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Postoperative blood salvage and reinfusion in spinal surgery: blood quality, effectiveness and impact on patient blood parameters

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Abstract Although reinfusion of salvaged shed blood has become popular in major orthopaedic procedures, this blood saving technique is still controversial. In an effort to assess the functional and metabolic status of shed blood erythrocytes and the impact of postoperative shed blood reinfusion on allogenic blood requirements and patient's blood parameters, analyses of perioperative blood samples were performed in 28 consecutive orthopaedic patients undergoing spinal fusion, in which postoperative shed blood was collected and reinfused with the ConstaVac CBC II device. In comparison with a previous series of 31 patients, this procedure reduced allogenic blood requirements by almost 30% ($P < 0.05$), without any increase in postoperative complications. Postoperative shed blood presented lower haematological values and higher plasma-free haemoglobin (PFHB) levels than preoperative blood, without any disturbance in morphology, median corpuscular fragility (MCF) or erythrocyte adenosine triphosphate (ATP) and diphosphoglycerate (DPG) content. Serum concentra-

tions of enzymes – glutamate-oxalacetate aminotransferase (GOT), glutamate-pyruvate aminotransferase (GPT), creatine kinase (CK), lactate dehydrogenase (LDH) – and inflammatory cytokines (IL-1 β , IL-6) were elevated in shed blood. After reinfusion, there was no alteration in coagulation parameters or cytokine levels. Serum levels of some enzymes increased at the end of surgery and remained elevated at postoperative day 2 (CK) or 7 (GOT, LDH), with a higher increase if postoperative autotransfusion was used as a blood saving method. Therefore, caution should be taken when these serum enzyme levels are used for diagnosis. In conclusion, salvaged shed blood in orthopaedic procedures of the spine seems to be an excellent source of red cells which are not significantly damaged, keeping a normal functional and metabolic status, and reduces allogenic blood requirements without significant side effects.

Key words Shed blood salvage · Blood quality · Serum enzymes · Cytokines · Spinal fusion

Introduction

The development of complex surgical procedures for the treatment of orthopaedic diseases and the increasing number of traffic accidents have raised the demand for allo-

genic blood to a level that often exceeds supply. In addition, the increased awareness of the potential hazards of allogenic blood transfusion, such as incompatibility reactions, metabolic and immunological disorders or transmission of viral diseases, has led to an emphasis on blood saving techniques [10, 18, 25].

Among these techniques, the use of intraoperative and postoperative autotransfusion, together with autologous blood predeposit, has been popularized in recent years due to its great potential for reducing donor blood requirements and for eliminating the risks that may accompany the use of homologous blood [21].

However, although the safety of reinfusion of unwashed shed blood after orthopaedic [2, 3, 5, 11, 12, 13, 14, 15, 20, 30] operations is well documented, the controversy about its effectiveness still exists regarding the functionality and viability of the reinfused erythrocytes, the nephrotoxic effects of free haemoglobin, the alteration of the immune response and the alteration of the haemostasia [5, 21].

Accordingly, this study was initiated:

1. To examine the haematological, biochemical and immunological characteristics and the metabolic and functional erythrocyte status of shed blood samples collected in the postoperative period from patients undergoing elective spinal surgery
2. To assess the effectiveness of drainage blood reinfusion in reducing allogenic blood requirements
3. To evaluate the impact of such a procedure on patient blood parameters

Materials and methods

Patients

The study group consisted of 28 consecutive orthopaedic patients receiving postoperative shed blood after instrumented spinal fusion (Allospine, Sulzer), comprising 2–4 levels with or without decompression of the neurologic elements by microsurgery (group B). In all patients, a spondylodesis with autologous bone from the iliac crest was added to the instrumentation.

A previous series of 31 patients (group A) with matched age and pathology, but not receiving postoperative shed blood, served as a control group to evaluate the efficacy of the device in the reduction of allogenic blood requirements (Table 1). All patients received oral iron (120 mg Fe²⁺/day) from the 3rd preoperative week, until 1 month after the operation.

