Microvascular response to red blood cell transfusion in patients with severe sepsis*

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Objectives: **Microvascular alterations may play a role in the development of multiple organ failure in severe sepsis. The effects of red blood cell transfusions on microvascular perfusion are not well defined. We investigated the effects of red blood cell transfusion on sublingual microvascular perfusion in patients with sepsis.**

Design: **Prospective, observational study.**

Setting: **A 31-bed, medical-surgical intensive care unit of a university hospital.**

Patients: **Thirty-five patients with severe sepsis requiring red blood cell transfusions.**

Interventions: **Transfusion of one to two units of leukocyte-reduced red blood cells.**

Measurements and Main Results: **The sublingual microcirculation was assessed with an Orthogonal Polarization Spectral device before and 1 hr after red blood cell transfusion. Red blood cell transfusions increased hemoglobin concentration from 7.1 (25th–75th percentile, 6.7–7.6) to 8.1 (7.5–8.6) g/dL ($p < .01$), mean arterial pressure from 75 (69–89) to 82 (75–90) mm Hg ($p < .01$), and oxygen delivery from 349 (278–392) to 391 (273–473) mL/min·M^2 ($p < .001$). Microvascular perfusion was not significantly altered by transfusion, but there was considerable interindividual variation. The change in capillary perfusion after transfusion correlated with baseline capillary perfusion (Spearman-rho = −.49; $p = .003$). Capillary perfusion was significantly lower at baseline in patients who increased their capillary perfusion by >8% compared with those who did not (57 [52–64] vs. 75 [70–79]; $p < .01$), while hemodynamic and global oxygen transport variables were similar in the two groups. Red blood cell storage time had no influence on the microvascular response to red blood cell transfusion.**

Conclusions: **The sublingual microcirculation is globally unaltered by red blood cell transfusion in septic patients; however, it can improve in patients with altered capillary perfusion at baseline. (Crit Care Med 2007; 35:1639–1644)**

KEY WORDS: tissue perfusion; sepsis; microcirculation; red blood cell transfusion.

Patients with severe sepsis frequently develop microvascular alterations, and these can play a major role in the development of organ failure. Using the Orthogonal Polarization Spectral imaging technique, we previously observed major microvascular blood flow alterations in patients with severe sepsis (1). These alterations were not affected by global hemodynamic variables or the use of vasopressor agents and were totally reversible with the topical application of acetylcholine (1). We also demonstrated that the microcirculation improved in survivors of septic shock but failed to do so in patients dying from acute circulatory failure or with multiple organ failure after shock resolution (2). Although these data suggest that microcirculatory alterations are implicated in the development of multiple organ failure, they do not prove that resuscitation procedures based on assessment of the microcirculation can improve outcome. This would require interventional studies aimed at improving microcirculatory oxygen delivery. Hence, it is important to study the effects of interventions that may affect the microcirculation, and red blood cell (RBC) transfusion is one of these potential interventions.

Indeed, RBC transfusions are commonly used to improve convective oxygen delivery (3) in acutely ill patients with anemia. However, a number of factors that determine oxygen availability to the cells may not be reliably assessed from global hemodynamic assessment (1, 4, 5). In addition, hematocrit is lower in the capillaries than in large arteries and veins as a result of heterogeneous flow distribution (6–8), the Fahraeus effect, and interactions between a luminal glycocalyx and plasma macromolecules. Furthermore, the rheologic properties of the transfused RBCs may be altered (9). In particular, a reduction in RBC deformability can occur during RBC storage (10). This may adversely affect capillary flux into the capillaries (11). In a rat model of hemorrhagic shock, the transfusion of stored RBCs did not restore microcirculatory oxygenation in contrast to fresh blood cells (12). However, RBC deformability is already altered in sepsis (13), so the impact of transfusion of altered RBCs may be more limited (14).

*See also p. 1773.

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Indeed, the alterations owing to storage may be less than those resulting from sepsis, so that RBC transfusion, even of stored RBCs, in septic patients may provide RBCs with more normal RBC deformability (14). Because many septic patients require blood transfusions and transfusions may have beneficial effects in early resuscitation (15), patients with sepsis may be an important group of patients to study.

The aim of our study was to evaluate the effects of RBC transfusion on sublingual microvascular perfusion, as assessed by the Orthogonal Polarization Spectral technique, and to evaluate a possible correlation of this effect with the RBC storage time in patients with severe sepsis.

