IgG anti-GM1 antibodies from patients with acute motor neuropathy are predominantly of the IgG1 and IgG3 subclasses

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Abstract

Increased titers of IgG anti-GM1 and anti-asialo GM1 (GA1) ganglioside antibodies are present in some patients with the Guillain-Barré syndrome, particularly with the motor axonal variant, and following infection with Campylobacter jejuni or parenteral administration of gangliosides. The subclass distribution of IgG anti-GM1 or GA1 antibodies from 19 patients with acute motor neuropathy and elevated antibody titers were measured by ELISA using mouse monoclonal antibodies specific for human IgG subclasses. The anti-GM1 or GA1 antibodies were predominantly of the IgG1 and IgG3 subclasses, which are capable of complement fixation, and are characteristic of a T cell-dependent antibody response.

Keywords: Anti-GM1 antibody; Anti-asialo GM1 antibody; Immunoglobulin G subclass; Acute motor neuropathy; Guillain-Barré syndrome

1. Introduction

Anti-GM1 ganglioside and GA1 antibodies of the IgG or IgA isotype have been reported to be increased in some patients with the Guillain-Barré syndrome (GBS), particularly with the motor axonal variant of the disease (Yuki et al., 1990; Gregson et al., 1991; Latov et al., 1991; Walsh et al., 1991; Garcia-Guigo et al., 1992; Ilyas et al., 1992; Nobile-Orazio et al., 1992; Latov, 1993; Vriesendorp et al., 1993). Increased titers have been reported to occur in approximately 67% of patients who developed GBS following parenteral administration of gangliosides which contain GM1 (Landi et al., 1993), and in 20–68% of patients with the Chinese paralysis syndrome or with GBS following gastrointestinal infection with Campylobacter jejuni which contain lipopolysaccharides with GM1-like oligosaccharides (Walsh et al., 1991; Aspinall et al., 1992; Obayashi et al., 1993; Vriesendorp et al., 1993; Kornberg et al., 1994). In both cases, the antibody response is directed at oligosaccharide determinants in GM1, GA1 or the cross-reactive lipopolysaccharides.

Immunoglobulins are divided into subclasses IgG1 to IgG4. The subclasses differ in their heavy chain constant regions and in their regulatory and biological properties (Flanagan and Rabbitts, 1982; Papadea and Check, 1989). We therefore determined the subclass distribution of the IgG anti-GM1 and GA1 antibodies from patients with acute motor neuropathy to help understand their relation to the disease.

2. Materials and methods

The subclass distribution of IgG anti-GM1 or GA1 antibodies from 19 patients with acute motor neuropathy and elevated autoantibody titers of > 1600 (Kinsella et al., 1994) was determined by enzyme-linked immunosorbent assay (ELISA) using mouse monoclonal antibodies specific for each IgG subclass.

Five of the patients developed symptoms after parenteral injection of gangliosides; six developed GBS following infection with Campylobacter jejuni or a diarr-
rheal illness, and the remaining eight had no identifiable prodrome.

ELISA was performed as described by Sadiq et al. (1990) with the following modifications. Microwells coated with 0.5 µg of GM1 or GA1/well were incubated overnight at 4°C with patient serum at dilutions of 1:100, 1:500, or 1:1000, depending on the total anti-GM1 antibody titer. For each serum, the optical density values for the four subclasses were determined at the same serum dilution for comparison. After washing, the wells were incubated with biotinylated mouse monoclonal subclass-specific antibodies to Human IgG1, IgG3 or IgG4 diluted 1:1000, IgG2 diluted 1:5000 (Sigma Chemical Co., St. Louis, MO), or affinity-purified antibodies to human total IgG at a dilution of 1:200, followed by peroxidase-conjugated avidin (Sigma Chemical Co.). Dilution of the mouse monoclonal antibodies to each IgG subclass was determined by ELISA using human IgG myeloma protein of each IgG subclass at 1 µg/ml as antigen.

3. Results

The results of IgG anti-GM1 subclass determinations for the 19 patients tested are given in Table 1. In 16 of the cases, including in patients whose neuropathy followed infection with Campylobacter jejuni, or parenteral injection of gangliosides, the IgG anti-GM1 antibodies were predominantly IgG1 or IgG3. In general, the optical densities obtained using the anti-IgG subclass antibodies were lower than those obtained using anti-total IgG antibodies, and in three cases (Patient No. 1, 3 and 11), there was only weak reactivity to GM1 or GA1 detected using the subclass-specific antibodies. None of the patients had predominance of IgG2 or IgG4 antibodies. The subclass-specific antibodies were equally sensitive in detecting their respective subclasses as shown in preliminary studies using class-specific myeloma IgGs as antigens. In most cases, the anti-GM1 antibodies were of the same predominant subclass as the GA1 antibodies, although in one patient the anti-GA1 antibodies were predominantly IgG1 and IgG2.