All surgical procedures were performed under general anaesthesia and all patients received 1.5 g cefuroxime intraoperatively plus low-molecular-weight heparin (dalteparine 5000 UI/24 h, 2–3 days) and antibiotics (cefuroxime 750 mg/6 h, 3–4 days) postoperatively to prevent thrombo-embolic and infectious complications.

The anaesthesiologist, who was unaware of the study, estimated intraoperative blood loss (based on blood aspirated from the surgical field and blood on sponges and drapes) and made decisions on intraoperative transfusions. Measurement of postoperative blood loss and decisions on postoperative transfusions were made by the attending surgeon.

Postoperative blood salvage

At the end of surgery, the collection blood canister (ConstaVac CBC II, Stryker, USA) was connected to both wound and donor sites and shed blood was collected without anticoagulant, at a negative pressure of 25 mmHg. Salvaged shed blood was reinfused within the first 6 postoperative hours. A 40- μ m screen filter (SQ40SJKL, Pall, UK) was intercalated in the reinfusion line to

Table 1 Characteristics of patients

	Group A	Group B
No. of patients	31	28
Age (years)	48 \pm 3	52 \pm 3
Sex (M/F)	12/19	14/14
Operation length (h)	5.3 \pm 0.4	5.3 \pm 0.1
Haemoglobin level (g/dl)		
Preoperative	13.5 \pm 0.3	14.2 \pm 0.3
Postoperative	10.7 \pm 0.3	10.5 \pm 0.3
Hospitalization (days)	9.6 \pm 1.4	8.4 \pm 0.5
Complications	4	3

eliminate microaggregates. The number of shed blood units recovered was calculated according to the expression:

$$U = \frac{\text{Shed blood volume (ml)} \cdot \text{Shed blood haematocrit (\%)}}{400 \text{ (ml)} \cdot \text{Preoperative haematocrit (\%)}}$$

Blood samples

To evaluate the impact of reinfused drainage blood on the recipient's biochemical and haematological parameters, besides the sample taken from the shed reservoir at the 6th hour of the postoperative period (sample 3), another five blood samples were obtained from the patients at different perioperative stages in the study: in the preoperative period (sample 1, day -7), at the end of operation (sample 2, day 0), and at the 1st, 2nd and 7th postoperative days (samples 4, 5 and 6; days 1, 2, and 7). An additional sample was collected from the patients 1–2 h after shed blood reinfusion, to evaluate coagulation parameters.

Haematological parameters

Red cell count, haematocrit, haemoglobin, white cell count and platelet count were determined using a Technicon III cell counter (Bayer, Germany) in blood samples collected in ethylenediaminetetraacetic acid (EDTA). Plasma-free haemoglobin (PFHB) was measured in 12,000 \times g centrifuged plasma according to the bicidine reaction. Red cell morphology was analysed under a light microscope (\times 600, Nikon Microphot FXA, Japan) after May-Grünwald-Giemsa staining. At least 100 cells in each duplicated specimen were evaluated.

Erythrocyte osmotic fragility

Fifty microlitres of blood collected in EDTA, either fresh or incubated at 37 °C for 24 h, was added to 10 ml of different NaCl concentrations (0.0–0.9%). After 30 min standing at room temperature, samples were centrifuged at 2000 \times g for 5 min and the optical density of the supernatants was measured at 540 nm using the 0.9%-NaCl sample as a blank. The percentage of lysis was calculated by relating the optical density at 540 nm of each supernatant to that obtained for the 0.0% NaCl one, taken as 100%. A computerized sigmoidal plot was used to estimate median corpuscular fragility (MCF: NaCl concentration for 50% haemolysis).

Coagulation parameters

Prothrombin time (PT, in seconds), activated partial thromboplastin time (APTT, in seconds), and prothrombin activity (PA, %)

were measured blood samples collected in 0.129 M sodium citrate before the operation and again 1–2 h after shed blood reinfusion using a Sysmex CA 6000 (Dade-Behring, Germany).

