ORIGINAL ARTICLE

# Short-term effects of extracorporeal graft rinse versus circulatory graft rinse in living donor liver transplantation. A prospective randomized controlled trial

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## SUMMARY

Living donor liver transplantation has shorter cold ischemia time, less preservative volume, and lower metabolic load compared to transplantation from deceased donors. We investigated the impact of rinsing the graft contents into the systemic circulation on operative course and postoperative outcomes. Donors had right hepatectomy, and grafts were preserved with cold histidine-tryptophan-ketoglutarate solution. On ending portal vein anastomosis, grafts were flushed by patient's portal blood either through incompletely anastomosed hepatic vein (extracorporeal rinse group, EcRg, $n = 40$) or into systemic circulation (circulatory rinse group, CRg, $n = 40$). The primary outcome objective was the lowest mean arterial blood pressure within 5 min after portal unclamping as a marker for postreperfusion syndrome (PRS). Secondary objectives included hemodynamics and early graft's and patient's outcomes. Within 5 min postreperfusion, mean arterial blood pressure was significantly lower in the CRg compared to the EcRg, yet this was clinically insignificant. Postoperative graft functions, early biliary and vascular complications, and three-month survival were comparable in both groups. Rinsing the graft into the circulation increased the incidence of PRS without significant impact on early graft or patient outcome in relatively healthy recipients.

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**Key words**
graft flushing, graft rinse, ischemia/reperfusion injury, postreperfusion syndrome, preservative solution

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## Introduction

Liver transplantation (LT) is the principal treatment for end-stage liver diseases and selected cases of liver neoplasms [1]. Living donor liver transplantation (LDLT) either supplements liver transplantation from deceased donors by increasing the donor pool or serves as a sole source of liver graft in some countries that do


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not allow donation from deceased donors for cultural, social, or religious reasons [2]. Postreperfusion syndrome (PRS) and ischemia reperfusion injury (IR) are well-known complications in LT with a major impact on graft and patient outcomes [3–6]. Previous investigators tested several techniques for graft reperfusion aiming to reduce the impact of graft preservative solution and ischemic products of the congested gut on the hemodynamics and graft function after portal unclamping. These techniques were tested in liver transplantation from deceased donors with no consensus on a preferred technique [7–10]. In our exclusively LDLT program, we adopted graft extracorporeal rinse with patient's own portal blood as a default practice for 6 years. This technique consumes a considerable blood volume over a swift time leading to acute hemodynamic derangement and may necessitate avoidable blood products transfusion. LDLT has a shorter preservation and cold ischemia times (CIT), and less preservative fluid volume is used compared to full graft transplantation from deceased donors. This advantage should be reflected as ameliorated hemodynamic turbulence and PRS. The impact of graft and portal extracorporeal rinse techniques on hemodynamics and patient outcome exclusively in living LDLT has not been previously tested in a RCT to the best of our knowledge. As both extracorporeal rinse and circulatory rinse techniques have their own merits and mechanisms for inducing hemodynamic changes, we conducted this prospective double-blind randomized controlled trial to examine the effects of introducing graft preservative solution and mesenteric blood into the systemic circulation versus our routine practice purging them out for suctioning exclusively in LDLT. Our null hypothesis assumed that there will be no statistically significant differences between both extracorporeal rinse and circulatory rinse techniques concerning the severity of postreperfusion syndrome as a primary outcome measure in this trial.

## Patients and methods

### Design

This study prospectively involved 80 adult recipients between 20 and 60 years old who underwent LDLT with right lobe liver graft in Mansoura university liver transplantation program, through prospective double-blinded randomized controlled design. The study was approved by the institutional review board in Faculty of Medicine, Mansoura University (R/15.08.49), and informed consents were secured from all patients. The trial registration code in clinicaltrials.gov is NCT02540447. Patients operated for retransplantation, patients with previous upper abdominal operation, with Budd–Chiari syndrome, and with mild porto-pulmonary hypertension were excluded from this study. All operations were performed by the same anesthesia and surgery teams.

