

## Results of Use of Ringer Solution and Preservation Solution as Kidney Washing Solution in Living Donor Renal Transplantation.

*Resultados del uso de la solución Ringer y la solución de conservación como solución de lavado de riñón en el trasplante renal de donante vivo.*

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### RESUMEN

**Antecedentes y Objetivos:** Las soluciones de conservación utilizadas como soluciones de lavado de riñón en trasplantes son necesarias para una conservación más prolongada del riñón. El estudio tiene como objetivo comparar diferentes soluciones de lavado de riñón utilizadas en el trasplante renal vivo. **Métodos y Resultados:** Cuarenta y nueve pacientes sometidos a trasplante renal de donante vivo incluidos en el estudio retrospectivo. La solución de Ringer se utilizó para lavar el injerto renal en 37 pacientes (Grupo1) y la solución de conservación se utilizó en 12 pacientes (Grupo2). Se incluyeron en el estudio pacientes del Grupo 1 y del Grupo 2. Había 22 (59,5%) hombres en el Grupo 1 y 9 (75%) hombres en el Grupo 2. Veintisiete (73%) pacientes que usaban Ringer y 7 (58,3%) pacientes que usaban solución de conservación tenían comorbilidades. No hubo diferencias significativas entre el Grupo 1 y el Grupo 2 con respecto a la isquemia caliente, los tiempos de isquemia fría y los niveles de desajuste ( $p > 0,05$ ). El valor de creatinina preoperatorio fue significativamente mayor en la solución de conservación ( $p = 0,003$ ).

No hubo diferencia significativa entre los dos grupos en términos de niveles de creatinina en el postoperatorio ( $p > 0,05$ ).

**Conclusión:** En el trasplante renal vivo, se puede utilizar una solución económica de Ringer en lugar de la costosa solución de conservación para lavar el injerto.

**PALABRAS CLAVE:** Trasplante de riñón, donante vivo, la solución de lavado, solución de conservación, la solución de ringer.

### ABSTRACT

**Background and Aims:** Preservation solutions used as kidney washing solutions in transplantation are necessary for the longer preservation of the kidney. The study aims to compare different kidney-washing solutions used in living renal transplantation. **Methods and Results:** Forty-nine patients who underwent renal transplantation from live donors were included in the retrospective study. The Ringer's solution flushed the renal graft in 37 patients (Group 1), and the preservation solution was in 12 patients (Group 2). Group 1, and Group 2 patients were included in the

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study. There were 22 (59.5%) males in Group 1 and 9 (75%) males in Group 2. Twenty-seven (73%) patients using Ringer's and 7 (58.3%) patients on preservation solution had comorbidities. There was no significant difference between Group 1 and Group 2 in warm ischemia time, cold ischemia time, and HLA mismatch levels ( $p > 0.05$ ). The preoperative creatinine value was significantly higher in the preservation solution group ( $p = 0.003$ ). There was no significant difference between the two groups in values of creatinine levels on the postoperative ( $p > 0.05$ ). **Conclusion:** In living renal transplantation, an inexpensive Ringer's solution, may be used instead of the expensive preservation solution to wash the graft.

**KEYWORDS:** Kidney transplantation, living donor, the washing solution, preservation solution, ringer solution.

## INTRODUCTION

Renal transplantation is more advantageous than alternative treatment methods in patients with end-stage renal disease in terms of patient survival and cost<sup>(1, 2)</sup>. Many factors are efficacy in patient and graft survival, and the importance of early graft function emphasizes among these factors<sup>(3)</sup>.

In preserving the viability of the graft, simple hypothermia should be the first objective, and in this way, cooling the graft reduces the metabolic and oxygen demand of the renal graft<sup>(4, 5)</sup>. However, due to anaerobic glycolysis, lactate levels rise, hypothermia cannot maintain cellular homeostasis, and cell death occurs. Therefore, various organ preservation solutions have been developed<sup>(4, 5, 7)</sup>. The first static cold storage preservation solution was discovered by GM Collins in 1969, then replaced by the Eurotransplant Foundation in 1976 by eliminating magnesium<sup>(8)</sup>. Meanwhile, several preservation solutions emerged; the University of Wisconsin (UV) solution, which was efficient in organ protection and still used today, was developed in 1987<sup>(9)</sup>.

