TRANSFUSION COMPLICATIONS

Release of mediators of systemic inflammatory response syndrome in the course of a severe delayed hemolytic transfusion reaction caused by anti-D

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BACKGROUND: In vitro studies suggest that mediators of systemic inflammatory response syndrome are generated in the course of hemolytic transfusion reactions. Evidence for the in vivo significance of these findings is given by the present clinical and laboratory analysis of a severe delayed hemolytic transfusion reaction (DHTTR).

CASE REPORT: A 67-year-old patient (blood group O, D-negative) with a negative pretransfusion antibody screen received a massive transfusion because of arterial bleeding (Day 1). The transfusion of group O, D-positive red cell concentrates was unavoidable because of limited supplies. At Day 10, the patient developed a DHTTR with symptoms of septic-toxic syndrome and signs of hemolysis; he received an exchange transfusion. Serologic markers, as well as proinflammatory and anti-inflammatory mediators, were monitored at the onset of the DHTTR and during the exchange transfusion.

RESULTS: At Day 10, the direct antiglobulin test was positive; anti-D was present, most likely as the result of an anamnestic immune response. Interleukin (IL)-1 was not detectable; all other mediators monitored were elevated: IL-1 receptor antagonist, tumor necrosis factor, IL-6, IL-8, IL-10, neopterin, elastase, C3a-desArg, C-reactive protein, and fibrinogen. Most of the values declined during the exchange transfusion, which was followed by an improvement of the clinical presentation.

CONCLUSIONS: Mediators of systemic inflammatory response syndrome were released in the course of a DHTTR caused by anti-D. Severe clinical symptoms could be treated successfully by exchange transfusion.

ABBREVIATIONS: CRP = C-reactive protein; DAT = direct antiglobulin test; DHTTR(s) = delayed hemolytic transfusion reaction(s); FFP = fresh-frozen plasma; IL = interleukin; IL-1ra = IL-1 receptor antagonist; LDH = lactate dehydrogenase; PMN(s) = polymorphonuclear leukocyte(s); RBC(s) = red cell(s); SIRS = systemic inflammatory response syndrome; TNFα = tumor necrosis factor-α.

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factor-α (TNFα); interleukin (IL)-1, IL-6, and IL-8; white cell (WBC) contents (elastase, neopterin); complement fragments (e.g., C3a); and other substances such as arachidonic acid products. Anti-inflammatory mediators, such as IL-10, IL-1 receptor antagonist (IL-1ra), and soluble TNFα receptor, are also detectable. In SIRS, the balance between the proinflammatory and anti-inflammatory potential seems to be disturbed.

Davenport et al. tested several in vitro models of hemolytic transfusion reactions for the release of inflammatory mediators. One experimental approach was to incubate heparinized whole blood with ABO-incompatible RBCs. TNFα was released with an early peak at 2 to 4 hours, whereas IL-8 showed a continuous increase over 24 hours; furthermore, procoagulant activity was generated. Heating of plasma destroyed the cytokine- and procoagulant-producing capacity of the system; therefore, heat-labile plasma factors, such as complement, may play a role in the initiating process. IL-1 release was not detectable. In a further experiment, anti-D-coated RBCs were incubated with heparinized blood. IL-6, TNFα, and IL-1 were released at low levels, whereas high levels of IL-8 and IL-1ra were observed.

Few clinical data are available to date on the question of whether the results of these in vitro studies can be of significance in vivo. A report by Butler et al. showed that TNFα and elastase were released in an ABO-incompatible transfusion. We describe here the clinical picture and laboratory findings of a severe DHTR caused by anti-D. The data give evidence for the release of mediators, as is typical for SIRS, in this condition.

**CASE REPORT**

A 67-year-old male patient with coronary heart disease (high-grade stenosis of ramus interventricularis anterior) and nephrolithiasis underwent bilateral percutaneous nephrolitholapaxy (nephrolithotripsy), followed by the insertion of a kidney fistula catheter on both sides. He developed prolonged macrohematuria. Five days later, the kidney fistula catheters were removed; on the left side, arteriovenous bleeding occurred from the puncture canal, and an extended retroperitoneal hematoma was detected. The patient underwent cardiac failure of short duration because of volume loss. The hematoma was removed and bleeding was stopped by surgical intervention (Day 1). The next day, another surgery was necessary because of excessive bleeding, and the spleen was removed. The blood group of the patient was O, D-negative. Because of the shortage of blood of this type, the patient was predominantly transfused with group O, D-positive RBCs after it was ensured that the antibody screening test was negative. In the course of Days 1 and 2, the patient received 68 RBC units and 63 units of fresh-frozen plasma (FFP).

