大韓不妊學會誌: 第 32 卷 第 1 號 2005
Kor. J. Fertil. Steril., Vol. 32, No. 1, 2005, 3

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Principle and Application of Immunotherapy in Women with Recurrent Pregnancy Losses and Infertility of Implantation Failures: Intravenous Immunoglobulin G Infusion Treatment

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INTRODUCTION

Reproductive immunology has become a mature principle and opened a clinical application of immunotherapy for reproductive failures. Recurrent pregnancy losses and multiple implantation failures after repeated assisted reproductive technology (ART) cycles of unknown etiologies are conditions that immunotherapy is often considered. Recurrent spontaneous abortion occurs in 2~5% of couples attempting to reproduce1 and a significant proportion of recurrent aborters are either subfertile or infertile. The observed frequency of recurrent pregnancy losses is reported to be higher than expected frequency by chance (0.3%). In a 1998 Center for Disease Control report, 8.7% of infertile couples received a diagnosis of unexplained infertility. These patients often receive multiple assisted reproductive cycles including IVF/ET arbitrarily. Although they do not have any abnormalities per commonly accepted infertility work up, their live birth rates (25.4%) with ART cycle are not significantly different than those who have a known cause(s) for infertility. These imply that recurrent pregnancy losses, infertility and assisted reproductive failures have underlying causes, which have not been properly explored.

Intravenous immunoglobulin G (IVIg) infusion, anticoagulation, immuno-suppression using steroid, or paternal lymphocyte immunization have been reported as potential immunotherapy for women with recurrent pregnancy losses or multiple implantation failures after repeated ART cycles. IVIg infusion treatment has been utilized as an alternative treatment for the paternal lymphocyte immunization in women with recurrent pregnancy losses.[2] Later it was demonstrated that IVIg contains anti-idiotype antibodies against autoantibodies,[3] which suppresses antiphospholipid antibody production in women with RSA.[4,5] IVIg has been reported to down regulate the elevated numbers of CD56+ NK cells found in women with RSA.[6,5] In this review, IVIg treatment for recurrent pregnancy losses and multiple implantation failures after repeated ART failures, its rationale and clinical trials are discussed.

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RATIONALE

Recent studies of women with recurrent spontaneous abortions and infertility women with repeated IVF failures show them to be T helper 1 activated by a variety of in vitro studies and flow cytometry[7,8] T helper 2 activation and T helper 1 suppression is the rule in healthy women with healthy pregnancies.[9] It has become clear that a large percentage of patients with recurrent spontaneous abortions and infertility are autoimmune and T helper 1 activated. The effectors that damage the conceptus are auto-antibodies and T helper 1 cytokines produced by natural killer cells. Because of this progress the American Society for Reproductive Immunology had a consensus conference about diagnosis and treatment of these autoimmune disorders.[10]

The underlying causes for recurrent pregnancy losses, infertility and assisted reproductive failures are not entirely clear. American Society for Reproductive Immunology had consensus conference on June 23, 1996. The committee presented options for diagnosis and treatment of recurrent pregnancy losses.[10] The committee report specifies clinical information being useful, including maternal age, number of prior abortions, number of previous live births, history of infertility and history of autoimmune disease. The committee also specifies that autoimmune parameters including anticardiolipin antibody, antiphospholipid antibody, antinuclear antibody and peripheral blood NK cell level have "evidence of value", and leukocyte antibody detection assay, NK functional assay and embryocytotoxicity assay have "value being tested". Commonly used clinical evaluation, such as hysteroscopy, ultrasound, microbiologic test such as Chlamydia and Ureaplasma culture, endocrine tests (prolactin, thyrotropin, endometrial biopsy, day 8 serum LH, 2 hour pp blood sugar) is specified as "value being tested". Prior to initiating any treatment for women with recurrent pregnancy losses, infertility and assisted reproductive failures, proper clinical evaluation is crucial. It is suggested that the evaluation of autoantibody abnormalities in all cases of suspected autoimmune-associated reproductive failure is valuable and will improve clinical care of affected patients.[11]