In live donor nephrectomies, when the transition time between nephrectomy and transplantation is minimal, it has been thought that only the cooling of the kidney would be adequate, and Ringer's lactate solution was also used as the perfusion solution to wash the kidney<sup>(6)</sup>. It is known that organ preservation solutions protect the kidney

from tissue damage during warm ischemia. In addition, much evidence reports the effect of preservation solutions alone on the physiological consequences of warm ischemia<sup>(10, 11)</sup>. However, the cost-efficacy problem of these solutions continues.

This study aimed to present the early results of the patients using high-costly organ preservation solutions and inexpensive Ringer-containing simple preservation solutions in the light of the literature.

## METHODOLOGY

The results of transplant operations performed in Sakarya University Training and Research Hospital Renal Transplantation Center between April 2019, and November 2020, were evaluated retrospectively. Induction therapy (Antithymocyte globulin or Simulect) and maintenance therapy (steroid + calcineurin inhibitor + antiproliferative) were administered to the recipients of renal transplants.

All patients were dispensed 1 gr flacon cefazolin sodium intravenously one hour before the first incision. We recorded demographic characteristics and laboratory data of kidney recipients and donors.

The patient's evaluation included dialysis time duration, renal replacement method, primary disease, comorbidity, surgical and clinical complications in the acute period, hospitalization day, graft functions, blood levels of calcineurin inhibitor drugs, anastomotic renal artery and veins, warm ischemia, and cold ischemia times. Live donor nephrectomies were performed laparoscopically. Renal transplantation was performed in 60 patients between the specified dates.

In the preservation solution group, four patients were excluded from the study, three because of cadaveric transplantation, and one due to treatment inconsistency. In Ringer's solution group, four out of 44 patients were excluded from the study since they had a second kidney transplant operation, and three patients were excluded due to the inaccessibility of follow-up data because they were continuing in other centers.

Ringer's solution was the principal solution for kidney perfusion, and the other preservation solutions were Institute Georges Lopez-1 (IGL-1)<sup>®</sup> and Histidine-tryptophan-ketoglutarate (HTK) (PlegiStore)<sup>®</sup>.

All perfusion solutions were stored at 4-6 ° C and positioned approximately 100-120 cm in height on the back table. The solutions were

applied through the renal artery; when the liquid extracted from the renal vein was cleared of blood completely, the washing process was concluded. Approximately, 300-400 cc solution was necessary for this procedure.

Polifarma® 500 ml polyflex was used for Ringer's solution. The electrolyte ingredient of the solution is Sodium 147 mEq / L, Chloride 155.5 mEq / L, Potassium 4 mEq / L, Calcium 4.48 mEq / L, and the osmolarity was 308.8 mOsm / L. The preservation solution, PlagiStore® solution in 1000ml packages, was used. Ingredients of the solution are Histidine 180mMol / L, Tryptophan 2mMol / L, Mannitol 30mMol / L, KCL 9mMol / L (9mEq / LK and 9mEq / L Cl), NaCl 15mMol / L (15mEq / L Na and 15mEq / L Cl), and the osmolarity was 310 mOsm/kg. The other preservation solution was IGL-1® solution in 1000 ml, and the ingredient of this preservation solution was Adenosine 5 nmol / L, Glutathione 3 mmol / L, Sodium 125 mmol / L, Potassium 25 mmol / L, Magnesium 5 mmol / L, and the osmolarity of the solution was 290 mOsm / kg.

We evaluated the Ringer 500 cc solution and the PlagiStore 1000 cc solution as cost, which are used actively as preservation solutions.

### Statistical analysis

In the statistical analysis, the frequency of socio-demographic and clinical data and descriptive statistics were calculated as numbers, percentages, mean±S.D, and median (IQR:25th percentile-75th percentile) using the SPSS 26 package program. Continuous variables, with normal distribution,

were compared with the independent sample t-test in determining the statistical significance between the case and control groups, variables that did not show a normal distribution, were compared with the Mann-Whitney U test, and the proportions of independent groups, were compared with the chi-square and Fisher's exact test. The level of significance was accepted as 95% ( $p < 0.05$ ).

This study was approved by the Sakarya University Medical Faculty Non-Invasive Ethics Committee (No:71522473/050.01.04/564) on 04/11/2020 and complied with the 1964 Helsinki declaration and its later amendments.

### RESULTS

The Ringer's solution group (Group 1) had 37 patients included, and the preservation solution group (Group 2) included 12 patients. Group 2 used PlagiStore® for 10 patients (83.3%), and in two patients (16.7%) the IGL-1® preservation solution was administered.