The patient, intubated via tracheostoma, developed septic temperatures (39.8°C) on Day 10. Approximately 8 hours later, he developed circulatory instability and a greater need for volume substitution. He was treated with catecholamines, dopamine, dobutamine, and volume replacement solution. The same day, a DHTR was diagnosed on the basis of a strongly positive direct antiglobulin test (DAT) and the presence of anti-D in the serum of the patient. Because of a drop in hemoglobin over Days 10 and 11, the patient was first transfused with 2 RBC units and 6 FFP units; thereafter, he received an exchange transfusion, extending over 24 hours (Day 11 to Day 12), with 27 RBC units (O, D-negative, K-) and 21 FFP units until the DAT was negative (Day 12). The recurrence of a slightly positive DAT was the reason for a second exchange transfusion on Day 14 (9 RBC units and 10 FFP units within 8 hours), which, seen retrospectively, was not necessary. The following discussion of data refers to the first exchange transfusion unless stated otherwise. Starting from Day 15, the patient experienced repetitive macrohematuria. Because of excessive arterial bleedings, his left kidney was removed on Day 17. In the further course, the patient experienced repeated bleeding from the right kidney; because of obstruction of the kidney and the bladder by coagula, he temporarily developed postrenal failure and had to undergo dialysis. After repeated infections and bronchopneumonia, the patient was discharged from hospital approximately 3 months after the DHTR. At that time, his kidney function was at the stage of compensated retention.

**MATERIALS AND METHODS**

Serologic testing was performed with column agglutination technology (BioVue, Ortho Diagnostic Systems, Baritan, NJ, or Micro Typing, DiaMed Diagnostika, Bensheim, Germany). For antibody screening, the anti-human globulin test and bromelain-treated RBCs were used. DATs that were positive with polyclonal antihuman globulin reagents were further investigated with monospecific anti-IgG, anti-IgM, and anti-C3d. Anti-D test reagents were used to determine anti-CDE (Fresenius Diagnostik, Bad Homburg, Germany), Seraclene anti-D blend (Biotest AG, Dreieich, Germany), and anti-D fast (Immucor Med. Diagnostik, Rödermark, Germany). For antibody titrations, an anti-human globulin tube test was used. Acid elution of the patient's RBCs was performed according to the instructions of the manufacturer (Biologische Arbeitsgemeinschaft, Lich, Germany); heat eluates were prepared from washed RBCs in 0.95-per cent NaCl by agitation for 10 minutes at 56°C; the supernatant was obtained after rapid centrifugation. Before and during the exchange transfusion, serum and plasma of the patient (anticoagulated with citrate or EDTA) were immediately frozen either at −20°C for various laboratory measurements or at −70°C for complement determinations.

The following measurements were made by enzyme-linked immunosorbent assay: C3a-desArg (Progen Biotechnik, Heidelberg, Germany); TNFα, IL-6, IL-8, and IL-10...
(Milenia, Diagnostic Products Corp., Los Angeles, CA); IL-1β and IL-1ra (Quantikine, R&D Systems, Minneapolis, MN); and neopterin (Gesellschaft für Immunchemie und Immunbiologie, Hamburg, Germany). Routine laboratory measurements (urea, creatinine, fibrinogen, total and free hemoglobin, lactate dehydrogenase (LDH), bilirubin, polymorphonuclear leukocyte (PMN) elastase-α1-antiprotease complexes, C1 inhibitor) were performed by standard methods. C3c, C4, C-reactive protein (CRP), haptoglobin, and hemopexin were estimated by nephelometric determination (Behringwerke AG, Marburg, Germany).

For analysis of antibody-coated RBCs by flow cytometry, samples of the patient’s RBCs were incubated with one of the following antibodies for 30 minutes at 4°C: fluorescein isothiocyanate-conjugated Fab', fragments of rabbit anti-human IgG, IgM, IgA, or IgD (DAKO Diagnostika, Hamburg, Germany); fluorescein isothiocyanate-conjugated mouse anti-human-k-light chain; or phycoerythrin-labeled mouse anti-human-λ-light chain (Becton Dickinson, Heidelberg, Germany). The samples were washed twice and analyzed in a flow cytometer (FACScan, Becton Dickinson).