4. Discussion

In this study, the IgG anti-GM1 and GA1 antibodies from patients with acute motor neuropathy were predominantly IgG1 and IgG3. The weaker reactivity seen with subclass-specific antibodies, as compared with anti-total IgG antibodies, may be due to the fact that the monoclonal anti-subclass antibodies recognize only a single epitope, or that the reactive epitopes are lacking in some of the patients’ anti-GM1 antibodies. IgGs of these subclasses differ from IgG2 and IgG4 antibodies as they are capable of fixing complement and bind to Fc receptors in monocytes, neutrophils, and lymphocytes (Papadea and Check, 1989). They are therefore more likely to cause tissue destruction and may contribute to the neurological disease. Pathologi-

### Table 1

<table>
<thead>
<tr>
<th>Patient</th>
<th>IgG anti-GM1 antibodies Titer</th>
<th>IgG anti-GA1 antibodies Titer</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 B.O.R.</td>
<td>&gt; 1/100,000 0.0 0.1 0.1 0.1</td>
<td>&gt; 1/100,000 2.2 0.1 0.1 0.1</td>
</tr>
<tr>
<td>2 P.R.A.</td>
<td>1/10,000 1.3 0.2 0.2 0.2</td>
<td>1/10,000 0.9 0.1 0.1 0.2</td>
</tr>
<tr>
<td>3 P.R.Y.</td>
<td>1/100,000 0.2 0.0 0.0 0.0</td>
<td>1/100,000 0.1 0.1 0.1 0.1</td>
</tr>
<tr>
<td>4 D.E.M.</td>
<td>1/10,000 2.0 0.3 1.7 0.2</td>
<td>1/10,000 2.0 0.2 1.7 0.1</td>
</tr>
<tr>
<td>5 K.U.M.</td>
<td>1/10,000 0.7 0.2 0.5 0.0</td>
<td>1/10,000 1.3 0.2 0.4 0.0</td>
</tr>
<tr>
<td>6 S.H.I.</td>
<td>1/10,000 0.5 0.3 0.3 0.1</td>
<td>1/10,000 0.2 0.2 0.1 0.0</td>
</tr>
<tr>
<td>7 P.A.S.</td>
<td>1/6400 1.1 0.1 0.3 0.3</td>
<td>1/100,000 1.4 0.2 0.4 0.2</td>
</tr>
<tr>
<td>8 A.N.D.</td>
<td>1/10,000 0.8 0.3 0.5 0.1</td>
<td>&gt; 1/100,000 1.6 0.3 0.3 0.2</td>
</tr>
<tr>
<td>9 P.A.M.</td>
<td>&gt; 1/100,000 0.8 0.0 0.1 0.1</td>
<td>&gt; 1/100,000 1.6 0.3 0.7 0.0</td>
</tr>
<tr>
<td>10 T.U.C.</td>
<td>1/100,000 0.3 0.0 0.2 0.2</td>
<td>1/100,000 0.4 0.1 0.2 0.1</td>
</tr>
<tr>
<td>11 S.E.A.</td>
<td>&gt; 1/100,000 0.2 0.0 0.0 0.0</td>
<td>1/10,000 0.1 0.1 0.0 0.0</td>
</tr>
<tr>
<td>12 P.A.L.</td>
<td>&gt; 1/100,000 0.5 0.1 0.1 0.1</td>
<td>1/100,000 1.5 0.5 0.2 0.1</td>
</tr>
<tr>
<td>13 C.O.L.</td>
<td>1/3200 0.7 0.1 0.1 0.0</td>
<td>1/12800 1.9 1.2 0.3 0.0</td>
</tr>
<tr>
<td>14 O.Z.A.</td>
<td>1/10,000 0.8 0.1 0.2 0.0</td>
<td>1/10,000 1.4 0.1 0.2 0.1</td>
</tr>
<tr>
<td>15 K.O.I.</td>
<td>1/100,000 0.9 0.4 0.4 0.2</td>
<td>1/100,000 0.8 0.2 0.4 0.2</td>
</tr>
<tr>
<td>16 K.O.C.</td>
<td>&gt; 1/100,000 0.5 0.2 0.4 0.2</td>
<td>&gt; 1/100,000 0.3 0.2 0.2 0.1</td>
</tr>
<tr>
<td>17 C.A.P.</td>
<td>&gt; 1/100,000 0.3 0.0 0.0 0.0</td>
<td>&gt; 1/100,000 0.1 0.1 0.0 0.0</td>
</tr>
<tr>
<td>18 R.U.F.</td>
<td>1/100,000 0.3 0.2 0.1 0.0</td>
<td>1/10,000 2.5 0.5 0.4 0.0</td>
</tr>
<tr>
<td>19 P.E.R.</td>
<td>&gt; 1/100,000 0.5 0.3 0.1 0.3</td>
<td>&gt; 1/100,000 0.5 0.2 0.5 0.2</td>
</tr>
</tbody>
</table>