METHODS

After approval by the ethical committee of Erasme hospital, informed consent was obtained from each patient's next of kin. We studied 35 consecutive patients who met four criteria. First, they required RBC transfusion. The indication for RBC transfusion was a hemoglobin concentration either <7 g/dL or between 7 and 9 g/dL in the presence of signs of altered tissue perfusion (i.e., elevated lactate levels) or coronary artery syndromes. Second, the patients had a diagnosis of severe sepsis, defined by the usual criteria (16). The diagnosis of infection was established by standard criteria (17). Organ failure was defined by a sequential organ failure assessment score >2 for each organ (cardiovascular, central nervous system, respiratory, hepatic, renal, and hematologic) (18). Third, the patients were considered to be euvolemic. Fourth, they were mechanically ventilated, as this facilitates the use of the Orthogonal Polarization Spectral system. Exclusion criteria were liver cirrhosis, RBC transfusion in the preceding 72 hrs, shock owing to any other cause (cardiogenic, hemorrhagic, obstructive), oral injuries, rapid deterioration of hemodynamic status with the need to increase vasopressor dose in the 2 hrs preceding transfusion, and previous inclusion in the study.

Hemodynamic and microvideoscopic assessments were carried out immediately before (baseline) and 1 hr after transfusion of one or two units of packed RBCs. During the study period, all bedside procedures were withheld, and the patient's position was maintained. The doses of vasopressor/inotropic and sedative/analgesic agents were kept constant.

Red Blood Cell Transfusion Characteristics. Packed red blood cell units were obtained from the blood bank (Blood Transfusion Center, Erasme Hospital, Belgian Red Cross). All RBC units transfused in intensive care unit (ICU) patients in our unit are leukocyte reduced. Leukocyte reduction is obtained by filtration before storage. Storage solution (saline-adenine-glucose-mannitol) is added to RBCs before storage. The storage period is allowed up to 42 days, and there is no blood bank policy to preferentially transfuse fresh RBC.

General Management. All patients were monitored with an arterial and a central venous catheter; 25 patients were also monitored with a pulmonary artery catheter (Swan Ganz catheter; Edwards, Irvine, CA). Standard hemodynamic targets included a mean arterial pressure >65 mm Hg. Vasopressors consisted of dopamine (at a dose up to 20 μg/kg/min) and/or norepinephrine, in addition to repeated fluid challenges with crystalloids and colloids (albumin, gelatin, or hydroxyethyl starch solutions) to optimize stroke volume and to attempt to limit the dose of vasopressors. If needed, dobutamine was added (up to a dose of 20 μg/kg/min). All patients were mechanically ventilated. Light sedation (with midazolam up to 4 mg/hr) and analgesia (with morphine or remifentanil) was provided according to individual needs.

Measurements. We recorded temperature, heart rate, arterial pressure, and central venous pressure in all patients; in addition, complete hemodynamic measurements were obtained in patients monitored with a pulmonary artery catheter. Arterial and central venous or mixed venous blood samples were withdrawn simultaneously, and blood gases, hemoglobin saturation, and hemoglobin and lactate concentrations were measured (ABL700, Radiometer, Copenhagen, Denmark). Oxygen delivery, oxygen consumption and oxygen extraction ratio (O_2ER) were calculated using standard formulas (19). The Acute Physiology and Chronic Health Evaluation II score (20) was obtained at admission and the sequential organ failure assessment score (18) on the study day. The RBC storage time was recorded only after all measurements were obtained. If two units of RBC were transfused, data were collected and measurements obtained after the second transfusion and the storage time of the oldest unit was considered in the analysis.