Double artery anastomosis was performed in 9 patients (24.3%) using Ringer's and 2(16.7%) patients using preservation solution.

There were comorbid diseases in 27 patients (73%) in group 1 and 7 individuals (58.3%) in group 2.

Comorbidities were often hypertension and diabetes.

When evaluating kidney recipients and donors together, there was no significant difference between age, body mass index, and gender between group 1 and group 2 ( $p > 0.05$ ) in **Table 1**.

**Table 1**

		<b>Group 1</b>	<b>Group 2</b>	<b>p</b>
Recipient	Gender (M/F)	22(59.5%)/15(40.5%)	9(75%)/3(25%)	0.494***
	Age (year)	46.11±12.70	38.33±15.55	0.08**
	BMI (kg/m <sup>2</sup> )	26.76±5.40	25.50±5.29	0.485**
Donor	Gender (M/F)	18(48.6%)/19(51.4%)	8(66.7%)/4(33.3%)	0.277*
	Age (year)	46.32±12.27	43.17±9.18	0.418**
	BMI (kg/m <sup>2</sup> )	27.44±4.71	28.72±4.79	0.419**
	GFR	110.93±12.42	107.62±10.23	0.409**

(Mean±Std), \*, Chi-Square Tests,\*\*; independent group t-test, \*\*\*; Fisher's exact test

There was no significant difference in the cold ischemia time, mismatch, and haplotype numbers between group 1 and group 2 ( $p > 0.05$ ).

Warm ischemia time was significantly longer in group 2 ( $p = 0.026$ ) (**Table 2**).

There were no significant differences in postoperative arterial blood gas and electrolyte levels of the patients in both groups ( $p > 0.05$ ).

The creatinine levels, measured at

pre-transplantation, were significantly higher in group 2 ( $p = 0.003$ ).

There was no statistical difference between the two groups in creatinine levels on the 1st day, 7th day, 1st month, and 3rd month post-transplantation.

Ringer's solution cost was 500 cc 0.8 €, and the preservation solution was 250 € (**Table 2**).

**Table 2**

	<b>Group 1</b>	<b>Group 2</b>	<b><i>p</i></b>
HLA-compatibility (Mismatch) Median (%25-%75)	3 (2-5)	3(2.25-3.75)	0.483*
HLA-compatibility (Haplotype) Median (%25-%75)	1 (0-1)	1(0.25-1.0)	0.320*
Hospitalization day (%25-%75)	7.0 (6-7.5)	7.0(7.0-7.75)	0.574*
First 24-hour urine output (L) (%25-%75)	10.3 (8.8-13.2)	10.1(7.8-13.0)	0.954*
Warm ischemia (sec) (%25-%75)	145 (117.0-171.5)	180(145-229.75)	0.026*
Cold ischemia (min) (%25-%75)	66 (75-80.5)	75(61.25-81)	0.415*
Post-transplantation day 1 <sup>th</sup> arterial blood gases pH	7.28±0.05	7.2±0.06	0.634**
Post-transplantation day 1 <sup>th</sup> arterial blood gases lactate (mmol/L) (%25-%75)	2.3(1.3-3.9)	2.8(1.5-3.6)	0.718*
Post-transplantation day 1 <sup>th</sup> blood bicarbonate (mmol/L) (%25-%75)	18.3(16.8-20.1)	16.5(16.1-19.6)	0.295*
Post-transplantation day 1 <sup>th</sup> blood calcium (mg/dL) (%25-%75)	9.1(8.5-9.5)	8.8(8.5-9.4)	0.584*
Post-transplantation day 1 <sup>th</sup> blood potassium(mmol/L) (%25-%75)	4.7(4.3-4.9)	4.7(4.0-5.2)	0.972*
Post-transplantation day 1 <sup>th</sup> blood phosphate (mg/dL) (%25-%75)	4.2(3.4-4.7)	4.2(3.7-4.8)	0.816*
Post-transplantation day 1 <sup>th</sup> blood sodium (mmol/L) (%25-%75)	139(138-141)	138.5(136.3-141)	0.565*
Pre-transplantation creatinine (mg/dl) (%25-%75)	5.7(4.8-6.8)	8.3(6.5-9.6)	0.003*
Post-transplantation day 1 <sup>th</sup> creatinine (mg/dl) (%25-%75)	2.7(2.0-4.2)	2.5(2.1-3.3)	0.766*
Post-transplantation day 7 <sup>th</sup> creatinine (mg/dl) (%25-%75)	1.22(1.05-1.52)	0.95(0.84-1.23)	0.054*
Post-transplantation month 1 <sup>th</sup> creatinine (mg/dl)	1.21±0.30	1.11±0.30	0.349**
Post-transplantation month 3 <sup>rd</sup> creatinine (mg/dl)	1.15±0.30	1.15±0.20	0.925**
<b>Solution Cost Per Patient (€)</b>	<b>Group 1</b>	<b>Group 2</b>	<b><i>p</i></b>
0.8 €	0(0%)	37(100%)	<0.001***
250 €	10 (83.3%)	0(0%)	
200 €	2 (16.7%)	0(0%)	