NK cell and IVIg treatment

The role of peripheral blood NK cell (CD16+/56+) has been reported as the basis for failure of early pregnancy. NK cell recognized the trophoblast cells.[12] Although NK cells cannot kill trophoblastic cells in vitro, NK cells activated by cytokines (TNF-alpha, Interferon gamma or IL-2) may kill trophoblast cells in vitro.[13] Indeed recent study showed that women with recurrent pregnancy losses, infertility and assisted reproductive failures have significantly increased activated peripheral blood NK cell levels than normal fertile controls.[14] In addition, activated NK cells can produce cytokines that are abortogenic.[15] Endometrial bed biopsies from women experiencing recurrent pregnancy losses and infertility of unknown etiology reveal an increase in conventional NK cell (CD56+/16+, CD57+ cells).[16,17] Peripheral blood NK cells (CD56+, CD56+/16+) are significantly elevated in women with recurrent pregnancy losses, infertility and assisted reproductive failures as compared with normal control women.[18,19] Quantitation of peripheral blood NK cells of women with recurrent pregnancy losses and infertility of assisted reproductive failures have shown a significant elevation associated with spontaneous abortion of a conceptus of normal karyotype, and abnormal level associated with loss of embryos that are karyotypically abnormal.[5,20] Further more, increased peripheral blood NK cell cytotoxicity has predictive value of pregnancy losses.[19,21] Thus, conventional NK cells and failure to suppress NK cell activation plays an

important role in immunologically preventable spontaneous abortions. Intravenous immunoglobulin G infusion treatment has been shown to down-regulate NK cell killing capacity[18,22] and enhances CD8+ cell activity.[16] Both of these events may be necessary for a successful pregnancy to occur.

Currently four randomized, controlled trials of IVIg for treatment of recurrent pregnancy losses have been published or finalized. A European-based study showed a positive trend but did not achieve statistical significance due to too few patients for adequate statistical power given the magnitude of the effect.[23] U.S. based trial, however, showed a significant benefit. This prospective trial showed 64% success rate in IVIg group and 34% in controls ($p<0.04$).[20] Although the number of study subjects in the U.S. based trial was not greater than European-based study, positive results are interpreted due to the greater magnitude of study effect in the U.S. based study. The greater magnitude of study effect could be related with study design. Patients started IVIg treatment prior to conception in U.S. based trial, but after implantation in European based trials. Previously, we have reported that pre-conceptional immunotherapy has 2 times higher success rate than that of post conception treatment in women with recurrent pregnancy losses.[24] The updated-third trial was presented in International Congress of Reproductive Immunology Meeting, 2001 in Croatia. The study was limited to secondary aborters only and recurrent second trimester abortions. The IVIg trial groups have significantly higher success rate than placebo controls.[25] This study was an extension of the previous trial,[26] which reported that an expected 55% therapeutic gain of IVIg in recurrent spontaneous abortion could not be confirmed using the treatment regimen tested.

A meta-analysis of the randomized spontaneous abortion revealed an overall relative risk of 1.38 ($p=0.02$)[27] and these results show that IVIg is effective in treatment of recurrent spontaneous abortion. With the addition of updated data from Christiansen, it is expected to have a more significant IVIg outcome data for women with recurrent pregnancy losses.

Antiphospholipid antibody and IVIg treatment

Recent data demonstrated that IVIg inhibits the thrombogenic effects of antiphospholipid antibodies in vivo and reduces the levels of anti-cardiolipin antibodies in the circulation. Blockade of stimulatory Fc gammaR on inflammatory cells is not necessary for this effect. The mechanism of action on IVIg is more likely saturation of the IgG transport receptor, leading to accelerated catabolism of pathogenic antiphospholipid antibodies. These results have implications in the management of thrombosis in antiphospholipid antibodies and may have applications for pregnant patients with a history of antiphospholipid antibody syndrome.[28] Each treatment with IVIg resulted in a reduction of anti-cardiolipin antibodies. A partial transient reduction of antiphospholipid antibody levels was observed immediately following each treatment course resulting in an accelerated fetal outcome.[5,29]

