Intravenous immunoglobulin infusion therapy in women with recurrent spontaneous abortions of immune etiologies

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Abstract

We have investigated clinical effectiveness of intravenous immunoglobulin G infusion (IVIg) on antiphospholipid antibody titers in five women with evidence of antiphospholipid antibody-associated recurrent spontaneous abortions and one with antinuclear antibody who became refractory to conventional autoimmune treatment during pregnancy and experienced pregnancy complications. Three women developed intrauterine growth retardation and three had complicated twin pregnancies with rising autoantibody titers. Antiphospholipid antibody and antinuclear antibody titers were tested pre and 2 weeks after each IVIg infusion. We report that: (i) IgG antiphospholipid antibody titers were significantly suppressed after each IVIg infusion (P < 0.05); (ii) IgM antiphospholipid antibody titers were also significantly suppressed after each IVIg infusion (P < 0.0001); (iii) decreased titers of autoantibodies paralleled increased levels of maternal IgG which lasted for at least 30 days; the autoantibodies showed a definite rise again prior to the next infusion; (iv) antinuclear antibody titers were effectively suppressed; and (v) rising autoantibody titers combined clinical manifestation of intrauterine growth retardation and women with complicated twin pregnancies. We conclude that IVIg infusion effectively suppresses IgM and IgG autoantibodies to phospholipids and antinuclear antibody in autoimmune women with a history of recurrent spontaneous abortions and refractory to conventional anticoagulation or immunosuppressive treatment.

Keywords: Recurrent spontaneous abortion; Antiphospholipid antibody; Antinuclear antibody; Immunoglobulin G

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1. Introduction

High dose intravenous immunoglobulin therapy is reported to be clinically beneficial in a variety of immune disorders associated with pregnancy, such as Rh-sensitization (de la Cámara et al., 1988), hypogammaglobulinemia (Smith et al., 1985), recurrent fetal loss caused by antiphospholipid antibodies (Scott et al., 1988; Parke et al., 1989), and idiopathic thrombocytopenia (Rose and Gordon, 1985). However, contradictory results of intravenous immunoglobulin G infusion (IVIg) therapy on Rh-sensitization and recurrent fetal loss (Wapner et al., 1989) are also reported.

The effects of high dose immunoglobulin G infusion includes feedback inhibition of autoantibody synthesis (Bussell et al., 1988), down-modulation of the IgG Fcγ receptor (FcγR) (Mannhalter et al., 1987), blockade of placental transport of maternal endogenous IgG, regulation of the idiotype network (Sultan et al., 1984), alteration of T-cell and B-cell functions, and natural killer cell activity in the mother and infant (Tsubakio et al., 1983). IVIg infusion is reported to effectively lower anti-cardiolipin antibody levels in a woman with recurrent spontaneous abortions (Scott et al., 1988).

A subset of women with recurrent spontaneous abortions (RSAs) who receive alloimmune and autoimmune therapy reported by us are refractory to conventional autoimmune treatment and continue to experience intrauterine growth retardation or pregnancy losses (Kwak et al., 1992b). Antiphospholipid antibody titers are up-regulated in women with poor pregnancy outcome (Kwak et al., 1994a). In normal pregnant women, antiphospholipid antibody titers remained persistently negative throughout the pregnancy (Kwak et al., 1994b). These findings suggest that suppression of autoantibodies to phospholipids is essential for the successful pregnancy outcome. Anticoagulation and/or immunosuppression were widely applied in women with a history of RSA and autoimmune abnormalities. In women who are refractory to conventional autoimmune treatment, IVIg therapy can be an alternative treatment for suppressing autoantibodies. In addition, potential side-effects of high dose heparinization or immunosuppression can be avoided with IVIg infusion treatment. In this study we followed six women with RSAs who were refractory to conventional autoimmune treatment and received IVIg infusion treatment. Antiphospholipid antibody titers, antinuclear antibody titers and serum concentration of immunoglobulin G, M and A were serially measured.

2. Materials and methods

2.1. Patients

Five women with evidence of antiphospholipid antibody-associated recur-
rent spontaneous abortions and one with antinuclear antibody who were refractory to conventional autoimmune treatment comprised the study group. The patients were enrolled at the Reproductive Immunology Clinic, University of Health Sciences, The Chicago Medical School. IVlg infusion treatment was started when autoantibody titers were rising even with the autoimmune treatment.

All had 3 or more RSAs of clinically unexplainable causes. None had genetic, anatomical, infectious, or hormonal causes for previous pregnancy losses. None had documented abnormal fetal karyotypes on any of prior abortus tested. None had histories of Raynaud’s phenomena, blood transfusion, photosensitivity, arthralgia, arthritis, serositis, thrombosis, hemolytic anemia, thrombocytopenia or neutropenia. All patients had significant titers of antiphospholipid antibodies or antinuclear antibodies and demonstrated a lack of alloimmune recognition by flow cytometric analysis of maternal anti-paternal lymphocyte antibody before the index pregnancy. Paternal lymphocyte immunization was given before the index pregnancy and all achieved an adequate level of maternal anti-paternal lymphocyte antibodies before the index pregnancy as previously reported (Kwak et al., 1992b).

