POSSIBLE MECHANISMS OF IMMUNOTHERAPY FOR MAINTAINING PREGNANCY AGAINST RECURRENT SPONTANEOUS ABORTERS

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INTRODUCTION
An immunotherapy has been performed on patients who repeated spontaneous abortions of unknown causes, and this immunization has shown favorable results. With this therapy the immunological response to the fetus must have been artificially controlled. Thus, these patients are convenient for the investigation of the immune mechanism of maintaining pregnancy.

This study targets the anti-HLA antibodies developed in the maternal serum. Analyses were made on the specificity of the alloantibodies and the change in activities of these antibodies. Investigation of difference in the immune response between the immunized patients and the non-immunized controls may explain the mechanism of retaining pregnancy.

MATERIALS AND METHODS
The patients were consisted of three groups, that were normal pregnancy group (229 cases), spontaneous abortion group (60 cases) and immunotherapy group (44 cases, 48 pregnancies).

A schedule of paternal lymphocyte immunization and sampling of the test serum is shown in Fig.1.

The method for detecting the anti-HLA antibodies was based on the complement-dependent cytotoxicity of the maternal serum against the paternal B-lymphocytes. In order to investigate the specificity of detected anti-HLA antibodies, the serum samples were treated with the procedure of ultra-centrifugation and platelet-absorption. The obtained serum before and after the procedure of platelet-absorption were used for the detection of specificity of anti-HLA class I and II antigens, respectively. Panel cells used for cytotoxicity test consisted of the cells with known antigens that covered all of paternal HLA class I and II antigens. For class II antigens, 13 kinds of homozygous typing cells (HTC) were applied.

RESULTS
1. Reproductive outcomes of immunotherapy (Tab.1)
The success ratio was about 80% or more in subgroups (A), (B) and (D), while it was very low at about 30% in subgroup (C).
2. Incidence of anti-HLA antibodies (Tab. 2)

The total incidence of antibodies showed significant differences among the spontaneous abortion group, the normal pregnancy group and the immunotherapy group. The most important finding was that comparing the cases of successful pregnancy and failed pregnancy in this group, the incidence was much higher in the former than in the latter.

Tab. 2

| INCIDENCE OF ANTI-HLA ANTIBODIES IN THE LAST HALF OF THE 1ST TRIMESTER OF PREGNANCY |
|---------------------------------|------------------|------------------|
| positive no./pregnant no. (× 100) | normal pregnancy group | spontaneous abortion group |
| progamda                         | 14/82 (17.1)      | 0/12 (0.0)       |
| progamda                         | 37/142 (26.1)     | 2/42 (4.8)       |
| Immunotherapy group              |                   |                  |
| success                          | 29/36 (80.6)      | 5/12 (41.7)      |
| secondary abortion               | 2/4 (50.0)        | 3/24 (12.5)      |
| primary abortion                 | 3/8 (37.5)        | 34/48 (70.8)     |
| secondary abortion               | 2/6 (33.3)        |                  |

A significant difference (*p < 0.05 by chi-square test*) is found in a.d.e.f.g.h.i.j.k.l.m.n.o.p.q.r.s.t.u.v.w.x.y.z combination. No significant difference is found in a.b.e.g.h.i.j.k.l.m.n.o.p.q.r.s.t.u.v.w.x.y.z.

3. Titration of anti-HLA antibodies

Primary aborters maintaining pregnancy showed increased incidence and titer of anti-HLA antibodies with increasing times of immunotherapy and advancement of pregnancy. In the cases who failed in abortion, all showed no antibody during and after the immunotherapy. One case aborted at 12 weeks of pregnancy developed the antibodies with a low titer only 2 to 4 folds (Fig. 2).
Secondary aborters maintaining pregnancy showed high incidence of antibodies after the 1st immunization with relatively high titer. This condition was maintained until 12 weeks of pregnancy. In aborted cases, similar to the cases with successful pregnancies, antibodies had already been developed after the 1st immunization. This finding was different from the pattern of failed cases in primary aborter. ( Fig.3 )

4. Specificity of anti-HLA antibody (Tab.3)
Representative 5 cases in the immunotherapy group who showed positive seroconversion of anti-HLA antibody were investigated. The anti-HLA antibodies against the incompatible paternal antigens were not always developed in the patient's sera after paternal lymphocyte immunization. However it should be emphasized that the developed antibodies did not react with the panel cells that were incompatible with the husband's type but react only with the panel cells compatible with the husband's type.
DISCUSSION

Although an allograft as the fetus should be rejected with the immune response, the fetus survives and grows up in the maternal uterus. Therefore there should be some kinds of immunological mechanism in order to overcome this rejection.

HLA antigens play an important role in the rejection against allograft. This is why attention has become to be paid to HLA antigens to clarify the mechanism with which pregnancy is safely maintained.

The focal point in this study is a significance of anti-HLA antibodies in respect to maintenance of pregnancy. On the occasion of discussing the anti-HLA antibodies developed by paternal lymphocyte immunization, essentially important factor is whether the detected antibody is true anti-HLA antibody or not. Antibody detected in this experimental system was confirmed to be husband-specific anti-HLA antibody. Then our results of incidence of the antibody indicate that spontaneous aborters, specially recurrent aborters, have lower immune response against fetal antigens than mothers with normal pregnancy. However the immunotherapy makes recurrent aborter to show high immune response. Thus anti-HLA antibody developed might play a key role in immunological maintenance of pregnancy. It is not still unclear how anti-HLA antibody acts on the mechanism of maintenance of pregnancy, but monitoring of the activities of anti-HLA antibody is a useful tool to predict the prognosis of pregnancy after immunotherapy.

REFERENCES

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