(Mean±Std), \*: Man Whitney U test, Z \*\*, independent group t-test, \*\*\*, Fisher's exact test

When the patients treated with Ringer's solution were evaluated in two groups, meaning single artery anastomosis and double artery anastomosis, there was a significant difference in cold ischemia time between the groups ( $p = 0.002$ ).

There was a significant difference in

hospitalization days between the two groups ( $p = 0.008$ ).

There was no significant difference in postoperative electrolyte and creatinine levels between the groups ( $p > 0.05$ ) (**Table 3**).

**Table 3**

	Single Artery Anastomosis	Double Artery Anastomosis	<i>p</i>
Patient (n) (%)	28(75.7)	9(24.3)	
HLA-compatibility (Mismatch) Median (%25-%75)	3(3-5)	3(0.5-6)	0.498*
HLA-compatibility (Haplotype) Median (%25-%75)	1(0-1.0)	1(0-1.5)	0.391*
Hospitalization day (%25-%75)	7(6-7)	9(7-12)	0.008*
First 24-hour urine output (mL) (%25-%75)	9.6(9.0-15.5)	11.0(7.0-12.0)	0.645*
Warm ischemia (sec) (%25-%75)	144(113-.161)	160(120.0-201.5)	0.215*
Cold ischemia (min) (%25-%75)	64.0(57.0-78.0)	81(72-96)	0.002*
Post-transplantation day 1 <sup>th</sup> arterial blood gases pH	7.28±0.05	7.30±0.05	0.406**
Post-transplantation day 1 <sup>th</sup> arterial blood gases lactate (mmol/L) (%25-%75)	2.6(1.4-4.1)	1.5(0.9-3.2)	0.136*
Post-transplantation day 1 <sup>st</sup> blood bicarbonate (mmol/L)	18.09±2.87	17.98±2.77	0.930**
Post-transplantation day 1 <sup>th</sup> blood calcium (mg/dL) (%25-%75)	9.2(8.5-9.5)	8.8(7.8-9.4)	0.172*
Post-transplantation day 1 <sup>th</sup> blood potassium (mmol/L)	4.70±0.55	4.56±0.44	0.551**
Post-transplantation day 1 <sup>th</sup> blood phosphate (mg/dL) (%25-%75)	3.8(3.3-4.7)	4.3(4.1-5.3)	0.127*
Post-transplantation day 1 <sup>th</sup> blood sodium (mmol/L)	139.25±2.40	139.67±2.12	0.641**
Pre-transplantation creatinine (mg/dl) (%25-%75)	5.8(5.1-6.8)	5.0(4.5-.7.5)	0.457*
Post-transplantation day 1 <sup>th</sup> creatinine (mg/dl) (%25-%75)	2.7(1.9-4.2)	2.7(2.5-4.1)	0.298*
Post-transplantation day 7 <sup>th</sup> creatinine (mg/dl) (%25-%75)	1.2(1.1-1.3)	1.4(1.1-2.1)	0.080*
Post-transplantation month 1 <sup>th</sup> creatinine (mg/dl)	1.20±0.31	1.27±0.30	0.509**
Post-transplantation month 3 <sup>rd</sup> creatinine (mg/dl)	1.12±0.30	1.26±0.27	0.225**

(Mean±std), \*: Man Whitney U test, Z \*\*; independent group t-test



## DISCUSSION

The principal purpose of organ preservation is to protect the tissue function until the organ for transplantation reaches the recipient and to ensure that the graft might be transplanted even to far distances, especially in cadaver transplants. Several preservation solutions have been developed and used for this purpose <sup>(7)</sup>.