2.2. Treatment

Patients began autoimmune treatment and preconceptionally utilized ASA 80 mg/day and heparin 5000 U from 48 h after ovulation (Table 1) (Kwak et al., 1992b). Heparin was discontinued when the contraindication was detected. Heparin therapy was contraindicated in women with subchorionic hematoma, severe vaginal bleeding, or vanishing twin with hematoma in the gestational sac. Otherwise heparin was stopped at 34 weeks of gestation. Prednisone was started 5 mg twice daily from 48 h after ovulation and increased to 10 mg twice daily with positive pregnancy test in women with positive antinuclear antibody. All patients received intravenous immunoglobulin G 400 mg/kg/day for 3 days when indicated and repeated monthly up to 34 weeks gestation. The mean IVIg initiation time was 17.2 ± 7.9 weeks gestation.

2.3. Laboratory evaluation

Antiphospholipid antibody test was done by ELISA which includes autoantibodies to cardiolipin, phosphatidylethanolamine, phosphatidylserine, phosphatidylinositol, phosphatidic acid and phosphatidylglycerol as previously reported (Kwak et al., 1992a). Titers were calculated by using the standard titration curve as previously reported (Kwak et al., 1994a). Titer equal to or greater than 1:50 was considered as a positive result. Only positive autoantibodies to phospholipids were traced on the graph of each patient (Figs. 1–5).
Table 1
Status of patients enrolled in the study

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age</th>
<th>G</th>
<th>SAB</th>
<th>IVIg initiated (wks gestation)</th>
<th>Clinical findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>33</td>
<td>5</td>
<td>5</td>
<td>24</td>
<td>Intrauterine growth retardation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Intrauterine growth retardation,</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>oligohydroamnios</td>
</tr>
<tr>
<td>2.</td>
<td>23</td>
<td>5</td>
<td>5</td>
<td>12</td>
<td>Intrauterine growth retardation,</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>gestational hypertension</td>
</tr>
<tr>
<td>3.</td>
<td>42</td>
<td>4</td>
<td>4</td>
<td>12</td>
<td>Twin pregnancy, subchorionic hematoma,</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>vaginal bleeding</td>
</tr>
<tr>
<td>4.</td>
<td>37</td>
<td>6</td>
<td>6</td>
<td>4</td>
<td>Twin pregnancy, subchorionic hematoma,</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>vaginal bleeding, vanishing twin</td>
</tr>
<tr>
<td>5.</td>
<td>33</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>Twin pregnancy, subchorionic hematoma,</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>vaginal bleeding</td>
</tr>
<tr>
<td>6.</td>
<td>33</td>
<td>8</td>
<td>8</td>
<td>12</td>
<td>Twin pregnancy, subchorionic hematoma,</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>vaginal bleeding</td>
</tr>
</tbody>
</table>

IVIg, intravenous immunoglobulin G; SAB, spontaneous abortion.

ANA was tested by immunofluorescence assay using HEp2 cells as previously reported (Kwak et al., 1992a). Maternal IgG, IgM and IgA serum concentration was measured pre and 2 weeks post IVIg infusion.

2.4. Statistical analysis

Pre and post infusion antiphospholipid antibody titers were compared with Wilcoxon matched-pairs signed-rank-test. Paired t-test was applied for pre and post infusion IgG, IgM and IgA level. A $P$ value of less than 0.05 was reported to be significant.

3. Results

Age, gravidity, number of spontaneous abortions, clinical findings, and time of IVIg initiation are listed in Table 1.

3.1. Antiphospholipid antibody and antinuclear antibody

Antiphospholipid antibodies were effectively suppressed after IVIg in-
sion (Figs. 1–5). IgG antiphospholipid antibodies were significantly suppressed after IVIg infusion ($P < 0.05$). IgM antiphospholipid antibodies were also significantly suppressed after IVIg infusion ($P < 0.0001$).

Antinuclear antibody titer was suppressed effectively with repeated IVIg infusion, however it mostly remained pre-conception titer (Fig. 6).