### Patients

Eighty-nine patients were assessed for over 18 months, and 80 patients were enrolled in the trial and randomized into two groups. All enrolled patients completed the study protocol, and their results were included in the statistical analysis (Fig. 1).

### Anesthesia technique

On admission to the operating suite, patients received intravenous (IV) pantoprazole sodium (40 mg) and midazolam (3 mg). Anesthesia team started anesthesia with IV propofol 1–1.5 mg/kg, fentany 1.2 μg/kg, and rocuronium bromide 0.6 mg/kg. A 7.5 French continuous thermal fiber-optic pulmonary artery catheter (CCO/SvO2 Edwards Life Science, Irvine, CA, USA) was inserted via the right internal jugular vein under fluoroscopic guidance then connected it to a dedicated monitor (Vigilance monitor; Edwards Life Science). An arterial catheter was inserted in the left radial artery after performance of modified Allen's test. Anesthesia was maintained with sevoflurane, fentanyl (1–2 μg/kg/h), and rocuronium infusion (200–300 μg kg/h). We adopted a dynamic goal directed fluid replacement protocol. Blood products transfusion followed the American society of Anesthesiologists (ASA) guidelines for blood transfusion [11]. Both the patient and the primary anesthetist were blinded about the study intervention as a second anesthetist took over patient management from the end of anhepatic period (marked by the surgeon just before the last stitch in the hepatic vein is tied) to the portal unclamping (maximum 10 min), and then, the first anesthetist was allowed access to the patient management again while not informed about the nature of the intervention till the end of the study period. During this window, data recording was achieved by a senior nurse who is not included in the study. Meanwhile, a surgeon who was not involved in the recipient surgery and was blinded for the flushing technique was responsible for reporting postoperative data including the complications.


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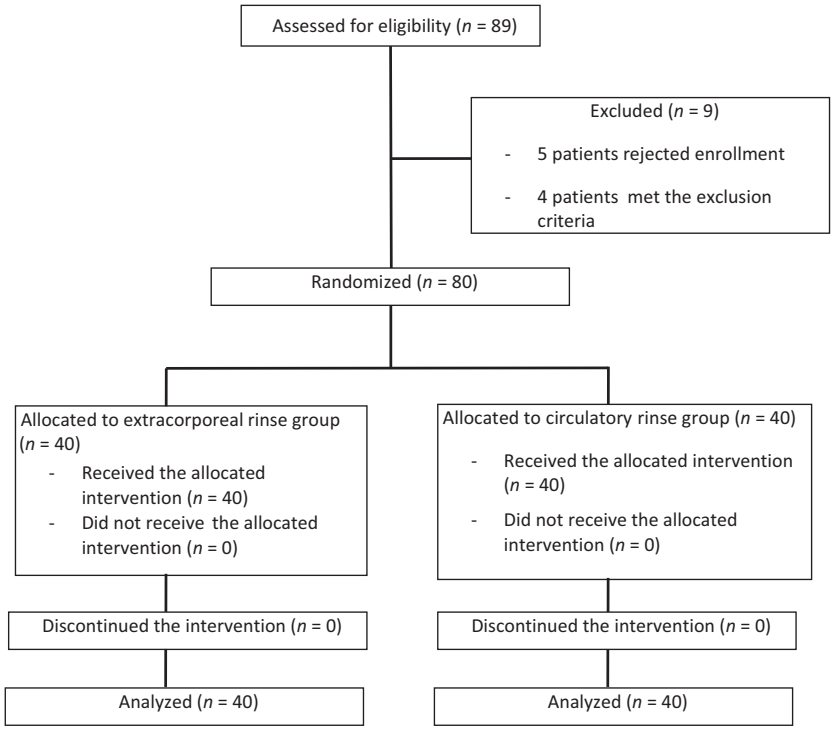