Microvideoscopic Measurements and Analysis. We used the Cytoscan ARII (Cytometrics, Philadelphia, PA) to study the sublingual microvascular network with a 5× objective providing a 167× magnification. The details and potential applications of this technique have been detailed elsewhere (21). Importantly, although the visualization of the microcirculation is based on light absorption by the hemoglobin contained in RBCs, this technique remains valid in anemia, as well as during acute changes in hemoglobin concentration (22). After the removal of saliva and other secretions using gauze, the device was gently applied (without any pressure) on the lateral side of the tongue, in an area approximately 1.5–4 cm from the tip of the tongue. Five sequences of 20 secs each from different adjacent areas were recorded using a computer and a Videocard (MicroVideo; Pinnacle Systems, Mountain Views, CA) and stored under a random number for later analysis. Every effort was made to avoid movement artifacts. An investigator, blinded to the patient's clinical course and the order of the sequences, analyzed the sequences semiquantitatively (1, 4). Three investigators (MC, YS, and CV) analyzed series of images, and the same investigator analyzed the images before and after transfusion. In addition, the senior investigator (DD) reviewed 10% of the images to ensure an absence of significant variation in the analysis of the measurements. The analysis was conducted as follows. Briefly, three equidistant horizontal and three vertical lines were drawn. The vascular density was calculated as the number of vessels crossing these lines divided by the total length of the lines. The type of flow was defined as continuous, intermittent, or absent. The vessels were separated into large and small vessels, using a cutoff value of 20 μm in diameter. For each type of vessel, vessel perfusion was defined as the percentage of continuously perfused vessels divided by the total number of vessels. Perfused capillary density was calculated as the product of vessel density and vessel perfusion of capillaries. In each patient, the data from the five areas were averaged.

Statistical Analysis. Data were analyzed using SPSS 12.0 for windows (SPSS, Chicago, IL). Descriptive statistics were computed for all study variables. The Klomogorov-Smirnov test was used to verify the normality of distribution of continuous variables. Nonparametric measures of comparison were used, as all variables evaluated were not normally distributed. Differences between groups were assessed using a chi-square, Fisher's exact test, and Mann-Whitney U test, as appropriate. A Wilcoxon's test was used to compare the baseline and posttransfusion values. We arbitrarily stratified patients into two groups according to the change in capillary perfusion from the baseline value after transfusion with a cutoff of 8% (>8% classified as group A and <8% as group B). This value was selected as it corresponds to the best cutoff value in changes in sublingual microvascular perfusion to separate survivors and nonsurvivors in septic shock (2); this value is higher than the intraobserver variability of the measurements with this method (1). Analysis of variance was used to evaluate differences in the evolution of microcirculatory variables between the two groups during transfusions.

Correlations between variables were investigated by Spearman-rho, and a linear regression line was computed with 95% confidence interval (CI). $p < .05$ was considered to be significant. Data are presented as median (25th–75th percentiles).

RESULTS

The study included 35 patients with severe sepsis (Table 1), including eight

Table 1. Characteristics of the study group

	All Patients (n = 35)
Age, yrs	69 (51–78)
Male gender, %	23 (65.7)
APACHE II score	25 (18–28)
SOFA score	10 (6–13)
Source of infection, (%)	
Lung	16 (46)
Abdomen	6 (17)
Urinary tract	4 (11)
Miscellaneous	9 (26)
Adrenergic dose	
Dopamine[a]	8;10 (5–20)
Norepinephrine[a]	8;0.2 (0.2–0.3)
Dobutamine[a]	18;10 (10–24)
Analgo-sedation	
Midazolam[b]	18;2 (1–4)
Morphine[b]	19;1 (1–2)
ICU length of stay, days	14 (10–35)
28-day mortality, %	37

APACHE, Acute Physiology and Chronic Health Evaluation; SOFA, Sequential Organ Failure Assessment; ICU, intensive care unit.

[a]n; dose in μg/kg·min; [b]n; dose in mg/hr.

patients with septic shock. The median ICU length of stay was 14 days (interquartile range, 10–35), and the 28-day mortality was 37% (Table 1). Four patients received two units of RBCs. The time from sepsis onset to transfusion was 2 (1–3) days (minimum, 0; maximum, 13). No transfusion-related adverse reaction was observed during the time of the study. Blood transfusion resulted in increases in hemoglobin (from 7.1 [6.7–7.6] to 8.1 [7.5–8.6] g/dL; $p < .01$) and in oxygen delivery (from 349 [278–392] to 391 [273–473] mL/min/M^2; $p < .001$). In addition mean arterial pressure (from 75 [69–89] to 82 [75–90] mm Hg; $p < .01$), central venous pressure (from 10 [8–14] to 12 [10–16] mm Hg; $p < .01$), and pulmonary artery occlusion pressure (from 16 [12–18] to 17 [13–20] mm Hg; $p = .03$) increased (Table 2). However, oxygen consumption and microcirculatory variables were unaltered (Table 2). The change in hemodynamic, microcirculatory, and blood gas variables did not differ between patients who received one (n = 31) or two units (n = 4) of RBCs.