The APACHE II score was calculated from clinical values as described by Knaus et al.12

RESULTS

Serologic investigations

The antibody screening test was negative with the serum of the patient at four time points ranging from 4 months before the massive transfusion to shortly before the massive transfusion (Day 1). On the day of onset of clinical symptoms of the DHTR (Day 10), anti-D was detectable with a titer of 256, which rose to 1536 the next day, despite the transfusion of 6 FFP units. The peak was reached approximately 10 days later, and thereafter the titer fell, to less than 15 at the time the patient was discharged from hospital. In addition to anti-D, anti-C and anti-K appeared on Day 12, and their peak and decline essentially followed the profile for anti-D, although the maximum values were much lower (Fig. 1). Finally, anti-E was observed, though not before Day 26, reached a maximum titer of 2, and became undetectable 3 months after the massive transfusion (not shown).

The DAT was strongly positive at the time of diagnosis of the DHTR. Investigation by the column agglutination method showed the presence of IgG on the patient’s RBCs; C3d was not detectable, and traces of IgM were demonstrable only on Days 10 and 11, before the exchange transfusion. Further, the RBCs showed a weak reaction with anti-CDE (NaCl phase), and no reaction with several IgM-type anti-D reagents. Eluates, obtained by either acid or heat, gave evidence of the presence of a single antibody, anti-D. When the DAT was performed before the exchange transfusion, almost all of the RBCs were trapped on top of the gel column. As the exchange progressed, the DAT, tested either with polyclonal reagents or with monospecific anti-IgG, showed a typical mixed-field agglutination with increasing predominance of unagglutinated cells.

Analysis of cell-fixed antibody by flow cytometry

Flow cytometric analysis gave evidence of the initial presence of 73-percent RBCs coated with IgG. As a control, RBCs of a D-positive donor were incubated with a surplus of the patient’s serum for 1 hour at 37°C in the presence of 10 mM EDTA; 99 percent of these RBCs were determined to be IgG-positive. During the exchange transfusion, an exponential reduction of antibody-coated RBCs was observed (Fig. 2). At the end of the first exchange transfusion, 3.9 percent of the IgG-coated RBCs were left, which is equivalent to approximately 160 mL of D-positive blood in the patient’s circulation; the corresponding value after the second exchange was 0.97 percent, which is equivalent to 41 mL. The analysis showed only the presence of IgG on the RBCs; IgM,

Fig. 1. Development of irregular antibodies after massive transfusion. Titers of anti-D, -C, and -K. TR = clinical and laboratory diagnosis of DHTR; MT = massive transfusion, Day 1.
Bilirubin (total and direct bilirubin, with little difference) increased from Day 10 to Day 11 by approximately twofold and, as did the levels of LDH, declined in the course of or after the exchange transfusion (Figs. 3 and 4). Hemopexin and haptoglobin were below normal values at the time of diagnosis of the DHTR. Probably because of the transfusion of plasma during the exchange transfusion, both values increased; a recovery to low normal levels was observed within 8 days (Fig. 3, haptoglobin data not shown). Serum creatinine levels continuously increased after the onset of the DHTR (Fig. 4), slowly decreased after Day 13, and rose further after nephrectomy. Levels of urea showed essentially the same course (data not shown).

**Total WBCs, differential, and platelet count**

The WBC count was in the low normal range after the massive transfusion and rose until Day 7. On Day 8, the WBC numbers dropped, and there after they showed a sharp rise in correlation with the DHTR. During the exchange transfusion, the WBCs fell again and stayed just above the higher limit until the end of the observation period, with an inter-

IgD, and IgA were negative. After fluorescence staining with anti-κ and anti-λ, the RBCs showed a homogeneous peak of κ and λ double-labeling.

**Clinical presentation**

During the patient's stay in the intensive care unit, the APACHE II score was determined. In parallel with the DHTR, a rise in the score was observed, reaching a maximum 2 days after the onset of symptoms (Fig. 2). The rise was mainly due to an increase in body temperature to 39.8°C, lowered mean arterial pressure, a rise in the heart rate, and an elevated Glasgow coma score, as controlled ventilation made it necessary to place the patient under sedation.

**Signs of hemolysis and measures of kidney function**

Starting from Day 8, a decline in the values of total hemoglobin was observed (Fig. 3), with the nadir on Day 11, before the exchange transfusion. Most of the other laboratory values were not available before the diagnosis of the DHTR. The highest levels of free hemoglobin were observed at the first time point monitored, on Day 10.