**Figure 1** CONSORT chart for the trial.

## Surgical technique

The surgical technique was previously described [12,13]. The donor surgical team excised the right liver lobe (without inclusion of the middle hepatic vein) and flushed for preservation on the back table with 3–4 l of cold histidine-tryptophan-ketoglutarate (HTK) (Custodial, Bensheim, Germany) via antegrade flushing of the portal vein to get completely clear effluent fluid without flushing via the hepatic artery [14,15]. Both recipient and donor operations were synchronized to minimize graft preservation time. Upon starting recipient portal vein anastomosis, a closed envelope containing a computer-generated random allocation code was opened to allocate the patient into one of two groups, a control group (extracorporeal rinse group, EcRg, $n = 40$) or a treatment group (circulatory rinse group) (CRg, $n = 40$).

In the extracorporeal rinse group, the right hepatic vein (RHV) anastomosis was completed (apart from the last knot) using continuous Proline 4/0 sutures. After completion of the portal vein (PV) anastomosis and before tying the continuous Proline 6/0 sutures used, the donor portal vein was irrigated with heparinized saline. The recipient PV was totally unclamped, while the donor PV is occluded using vascular forceps, to wash off any thrombi and confirm good portal flow. The PV anastomosis was finished by tying the ends of the continuous sutures. The anesthetist was notified about the upcoming PV unclamping. The surgical field was completely dried then, the portal blood was used to wash the graft off the preservative solution escaping through the incomplete RHV anastomosis and sucked through a separate external sucker, and its volume was recorded. The volume of portal blood used was based on the graft volume (0.4–0.6 ml/g) including the washed preservative solution held in the graft's portal and venous systems that cannot be actually quantified, yet it is generally a minor volume in partial liver grafts. The RHV anastomosis was finished by tying the last knot and unclamped followed by unclamping of the PV over 10 s. In the circulatory rinse group, the RHV anastomosis was finished. The PV anastomosis was performed as described previously. The RHV was unclamped then the PV was unclamped over 10 s, and the graft preservative contents were washed into the systemic circulation by the portal blood at portal unclamping.

## Rescue therapy

During anhepatic phase, fluid (4% Albumin) and blood product transfusion commenced to keep (SV) stroke volume within 20% of the basal value, CVP around 5 mmHg, and hemoglobin more than 8 g/dl. In the extracorporeal rinse group, patients received a rapid

infusion of equal volume to the purged solution in the external sucker. Either albumin 4% or RBCs (cell saver or packed homologous) based on the end-anhepatic hemoglobin level. The infusion of this volume is finished before commencing to portal unclamping. At portal reperfusion, the initial treatment of hypotension (systolic arterial blood pressure <30% of the end-anhepatic reading) was IV infusion of 500 ml bolus albumin 4% over 5 min as long as the mean arterial blood pressure (MAP) was above 60 mmHg. Whenever MAP was <60 mmHg, ephedrine in increments of 6 mg IV to a maximum of 30 mg was given. If MAP remained <60 mmHg after 5 min from portal unclamping time, noradrenaline 50 ng/kg was given as IV injection followed by intravenous infusion of noradrenaline 100–200 ng kg/min until we had MAP readings over 70 mmHg for complete 5 min when we gradually withdrew noradrenaline.