Biochemical parameters

Serum levels of glucose, triglycerides, total cholesterol, phospholipids, total proteins, glutamate-oxalacetate aminotransferase (GOT), glutamate-pyruvate aminotransferase (GPT), gamma-glutamyl transpeptidase (GGT), lactate dehydrogenase (LDH), creatine kinase (CK), alkaline phosphatase (ALP), Na⁺, K⁺, and Cl⁻ were measured using the Hitachi 747 (Japan) and Falcor 200 (Menarini, Italy) multianalysers. Measurements of haptoglobin, complement C3 and C4 fractions, and immunoglobulin levels were performed using a nephelometer NB-II (Behring, Germany). Serum enzyme, complement fraction, immunoglobulin and haptoglobin levels were corrected for haemodilution using total protein concentration of preoperative blood sample as control.

Enzyme immunoassays for cytokines

The levels of plasma immunoreactive interleukin-1 β (IL-1 β), IL-6, and tumour necrosis factor- α (TNF α) were determined by immunoenzymatic assay (ELISA) according to the manufacturer's procedure. The detection limits (pg/ml) were < 1 for IL-1 β , 1.4 for IL-6, and < 5 for TNF- α . Results were corrected for haemodilution using total protein concentration of preoperative blood sample as control.

Adenosine triphosphate (ATP) and 2,3-diphosphoglycerate (DPG) determinations

One millilitre of heparinized shed blood was mixed with either 1 ml 12% TCA (ATP) or 3 ml 8% TCA (DPG), vortex mixed and centrifuged after 5 min standing at 4°C. Supernatants were assayed for ATP by the phosphoglyceric/glyceraldehydephosphate dehydrogenase method and for DPG by the phosphoglycerate mutase reaction method using standard kits. Results were expressed in $\mu\text{mol/g Hb}$.

Chemicals

Standard kits for ATP and DPG were purchased from Sigma Chemical Co. (USA). IL-1 β , IL-6, and TNF- α enzyme-linked immunosorbent assay kits were from Bender Med Systems (Austria). Standard kits for serum enzymes and haptoglobin were from Menarini (Italy). All other chemicals were of analytical grade and purchased from Merck (Germany).

Statistical analysis

All results are presented as the mean \pm SE (n) and statistical analysis was carried out using paired Student's t -test for comparisons within a group and unpaired Student's t -test for comparisons between groups. All statistical analysis was performed with an SPSS-PC+ package.

Results

Effectiveness of salvaged shed blood reinfusion

In this series of patients, the overall transfusion rate was similar in both groups (1.90 units/patient group A vs

2.13 units/patient group B, $P = \text{NS}$). However, in group B the reinfusion of 18 units of shed blood resulted in a 60% reduction in postoperative allogenic blood requirements as compared to group A (0.25 vs 0.64 units/patient, $P < 0.05$). In fact, 5 out of 28 patients (18%) in group B, compared to 14 out of 31 (45%) in group A, needed postoperative allogenic blood. Thus, the rate of allogenic blood transfusion in group B was almost 30% lower than that of group A (1.42 vs 1.90 units/patient; $P < 0.05$), despite the higher blood loss of the former group (Table 1, Table 2). However, no significant differences were found in operation length, preoperative and postoperative haemoglobin, complications or days of hospitalization (Table 1).