![Fig. 2. APACHE II score in relation to IgG-coated RBCs in the course of the DHTR.](image)

![Fig. 3. Levels of total and free hemoglobin, LDH, and hemopexin. Normal: total hemoglobin, 8.6-12 mmol/L; free hemoglobin, <4.5 μmol/L; hemopexin, 0.5-1.15 g/L; LDH, 3.8-7.7 μmol/L. (1) = first exchange transfusion; (2) = second exchange transfusion; MT = massive transfusion; TR = clinical and laboratory diagnosis of DHTR; N = nephrectomy.](image)
Bilirubin (total) • Bilirubin (direct) --- Creatinine

- Bilirubin (total) <17 μmol/L; direct bilirubin <5 μmol/L; creatinine, 55-100 μmol/L.

MT = massive transfusion; TR = clinical and laboratory diagnosis of DHTR; N = nephrectomy.

Acute-phase proteins
Fibrinogen levels increased after the massive transfusion, reaching a peak on Days 6 and 7 (Fig. 6). At the time of onset of the DHTR, the levels were still above normal, and a moderate increase was observed, followed by a drop to normal levels in the course of the exchange transfusion. CRP showed a sharp rise after the massive transfusion and stayed at high levels during the observation period. The onset of the DHTR was accompanied by a further moderate increase (Fig. 6). A drop on Day 17 was followed by a sharp increase after nephrectomy.

Inflammatory mediators and anti-inflammatory substances
During the observation period, IL-1 was not detectable, while levels of IL-1ra exceeded 3000 pg per mL throughout. TNFα, IL-6, IL-8, and IL-10, as well as PMN elastase and neopterin, were elevated at the time of diagnosis of DHTR, with peak values at the beginning of (TNFα, IL-10) or during (IL-6, IL-10) the exchange transfusion; this was followed by a decline (Figs. 6-8). IL-6 and IL-10 increased after nephrectomy, while a short-lived second peak in TNFα was observed during the second exchange transfusion. IL-8 showed a second peak on Days 15 and 16. Neopterin levels increased sixfold from Day 10 to Day 11—that is, at the onset of the DHTR—with a peak at Day 12, but a more prominent rise occurred on Day 14 (Fig. 6).
when undergoing partial lung resection in 1958 and that the DHTR was due to an anamnestic secondary response. Repeated antibody screening tests during the last months before the massive transfusion were negative, and it may be assumed that the antibody titer had fallen to undetectable levels. This is in agreement with the fact that, after reaching peak levels within 20 days after the massive transfusion, anti-D and other antibodies (anti-C, anti-K, anti-E) declined quickly within the next 2 months. The antibodies other than anti-D seem to be without significance for the DHTR, as they were first detected after the exchange transfusion, in which the patient had received RBCs that were negative for the D, C, E, and K.

The anti-D belonged to the IgG class; it was polyclonal, as both κ- and λ-light chains were detectable on the patient's RBCs by flow cytometry. With column agglutination technology, traces of IgM were detectable before the exchange transfusion was initiated; it is unclear whether this finding

The protein measurements of complement components C3 and C4 showed a similar profile; the level was below or in the lower part of the normal range. An elevation in terms of an acute-phase response was not evident, and a decline occurred in the second phase of the exchange transfusion (Fig. 9). C1 inhibitor levels were just above or in the upper part of the normal range and otherwise followed the same profile as C3 and C4. C3a-desArg was elevated at the beginning of the exchange transfusion and showed a peak toward the end, which correlated with a decline in the C3 protein (Fig. 9). A further small peak was observed before the second exchange transfusion.

**DISCUSSION**

On Day 10, after a massive transfusion of D-positive blood to a D-negative patient, the appearance of clinical symptoms coincided with a sharp rise in the titer of anti-D. It was not clear whether the patient had received transfusions before; nevertheless, it is likely that he was immunized
MEDIATORS OF SIRS IN DHTR

Fig. 8. Levels of IL-8 and IL-10 during the DHTR. Normal ranges: serum IL-8, 0-40 pg/mL; serum IL-10, 2-24 pg/mL. (1) = first exchange transfusion; (2) = second exchange transfusion; TR = clinical and laboratory diagnosis of DHTR; N = nephrectomy.