3.2. Immunoglobulin level

Figs. 1–6 illustrate IgG, IgM and IgA serum concentrations pre and post IVIg infusion. The serum IgG level was significantly elevated post IVIg as

Fig. 1. Tracing of antiphospholipid antibody and immunoglobulin levels during the index pregnancy with IVIg therapy on patient 1. The patient had 5 previous spontaneous abortions. Only positive antiphospholipid antibodies were traced on the graph. IgM CL, IgM anti-cardiolipin antibody; IgM PA, IgM anti-phosphatidic acid antibody; IgM PE, IgM anti-phosphatidylethanolamine antibody; IgM PG, IgM antiphosphatidylglycerol antibody; IgM PI, IgM phosphatidylinositol antibody.
Fig. 2. Tracing of antiphospholipid antibody and immunoglobulin levels during the index pregnancy with IVIg therapy on patient 2. The patient had 5 previous spontaneous abortions. Only positive antiphospholipid antibodies were traced on the graph. IgG CL, IgG anticardiolipin antibody; IgG PA, IgG antiphosphatidic acid; IgG PI, IgG antiphosphatidylinositol antibody; IgG PS, IgG antiphosphatidylserine antibody.

compared with the pre IVIg serum IgG level ($P = 0.007$) (Table 2). IgM and IgA levels were not different when compared with pre and post IVIg serum level.

3.3. Reproductive outcome

Six pregnancies were reported. Three women developed intrauterine growth retardation with rising antiphospholipid antibody titers. Three women with complicated twin pregnancies demonstrated rising antinuclear antibody titers with conventional autoimmune treatment (Table 3). The
mean gestational age of the live born infants was 35.8 ± 1.9 weeks and mean birth weight was 2374.7 ± 716.4 g. Five women delivered prematurely. Three infants were small for gestational ages.

4. Discussion

IVIg therapy appears to be of therapeutic benefit for the fetoplacental unit experiencing severe compromise early in pregnancy for women with a history of RSA and autoimmune serological abnormalities. Autoantibodies to adhe-
Fig. 4. Tracing of antiphospholipid antibody and immunoglobulin levels during the index pregnancy with IVIg therapy on patient 4. The patient had 6 previous spontaneous abortions. IgG CL, IgG anti-cardiolipin antibody; IgG PG, IgG phosphatidylglycerol antibody; IgG PI, IgG phosphatidylinositol antibody; IgG PS, IgG antiphosphatidylserine antibody.

sion molecules such as phosphatidylserine and phosphatidylethanolamine were reported to inhibit the process of cytotrophoblast to syncytiotrophoblast formation. Phosphatidylinositol is an important placental adhesion molecule for amnion formation (Ohno et al., 1992). Antibodies to these fusion molecules may ultimately result in early organ failure, defective placentation as well as hypercoagulation. IVIg therapy may down-regulate the levels of these autoantibodies. We have reported a high rate of successful deliveries in most of these women following achievement of alloimmune recognition and autoimmune treatment started during the pre-implantation period of pregnancy (Kwak et al., 1992b). These women showed a greater in-
Fig. 5. Tracing of antiphospholipid antibody and immunoglobulin levels during the index pregnancy with IVIg therapy on patient 5. The patient had 6 previous spontaneous abortions. IgM CL, IgM anti-cardiolipin antibody; IgM PA, IgM phosphatidic acid antibody; IgM PI, IgM phosphatidylinositol antibody; IgM PS, IgM antiphosphatidylserine antibody.

cidence of placental immunopathology and incomplete transformation of myometrial spiral arteries to fully developed uteroplacental arteries which could seriously impair the normal increase in placental perfusion required to meet the needs of the growing fetus (Bronsens et al., 1967).

Our study focuses on a subgroup of patients whose pregnancies seemed destined for failure early in the second trimester despite what could be described as heroic therapy to that point (Branch et al., 1985). In most women, we observed post IVIg therapy decreased titers of autoantibodies that paralleled increased levels of maternal IgG which lasted for at least 30
Fig. 6. Tracing of antinuclear antibody and immunoglobulin levels during the index pregnancy with IVIg therapy on patient 6. The patient had 8 previous spontaneous abortions.

Table 2
Pre and post immunoglobulin G infusion serum IgG, IgM and IgA concentration in women with a history of recurrent spontaneous abortions

<table>
<thead>
<tr>
<th></th>
<th>Pre infusion serum concentration (mg/dl) (mean ± S.D.)</th>
<th>Post infusion serum concentration (mg/dl) (mean ± S.D.)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgG</td>
<td>1393.6 ± 291.8</td>
<td>2075.6 ± 573.2</td>
<td>0.007</td>
</tr>
<tr>
<td>IgM</td>
<td>267.9 ± 137.4</td>
<td>248.9 ± 135.2</td>
<td>NS</td>
</tr>
<tr>
<td>IgA</td>
<td>273.7 ± 123.9</td>
<td>223.6 ± 109.8</td>
<td>NS</td>
</tr>
</tbody>
</table>

NS, not significant.
Table 3
Reproductive outcome after intravenous immunoglobulin G therapy