RBC transfusion globally failed to affect microvascular perfusion (Table 2), but there was considerable interindividual variation, with improvement in some patients and deterioration in others. The effects on global hemodynamic variables were also variable. There was no significant correlation between microvascular changes and the other hemodynamic and oxygen transport variables, vasopressor dose, or hemoglobin levels ($p > .2$ for all correlations). At baseline, group A and group B patients had similar hemodynamic and global oxygen transport variables (Table 3). However, group A patients had lower baseline capillary perfusion and perfused capillary density than group B patients (57 [52–64] vs. 75 [70–79]%; $p < .01$; and 2.7 [1.9–3.0] vs. 3.5 [3.2–4.5]). Following transfusion, the perfused capillary density increased in

Table 2. Physiologic and microcirculatory variables before and 1 hr after transfusion of packed red cells

	All Patients (n = 35)	
	Baseline	Transfusion
Temperature, °C	36.7 (36.3–37.4)	36.9 (36.4–37.4)
Heart rate, beats per min	99 (90–111)	98 (89–110)
Mean arterial pressure, mm Hg	75 (69–89)	82 (75–90)[a]
Central venous pressure, mm Hg	10 (8–14)	12 (10–16)[a]
Mean pulmonary artery pressure, mm Hg[b]	29 (25–34)	32 (28–35)
Pulmonary artery occlusion pressure, mm Hg[b]	16 (12–18)	17 (13–20)[c]
Cardiac index, L/min·M^{2b}	3.6 (3–4.2)	3.7 (2.6–4.4)
Hemoglobin concentration, g/dL	7.1 (6.7–7.6)	8.1 (7.5–8.6)[a]
$Paco_2$, mm Hg	37 (34–42)	37 (35–42)
Pao_2, mm Hg	100 (77–132)	101 (75–116)
pH	7.40 (7.30–7.43)	7.37 (7.29–7.43)
Sao_2, %	98 (95–100)	99 (97–100)
Lactate, mmol/L	1.3 (0.8–1.8)	1.3 (1.0–1.7)
Mixed venous oxygen saturation, %[b]	64 (59–73)	67 (60–79)
Oxygen delivery, mL/min·M^{2b}	349 (278–392)	391 (273–476)[a]
Oxygen consumption, mL/min·M^{2b}	105 (84–146)	108 (67–159)
Oxygen extraction ratio, %[b]	33 (26–39)	32 (20–38)
Total vascular density, n/mm	5.7 (4.3–8.4)	5.5 (4.4–8.8)
% all vessels perfusion	85 (80–89)	87 (82–93)
Perfused capillary density, n/mm^3	2.4 (1.8–3.2)	2.3 (1.8–2.8)
% capillary perfusion	74 (58–78)	71 (61–80)

[a]$p < .01$ compared with baseline; [b]measured in 25 patients; [c]$p < .05$ compared with baseline.

Table 3. Baseline physiologic variables in both groups

	Group A (n = 10)	Group B (n = 25)
Temperature, °C	37 (36.4–38)	36.6 (36.3–37.1)
Heart rate, beats per min	97 (90–109)	100 (91–111)
Mean arterial pressure, mm Hg	77 (71–89)	75 (69–86)
Central venous pressure, mm Hg	9 (8–12)	11 (8–15)
Mean pulmonary artery pressure, mm Hg[a]	30 (25–32)	29 (26–34)
Pulmonary artery occlusion pressure, mm Hg[a]	15 (13–17)	16 (12–18)
Cardiac index, L/min·M^{2a}	3.5 (2.7–4.1)	3.8 (3.2–4.2)
Hemoglobin concentration, g/dL	7.0 (6.8–7.2)	7.2 (6.7–7.6)
Change in hemoglobin concentration, g/dL	0.8 (0.6–1.5)	1.0 (0.7–1.2)
Relative change in hemoglobin concentration, %	11 (8–21)	13 (9–18)
$Paco_2$, mm Hg	38 (36–41)	37 (34–42)
Pao_2, mm Hg	117 (81–146)	85 (76–119)
pH	7.40 (7.30–7.49)	7.40 (7.30–7.40)
Sao_2, %	98 (96–100)	97 (95–99)
Lactate, mmol/L	1.4 (1–1.5)	1.2 (0.8–2.2)
Mixed venous oxygen saturation, %[b]	70 (57–76)	63 (59–72)
Oxygen delivery, mL/min·M^{2b}	340 (250–381)	359 (316–414)
Oxygen consumption, mL/min·M^{2b}	91 (84–136)	118 (85–154)
Oxygen extraction ratio, %[b]	30 (23–42)	34 (27–37)
APACHE II score	25 (19–31)	25 (18–28)
SOFA score	10 (5–11)	11 (7–15)
Time between sepsis onset and RBC transfusion, days	2 (1–2)	2 (1–3)

APACHE, Acute Physiology and Chronic Health Evaluation; SOFA, Sequential Organ Failure Assessment; RBC, red blood cell count.