The titer of total IgG anti-GM1 and GA1 antibodies, and the optical density obtained for each IgG subclass are presented for each patient. Patients Nos. 1–6 developed GBS following infection with Campylobacter jejuni or a diarrheal illness. Patients Nos. 7–11 developed GBS following parenteral injection of gangliosides. Patients No. 12–19 had no identifiable prodrome.
ical studies of nerves from patients who died with acute motor axonal neuropathy following *Campylobacter jejuni* infection indicate that the lesion is non-inflammatory, consistent with an antibody-mediated disease (Mckhann et al., 1993). A similar predominance of the IgG1 or IgG3 subclasses were also reported for anti-glycolipid autoantibodies in other patients, including in two patients with neuropathy and anti-GM1 antibodies reported by Garcia-Guijo et al., (1992), and in patients with anti-GQ1b antibodies and the Miller-Fisher variant of GBS (Willison and Veitch, 1994). Other autoantibodies including Anti-Hu antibodies in paraneoplastic encephalomyelitis (Jean et al., 1994), and autoantibodies in non-neurological immune diseases have also been reported to be predominantly of the IgG1 or IgG3 subclasses (Rubin et al., 1986b; Watson et al., 1986; Forouhi et al., 1987; Mahmoud et al., 1990; Sokol et al., 1990; Loizou et al., 1992; Prada and Strife, 1992; Rothfield, 1992; Sammaritano and Gharavi, 1992).

The IgG1 and IgG3 antibodies which are directed at the GM1 and GA1 oligosaccharides are atypical for anti-carbohydrate antibodies which are typically T cell-independent. Antibodies to exogenous carbohydrate antigens including to dextran, teichoic acids, group-A streptococcal polysaccharide, and pneumococcal capsular polysaccharide, are predominantly IgG2, although in some cases there is also an IgG1 response. In contrast, IgG antibodies of the IgG1 and IgG3 subclasses are typically T cell-dependent, and directed at protein antigens such as tetanus toxoid. IgG4 antibodies are frequently induced by allergens originating in parasites, insect venoms, and foods (Zouali et al., 1984; Rubin et al., 1986a; Rubin et al., 1986b; Shackelford et al., 1987; Papadea and Check, 1989). The anti-GM1 and GA1 antibodies in acute motor neuropathy might therefore also be derived by T cell-dependent mechanisms, possibly directed at the carbohydrate antigen in association with a carrier protein.

The subclass distribution of the IgG antibodies in a T cell-dependent response is determined in part by the associated T cells and cytokines released during the immune response (Noma et al., 1986; Papadea and Check, 1989; Coutelier et al., 1991; Snapper et al., 1992). Alternatively, the IgG subclass distribution may be restricted by the responding B cell. For example, human leukemic B cells which are CD5+ preferentially transcribe and express IgG1 and IgG3 genes (Hashimoto et al., 1992; Sideras et al., 1992), which are closely linked on chromosome 14 (Flanagan and Rabbitts, 1982; Papadea and Check, 1989). Since anti-GM1 antibody-secreting B cells may also belong to the CD5+ subpopulation (Graves and Ravindranath, 1992), they too may be similarly restricted. Of interest, lipopolysaccharide has been reported to preferentially activate CD5+ B cells in the mouse (Su et al., 1991). Further studies into the mechanisms regulating the IgG antibody response would further help our understanding of the pathogenesis of the disease.

References


