

Hemolytic Disease of the Newborn Associated with Anti-Jr^a Alloimmunization in a Twin Pregnancy: The First Case Report in Korea

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Jr^a is a high-frequency antigen found in all ethnic groups. However, the clinical significance of the anti-Jr^a antibody has remained controversial. Most studies have reported mild hemolytic disease of the newborn and fetus (HDNF) in Jr^a-positive patients. Recently, fatal cases of HDNF have also been reported. We report the first case of HDNF caused by anti-Jr^a alloimmunization in twins in Korea. A 33-yr-old nulliparous woman with no history of transfusion or amniocentesis was admitted at the 32nd week of gestation because of vaginal bleeding caused by placenta previa. Anti-Jr^a antibodies were detected in a routine laboratory examination. An emergency cesarean section was performed at the 34th week of gestation, and 2 premature infant twins were delivered. Laboratory examination showed positive direct antiglobulin test and Jr(a+) phenotype in the red blood cells and the presence of anti-Jr^a antibodies in the serum in both neonates. The infants underwent phototherapy for neonatal jaundice; this was followed by conservative management. They showed no further complications and were discharged on the 19th postpartum day. Preparative management to ensure the availability of Jr(a-) blood, via autologous donation, and close fetal monitoring must be performed even in cases of first pregnancy in Jr(a-) women. (*Korean J Lab Med* 2010;30:511-5)

Key Words : Anti-Jr^a, Neonatal jaundice, Fetal erythroblastosis, Hematologic pregnancy complications

INTRODUCTION

Jr^a is a high-frequency antigen that has been included in the 901 series (antigen number, 901005) of the International Society of Blood Transfusion blood group classification [1]. The frequency of occurrence of this antigen is known to be over 99% in most ethnic groups [2]. Although the Japanese population is known to have the highest

number of Jr(a-) individuals, the incidence of the Jr(a-) phenotype is estimated to be only 0.03-0.12% even in the Japanese population [3], and this percentage is thought to be lower in Caucasians, Mexicans, and Arabs [4]. The incidence of the Jr(a-) phenotype in the Korean population has not yet been reported.

Jr^a was first reported in 1970 by Stroup and MacIlroy [5], who identified 5 cases in which anti-Jr^a antibody was detected during the assessment of a high frequency antigen in the Caucasian population. Although case reports regarding anti-Jr^a antibody have been published occasionally over the past 40 yr, its clinical significance has not been well established. This is mainly because of the diversity of patient outcomes and scarcity of reported data. The 2 main clinical scenarios associated with

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anti-Jr^a positivity are incompatible transfusions and hemolytic disease of the newborn and fetus (HDNF). In the former scenario, no biologic or clinical evidence of hemolysis due to mild, acute, or delayed transfusion reactions has been reported for individuals with anti-Jr^a antibodies [6]. In the latter scenario, limited availability of data has led to conflicting results [6]. We present a case of neonatal jaundice associated with anti-Jr^a alloimmunization in premature twins; this case occurred during the first pregnancy of a Korean woman. This is the first report documenting HDNF related to anti-Jr^a alloimmunization in Korea.

CASE REPORT

A 33-yr-old nulliparous woman in her 32nd gestational week visited the emergency room because of vaginal bleeding. Her medical history was unremarkable except for the fact that she was a hepatitis B carrier. She had no history of blood transfusion or amniocentesis. She had a twin pregnancy after in vitro fertilization and embryo transfer (IVF-ET). A routine obstetric examination revealed that vaginal bleeding was caused by placenta previa, and the patient was admitted to undergo conservative perinatal management.

Initial laboratory tests showed a low hemoglobin level (10.9 g/dL). The antibody-screening test (Search-Cyte; Medion Diagnostics AG, Düringen, Switzerland) yielded weakly positive, irregular results in both the reagent cells only in the antiglobulin phase. The patient's ABO and RhD blood type was O+. Antibody identification test (Data-Cyte; Medion Diagnostics AG) revealed the presence of a pan-reactive antibody that showed weak positivity against all the 11 reagent panel cells in the antiglobulin phase. The results of the antibody identification tests conducted at room temperature and at 37°C with albumin enhancement were negative. The indirect antiglobulin test (IAT) with autologous control and direct antiglobulin test (DAT) also yielded negative results. The antibody characteristics indicated that the corresponding antigen was a high-frequency red blood cell (RBC) anti-

gen. Screening of the patient's RBC phenotype with a panel of antisera showed that the RBCs were positive for k, Di^b, and Lu^b. A negative reaction against Jr^a antiserum prepared in-house confirmed the patient's Jr(a-) phenotype. Crossmatching of the patient's serum and previously confirmed Jr(a-) O+ RBCs yielded negative results.