Table 2 Blood lost and blood units transfused

	Group A	Group B
Blood lost (ml)	1125 \pm 63	1399 \pm 68*
Intraoperative	670 \pm 46	852 \pm 55*
Postoperative	480 \pm 40	542 \pm 41
Allogenic blood (units/pt)	1.90 \pm 0.22	1.42 \pm 0.13*
Intraoperative	1.35 \pm 0.13	1.22 \pm 0.11
Postoperative	0.64 \pm 0.5	0.25 \pm 0.19*
Autologous shed blood (units/pt) ^a	0	0.65 \pm 0.05**
Overall transfusion rate	1.90 \pm 0.22	2.13 \pm 0.19

* $P < 0.05$; ** $P < 0.01$

^aCalculated according to the expression described in Materials and methods

Table 3 Haematological characteristics of reinfused shed blood in comparison to preoperative blood. Data are the mean \pm SE of 20 determinations (MCF median corpuscular fragility, f fresh, i incubated, PFHB plasma-free haemoglobin, ATP adenosine triphosphate, DPG diphosphoglycerate)

	Preoperative blood sample	Shed blood sample
Erythrocytes ($10^6/\mu\text{l}$)	4.73 \pm 0.4	3.00 \pm 0.2**
Haemoglobin (g/dl)	14.2 \pm 0.3	9.6 \pm 0.8**
Haematocrit (%)	41.3 \pm 3.2	28.5 \pm 1.8**
MCF f (NaCl %)	0.406 \pm 0.01	0.422 \pm 0.01
MCF i (NaCl %)	0.498 \pm 0.01	0.486 \pm 0.01
PFHB (mg/l)	49 \pm 8	2029 \pm 146**
Haptoglobin (mg/dl) ^a	160 \pm 34	101 \pm 14
ATP content ($\mu\text{mol/g Hb}$)	3.5 \pm 0.8	4.3 \pm 0.7
DPG content ($\mu\text{mol/g Hb}$)	16.1 \pm 2.6	11.5 \pm 2.6
Leukocytes ($10^3/\mu\text{l}$)	6.8 \pm 1.6	6.7 \pm 0.6
Histamine (nmol/l) ^a	1.4 \pm 0.3	2.4 \pm 1.4
Platelets ($10^3/\mu\text{l}$)	186 \pm 71	63 \pm 5**

** $P < 0.01$

^aHaptoglobin and histamine levels were corrected for haemodilution using total protein in preoperative blood sample as control

Characteristics of salvaged shed blood

Blood haematology

As shown in Table 3, blood samples obtained from the drainage reservoir in the 6th hour of the postoperative period contained fewer erythrocytes, less haemoglobin and haematocrit, and fewer platelets than the preoperative control sample, while PFHB levels were significantly raised. There were no important morphological abnormalities in the red cells from any of the perioperative samples, and no significant differences were found in either MCF determined from the erythrocyte osmotic fragility curves or in red cell ATP and DPG contents (Table 3). In comparison with the control sample, platelet counts, but not white cell counts or histamine levels, were significantly lower in shed blood (Table 3).

Serum biochemistry

Serum concentrations of total cholesterol, phospholipids, triglycerides, and total protein were decreased in drainage blood, while those of glucose remained unchanged. In contrast, serum concentrations of most enzymes were higher (Table 4). No significant differences were observed in Cl^- or Na^+ concentrations. However, K^+ concentrations were significantly increased in shed blood, correlating with PFHB levels ($r = 0.93$, $P < 0.02$) (Table 4).

Table 4 Biochemical characteristics of reinfused shed blood in comparison to preoperative blood. Data are the mean \pm SE of 20 determinations (*GOT* glutamate-oxalacetate aminotransferase, *GPT* glutamate-pyruvate aminotransferase, *GGT* gamma-glutamyl transpeptidase, *LDH* lactate dehydrogenase, *CK* creatine kinase, *ALP* alkaline phosphatase)

