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Analysis of the use of cyclosporin A to treat refractory immune recurrent spontaneous abortion

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Summary

Purpose: This study aims to determine the curative effect of cyclosporin A (CsA) in treating refractory immune recurrent spontaneous abortion (RSA). Patients with recurrent abortion caused by dysimmunity were enrolled. *Materials and Methods:* The patients were given aspirin, prednisone, heparin, immunotherapy with their husband's leukomonocyte, and intravenous immunoglobulin (IVIG) treatment, but treatment outcomes were unsuccessful. Therefore, CsA was added to treat the women before and after pregnancy. During treatment, CsA concentration was maintained at 80 ng/ml to 150 ng/ml. The clinical effect and pregnancy outcome were observed. *Results:* Of the 26 patients, 20 cooperated and accomplished complete pregnancy. Twelve cases showed hypertensive disorders during pregnancy but did not exhibit symptoms of preeclampsia. Three cases were lost to follow-up. The success rate was 76.92%. Twenty patients underwent premature labor (34 weeks to 37 weeks). Nevertheless, the mothers and their children were all healthy. *Conclusion:* An appropriate dose of CsA has good curative effects and pregnancy results in the treatment of RSA.

Key words: Cyclosporine; Recurrent spontaneous abortion; Pregnancy.

Introduction

Reproductive immunology studies recently suggested that approximately 50% to 60% of etiologies for recurrent spontaneous abortion (RSA) are related to immunologic derangement. Successful rate of treatment has exceeded 90% because of the development of mechanism research and treatment of recurrent abortions. Immune abortion is generally classified into two types: autoimmunity and alloimmunity.

Among the RSAs caused by immune factors, applications of prednisone, aspirin, low-molecular-weight heparin, and human gammaglobulin achieved the most successful pregnancy rate in our hospital (>85%). The immune antibody reaction of women before and after pregnancy accounted for the unsuccessful pregnancy rate of 15%. Thus, a question arises as to how immune antibodies can be made negative or how to reduce immune antibody content in the blood of pregnant woman. Du *et al*. [1] reported that an appropriate dose of cyclosporine A (CsA) contributed to the dual-regulation role in mother-fetus immunoloregulation, not only by suppressing immunologic rejection of the mother's body to the embryonic antigen, which causes failed pregnancies, but also by promoting the growth, movement, and invasiveness of cytotrophoblast cells. Therefore, CsA is a potential drug for treating pregnancy diseases, such as the RSA. CsA is a widely used drug for patients receiving organ transplantation and can effectively reduce autoimmunity. Many researchers believe that no adverse pregnancy effect can result from using conventional doses of CsA and prednisone for the prevention of immunologic response during organ transplantation. Therefore, these immunosuppressive agents are additionally used before and after pregnancy for women who cannot be successfully treated by four drugs.

Combining CsA with current treatment approaches reduces the immune antibody content in blood and thereby decreases injuries of gestational sac, fetus, placenta, and uterus caused by the antibody for pregnancy success, thus ensuring a normal newborn.

RSA is a medical condition characterized by three or more consecutive pregnancy losses and has an incidence rate of approximately 5%. Since the 1990s, the present hospital has applied prednisone, aspirin, heparin, and intravenous immunoglobulin (IVIG) treatments for RSAs caused by dysimmunity, and the success rate has exceeded 85%. However, treatment for 15% of the patients still failed. This condition is defined as refractory RSA and is the focus of this study. The treatment failures for these patients can mainly be attributed to the immune antibody titers of the currently applied treatment regimens, which cannot reduce pregnancy losses. In this study, CsA was found to be an effective immunosuppressive agent that serves a double-regulation function in maternal-fetal immunoregulation. On one hand, CsA inhibits maternal immunologic rejection against fetal antigen. On the other hand, CsA promotes the growth, migration, and invasive-

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Table 1. — *The general data of 26 cases pregnant women with refractory recurrent miscarriage.*

Clinical	Features	Cases
Age (years old)	< 30	6
	≥ 30-40	13
	≥ 40	7
Times of abortion	5	6
	6-7	14
	≥ 8	6
Miscarriage or premature birth time	< 13	6
	≥ 13-30	11
	≥ 30	9
Abortion cause	Embryo growth stop	10
	FGA and less amniotic fluid	7
	Preeclampsia and its complications[a]	9
The checked RSA reasons (positive)	ACA	6
	LA	10
	Resistance to beta 2-GP1 antibodies	9
	Resistance to any of nuclear antibodies	1

[a] Complications included abruption, stillbirth, oligohydramnios, HELLP syndrome, and so on.
FGA= fetal growth restriction; ACA= anti-cardiolipin antibodies; LA= lupus anticoagulant

ness of Langhans cells. Thus, CsA may become an effective drug for pregnancy-associated diseases, such as RSA. CsA has long been used for treating pregnant women receiving organ transplantation during gestation. These patients are subject to long-term exposure to normal doses of CsA and prednisone in case of immunologic rejection after organ transplantation. No report exists on the influence of CsA on fetuses and newborns. Therefore, the results of CsA combination for 26 RSA patients are reported in this study.