Because of frequent uterine contraction and vaginal spotting, an emergency cesarean section was performed at the 34th week of gestation. The patient delivered 2 premature male infants. Their body weight and Apgar scores at 1 and 5 min after birth were as follows: baby 1, 1.91 kg, 4, and 7; baby 2, 2.2 kg, 5, and 8. Both babies underwent phototherapy for the treatment of neonatal jaundice. The results of the laboratory tests conducted on the 4th day of birth are shown in Table 1. The ABO/RhD blood type of baby 1 and baby 2 were O+ and A+, respectively. The antibody screening and identification test results of the 2 infants were similar to those of their mother. In both tests, the serum of the 2 infants weakly reacted with all the reagent-panel RBCs only in the antiglobulin phase. IAT with autologous control yielded positive results in both infants. The RBCs of both infants showed reactivity with Jr^a antiserum; this confirmed the Jr(a+) phenotype (the infants' RBCs were treated with chloroquine to dissociate the antibodies on the RBC surface before testing). Crossmatching with the infants'

Table 1. Results of laboratory tests conducted on the neonates on the 4th day of birth

	Baby 1	Baby 2
Hemoglobin (g/dL)	14.6	13.8
Hematocrit (%)	43.6	41.3
WBC count ($\times 10^3/\mu\text{L}$)	14.78	10.74
Platelet count ($\times 10^3/\mu\text{L}$)	279	363
Bilirubin, total (mg/dL)	8.4	8.1
Bilirubin, direct (mg/dL)	1.1	0.9
LDH (IU/L)	522	394
ABO cell type	O	A
RhD	Positive	Positive
Direct Coombs' (IgG)	Positive (3+)	Positive (1+)
Direct Coombs' (IgA)	Negative	Negative
Direct Coombs' (IgM)	Negative	Negative
Direct Coombs' (C3d)	Negative	Negative
Direct Coombs' (C3c)	Negative	Negative

Abbreviations: WBC, white blood cell; LDH, lactate dehydrogenase.

serum and previously confirmed Jr(a-) O+ RBCs showed negative results. Antibody-identification test with the eluate of the infants' RBCs was not performed because of the inadequate quantity of the referred blood sample. Taken together, these data indicated the presence of anti-Jr^a antibodies of maternal origin in the twins' serum and the role of these antibodies in the positive DAT results in the neonates. The infants were discharged after conservative management on the 19th postpartum day, without further complications.

DISCUSSION

Although the anti-Jr^a antibody was identified nearly half a decade ago, its clinical significance with regard to hemolytic reactions has not been clearly established. This is mainly because of the paucity of reported data. In most of the cases described in the literature, HDNF did not occur, and when it did occur, its intensity was mild to moderate, and hence, no treatment beyond phototherapy was required [3, 4, 7-13]. A few cases of incompatible Jr(a+) RBC transfusion with no or delayed hemolytic transfusion reactions have also been reported [14-16].

However, recent evidence has shown that anti-Jr^a antibodies have the potential for causing fatal HDNF and acute hemolytic reactions. Acute hemolytic transfusion reaction caused by anti-Jr^a antibodies has been reported in a 45-yr-old Japanese woman [15]. Peyrard et al. [6] reported the first case of fatal HDNF in a 28-yr-old Caucasian woman of Spanish Gypsy origin. She had an obstetric history of 2 abortions and 1 full-term pregnancy (G4P1). At the time of the first abortion, she developed massive hemorrhage that required transfusion of 10 units of RBCs. Fetal cardiomegaly associated with intrauterine growth retardation was confirmed at the 35th week of her 4th pregnancy. An emergency cesarean section was performed at the 36th week. The newborn was hydropic, had severe anemia, and died 30 hr after birth. Anti-Jr^a antibodies, which were later identified in the mother's serum, were thought to be the cause of severe fetal anemia. Arriaga et al. [17] also reported a similar

fatal case of hydrops fetalis associated with anti-Jr^a antibodies in a 39-yr-old Caucasian woman with a history of multiple pregnancies. An earlier case reported in 2006 also described severe fetal anemia in a 40-yr-old Japanese woman (G4P3), but the infant was born healthy after 4 intravascular transfusions via the umbilical cord [18].