is specific or whether it may be due to unspecific adsorption of IgM to cells that are heavily loaded with IgG. With flow cytometry, no IgM was detectable; however, IgM with low affinity may have been missed because washing steps preceded the analysis of RBCs. It may be assumed that virtually all the D epitopes were covered with antibodies and, therefore, were unaccessible for antigen determination at onset of the DHTR. By the column agglutination method, several IgM-type anti-D reagents were not able to detect the D-positive RBCs in the patient's circulation, while anti-CDE agglutinated the RBCs. This finding underlines the fact that results of antigen determination in DHTRs have to be regarded with care; the elution of antibodies before testing is not always successful in solving this problem. According to flow cytometry, 73 percent of the RBCs were coated with IgG at the time of diagnosis of the DHTR. On the other hand, DAT performed by the column agglutination method showed a retention of almost all of the RBCs; only traces of sedimenting free RBCs were detectable. The column agglutination method may underestimate the number of free RBCs, as these are captured in agglutinates lying on top of the gel or glass beads.

No major bleeding complications were observed, and no blood components were transfused between Days 5 and 10 after the massive transfusion. The progressive drop in hemoglobin starting from Day 8 may, therefore, indicate the onset of hemolysis as the first sign of the DHTR. Other laboratory measures to prove this assumption are not available before the day of diagnosis of the DHTR (Day 10). Fever was the first clinical sign and was followed by circulatory instability; the increased requirement for volume substitution probably reflected the beginning of a vasodilatation and permeability syndrome. On Days 10 and 11, a sharp rise in anti-D was accompanied by laboratory signs of progressive hemolysis. The decision for an exchange transfusion was made on the basis of an unstable circulatory and respiratory situation as well as a progressive impairment of renal function in a patient affected by heart and kidney disease.

Fig. 9. Levels of serum complement component C3 and C4 in relation to plasma C3a-desArg. Normal ranges or values: serum C3 (measured with anti-C3c and denoted as C3c), 0.9-1.8 g/L; serum C4, 0.1-0.4 g/L; plasma C3a-desArg, <200 ng/mL, plasma C1 inhibitor, 80-125%. (1) = first exchange transfusion; (2) = second exchange transfusion; TR = clinical and laboratory diagnosis of DHTR; N = nephrectomy.
Of the inflammatory mediators and anti-inflammatory substances tested, most were elevated at the time the DHTR was diagnosed and showed peaks at the beginning or during the exchange transfusion. Among these were the proinflammatory cytokines TNFα, IL-8, and IL-6, the anti-inflammatory mediators IL-10 and IL-1ra, elastase, and neopterin, a metabolic product of monocytes. The description of the pattern of mediators in this single, clinically complicated case does not allow firm general conclusions about cause-and-effect relationships; nevertheless, it raises questions about pathophysiological mechanisms that may stimulate further investigations.

TNFα is known to stimulate the release of other inflammatory cytokines. The injection of recombinant TNFα into humans causes the appearance of IL-6 and IL-8 and an increase in IL-1ra, whereas IL-1 is not detectable. Furthermore, characteristic changes in the WBC count are induced: lymphocytopenia, monocytopenia, and a biphasic behavior of neutrophil counts, with an initial decrease followed by neutrophilia. TNFα also induces an increase of plasma elastase-α,-antiprotease complexes, due to neutrophil activation, and is known as a pyrogen by direct action on the hypothalamus. Although detectable levels of the short-lived cytokine TNFα appear to be low in the case reported here, there was a stepwise reduction during the exchange transfusion, and many of the properties characteristic of TNFα were found in conjunction with the DHTR: fever as the initial sign; peaks of IL-6 and IL-8 that are somewhat delayed with respect to the appearance of TNFα; high levels of IL-1ra in the absence of IL-1; and elevated levels of elastase, monocytes, and lymphocytes and a biphasic change in the WBC count, whereby the major part of the WBCs was represented by the neutrophils. It is, therefore, tempting to speculate that TNFα, which may have been released much earlier than the monitoring showed, played a role in the inflammatory response that accompanied the DHTR. Butler et al. report the release of TNFα together with elastase-α,-antiprotease complexes after an erroneous transfusion of 100 mL of group A, D-positive blood to a blood group 0, D-positive recipient. The experiments by Davenport et al. also give evidence for the appearance of TNFα and IL-8 in an in vitro model of ABO- and D-incompatible blood transfusion.

Like TNFα, IL-8 has proinflammatory properties; it functions as an activator and chemoattractant of neutrophils; and elevated levels are found in a number of inflammatory disease states as well as in trauma and sepsis. In the DHTR described here, the order of cytokines released was the same as that in the baboon model of endotoxemia: IL-8 appeared simultaneously with IL-6, but somewhat after the appearance of TNFα, which is known as an inducer of these two cytokines.