Materials and Methods

Clinical data

General data: Two hundred fifty-six cases of patients with RSA were selected from February 2008 to June 2011. All patients were examined for etiologies of RSA in the present hospital and other hospitals. Cases with genital malformation and dysfunction, chromosome abnormality, cryptorrhea, infection, and hereditary diseases were excluded. Among the immune reasons (ACA, LA, β2-GP1, and ANA), 156 cases exhibited a positive antibody. Conventional systematic treatment was applied to 132 cases (prednisone, aspirin, heparin, and IVIG). However, pregnancy occurred again and 26 newborns did not survive. The general conditions of the 26 cases of pregnant women with treatment failure are shown in Table 1.

Treatment method

Cases with positive APS (ACA; LA; anti-β2-GP1 antibody) or anti-nuclear antibody (ANA) were examined. Some patients had a treatment history with more than two of following treatment regimens, whereas for patients with one treatment before CsA, Regimen (3) was implemented. Four treatment regimens were employed (if the blocking antibody of the patients was negative, immune therapy using the lymphocytes of the husband was additionally performed before gestation until the antibody turned positive).

Regimen (1): Aspirin 80 mg and prednisone 60 mg were jointly administered for two to three months before pregnancy. If at this time, the antibody became negative (within the normal range), administration of all drugs was stopped, and the antibody was examined weekly. However, after the antibody became positive (beyond the normal range), drug administration was immediately conducted. Moreover, prednisone 30 mg and aspirin 80 to 100 mg were jointly administered until one to three days before the end of pregnancy.

Regimen (2): Hypodermic injection of low-molecular weight heparin 5,000 IU for q12 h + Regimen (1).

Regimen (3): IVIG 400 mg/kg·d for five consecutive days once monthly + Regimen (1) + Regimen (2).

Regimen (4) (for the 26 patients in this study): This regimen was discussed with the patients after Regimen (3) (prednisone, aspirin, heparin, and IVIG) failed. With the consent of patients, CsA 50 mg bid or tid was additionally administered with Regimen (3) 30 d to 60 days before pregnancy. CsA concentration in the blood was then examined at 15 and 30 days. The dosage of this drug was maintained between 80 and 150 ng/ml. This dosage was continuously administered until after pregnancy. If the antibody became negative, the dose of CsA was reduced gradually. If the positive antibody was recurrent, CsA was again administered until childbirth.

Results

Among the 26 patients treated with Regimen (4), three lost visits occurred at the medium stage of pregnancy, 18 patients exhibited increased blood pressure at 15 to 34 weeks of pregnancy (gradually rose to 160/100 mmHg), one presented fetus malformation abortion at 26 weeks of pregnancy, and two intrauterine fetal deaths occurred at 13 and 22 weeks, respectively. Between 33 weeks and 37 weeks plus two days of pregnancy, 20 cases became parturient because of the premature rupture of membrane (PROM), and fetuses were prematurely born because of intrauterine distress. Cesarean section was conducted for 12 cases, and parturient parturition was conducted for five cases. Oxytocin odinopoeia was performed for the five cases caused by PROM. Results of the antibodies before and after CsA application are summarized in Table 2. Maternal conditions before and after parturition are shown in Table 3, and the results of newborns are shown in Table 4.

Discussion

The probability of RSA for patients with previous spontaneous abortions ≥ three times and positive anti-phospholipid antibody (ACA, LA, anti-β2-GP1) reaches up to 90%. Decidua and extensive intravascular thrombosis and infarction of placenta are possibly the main pathological bases of abortions caused by anti-phospholipid antibody. Damage to vascular endothelial cells and intravascular thrombosis may result from phospholipid antibody binding with the phospholipid/phospholipid binding protein (β2-GP1) compound. Aggravation of intravascular thrombosis of placenta thereby damages placental function and finally causes abortion [2, 3].