	Preoperative blood sample	Shed blood sample
Glucose (mg/dl)	100.0 \pm 14.9	107 \pm 15
Cholesterol (mg/dl)	218.0 \pm 43.0	152 \pm 9
Triglycerides (mg/dl)	184.6 \pm 102.0	114 \pm 11
Phospholipids (mg/dl)	249.6 \pm 41.6	131 \pm 22
Total proteins (g/dl)	7.67 \pm 0.96	6.05 \pm 1.12
GOT (U/l) ^a	26 \pm 3	1857 \pm 231**
GPT (U/l) ^a	15 \pm 2	314 \pm 67**
GGT (U/l) ^a	27 \pm 6	25 \pm 5
LDH (U/l) ^a	293 \pm 29	7452 \pm 662**
CK (U/l) ^a	62 \pm 13	58791 \pm 2168**
ALP (U/l) ^a	68 \pm 5	95 \pm 15
Na^+ (mmol/l)	135 \pm 1.6	140 \pm 1.4
K^+ (mmol/l)	4.9 \pm 0.2	6.4 \pm 0.5*
Cl^- (mmol/l)	106 \pm 1.3	106 \pm 1.7

* $P < 0.05$; ** $P < 0.01$

^aSerum enzyme levels were corrected for haemodilution using total protein in preoperative blood sample as control

Serum immunology

As shown in Table 5, IL-1 β and IL-6 levels were high in shed blood samples, while those of TNF α were always below the detection limit. Complement fractions and immunoglobulin concentrations remained unchanged.

Impact of reinfused drainage blood on patient blood parameters

Coagulation parameters

No changes in standard coagulation parameter values were detected 1–2 h after shed blood reinfusion, as compared with those obtained in the preoperative period (Table 6).

Serum enzymes

Table 7 shows serum enzyme levels follow-up over 14 perioperative days in patients with or without reinfusion of postoperative salvaged shed blood. In both groups, serum levels of some enzymes were elevated at the end of surgery and remained raised at postoperative day 2 (CK) or

Table 5 Immunological characteristics of reinfused shed blood in comparison to preoperative blood. All results were corrected for haemodilution using total protein in preoperative blood sample as control. Data are the mean \pm SE of 20 determinations (*TNF* tumour necrosis factor, *Ig* immunoglobulin, *ND* below the limit of detection)

	Preoperative blood sample	Shed blood sample
IL-1 β (pg/ml)	4.6 \pm 1.1	10.7 \pm 1.5*
IL-6 (pg/ml)	2.5 \pm 1.1	1335 \pm 49**
TNF α (pg/ml)	ND	ND
C3 (mg/dl)	185 \pm 9	134 \pm 8
C4 (mg/dl)	35 \pm 5	25 \pm 2
Ig A (mg/dl)	272 \pm 44	191 \pm 33
Ig G (mg/dl)	1128 \pm 110	895 \pm 72
Ig M (mg/dl)	164 \pm 22	107 \pm 19

* $P < 0.05$; ** $P < 0.01$

Table 6 Coagulation parameters after the reinfusion of postoperative shed blood. Data are the mean \pm SE of 20 determinations (*PT* prothrombin time, *APTT* activated partial thromboplastin time, *PA* prothrombin activity)

	Preoperative	1–2 h post-reinfusion of shed blood	P-value	Reference values
PT	12.2 \pm 0.2	13.2 \pm 0.4	NS	12.2 s
APTT	29.8 \pm 1.0	32.0 \pm 4.0	NS	29.2 s
PA	97.7 \pm 3.1	86.6 \pm 3.9	NS	75–124%

Table 7 Perioperative evolution of serum enzyme levels in patients with or without reinfusion of postoperative shed blood. All serum enzyme levels were corrected for haemodilution using total protein in preoperative blood sample as control. Data are the mean \pm SE of 20 determinations [normal ranges (U/l): GOT 10–34, GPT 10–44, GGT 7–32, LDH 210–420, CK 25–195, ALP 10–1500]