[a]Measured in 25 patients in total (10 with shock and 15 without shock). The two groups are separated according to the percent increase in capillary perfusion at a 8% cutoff value (2): capillary perfusion increased by >8% in group A and by <8% in group B. No significant difference between baseline values.

group A as a consequence of a combined increase in both capillary density and in the perfusion of the visualized capillaries (+17.1 [10.3–30.1]%). In contrast, perfused capillary density decreased in group B, mostly as a consequence of a decrease in the perfusion of visualized capillaries (−9.0 [−18.3 to −0.2]%) (Table 4). The change in capillary perfusion was negatively correlated to baseline capillary perfusion (Spearman-rho = −.49 [−0.27 to −0.77]; p = .003) (Fig. 1). Similar results were observed with perfused capillary density, with changes in perfused capillary density after transfusion being negatively correlated with the baseline perfused capillary density (Spearman-rho = −.38 [95% CI, 0.05 to −0.71]; p = .02).

The median RBC storage time was 24 days (interquartile range, 12–28), with a minimal value of 6 days and a maximal value of 35 days. There was no relation between the storage time and the change in capillary perfusion owing to RBC transfusion (Spearman-rho = −.01 [95% CI, −0.01 to −0.02]; p = .963) (Fig. 2). There was no difference in the age of the RBCs between the two groups (19 [11–27] in group A vs. 24 [16–28] days in group B; p = .42)

Table 4. Evolution of microcirculatory variables in the two groups before and after RBC transfusion

	Group A (n = 10)		Group B (n = 25)	
	Before	After	Before	After
Total vascular density, n/mm	4.9 (3.7–8.6)	5.3 (3.8–9.0)	5.8 (4.5–8.3)	5.5 (4.7–8.2)
Capillary vascular density, n/mm	4.5 (3.6–4.9)	4.8 (3.7–5.2)[a]	5.2 (4.3–5.7)[b]	5.1 (4.4–5.7)
% Perfused capillaries[c]	57 (52–64)	74 (68–80)[d]	75 (70–79)[b]	65 (59–80)[d]
Perfused capillary density, n/mm[e]	2.7 (1.9–3.0)	3.2 (2.7–3.9)[a]	3.5 (3.2–4.5)[b]	3.3 (3.0–3.8)

[a]$p < .05$ and [d]$p < .01$ compared with baseline; [b]$p < .05$ compared with other group; [e]and [c]$p < .01$ and $p < .001$ group time interaction by analysis of variance. The two groups are separated according to the percent increase in capillary perfusion at a 8% cutoff value (2).

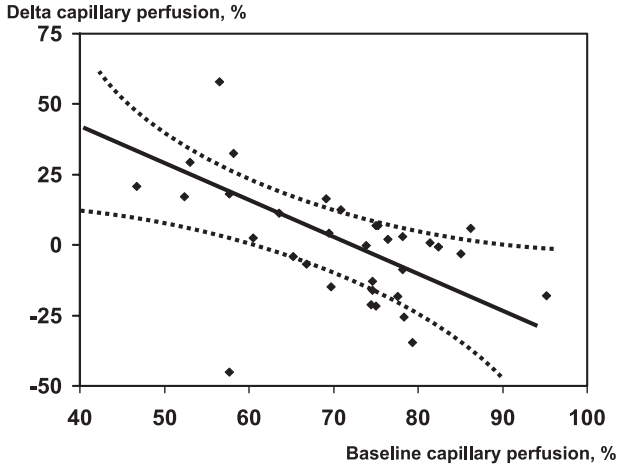

Figure 1. Scatter plot representing the baseline capillary perfusion (%; *x-axis*) and the relative change in capillary perfusion (delta capillary perfusion, %, *y-axis*). Linear regression (*solid line*) with 95% confidence intervals (CI; *dashed lines*) are fitted, where Y = 69 − 0.5× (p = .001) and Spearman-rho = −.49 (95% confidence interval, −0.77 [−0.27]; p = .003).

