune rheumatic fever occurring after Group A β-hemolytic *Streptococcus* (GABHS) infection (1). The concept of pediatric autoimmune neuropsychiatric disorder associated with *Streptococcus* (PANDAS) has been supported by temporary relief of symptoms in severe patients after plasmapheresis (1), the presence of antibasal ganglia antibodies in serum of TS/OCD patients (2), the cross-reactivity of antistreptococcal antibodies with neuronal epitopes (3–6), enhanced activity of T cell and NK cells in peripheral blood (7–9), and decreased numbers of regulatory T lymphocytes, the function of which is to suppress immune responses and prevent autoimmunity (10). This suggests enhanced activity of the immune system in TS/OCD patients, which is consistent with autoimmune processes. Other studies have demonstrated increased frequency of streptococcal infections and sinusitis in the patients, implying some form of immune deficiency (11,12). Simultaneous occurrence of autoimmunity and immune deficiency is not an uncommon scenario.

Neuronal circuits affected in TS/OCD involve both gray and white matter (striatum, associated limbic system, frontal cortex, and corpus callosum) (13). We hypothesized that TS/OCD patients may have increased levels of anti–basal ganglia antibodies previously shown to be elevated in SC (antibodies against ganglioside GM1, lysoganglioside, and tubulin) (6), as well as anti-myelin autoantibodies typically increased in multiple sclerosis, a white matter disorder (anti-myelin basic protein [MBP] and anti-myelin-associated glycoprotein [MAG] antibodies). We also hypothesized that the putative immune deficiency may be reflected by decreased levels of total immunoglobulins (Igs).

Methods and Materials

Subjects

Blood samples of TS/OCD ($n = 24$, Table 1) and healthy age-matched control subjects ($n = 22$, Table 1) were collected as part of three clinical studies to perform pilot investigations of immune system in TS/OCD. The Human Investigation Committee at Yale University approved these studies; all parents signed a permission statement, and each child signed a statement of informed assent. Clinical evaluation was performed as described previously using ordinal severity scales of the Yale Global Tic Severity Scale and Children's Yale–Brown Obsessive Compulsive Scale (7,10).

Blood Drawing and Analysis

Blood was drawn into heparinized vacutainer tubes (BD Biosciences, Bedford, Massachusetts) and placed on ice. Within 1 hour, blood was loaded on column of lymphocyte separation medium and spun at 400 g for 30 min to separate peripheral blood mononuclear cells and plasma. The upper

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Table 1. Demographic and Clinical Characteristics

Variable	*n*	TS/OCD Patients (*n* = 24)	Unaffected Control Subjects (*n* = 22)
Age in years (SD), range		12.9 (2.8), 7–17	13.6 (2.2), 9–17
Sex (% Female)		22%	43%
Ethnicity (% Caucasian)		96%	71%
Baseline Symptom Severity by Diagnosis			
TS (including chronic tics)[a]	20[c]	27.8 (14.1)	NA
OCD[b]	16[b]	9.5 (9.5)	
PANDAS subjects		39%	NA

OCD, obsessive-compulsive disorder; TS, Tourette's syndrome.
[a]Total tic severity score on the Yale Global Tic Severity Scale.
[b]Total score on the Children's Yale-Brown Obsessive Compulsive Scale.
[c]Number of subjects out of 24 patients who had TS (including chronic tics).

layer containing plasma was collected into Eppendorf tubes and stored at −80°C.

Analysis of Plasma Samples

The plasma samples were analyzed for total IgG, IgM, and IgA by nephelometry using the Immulite system (DPC, Los Angeles, California) and for specific antibodies to MBP, MAG, lysoganglioside, ganglioside GM1, and tubulin using the enzyme-linked immunosorbent assay (ELISA) technique as previously described (14). Coefficient of intraassay variation for IgG, IgM and IgA against all antigens was less than 6%, and coefficient of interassay variation was less than 15%.

Data Analysis

The Mann–Whitney *U* test was used to compare patients and healthy control subjects because the data did not follow normal distribution. The results are reported as medians with interquartile ranges (IQR). Multivariant comparison of PANDAS, non-PANDAS and healthy control groups was performed by Kruskal–Wallis test, and where relevant, subsequent analysis of differences between individual groups was performed by Mann Whitney *U* test. Values of $p < .05$ were considered significant.