### Outcome measures

The primary outcome objective for this trial was the lowest MAP within 5 min after portal unclamping as an indicator for the severity of PRS with and without graft extracorporeal rinse. For this objective, invasive MAP was recorded every minute during the first 5 min postreperfusion and the lowest reading is recorded as the lowest MAP within 5 min postreperfusion. The original definition of PRS described by Aggarwal *et al.* [16] included hemodynamic and fibrinolytic activity, yet some authors used only the hemodynamic disturbance to define mild PRS. In this trial, we described PRS as drop of the MAP by 30% with a decrease in the systemic vascular resistance compared to the end-anhepatic value sustained for at least 1 min within 5 min after portal unclamping. The incidence of PRS was recorded as well as the need for noradrenaline. Secondary objectives were to assess the hemodynamic, laboratory, and outcome parameters in the studied groups. Hemodynamic parameters assessed were heart rate (HR), central venous pressure (CVP), mean pulmonary artery pressure (mPAP), pulmonary artery occlusion pressure (PAOP), cardiac index (CI), pulmonary vascular resistance (PVR), systemic vascular resistance (SVR), and mixed venous oxygen saturation (SvO2). These variables were measured at the end of anhepatic phase as a basal value, immediately before portal reperfusion, and at 5, 15, 60 min after portal unclamping, and at skin closure. Laboratory assessment of graft and renal functions included measurement of pH, international normalized ratio (INR), serum levels of serum glutamic-pyruvic transaminase (SGPT), gamma glutamyl transferase (GGT), bilirubin, lactate, and creatinine at 1st, 3rd, 7th, and 28th postoperative days. Recorded outcome measures included lengths of intensive care stay and hospital stay, early postoperative complications (30 days), and three-month patient survival. Graft rejection and ischemia/reperfusion injury were both diagnosed and graded by histopathology examination of day 7 needle biopsy (analyzed by a single consultant liver pathologist).

### Statistical analysis

Based on a Pilot trial including 16 patients in our institute, a sample size was calculated to achieve 80% power

**Table 1.** Demographic and operative date for the studied groups, EcRg ($n$ = 40) and CRg ($n$ = 40).

| | EcRg ($n$ = 40) | CRg ($n$ = 40) | $P$ value |
|---|---|---|---|
| Age (years) | 51.5 ± 6.19 | 50.90 ± 7.62 | 0.348 |
| Sex (F/M) | 8/32 | 6/34 | – |
| HCC/HCV | 16/24 | 18/22 | – |
| MELD score | 15.23 ± 5.04 | 15.35 ± 3.89 | 0.175 |
| Operative time (min) | 600.68 ± 40.95 | 609.83 ± 46.79 | 0.452 |
| Warm ischemia time (min) | 37.98 ± 10.0.5 | 36.55 ± 8.97 | 0.249 |
| Cold ischemia time (min) | 35.15 ± 19.50 | 33.80 ± 12.55 | 0.097 |
| GRWR | 0.911 ± 0.164 | 0.935 ± 0.176 | 0.721 |
| ICU stay (day) | 6.13 ± 2.12 | 5.75 ± 1.82 | 0.632 |
| Hospital stay (day) | 25.55 ± 10.36 | 24.68 ± 7.19 | 0.074 |
| Donor Age (years) | 26.8 ± 4.6 | 25.9 ± 4.2 | 0.331 |

MELD, model of end-stage liver disease score; HCC, hepatocellular carcinoma; HCV, hepatitis C viral cirrhosis; GRWR, graft weight – recipient body weight ratio.

Data are expressed as mean ± SD, number.



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**Table 2.** Hemodynamic and mixed venous oxygen saturation date for the studied groups, Ec Rg ($n$ = 40) and CRg ($n$ = 40).