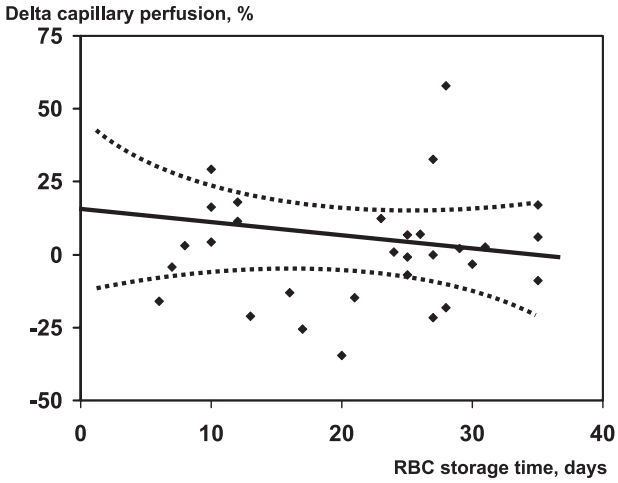

Figure 2. Scatter plot representing the red blood cell (*RBC*) storage time (days; *x-axis*) and the relative change in capillary perfusion (delta capillary perfusion, %; *y-axis*). Linear regression (*solid line*) with 95% confidence intervals (*dashed lines*) are fitted, where Y = 78 − 0.21× (p = .224) and Spearman-rho = −.01 (95% confidence interval, −0.01 [−0.02]; p = .963).

DISCUSSION

The principal finding of our study was that RBC transfusion had no straightforward effect on sublingual microvascular perfusion in a group of critically ill patients with severe sepsis. There was, however, considerable interindividual variability. Importantly, there was a dichotomous response, with an improvement in sublingual microvascular perfusion in patients with an altered perfusion at baseline and a deterioration in sublingual microvascular perfusion in patients with preserved baseline perfusion. The changes in sublingual microvascular perfusion were not related to the RBC storage time.

The effects of blood transfusions on the microcirculation are still largely unknown, especially in the context of sepsis. Some studies used gastric tonometry to study the effects of transfusions on gastric intramucosal pH (23–25). In these studies, a large interindividual variability in the response to RBC transfusions was also observed, as in the present study.

The critical question is why some patients show beneficial effects of RBC transfusions while others do not. RBC storage may be an important factor. Well-documented changes occur to RBC products during ex vivo storage (9). These changes include a reduction in RBC deformability (9), altered adhesiveness and aggregability, and reduction in 2,3-diphosphoglycerate (26) and adenosine triphosphate. However, these results were recently challenged by Raat et al. (27), who reported that storage of rat RBCs for up to 5 wks did not alter their deformability or their oxygen-carrying

properties. In patients, Marik and Sibbald (23) demonstrated the adverse effects of RBC storage on tissue oxygen variables (intramucosal pH), but others have failed to confirm this relationship (25, 28). The median RBC storage time in our study was 24 days, as reported in other studies (29, 30). We did not find a relationship between the changes in microvascular perfusion and RBC age.

Endogenous RBC deformability may be a critical factor. Friedlander et al. (14), indeed, observed that RBC transfusions improved RBC deformability in patients with sepsis, probably by replacing rigidified RBCs by more functional, or less dysfunctional, exogenous RBCs. Hence, transfusions may be deleterious when performed in patients with preserved RBC deformability but may have favorable effects when performed in patients with markedly altered RBC deformability. These findings may explain why we observed that RBC transfusion decreased the sublingual microcirculation when it was essentially normal at baseline but improved it when it was already decreased at baseline.

An important finding in our study is that patients who improved their microcirculation were characterized by a significant difference in the baseline capillary perfusion but had similar baseline hemodynamic and global oxygen transport variables to the other patients. Moreover, no significant correlation was observed between the change in capillary perfusion on the one hand and the global hemodynamic and oxygen transport variables and the vasopressor dose on the other. This observation corresponds well with previous experimental (5, 31) and clinical (4, 32) data. Animal studies suggest that microcirculatory alterations are independent of arterial hypotension. The perfusion of diaphragmatic capillaries (5) and of gut villi (31) was markedly decreased in septic rodents compared with hypovolemic controls with a similar degree of hypotension. In patients with septic shock, LeDoux et al. (32) reported that increasing the mean arterial pressure from 65 to 85 mm Hg with norepinephrine was associated with an increase in cardiac index while skin microvascular blood flow and gastric mucosal P_{CO_2} remained unchanged. We (4) also previously reported that in patients with severe sepsis, microcirculatory alterations were not correlated with blood pressure, cardiac index, or other global hemodynamic variables.