	Sample 1 Preoperative day-7	Sample 2 End of surgery, day 0	Sample 4 Postoperative day 1	Sample 5 Postoperative day 2	Sample 6 Postoperative day 7
GOT (U/l)					
Control	23 \pm 3	37 \pm 5	86 \pm 12**	59 \pm 8**	50 \pm 9*
Reinfused	26 \pm 3	46 \pm 7*	130 \pm 21**	75 \pm 14**	42 \pm 5*
GPT (U/l)					
Control	18 \pm 3	23 \pm 4	28 \pm 5	24 \pm 5	22 \pm 7
Reinfused	15 \pm 2	17 \pm 4	26 \pm 3	24 \pm 4	22 \pm 2
GGT (U/l)					
Control	10 \pm 2	15 \pm 4	16 \pm 3	21 \pm 10	11 \pm 3
Reinfused	27 \pm 6	28 \pm 9	26 \pm 6	29 \pm 7	41 \pm 8
LDH (U/l)					
Control	302 \pm 21	396 \pm 46	485 \pm 53**	506 \pm 63**	402 \pm 85
Reinfused	293 \pm 29	458 \pm 85*	780 \pm 118**	615 \pm 59**	526 \pm 66**
CK (U/l)					
Control	57 \pm 10	221 \pm 42*	710 \pm 134**	518 \pm 58**	127 \pm 26
Reinfused	62 \pm 13	216 \pm 46*	2558 \pm 339**	676 \pm 110**	100 \pm 27
ALP (U/l)					
Control	83 \pm 9	94 \pm 11	100 \pm 10	109 \pm 9	119 \pm 20
Reinfused	68 \pm 5	67 \pm 8	69 \pm 8	77 \pm 7	88 \pm 9

* $P < 0.05$; ** $P < 0.01$

7 (GOT, LDH), with a higher increase if postoperative autotransfusion was used as a blood saving method. No significant changes were observed in serum levels of GPT, GGT and ALP.

Cytokine levels

Perioperative serum levels of cytokines are depicted in Fig. 1, showing a 4-fold increase at the end of surgery and a 20-fold increase at postoperative day 1 in those of IL-6.

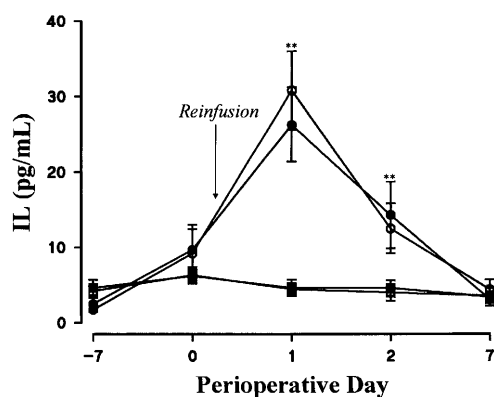


Fig. 1. Perioperative evolution of serum cytokine levels in patients with (solid symbols) or without (open symbols) reinfusion of postoperative shed blood. Serum levels of IL-1 β (\square , \blacksquare) and IL-6 (\circ , \bullet) were measured by ELISA and corrected for haemodilution using total protein in preoperative blood sample as control. Data are the mean \pm SE of 20 determinations (** $P < 0.01$)

Despite the high IL-6 content of shed blood (Table 5), IL-6 concentrations were not further increased by reinfusion of shed blood, and returned to control levels by postoperative day 7. In contrast, no significant changes were observed in IL-1 β levels, while those of TNF α were always below the detection limit.

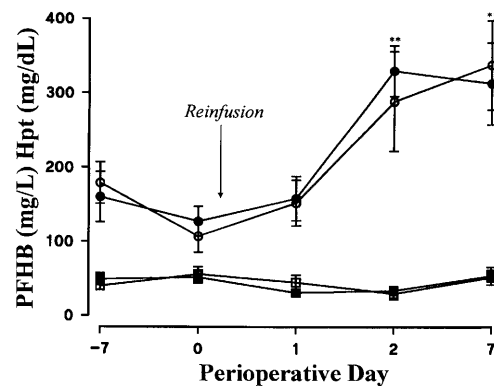


Fig. 2 Perioperative evolution of serum haptoglobin and plasma-free haemoglobin levels in patients with (solid symbols) or without (open symbols) reinfusion of postoperative shed blood. Nephelometric measurements of serum levels of haptoglobin (Hpt, \circ , \bullet) were corrected for haemodilution using total protein in preoperative blood sample as control. Plasma-free haemoglobin levels (PFHB, \square , \blacksquare) were determined according to the bencidine reaction. Data are the mean \pm SE of 20 determinations (** $P < 0.01$)