| | End-anhepatic | | Prereperfusion | | Within 5-min reperfusion | | 15-min reperfusion | | 60-min reperfusion | | Closure | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) |
| Heart rate (bpm) | 91.3 ± 11.6 | 89.3 ± 10.9 | 93.4 ± 31.0 | 96.8 ± 28.6 | 96.9 ± 9.4 | 104.2 ± 11.6 | 89.2 ± 7.3 | 87.9 ± 7.7 | 84.6 ± 4.7 | 82.8 ± 5.2 | 77.6 ± 5.2 | 78.3 ± 4.3 |
| Mean arterial blood pressure (mmHg) | 87.3 ± 11.2 | 89.4 ± 13.5 | 84.2 ± 22.4 | 85.6 ± 18.9 | 66.0 ± 6.1 | **57.8 ± 5.9*** | 80.7 ± 6.3 | 80.2 ± 6.4 | 84.4 ± 7.5 | 84.2 ± 9.1 | 88.6 ± 7.6 | 90.8 ± 8.8 |
| Mean pulmonary artery pressure (mmHg) | 11.6 ± 4.1 | 12.8 ± 2.6 | 10.5 ± 3.1 | 13.4 ± 4.2 | 16.0 ± 2.0 | **20.2 ± 3.3*** | 17.7 ± 2.6 | 18.3 ± 2.9 | 16.0 ± 2.1 | 16.3 ± 2.1 | 13.6 ± 2.3 | 14.6 ± 2.4 |
| Pulmonary artery occlusion pressure (mmHg) | 5.8 ± 1,6 | 5.3 ± 1.3 | – | – | 7.2 ± 1.4 | **6.1 ± 1.3*** | 8.9 ± 1.5 | 8.7 ± 1.5 | 9.2 ± 1.9 | 9.2 ± 1.9 | 7.6 ± 2.2 | 8.2 ± 2.4 |
| Cardiac index (l min $m^2$) | 3.8 ± 0.3 | 4.1 ± 0.7 | 4.1 ± 0.8 | 4.6 ± 1.0 | 4.4 ± 0.5 | **5.4 ± 0.7*** | 3.9 ± 0.5 | 3.8 ± 0.4 | 3.4 ± 0.4 | 3.4 ± 0.4 | 3.2 ± 0.3 | 3.3 ± 0.4 |
| Systemic vascular resistance index (dyne, s, $cm^5$ $m^2$) | 1847 ± 267 | 1798 ± 303 | 1755 ± 428 | 1701 ± 387 | 1138 ± 224. | **894 ± 180*** | 1558 ± 234 | 1617 ± 243 | 1911 ± 304 | 1900 ± 240 | 2128 ± 310 | 2179 ± 340 |
| Pulmonary vascular resistance index (dyne, s, $cm^5$ $m^2$) | 173 ± 61 | 169 ± 59.4 | – | – | 163 ± 38.5 | **214 ± 65.4*** | 180 ± 50.9 | 206 ± 69.8 | 161 ± .37.8 | 171 ± 37.0 | 151 ± 40.9 | 159 ± 38.2 |
| Mixed venous oxygen saturation (%) | 79.7 ± 6.3 | 78.4 ± 4.2 | 78.4 ± 3.7 | 78.9 ± 4.8 | 81.5 ± 3.0 | 79.5 ± 4.1 | 78.7 ± 3.3 | 80.2 ± 3.6 | 82.1 ± 2.9 | 82.9 ± 2.9 | 82.7 ± 3.2 | 83.4 ± 2.9 |

Data are expressed as mean ± SD.

*Statistically significant compared to the EcRg group.

to detect an effect size of 40% in the 5 min postreperfusion mean arterial blood pressure with an alpha level of 0.05. A per protocol population analysis was adopted. Thirty-six patients per arm were required, yet four patients were added per arm to compensate for drop-outs. Quantitative data distribution was tested by Shapiro–Wilk test and all exhibited normal distribution. Statistical differences between the control and treatment groups were verified using independent-samples Student's *t*-test (two-tailed) or Fisher's exact test as relevant. Pearson chi-square was used to test associations between binominal data.

## Results

The study included 80 adult LDLT recipients, all completed the study, and their data were subjected to statistical analysis. Patient's characteristics and operative data did not show any statistically significant differences among the studied groups (Table 1).