Results

Plasma Levels of Total Ig Isotypes

TS/OCD patients had significantly lower levels of total plasma IgA (median 115 mg/100 mL, IQR 86–151) than the age-matched control subjects (141 mg/100 mL, IQR 121–170 in control subjects; $U = 145$; $n_1 = 24$, $n_2 = 22$, $p = .02$), although there were no differences in total IgG (935 mg/100 mL, IQR 746–1064 in patients vs. 977 mg/mL, IQR 803–1332 in control subjects, $U = 200$; $p = .32$) or total IgM levels (199 mg/mL, IQR 152–259 in patients vs. 209 mg/100 mL, IQR 148–240 in control subjects, $U = 232$; $p = .81$; Figure 1).

Plasma Levels of Specific Ig Isotypes

TS/OCD patients had significantly lower levels of plasma anti-MBP, MAG, ganglioside GM1, or lysoganglioside IgA than their age-matched control subjects (MPB 0 ELISA units [EU], IQR 0–5 in patients vs. 13 EU, IQR 4–28 in control subjects, $U = 99$, $p = .001$; MAG 29 vs. 44 EU, $U = 153$; $p = .04$; ganglioside GM1 21 EU, IQR 17–30 vs. 35 EU, IQR 16–45, $U = 134$, $p = .01$; lysoganglioside 44 EU, IQR 37–58 vs. 56 EU, IQR 43–105, $U = 140$ $p = .03$; Figure 2A–2D). There was no difference in antitubulin IgA between patients and control subjects (44 EU, IQR 39–54 vs. 44 EU, IQR 42–55, $U = 231$, $p = .79$; Figure 2E). Also, no differences were observed in specific IgG (MBP 18 EU, IQR 5–27 vs. 16 EU, IQR 3–41, $U = 216$; $p = .54$; MAG 30 EU, IQR 24–38 vs. 39 EU, IQR 15–46, $U = 204$, $p = .37$; ganglioside GM1 55 EU, IQR 41–63 vs. 51 EU, IQR 44–73, $U = 232$, $p = .83$; lysoganglioside 49 EU, IQR 37–58 vs. 46 EU, IQR 43–105, $U = 227$, $p = .72$; tubulin 54 EU, IQR 65–98 vs. 50 EU, IQR 76–112, $U = 220$, $p = .6$) or specific IgM (MBP 7 EU, IQR 0–26 vs. 9 EU, IQR 0–31, $U = .9$, $p = .9$; MAG 28, IQR 24–38 vs. 23 EU, IQR 15–46, $U = 234$, $p = .86$; ganglioside GM1 28 EU, IQR 18–38 vs. 27 EU, IQR 6–52, $U = 231$, $p = .81$; lysoganglioside 44, IQR 33–67 vs. 47 EU, IQR 33–83, $U = 220$, $p = .61$; tubulin 85, IQR 65–98 vs. 99 EU, IQR 76–112, $U = 205$, $p = .38$) in patients versus control subjects, respectively.

Plasma Levels of PANDAS and Non-PANDAS Cases

Among our patients, there were 19 subjects with defined PANDAS status according to a history of association between TS/OCD symptoms and GABHS infection as defined by Swedo's recommendations (15). Multivariant comparison of PANDAS cases ($n = 9$), non-PANDAS cases ($n = 10$), and healthy subjects by Kruskal–Wallis test revealed a significant differences in total IgA ($\chi^2 = 6$, $df = 2$P, $= .05$) and anti-MBP IgA levels ($\chi^2 = 14$, $df = 2$, $p = .001$). We then compared the differences between the individual groups using Mann–Whitney test. Total IgA levels were significantly lower in PANDAS versus non-PANDAS cases (98 mg/mL, IQR 82–103 vs. 133 mg/m; IQR 118–222, $U = 16$; $p = .02$) or PANDAS versus healthy subjects (98 mg/mL, IQR 82–103 vs. 141 mg/mL, IQR 121–170; $U = 47$; $p = .02$; Figure 3A). Anti-MBP IgA was decreased in PANDAS versus non-PANDAS cases (1 EU, IQR 0–0 vs 6 EU, IQR 0–10; $U = 16$ $p = .02$) and PANDAS versus healthy subjects (1 EU, IQR 0–0 vs. 13 EU, IQR 4–28, $U = 35$; $p = .005$; Figure 3B). Levels of other Igs did not significantly differ.

Discussion

TS/OCD patients have decreased total and specific IgA plasma levels in comparison with healthy age-matched children (Figures 1 and 2). This could contribute to deviation of immune responses in TS/OCD patients by at least two mechanisms. First, inhibitory functions of IgA in plasma on immune responses may be reduced (16), which could increase the vulnerability of TS/OCD patients for developing autoimmune disorders (17). Second, IgA secretion on mucosal surfaces may be affected (18,19), and in this case, the very first steps of immune defense against mucosal pathogens would be affected. This could then explain increased frequency of streptococcal infections and sinusitis in TS/OCD patients (11,12).