At the time of diagnosis of the DHTR described here, IL-1ra was present in the absence of detectable IL-1. The absence of IL-1, or the presence of low levels, together with the presence of high levels of, is a common finding in patients with severe illness, sepsis, or trauma or after the injection of recombinant TNFα into humans. Only after the application of high doses of endotoxin into animals does IL-1 appear. The question has been raised as to whether IL-1 is really absent or whether measurements are interfered with by inhibitors. The release of IL-1ra is interpreted as a counterregulation of the proinflammatory stimuli of IL-1; because IL-1ra is a competitive inhibitor of IL-1 at the receptor level, relatively high amounts are required to counteract the function of IL-1.

IL-6 is a major inducer of hepatic synthesis of acute-phase proteins, and it is released after operation, trauma, sepsis and other states of SIRS. In the study presented here, the levels of the acute-phase protein CRP showed a correlation with the levels of IL-6. Stimuli for the release of CRP seemed to be the massive transfusion, the onset of DHT, and nephrectomy. Before the diagnosis of the DHTR, IL-6 levels were not available, but the onset of the DHTR and nephrectomy were both marked by an increase in IL-6. Fibrinogen showed, as expected, a slower response to the massive transfusion; only a small further augmentation was observed at the time of the DHTR, but the maximum of the peak was reached 1 day earlier than the peak of IL-6 and, hence, was not in obvious correlation to this cytokine. The coagulation disease and the exchange transfusion in conjunction with the DHTR probably influenced the course of fibrinogen levels.

Elevated levels of C3a throughout the observation period were an indicator of complement activation. Complement peptides are known to induce the release of TNFα, IL-1, IL-6, and IL-8 from WBCs. The main peak of C3a was not in correspondence to that of TNFα, and a correlation to other cytokines remains speculative, because it may be assumed that the levels measured were influenced by the exchange transfusion. Antibodies of the Rh system are generally considered not to be complement-activating.
tive DAT and the moderate decrease in C3, C4, and C1 inhibitor suggest that, in this DHTR, destruction of RBCs predominantly occurred in a slow process via the reticuloendothelial system.

It is well known that hemolytic transfusion reactions may cause renal dysfunction, and several hypotheses have been put forward for the mechanisms by which this occurs. The profiles for urea and creatinine were similar in the active DAT and the moderate decrease in C3, C4, and C1 inhibitor suggest that, in this DHTR, destruction of RBCs predominantly occurred in a slow process via the reticuloendothelial system.

The profiles for urea and creatinine were similar in the DHTR described here, and they suggest that the DHTR caused an impairment of kidney function. But it may be assumed that the underlying disease of the patient favored this process. As expected, nephrectomy caused a second rise in these two measures.

The APACHE II score showed a peak with 1 to 2 days' delay over the appearance of the maximum levels of cytokines. Because of its complexity, the overall clinical picture may require time to develop in response to biologic modifiers.

The patient underwent hemorrhage and massive transfusion 10 days before the DHTR. No data on the cytokine pattern are available from the time of the massive transfusion until the onset of the DHTR, but it has been reported that, in the animal model, hemorrhage may cause the release of cytokines and a dampening of the immune response. Because the WBC count, the platelet count, hemoglobin, and the acute-phase proteins CRP and fibrinogen showed a tendency to normalize after the massive transfusion, it is not likely that the effects ascribed to the DHTR were solely due to this previous event, but a modulation of the mediator response, qualitatively and/or quantitatively, is probable.

It may be assumed that the levels of the mediators, proteins, and fragments measured were influenced by the exchange transfusion, for example, by dilution effects, augmentation of the inhibitory potential, and the supplying of unactivated proteins. It is difficult to evaluate whether the exchange transfusion by itself caused mediator release. In this case, the second exchange transfusion, which it can be seen retrospectively could have been omitted, should have reproduced the pattern of the first. Although a second short-lived peak of TNFα appeared during the second exchange transfusion, the overall picture is not suggestive of a major release of mediators induced by the exchange transfusion.

D-incompatible transfusion, although potentially life-threatening, is unavoidable in emergency situations such as the need for massive transfusion at the same time that there is a shortage of supplies. Except for the exchange transfusion, the present treatment regimens for the rarely developing DHTRs mainly address the symptoms. Despite major obstacles in the treatment of sepsis or SIRS caused by the complexity of interactions of multiple immunologic mediators, the recognition of hemolytic transfusion reactions as a reflection of SIRS may guide the development of future treatment strategies.

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