Within 5 min postreperfusion, the hemodynamic profile exhibited statistically significant differences between both groups where the lowest MAP was significantly lower in the circulatory rinse group compared to the extracorporeal rinse group (57.8 ± 5.9 mmHg vs. 66.0 ± 6.1 mmHg, $P = 0.03$). Similarly, SVR and PAOP were significantly lower in the CRg compared to the EcRg. MPAP, PVR, and CI were significantly higher in CRg compared to EcRg during the lowest MAP reading within 5 min postreperfusion time frame (Table 2). Number of postreperfusion hypotensive episodes defined as MAP <30% of the end-anhepatic phase reading, number of patients with episodes of severe hypotension (MAP <60 mmHg) within 5 min postreperfusion as well as number of patients receiving nor-adrenalin were statistically more frequent in the CRg compared to EcRg (Figure 2). The mean reduction of 5 min postreperfusion MAP to the end-anhepatic MAP (basal) was statistically significant between EcRg and CRg (23.2 ± 4.3 vs. 33.2 ± 6.5 mmHg). Based on a pure hemodynamic definition of PRS, the incidence of PRS was 45% vs. 90% in EcRg and CRg groups respectively (Table 1).

No statistically significant differences between both groups in the assessed laboratory parameters until the 28th day (Table 3). Over 30 days, the encountered (early) complications episodes did not differ statistically in both groups, nor did the 3-month mortality [4 patients (10%) in EcRG vs. 3 patients (7.5%) in CRG] (Table 4).

## Discussion

Our null hypothesis was rejected. The magnitude of PRS was greater in the CRg group without difference in the early postoperative outcomes compared to the EcRg. This was manifested by lower MAP and SVR and higher SV, CI, mPAP, and PAOP in the CRg compared to the EcRg. Postreperfusion syndrome has been described around three decades ago [16]. In our trial, the incidence of PRS was statistically significantly higher in the EcRg compared to CRg (45% vs. 90%). Reports for incidence of PRS from 2% to 61.3% have been published, with this variability attributed to numerous definitions of PRS and retrospective designs in which confounders cannot be always nullified [17–22]. Hilmi and co-workers, in a retrospective study, reported a 100% incidence of varying degrees of PRS in recipients of liver graft from deceased donors [6]. They defined PRS using only the changes in the postreperfusion

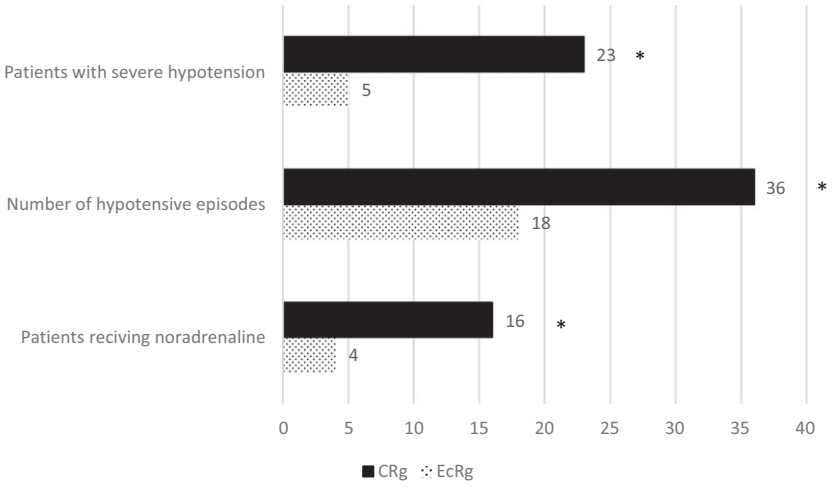


**Figure 2** Number of patients received receiving noradrenaline, number of postreperfusion hypotensive episodes mean arterial blood pressure (MAP <30% of end-anhepatic value), and number of patients with at least one severe hypotension episodes (MAP ≤60 mmHg 5 min postreperfusion in the studied groups (extracorporeal rinse group EcRg, $n = 40$ and circulatory rinse CRg group, $n = 40$). Data are in number. *Statistically significant compared to the EcRg group.