In women with antiphospholipid antibody syndrome, IVIg treatment improved pregnancy outcome, with significantly lower pregnancy complication rates, when compared with prednisone plus low dose aspirin treatment.[30] In the prednisone and low dose aspirin treated patients, gestational hypertension and gestational diabetes were reported to be significantly more often than in the IVIg-treated group. Recent review of Antiphospholipid Antibody Syndrome (APS) by Sherer, et al.[31] concluded that APS, an autoimmune disease whose main features are vascular thrombosis and pregnancy morbidity, is a good candidate for immunotherapy with IVIg that contains anti-idiotypes directed to wards patients' pathogenic antiphospholipid anti-

bodies.[31] Systemic lupus erythematosus and antiphospholipid antibody syndrome are associated with an increased risk of intrauterine growth retardation, miscarriage, stillbirth, and preterm delivery. Recent advances in therapy during pregnancy have improved the outcome but there is still significant fetal and maternal morbidity and mortality. Treatment of patients failing conventional therapy during the second half of pregnancy is difficult and may be complicated by the development of preeclampsia. The addition of intravenous immunoglobulin therapy offers a low risk strategy for reducing autoantibody-mediated disease and improving placental function in severely compromised growth restricted pregnancies.[32] Low dose IVIg treatment (200 mg/Kg) was also reported to be effective in women with antiphospholipid antibody, antithyroid antibody, antiovarian antibody, increased immunoglobulin M levels and increased natural killer cells.[33]

Controversial study has been also reported. IVIg treatment was reported to be no more effective than heparin and low-dose aspirin in the treatment of pregnancies complicated by antiphospholipid syndrome in a small size clinical trial, but has not been adequately evaluated in refractory cases.[34] In addition, there are no randomized studies using IVIg in women with elevated NK activity and presence of antiphospholipid antibodies. Although experience remains limited and uncontrolled, intravenous immunoglobulin (IVIg) therapy probably has a place in the management of selected patients with the antiphospholipid syndrome. It seems effective for the prevention of recurrent pregnancy losses when conventional strategies using subcutaneous heparin and low-dose aspirin have failed.[5] IVIg is currently investigated in the treatment of recurrent in vitro fertilization failure associated with antiphospholipid antibodies. It is speculated that IVIg associated with steroids and heparin might improve survival in the rare but life-threatening catastrophic antiphospholipid syndrome.[35]

Intravenous immunoglobulin is safe, but expensive. Despite its expense, if IVIG is shown to markedly decrease maternal and fetal morbidity, it may be the logical treatment of choice to prevent pregnancy loss in APS.[36]

IVF failures and IVIg

Idiopathic infertile women with multiple IVF failures demonstrate significantly higher level of CD56+ lymphocytes than normal fertile controls and the conception rate is much higher in those with CD56+ level less than 12%.[18] In addition, infertile and recurrent spontaneous abortion women who fail alloimmune and autoimmune therapy have significant alteration in cellular and humoral immunity involving NK cells and CD19+/CD5+ B cells.[18] IVIg treatment suppresses NK cells and CD19+ B cells and favorable pregnancy outcome is reported.[18]

In women with antiphospholipid antibodies, heparin/aspirin alone improved IVF birthrates. This benefit is selective in that it does not apply in cases in which IgG or IgM APAs are directed against phosphatidylethanolamine or phosphatidylserine. In such cases the addition of IVIg significantly improves the outcome.[37] In addition, in women with repeated IVF failures, IVF outcome is significantly improved when heparin/aspirin and IVIg are administered in women with positive antiphospholipid antibodies.[37] Intravenous Ig is useful in the treatment of unexplained IVF failure in women who have oocyte fertilization rates > or = 50% and generate at least three embryos per cycle.[38]

CONCLUSION

Randomized double blind study has been accepted as the standard for clinical trials for drug efficacy in Unites States. However, IVIg treatment

for recurrent pregnancy losses or infertility of implantation failures has its limitation for double blind randomized trial due to 1) heterogeneous nature of disease which requires significantly higher number of study participants; 2) cost concern for clinical trial due to high cost of IVIg; and 3) concerns about legal issues in relation to pregnancy outcome from clinical trial. Clinicians need, however, to recognize the limitations of IVIg clinical data and have to adjust their clinical management to the degree and quality of patient evaluation available to them in their community. Further investigation is warranted for IVIg treatment in women with recurrent pregnancy losses and in vitro fertilization failure associated with antiphospholipid antibodies and elevated NK cells.

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