With the exception of baseline microvascular perfusion, none of the other measured factors discriminated patients with a favorable response from the others. In particular, doses of vasoactive agents, severity indexes, or changes in hemoglobin were similar in both groups. Although no interventions other than RBC transfusion were allowed during the intervention period, we cannot rule out that these changes may have been influenced by other recently administered interventions.

Because of the short duration of the experiment, we were unable to study the relation with organ function or outcome. As we expected (4), there was no relationship between changes in sublingual microvascular and global hemodynamic variables. The cutoff value used to separate both groups was set at 8%, based on our previous observation showing that patients improving their sublingual microcirculation above this threshold had better chances of surviving than the others (2). Future studies should investigate whether monitoring the sublingual microcirculation, in association with other factors, may be useful to guide transfusion policy in patients with severe sepsis with moderate anemia.

Our study has its limitations. First, data could be analyzed only semiquantitatively as direct quantitative measurement of blood flow in each vessel was limited by the multiple projection of vessels in the area of interest and the movement artifacts. However, the analysis was performed by an investigator blinded to the patient's condition. Also, there was no systematic overestimation of the second measurement because of change in hemoglobin concentration, as there was no significant change in the entire population. In addition, we previously reported that the intraobserver variability with this semiquantitative analysis is only around 5% (1); similar results have been reported recently by another team of investigators using a similar analysis (33). Second, the study of perfusion in the sublingual mucosa may not reflect perfusion in other microcirculatory beds. However, in experimental models of sepsis, changes in sublingual and gut microcirculatory perfusion were of similar magnitude and occurred within the same time interval (34, 35). Clinical observations from our group (36) and others (37, 38) indicate parallel alterations in sublingual capnometry and gastric tonometry measurements, suggesting that both areas can be similarly and simultaneously affected. Third, we administered relatively small amounts of RBCs, and deleterious effects, unrelated to the microcirculation (volume overload), may occur with larger transfusions. It was ethically difficult to impose multiple transfusions unnecessarily, so that the indication for transfusion and the number of RBCs transfused were left to the discretion of the treating physician; these corresponded to current practices (39). Critical care physicians have modified their approach to transfusion in recent years (39). In a recent survey of Canadian physicians (39), single-unit transfusion was used in 56% of cases compared with 10% in an earlier similar survey (40) conducted by the same authors. Moreover, Suttner et al. (41) reported that transfusion of two units, instead of one, of allogenic RBCs did not affect the changes in skeletal muscle oxygen tension in patients after elective coronary artery bypass grafting. Fourth, our measurements were restricted to 1 hr after RBC transfusion, which may have overlooked possible alterations occurring later. Our study already lasted a total of about 2.5 hrs (including the time needed for RBC transfusion), and longer follow-up periods are practically difficult, because of the inevitable therapeutic alterations and patient manipulation. Also, spontaneous changes in the patient's condition may influence microcirculatory perfusion. Nevertheless, changes in the microcirculation are expected to occur early after the RBC transfusion, so that it is unlikely that extending the study period would have altered our conclusions. In the study by Marik and Sibbald (23), alterations in intramucosal pH occurred just after RBC transfusion and persisted up to 6 hrs thereafter. Using other interventions, it has been shown that the sublingual microcirculation responds rapidly to vasoactive interventions (4, 36). Fifth, the timing of RBC transfusions was not standardized in relation to the onset of sepsis with a possible difference in microvascular response in early vs. late sepsis. Finally, we did not measure the microvascular hematocrit so that we did not estimate changes in microvascular oxygen delivery. However, this limitation only applies to the perfused capillaries, and a key finding in our study was that the number of perfused capillaries decreased in some patients and increased in others, hence leading, by definition, to parallel changes in oxygen

delivery in the areas depending on these capillaries.

CONCLUSION

The microvascular effects of RBC transfusions are quite variable and are dependent on baseline microvascular perfusion. These effects cannot be predicted from systemic hemodynamics, biological variables, or severity of the disease. They are also independent of the age of the transfused RBCs.

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