**Table 3.** Laboratory date for the studied groups, EcRg ($n$ = 40) and CRg ($n$ = 40).

| | POD 1 | | POD 3 | | POD 7 | | POD 28 | |
|---|---|---|---|---|---|---|---|---|
| | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) | EcRg ($n$ = 40) | CRg ($n$ = 40) |
| SGPT (mg/dl) | 146.00 ± 41.81 | 150.58 ± 40.69 | 158.98 ± 31.44 | 153.83 ± 34.06 | 54.81 ± 13.83 | 53.33 ± 13.47 | 24.63 ± 5.39 | 24.15 ± 5.01 |
| Serum bilirubin (total) (mg/dl) | 3.71 ± 1.81 | 3.093 ± 1.44 | 3.17 ± 2.10 | 2.578 ± 1.45 | 2.62 ± 2.36 | 2.110 ± 1.76 | 2.38 ± 2.59 | 1.92 ± 2.58 |
| INR | 2.27 ± 0.59 | 2.313 ± 0.59 | 1.87 ± 0.58 | 1.703 ± 0.49 | 1.54 ± 0.71 | 1.390 ± 0.58 | 1.41 ± 0.79 | 1.26 ± 0.57 |
| Serum GGT (mg/dl) | 21.45 ± 6.42 | 26.21 ± 7.2 | 39.94 ± 11.34 | 41.90 ± 9,8 | 37.63 ± 7.40 | 35.31 ± 8.12 | 30.39 ± 7.22 | 28.53 ± 7.83 |
| Serum lactate (mg/dl) | 43.75 ± 16.16 | 47.950 ± 15.58 | 10.58 ± 4.06 | 12.125 ± 4.92 | 11.15 ± 4.88 | 12.075 ± 4.70 | 15.68 ± 3.41 | 16.25 ± 3.57 |
| Serum creatinine (mg/dl) | 1.69 ± 0.0 | 1.800 ± 0.41 | 1.55 ± 0.33 | 1.538 ± 0.38 | 1.47 ± 0.47 | 1.428 ± 0.51 | 1.42 ± 0.59 | 1.265 ± 0.46 |

SGPT, serum glutamic-pyruvic transaminase; GGT, gamma glutamyl transferase; POD, postoperative day.
Data are expressed as mean ± SD.

**Table 4.** Early complications episodes (30 days) for the studied groups, EcRg ($n$ = 40) and CRg ($n$ = 40).

| | EcRg ($n$ = 40) | CRg ($n$ = 40) | $P$ |
|---|---|---|---|
| Infectious complications | 9 | 8 | 0.632 |
| Acute cellular rejection | 12 | 15 | 0.478 |
| Moderate and severe ischemia reperfusion injury | 3 | 2 | 0.644 |
| Biliary complications | 10 | 6 | 0.264 |
| Vascular complications | 1 | 3 | 0.556 |

Data are expressed as number.

hemodynamic profile and omitted the fibrinolytic component of the classic definition for PRS. This definition may lead to overestimation of the incidence of PRS due to the multifactorial nature of the hemodynamic changes after reperfusion [17]. However, this definition remains of clinical value to the anesthetist, hence, its use in several trials [19,21,22]. We adopted similar definition, which explains the high incidence of PRS in both groups. Three active constituents contribute in PRS in liver transplantation: first is the graft contents of preservative solution with harmful ingredients like potassium. Added to it, vasoactive mediators released from the graft during the period of graft ischemia (from vascular clamping in the donor to the start of graft reperfusion in the recipient). Second are the mediators released in the recipients' mesenteric circulation due to intestinal congestion during portal clamping. The third factor is the oxygen free radicles generated on restoration of blood flow [10,17,20,23,24]. Several techniques are being used to flush the graft in attempts to wash the graft contents of preservative solution, vasoactive mediators, minor thrombi, and air emboli [9,25]. Numerous transplant centers use graft flushing as the most effective technique for amelioration of PRS, yet none of these techniques proved superiority in a recent meta-analysis [25]. LDLT has the advantage of short graft ischemia time and small volume of liver graft compared to the liver graft from deceased donors, while it has the same recipient intestinal ischemic metabolite load. This fact was enough justification for some centers to abandon graft flushing in LDLT. The acute surge in serum potassium with flushing of the graft preservative solution into the systemic circulation is a rational for rinsing the graft prior to reperfusion [9,25]. In our trial, liver grafts were flushed by HTK with a lower potassium concentration (10 mmol/l) compared to university of

