Comparison Between IGL-1 and HTK Preservation Solutions in Deceased Donor Liver Transplantation

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ABSTRACT

The effectiveness of liver preservation solutions remains in evidence. Cold ischemia time, steatosis, expanded criterion donors, operational cost, and survival represent important roles in its success. In a prospective cohort study between August 2009 and April 2014, 178 patients were allocated into an Institut Georges Lopez e (IGL-1) solution group (63.5%) or histidine-tryptophan-ketoglutarate (HTK) group (36.5%). There were no differences among recipient’s characteristics including age, skin color, gender, Model for End-stage Liver Disease score, acute rejection, cholestasis, and reperfusion syndrome incidences. Also, donors, age average, skin color, donor risk index, time in intensive care unit, hemodynamic variables, infections, and steatosis incidences were similar. The average cold ischemia time was 494 minutes in the IGL-1 group and 489 minutes in the HTK group (P = .77). Alanine aminotransferase and aspartate aminotransferase serum levels on the first postoperative day were 707 and 1185 mg/dL, respectively, with IGL-1 and 1298 and 2291 mg/dL, respectively, with HTK (P = .016) and similar at day 15 (P > .88). The incidence of delayed graft function was 4.5% with IGL-1 and 4.6% with HTK (P = .90). The incidence primary nonfunction was 2.7% with IGL-1 and 3.1% with HTK (P = .71). The incidence of perioperative death was 11.5% with IGL-1 and 13.8% with HTK (P = .94). The survival in 30 months was 86% in IGL-1 group and 82% in HTK group (P = .66). Both preservation solutions are efficient to liver transplantations with deceased donors. Major prospective trials are necessary to evaluate each preservation solution’s particularities. The preservation solution availability in each transplantation center must guide its use at the present moment.

NOWADAYS, transplantation centers in the world are able to rely on various types of organ preservation solutions. The question that remains is what solution is the best. We need to know which solution is the most reliable to extend ischemia time without increasing the risk of dysfunction and organ failure, to avoid thrombotic and biliary complications, and to maintain excellent patient survival. Histidine-tryptophan-ketoglutarate (HTK), also known as Bretschneider’s solution, has been fairly used in liver transplantation centers in Europe, especially, in Germany [1,3,6,13]. University of Wisconsin (UW) solution, once considered the gold standard preservation solution worldwide, has been intensively tested and compared to others in the last three decades [2,4,5,10,11,19]. Some experimental and observational studies have compared HTK to UW solution with similar results [7,9,12,14,15,20]. The main outcomes of patient and graft survival were similar in the majority of trials, but some differences regarding biliary and arterial complications, bile production, delayed graft function (DGF), and cost effectiveness were favorable to HTK in particular studies [17,18,26,29–32]. Contrary to the most clinical trials, the North American experience with HTK in the last decade has been discouraging, also
PATIENTS AND METHODS

Between August 2009 and April 2014, we performed 262 adult liver transplantations in our center (HDVS-ISCMPA) in recipients older than 18 years.

A prospective cohort was conducted with 178 patients submitted to DDLT, allocated in two groups according availability of each solution in our center during this time. We excluded for analysis transplantsations performed for severe acute hepatitis (5 cases), retransplantation surgery (13 cases), and DDLT performed with other preservation solutions, such as Celsior (Genzyme™, France) or UW (60 cases). Other exclusion criteria included liver grafts with steatosis greater than 50%, center-lobular ischemia, or moderate-to-severe polymorph nuclear infiltrate at the biopsy observed by the same pathologist.

The state transplantation center is responsible for offering a patient in brain death with no age limitations to liver retrieval, according ABO blood type compatibility and Model of End-stage in Liver Disease (MELD) score, which is the main criteria to enter in the state’s unique waiting list, defined by a national law.

Each liver transplantation was routinely developed, preserving the receptor’s vena cava (via the Piggyback technique), without administration of any rinse before reperfusion. Each liver retrieval surgery was performed at a credentialed state hospital by the same surgical team, specifically trained for this procedure, and whose members had also been part of the transplantation team. The standard technique was performed in all cases. The abdomen and mediastinum were exposed through a midline thoracoabdominal incision. The retroperitoneum and portal triad were dissected, the gallbladder was removed, and the common bile duct was flushed with normal saline solution. The supra-iliac aorta was clamped and the vena cava was sectioned at infra-renal and juxta atriun levels. Each liver was perfused simultaneously through the infra-renal aorta and portal vein. A total volume of 4000 mL of HTK or 2000 mL of IGL-1 were infused through the aorta while 2000 mL of either solution was infused through the portal vein.