Furthermore, normothermic machine perfusion and hypothermic machine perfusion were used, although they were not cost-effective, reporting that delayed graft function was lesser than with usual cold storage <sup>(12, 13)</sup>. Several studies in the literature compare preservation solutions used in cadaver donor renal transplantation. It has been reported that IGL-1 is superior to other solutions in delayed graft function, but when observing results annually, preservation solutions are not superior to each other <sup>(14)</sup>.

Another study reported that IGL-1 could be safely used to preserve kidney grafts, and further studies are needed to determine if it is superior to the University of Wisconsin (UW) or HTK <sup>(15, 16)</sup>. In the other study with cadaveric and living donors, the reported efficacy of UW and Celsior solutions was identically in kidney protection <sup>(17)</sup>.

Another study performed in living donor kidney transplantation compared the IGL-1 solution and UW, and the early graft function of IGL-1 was highlighted <sup>(18)</sup>. The IGL-1, and PlagiStore (HTK) solutions, were used in the current study. We discontinued using the IGL-1 solution since short-term bradycardia was observed post-perfusion in patients.

Roughly rates of normal renal function could occur in transplants from live donors with a short ischemia time in a few hours post-transplantation <sup>(19)</sup>.

If the time between nephrectomy and implantation is minimal in live donor transplants, the kidney is cooled using preservation solutions immediately after nephrectomy, where it is believed, that any preservative solution for washing could be used <sup>(6, 20)</sup>. HTK was used as a washing solution in a high-volume study in which a laparoscopic live donor nephrectomy was performed <sup>(21)</sup>. In a different study, Ringer's lactate, which is more affordable, was used as a perfusion solution for kidney washing and was recommended to use as a protective solution in cases where the renal revascularization period may

be prolonged <sup>(6)</sup>.

In the current study, the preservation solution (Group 2) was used for kidney washing in 24.5% of the patients. In group 2, preoperative creatinine values were significantly higher than group 1, and postoperative 7th-day creatinine values were lower than group 1. The preservation solution was thought to have a positive effect on early graft function, but this was not statistically significant. Also, first-month and 3rd-month creatinine levels were found similar in both groups.

Another way to decrease the cost of surgical procedures is to reduce the complications that may occur. Thus, delayed graft function, prolonged hospital stays, and increased related costs are prevented.

Surgical technique standardization is recommended to reduce the complications associated with surgery in renal transplantation <sup>(22)</sup>. We performed our surgical procedures with the same technique. Incompatible living donor kidney transplantation associated intravenous immunoglobulin and plasmapheresis treatments, and protocol biopsies are also associated with increased costs <sup>(23)</sup>.

We applied plasmapheresis for desensitization to a patient who underwent a second kidney transplant, not included in the study.

The delayed graft function observed in some patients could have depended on many distinct variables related to kidney donors and recipients. Multiple renal arteries are one of these variables. Although considered a relative contraindication because it prolongs ischemia time and increases the risk of complications, multiple renal artery allografts have been used successfully in kidney transplantation in experienced centers <sup>(24)</sup>.

The prolongation of warm and cold ischemia times, which can cause delayed graft function, may result in the death of the organ after a certain period <sup>(25)</sup>.

In the current study, double renal artery anastomosis had performed in group 1 on nine patients. When observing the results, although it had caused a significant increase in the cold ischemia time, there was no statistical difference in graft function.

The study has limitations: the relatively low number of patients, its short follow-up time, and its retrospective character.

## CONCLUSION

Ringer solution, which is cost-effective in living kidney donor transplants, may be used in experienced centers with similar graft survival results.

However, using expensive preservation solutions may be recommended in cases with long cold ischemia times, such as paired exchange transplants and cadaveric kidney transplants.

Besides this, Ringer's solution can be used in living donor kidney transplantation cases, as it is safe, effective, and inexpensive. There could be planned high-volume randomized controlled studies on this issue.

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## Consent to participate

Written informed consent was obtained from the patient.

## Authors' contributions

**NF:** study concept and design, acquisition of data, analysis, and interpretation of data, and drafting of the manuscript.

**EA, HD, FA:** acquisition of data, analysis, and interpretation of data

**MBK:** analysis and review.

**FC:** study conception, drafting of manuscript, analysis, and critical revision of the manuscript.

**Ethical standards:** This study was conducted following the Declaration of Helsinki. For the study, approval of the Ethics Committee of Sakarya University Medical Faculty and written informed consent from all of the participants were obtained. Ethics committee approval code; 71522473/050.01.04/564 and dated 04/11/2020 was obtained.

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