<table>
<thead>
<tr>
<th>Patient</th>
<th>Weeks gestation</th>
<th>Delivery method</th>
<th>Sex</th>
<th>Weight (g)</th>
<th>APGAR</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>32</td>
<td>C/S</td>
<td>M</td>
<td>1007.2</td>
<td>6/7</td>
<td>IUGR, PROM</td>
</tr>
<tr>
<td>2.</td>
<td>35</td>
<td>C/S</td>
<td>M</td>
<td>1675.1</td>
<td>7/8</td>
<td>IUGR, fetal distress</td>
</tr>
<tr>
<td>3.</td>
<td>34</td>
<td>C/S</td>
<td>M</td>
<td>1532.2</td>
<td>6/8</td>
<td>IUGR</td>
</tr>
<tr>
<td>4.</td>
<td>36</td>
<td>C/S</td>
<td>M</td>
<td>2748.8</td>
<td>9/9</td>
<td>Twin</td>
</tr>
<tr>
<td>5.</td>
<td>35</td>
<td>V/D</td>
<td>M</td>
<td>2807.7</td>
<td>9/9</td>
<td>Breech</td>
</tr>
<tr>
<td>6.</td>
<td>37</td>
<td>C/S</td>
<td>M</td>
<td>2664.9</td>
<td>9/9</td>
<td>Twin</td>
</tr>
</tbody>
</table>

AGA, adequate for gestational age; C/S, cesarean section; IUGR, intrauterine growth retardation; PROM, premature rupture of membrane; V/D, vaginal delivery.

days and showed a definite rise again before the next infusion. Rising autoantibody titer was noticed with the development of intrauterine growth retardation and in women with twin pregnancies and obstetrical complications while on conventional autoimmune therapy.

In this study, we documented that titers of autoantibodies to phospholipids and antinuclear antibodies decreased with IVIg. The suppression of autoantibody was transient and lasted for 4 weeks. IVIg preparation derived from large pools of plasma may contain anti-idiotypic antibodies which may down-regulate autoantibody synthesis (Dietrich et al., 1992). Recently, Vassilev et al. reported that IVIg contains antibodies to CD5 molecules (Vassilev et al., 1993). CD5+ B cells were reported to be elevated in patients with autoimmune diseases (Dauphinee et al., 1989) and related with polyreactive antibody production (Casali and Notkins, 1989). Human CD5 antibodies in IVIg allow therapeutic immunoglobulin preparations with the potential of modulating T-cell functions through CD5 and/or regulating the expression of B-cell subsets expressing CD5. This may have implications for the suppression of IgM and IgA class antibodies in treated women.

Since commercial IVIg preparation does not contain IgM fractions and a very low amount of IgA, lack of IgM or IgA increase after IVIg was the expected outcome. It is of interest to find that the IgM class autoantibodies were also suppressed after IVIg. Modulation of CD5+ B cells by CD5 antibodies in IVIg may be a major mechanism of IVIg on autoantibody suppression. It is also possible that IVIg centrally suppresses the production of activated lymphocytes of the NK series that may be involved in damage to or retardation of growth of trophoblast.
Decidual vascular lesions of a necrotizing or inflammatory nature in the placenta of patients with systemic lupus erythematosus (SLE) have been reported (Abramowski et al., 1980). In addition, antinuclear antibody can traverse the placenta to the fetus and can penetrate into the living cells. DNA/anti-DNA complexes can be detected on the trophoblast basement membrane of the placenta in women with SLE (Grennan et al., 1978). Kiutttu et al. (Kiutttu et al., 1994) reported an 18.6% perinatal mortality rate in the ANA-positive women. In the same study, the perinatal mortality of infants of women with negative ANA was 0%. Infants with congenital AV heart block are reported in women with high ANA titer of complement fixing activity. A congenital complete heart block, cardiomyopathy or a congenital heart disease and inflammatory changes, calcification and fibrosis have been detected in the conduction system of embryos from mothers with systemic lupus erythematosus (McCue et al., 1977; Chameides et al., 1979). Although there are controversial reports about the role of ANAs in women with RSAs (Rosenberg et al., 1986), we suggest that ANA should be measured in women with recurrent spontaneous abortions or other fetal losses of unknown etiology.

Although we applied IVIg on a monthly basis, since the catabolism of IVIg is both state and concentration dependent, the dosage cannot be based on half-life values. More constant IgG levels may reduce catabolism because the rate is concentration dependent at high IgG concentration. More consistent serum levels may be obtained by decreasing the interval between infusions, dampening the metabolic perturbation resultant from extreme peak and trough level (Wedgwood, 1991).

The observed results of IVIg infusion in women with RSA although very encouraging, have to be considered preliminary in nature. Women with a history of RSA and rising autoantibody titers even with conventional auto-immune therapy should be treated promptly to avoid the risk of developing intrauterine growth retardation. Careful monitoring of autoantibody titers while on autoimmune therapy in women with a history of RSAs and twin pregnancies is advocated.

References