Table 2. — *The decreased antibody contents beyond the maximum normal value in 26 pregnant women before and after the application of CsA.*

	Within the normal range	< 20%	< 50%	< 100%	> 100%
At the time of miscarriage after the last time of treatment failure (the non-CsA and control groups)	0	0	1	11	14
After CsA administration (until the time of childbirth)	8	6	7	1	1

Table 3. — *General date of 26 puerperas.*

	Cases			Pregnancy period (weeks)		Postpartum hemorrhage		Amniotic fluid	Delivery pattern	
	Succeed	Failure	Lost to follow up	33-35	36-37	<500 ml	>500 ml	Meconium staining	Cesarean section	Vaginal delivery
Cases	20	3	3	8	12	18	3	3	10	11
(%)	76.92	11.53	11.53	34.78	52.17	78.26	13.04	13.04	43.47	47.82

Table 4. — *Results of 20 newborns.*

Cases	Birth weight	Apgar score (χ±s)		CRP or breathing machine treatment[a]		Asphyxia[b]		Transfer NICU	
		1 min	5 min	Cases	%	Cases	%	Cases	%
20	2865±601	9.5±0.6	9.9±0.1	4	2.6	3	3.3	2	10

Note: [a] underwent CRP or breathing apparatus within 24 hours after birth; [b]included respiratory distress syndrome and transient tachypnea.

Pregnancy failure, such as RSA, is a problem that distresses gynecology and obstetrics doctors. The existing treatment mechanism is unclear, and the results reported by various researchers are inconsistent [4-6]. Therefore, more effective methods of treating pregnancy failures must be sought. After being treated by prednisone + anticoagulant, heparin combined with aspirin [7, 8] + simple IVIG or IVIG combined with anticoagulant, patients with RSA caused by dysimmunity and with positive anti-phospholipid and antinuclear antibodies achieved a successful pregnancy rate of > 85%. Many studies showed that IVIG was effective for treating RSA patients with positive anti-phospholipid antibody and that the treatment of low-molecular weight heparin combined with aspirin or prednisone was ineffective. Moreover, the above methods still failed in treating RSA caused by dysimmunity, with a resultant pregnancy success rate of approximately 10% to 15%. Although the gestational weeks increased, adverse pregnancy results, such as stillbirth in the early or medium stage of pregnancy or recurrent preeclampsia, still occurred. With the consent of the patients and their families, CsA was added for treatment. In 36 months, a total of 26 patients were involved in this therapy. As a result, 20 cases succeeded, three cases lost visits, and three cases presented stillbirth because of fetus umbilical hernia (one case) and FGA (two cases).

CsA can retard other pathways involved in immunosuppression, as discovered by Kuprash *et al*. [9] They found that CsA suppresses the expression of the LTα subunit of TNFα (member of TNF family) and then serves its immunosuppression function.

Immune RSA is caused by immune antibody, and CsA is an immunosuppressive agent that exerts treatment effects by inhibiting the generation or increase of immune antibodies. CsA effectively reduces autoimmunity and is widely used for patients receiving organ transplantation. According to foreign reports [10-12], a conventional dose of CsA and prednisone for long-term therapy to prevent immunologic rejection during organ transplantation can cause no adverse pregnancy result. Moreover, CsA within the effective dose range can stimulate the growth, morphological change, and invasiveness of cytotrophoblast cells during early pregnancy [13]. Among the patients for whom received prednisone, aspirin, heparin, and IVIG treatments were infective, 26 patients were selected to receive CsA in combination with their current treatment. Twenty cases showed successful results. After CsA application, the blood antibody content evidently decreased compared with that in non-CsA patients. Twenty-one cases presented an antibody content <50% beyond the normal maximum value, accounting for 80.76% of the studied group. This percentage also exhibited a significant difference when compared with the group that did not receive CsA (3.8%). Although all pregnant women presented severe gestational hypertension after 13 weeks of pregnancy during the treatment process, no kidney injury resulted in proteinuria and edema. The fetuses grew normally. Thus, the gestational weeks of the patients were delayed as much as possible. However, 20 patients entered the labor stage in less than 38 weeks of pregnancy and experienced premature birth. The premature newborns, particularly the two cesarean newborns, were transferred into the NICU for treatment. Their neonatal complications were more severe than those of the normal newborns. All 19 cases of newborns left hospital with their mothers five days postpartum, except for one,

who stayed in the hospital for 30 days because of hypoxic ischemic encephalopathy. Nevertheless, this newborn still left the hospital as a healthy child. The newborns did not exhibit any abnormality even at 42 days postpartum. Although the blood pressure of the ten cases of mothers was reduced, they were diagnosed as hypertensive patients. Other conditions returned to the state before pregnancy.

CsA is a better prospect in the treatment of RSAs resulting from dysimmunity and cases with unknown causes because of its immunoloregulation in vivo. However, a large-sample randomized controlled study result remains lacking. Thus, more extensive studies must be conducted.

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