PFHB-haptoglobin levels

PFHB and haptoglobin concentrations along the perioperative observation time displayed almost identical profiles in the two study groups (Fig. 2). Those of haptoglobin showed a decrease at the end of surgery, which was followed by a continuous increase until postoperative day 7, while those of PFHB remained unchanged along the observation time.

Discussion

Perioperative collection and reinfusion of autologous blood is beneficial for the patient, as it may decrease the allogenic blood requirements, thus reducing or avoiding the risk of transfusion-associated complications. Preoperative donations of several blood units is becoming a standard practice before major elective orthopaedic procedures, and there is a consensus on its safety and efficacy. However, it can be more expensive per unit than allogenic blood, while intraoperative red cell salvage seems to be cost-effective if a blood loss higher than 1000 ml is expected [21]. On the other hand, the safety of the reinfusion of postoperative unwashed filtered shed blood is well documented, but its efficacy has been questioned, mainly on the basis of the viability and functionality of these erythrocytes and the alteration of haemostasia leading to increased bleeding. Since the latter point of controversy has been addressed by several authors and recently reviewed [21, 31], this work aimed to assess the degree of haematological, biochemical and immunological normality of blood collected in the postoperative period in elective spine procedures and the functionality of its red cells, its efficacy in reducing allogenic blood requirements and its impact on patient blood parameters.

Effectiveness of shed blood reinfusion

In these series of patients, the reinfusion of shed blood resulted in a 60% reduction in postoperative allogenic blood requirements (0.25 vs 0.64 units/patient, $P < 0.05$). Thus, the rate of allogenic transfusion in group B was almost 30% lower than that of group A (1.42 vs 1.90 units/patient; $P < 0.05$) (Table 2). These results, which are almost identical to those reported by Behrman and Keim [3] in a series of spinal patients, confirm the effectiveness of the procedure as a tool towards the achievement of bloodless spinal surgery, especially when it is used in association with other blood conservation techniques. Moreover, since the last 50–100 ml of shed blood was scrapped and a microaggregated filter was intercalated in the line, no reactions were seen after shed blood reinfusion [20].

Characteristics of postoperative drainage blood

From a haematological point of view, shed blood samples collected in the postoperative period contained lower erythrocyte and platelet counts, haematocrit and haemoglobin than blood drawn from the patient in the preoperative period, but their erythrocytes did not show any significant morphological abnormality, presented a normal MCF and maintained a normal energy metabolism, since no changes were observed in intracellular concentrations of ATP and DPG. DPG is a red cell metabolite with some unique functions, and is derived from glucose metabolism. In addition to its well-known function in reducing haemoglobin- O_2 affinity, DPG modulates the mechanical properties of the erythrocyte membrane. In this sense, a critical role for the major linkages between the bilayer and the underlying skeleton in control of membrane stability has been demonstrated [22]. Recently, it has been reported that the two major attachment sites of the membrane skeleton (ankyrin and protein 4.1) are specifically dissociated by physiological concentration of DPG, suggesting that DPG may influence erythrocyte function far beyond its role in regulating haemoglobin- O_2 affinity [23]. The fact that DPG content in erythrocytes from shed blood is not significantly different from that of control blood adds more arguments in favour of the normal functionality of these cells.

Taken together, our results seem to show that these red cells are not significantly damaged, keeping all their functionality, and, hence, there is no reason to be concerned about their viability. In this regard, it has been reported that erythrocytes collected in the postoperative period in patients undergoing posterior spinal fusion have a viability comparable to those from intraoperative and preoperative blood collection [9, 17, 26].