Wisconsin (UW) solution (120 mmol/l) prevalently used in transplant centers [9,25,26]. This lower potassium concentration in HTK allowed for *in vivo* flushing of the preservative solution in the systemic circulation in clinical practice, including our center [26]. In our trial, the direct vasodilator effect of the released mediators from the graft and mesenteric circulation are incriminated for the sudden drop of the SVR, MAP, and PAOP and the subsequent increase in the SV and CI [6,17]. Increased pulmonary artery pressure is a part if the PRS, yet, in this trial, the significant rise of the mPAP in the CRg after portal unclamping was augmented by the acute increase in the circulating volume by the flushed preservative solution from the graft, in contrast to the EcRg where graft contents are washed externally with a significant volume of recipient's blood (average 0.5 ml/g) [27]. In consistence with the short-living nature of PRS in liver transplantation, all recorded hemodynamic parameters headed toward normalization in both group after initial reperfusion (5 min). While the recipient's circulation is the main vector in PRS, the transplanted liver graft sustains another form of injury, namely IR injury representing the cellular response to ischemia and reperfusion of the graft [9,20,28]. Both liver graft and the congested intestine during portal clamping might contribute in this injury that ranges from simple rise of the transaminases to graft nonfunction and graft loss [5]. In our trial, neither the graft nor the patient outcomes exhibited significant differences between both groups. Among several factors cooperating to generate IR injury are cold and warm ischemia times that are highly influential in determining the severity of graft injury [9,29]. In our trial, both warm and cold ischemia times were comparable in both groups contributing to the equivalent outcome parameters observed. Similarly, Fukazawa and his colleagues did not record any association between the PRS and the graft outcome [18]. We reported a higher incidence of PRS in both groups compared to several studies on graft transplant from deceased donors with significantly longer CIT, negating a major contribution of the CIT in the generation of PRS proposed in previous reports [21,22]. In these reports, purging the graft contents prior to portal unclamping might have reduced the incidence of PRS in spite of a longer CIT compared to our results. In our country, liver transplantation is exclusively from living donors until the time being. This fact hinders us from verifying the differential contribution of CIT and the extracorporeal rinse technique in generating PRS through a randomized controlled trial. MELD score in the present study was 15.23 and 15.35 in extracorporeal rinse and circulatory rinse groups, respectively, indicating relatively healthy recipients. This fact renders the extrapolation of the results of this study to recipients with high MELD scores inappropriate. Further randomized controlled trials recruiting recipients with high MELD scores (more than 20) are essentially required to determine the clinical safety of the circulatory rinse technique in this particular group. Meanwhile, the effects of donor factors as small for size grafts and longer cold preservation times due to surgical difficulties or donor–recipient operative asynchrony were also not evaluated in our trial, and consequently, a statement about safety and efficacy of the circulatory rinse technique in such conditions cannot be concluded from our results and necessitates a more focused study. In conclusion, the circulatory rinse technique induced early significant, short-lived turbulent hemodynamic changes after portal unclamping that were clinically controllable. Yet, this did not influence hemodynamics nor clinical outcomes beyond this point, justifying its use in relatively healthier recipients with lower MELD score.

## Authorship

AMY and WRE: designated the study. AMY and MAE: collected and analyzed data, and wrote the manuscript. MAE, MAW, TS, AMS, ANE, MME, ME, KZ and US: performed research.

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## Conflict of interest

The authors have declared no conflicts of interest.

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