The common feature of the 3 cases of HDNF described above is that the mothers had multiple pregnancies with or without transfusion history. In many previously published cases reporting no or mild to moderate clinical HDNF associated with anti-Jr^a antibody positivity, the Jr(a-) pregnant women had no transfusion history [6]. Thus, it can be presumed that the clinical significance of the anti-Jr^a antibody may depend on the frequency of exposure to the Jr^a antigen. Previous studies have shown that anti-Jr^a antibodies may develop during pregnancy and that they can cross the placenta [3, 9]. The Jr^a antigen has also been reported to be fully developed at birth [7]. Although anti-Jr^a antibodies can be stimulated by a first pregnancy, HDNF associated with anti-Jr^a antibody was thought to occur in subsequent pregnancies [6]. However, in our case, the mother was nulliparous and had no history of transfusion or amniocentesis. The present pregnancy was the only identifiable cause of alloimmunization. Although confirmative findings such as negative antibody screening test results before the present pregnancy were not available, it should be noted that the findings in this case suggest that HDNF can occur in a first pregnancy. This may probably be the reason for the low severity of HDNF. Another interesting finding was that the DAT results for both neonates were positive and that the positivity in the DAT result of baby 1, whose ABO/Rh blood type was O+, was greater than that in the DAT result of baby 2. The reasons for the differences in DAT results are not clear. The effect of having twins on anti-Jr^a alloimmunization requires further investigation. Although we could not evaluate the eluates of the twin's RBCs because of lack of specimens, the presence of anti-Jr^a antibodies in the twin's serum and the Jr(a+) phenotype of the twin's RBCs indicated that anti-Jr^a antibodies were the cause of the positive

DAT results.

Several in vivo and in vitro studies have been performed to assess the clinical significance of anti-Jr^a antibodies. Kendall [14] performed the traditional ⁵¹Cr RBC survival test and showed the moderately rapid destruction of incompatible Jr(a+) RBCs after transfusion to a Jr(a-) patient. Ogasawara and Mazuda [19] reported that the Jr^a antigen density was lower than the D-antigen density in RBCs with a weak D phenotype and was significantly lower than the density of high-incidence antigens such as Di^b. They concluded that the small number of antigens on the RBC membrane may account for the general lack of clinical significance of anti-Jr^a antibodies. Miyazaki et al. [20] described that the fluorescence intensity of Jr(a+) RBCs was significantly lower than that of E+ RBCs from E homozygotes; this finding suggested that the number of Jr^a antigen sites is substantially lower than that of Rh antigen sites. The monocyte monolayer assay (MMA) has also been used in some studies to predict the potential clinical significance of antibodies to high-frequency antigens [15, 21, 22]. However, the exact mechanism of the toxicity of anti-Jr^a antibodies remains unclear.

One of the difficulties in identifying anti-Jr^a antibodies is the lack of commercially available antisera. A human monoclonal anti-Jr^a antibody has been produced from a human-mouse heterohybridoma (HMR0921) [20]. However, the biochemical structure of the Jr^a antigen and the molecular basis of the Jr(a-) phenotype are currently unknown [6]. Our laboratory has been identifying anti-Jr^a antibodies since 1995 after donation of Jr^a antisera and Jr(a-) type O RBCs obtained from Osaka Red Cross Center. We are currently using in-house Jr^a antisera and Jr(a-) RBCs prepared by constantly storing confirmed patient samples. Although we have been identifying a few cases of anti-Jr^a alloantibodies annually, this is the first case of HDNF related to anti-Jr^a alloimmunization in Korea. The first case of anti-Jr^a alloimmunization in Korea was reported in 1995 in a 35-yr-old pregnant woman with a history of multiple pregnancies and abortions (G₆P₄L₁D₃A₁) [23]. Anti-Jr^a antibodies were

identified after an anamnestic response following the transfusion of 1 RBC unit. However, the infant did not show any evidence of HDNF. Because a large percentage of Jr(a-) individuals appear to be of Asian origin [18], further studies on the incidence of the Jr(a-) phenotype in the Korean population may reveal interesting findings.

In conclusion, the potential of the anti-Jr^a antibody as a clinically significant alloantibody should be considered even in cases of first pregnancy. Preventive management and close fetal monitoring are recommended, especially if the mother has a high antibody titer and a history of multiple pregnancies and/or transfusion. Because Jr(a-) RBC units are very difficult to obtain, measures must be taken to ensure the availability of appropriate blood types. Autologous donations should always be considered; siblings are also potential donors of Jr(a-) RBCs. In emergent situations where Jr(a-) RBCs are not available, transfusion of the least incompatible units is inevitable. Nevertheless, close monitoring of the patient is necessary while performing subsequent transfusions.

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