The observed reduction in the haematological parameters could be due to haemodilution, bleeding and haemolysis. Because of bleeding and the relative haemodilution, due to the administration of glycosaline solutions for volume reposition during surgery, there was also a decrease in cholesterol, phospholipids, triglycerides and protein concentrations in shed blood, but not in glucose and ions. In regard to haemolysis, measurement of PFHB has been used as an index of haemolysis and, certainly, its levels in shed blood samples were above the normal limits. Moreover, the increased serum concentrations of K^+ , GOT, and LDH in these samples also suggest a degree of haemolysis (increased levels of GPT and CK, as well as partially those of the LDH, are most probably due to enzyme release from muscles during surgery). However, it has been previously reported that if postoperative shed blood is reinfused up to 15% of the total blood volume [5] or 1000 ml [2], there seems to be enough haptoglobin (an acute phase reactant) in the general circulation to bind free haemoglobin, avoiding possible renal damage [3, 11, 13] (Fig. 2).

Impact on patient blood parameters

The changes in the coagulative capacity of the patients, the content of immunologically bioactive substances and the presence of confounding factors are among the concerns reported about the reinfusion of unwashed shed blood.

In regard to coagulation, levels of fibrinogen, AT-III, factor VIII and plasminogen are lower than those measured in fresh blood, while those of FDP or PAI-I are higher [5, 11, 16]. However, for these parameters there are no differences between blood samples drawn before and those drawn 1 or 24 h after the reinfusion of shed blood [11, 16]. In addition, in comparison to preoperative values, we did not find significant differences in PT or APTT (Table 6) measured 1–2 h after completion of shed blood reinfusion. Similar results have been reported within 15 min following completion of reinfusion [27]. Moreover, reinfusion of unwashed shed blood did not result in an increase in bleeding, since there were no differences in postoperative blood loss (Table 2).

On the other hand, high levels of GOT, GPT, LDH and CK, and normal levels of ALP and GGT, were measured in postoperative shed blood samples. In the control group, perioperative serum levels of these enzymes in the patient evolved in different ways: GPT levels were normal at postoperative day 1 and CK levels at postoperative day 7, while those of GOT and LDH were still high at postoperative day 7. These alterations in serum enzyme levels are most probably due to enzyme release from the injured muscles, to inflammation-induced postoperative intravascular haemolysis and to transfusion of stored allogenic blood. A very similar pattern, but more pronounced, was observed in those patients receiving postoperative shed blood, due to the extremely high enzyme content of this blood (Table 4).

Therefore, as reported for cardiac surgery [24], caution should be taken when serum levels of those enzymes are used for diagnosis (e.g. to determine postoperative myo-

cardial or hepatic injury) after spinal surgery, especially if postoperative autotransfusion is used as a blood saving method.

Another point of controversy regarding the reinfusion of unwashed postoperative shed blood is the presence of mediators, such as cytokines, complement fractions or histamine, which may modify the immune response of the patient. In our study, except for a dramatic increase in IL-6, there were no other major changes in these mediators in shed blood. Moreover, as has been reported for cardiac surgery and orthopaedic surgery [4, 19, 29], after the reinfusion of shed blood, cytokine levels were within the normal range from postoperative day 1 and they were no different to those measured in the control patients (Fig. 1).

Conclusions

In conclusion, postoperative salvage of blood seems to be an excellent source of functional and viable red cells, without many of the transfusion-related risks. Moreover, as in other studies in knee, hip or spine surgery [2, 3, 5, 6, 11, 13, 14, 15, 30, 32] and cardiac surgery [1, 7, 28], postoperative reinfusion of unwashed filtered shed blood after major spine procedures has proved to reduce homologous transfusion requirements, with only minor complications. A prospective study, using the Constavac CBC II plus short-time preoperative autologous blood predeposit, is currently in progress to evaluate the impact of this strategy, not only on the prevalence of homologous blood transfusion but also on the haemostatic and immunological status of the patients.

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