DGF was defined as a necessity of clotting factor support, prolonged intensive care, or persistent elevation of liver enzyme levels during the first week after transplantation. Despite this major illness, patients could achieve full recovery with proper support [10,16,21].
Table 2. Features of Liver Receptors

<table>
<thead>
<tr>
<th></th>
<th>IGL-1 (n = 113)</th>
<th>HTK (n = 65)</th>
<th>Total (n = 178)</th>
<th>P a</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>64.1 (± 6.6)</td>
<td>53.3 (± 10.5)</td>
<td>59.2 (± 7.3)</td>
<td>.13</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>76 (67.3%)</td>
<td>41 (63.1%)</td>
<td>117 (65.7%)</td>
<td>.62</td>
</tr>
<tr>
<td>Female</td>
<td>37 (32.7%)</td>
<td>41 (36.9%)</td>
<td>61 (34.3%)</td>
<td></td>
</tr>
<tr>
<td>Skin color</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>91 (80.5%)</td>
<td>53 (81.5%)</td>
<td>144 (80.9%)</td>
<td>.43</td>
</tr>
<tr>
<td>Black</td>
<td>16 (14.1%)</td>
<td>7 (10.8%)</td>
<td>23 (12.9%)</td>
<td></td>
</tr>
<tr>
<td>Others</td>
<td>6 (5.3%)</td>
<td>5 (7.7%)</td>
<td>11 (6.2%)</td>
<td></td>
</tr>
<tr>
<td>HCV</td>
<td>56 (49.6%)</td>
<td>34 (52.3%)</td>
<td>90 (50.6%)</td>
<td></td>
</tr>
<tr>
<td>HBV</td>
<td>6 (5.3%)</td>
<td>3 (4.6%)</td>
<td>9 (5.1%)</td>
<td>.86</td>
</tr>
<tr>
<td>OH</td>
<td>10 (8.8%)</td>
<td>3 (4.6%)</td>
<td>13 (7.3%)</td>
<td></td>
</tr>
<tr>
<td>OH + virus</td>
<td>23 (20.4%)</td>
<td>14 (21.5%)</td>
<td>37 (20.8%)</td>
<td></td>
</tr>
<tr>
<td>MELD</td>
<td>18 (15.9%)</td>
<td>11 (16.9%)</td>
<td>29 (16.3%)</td>
<td></td>
</tr>
<tr>
<td>HCC</td>
<td>65 (58%)</td>
<td>46 (71%)</td>
<td>111 (62.4%)</td>
<td>.11</td>
</tr>
<tr>
<td>RS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Presence</td>
<td>60 (53.1%)</td>
<td>42 (64.6%)</td>
<td>102 (57.3%)</td>
<td></td>
</tr>
<tr>
<td>RS Min</td>
<td>2.7 (± 0.6)</td>
<td>3.2 (± 0.6)</td>
<td>2.9 (± 0.5)</td>
<td>.62</td>
</tr>
<tr>
<td>AR</td>
<td>21 (18.6%)</td>
<td>8 (12.3%)</td>
<td>29 (16.3%)</td>
<td>.45</td>
</tr>
</tbody>
</table>

Abbreviations: IGL-1, Institut Georges Lopez – 1 solution; HTK, histidine-tryptophan-ketoglutarate solution; HCV, hepatitis C virus; HBV, hepatitis B virus; OH, alcoholic cirrhosis; MELD, Model for End-Stage Liver Disease; HCC, hepatocellular carcinoma; RS, reperfusion syndrome; AR, acute rejection.

PFN was defined as the irreversible hepatic failure that will invariably culminate in death if retransplantation is not performed. It occurs in proven absence of arterial or venous thrombosis and its cause remains uncertain [21].

Blood for biochemical analysis was collected daily during the first post-operative week, and two or three times a week thereafter. We evaluated AST and ALT in serum on PODs days 1, 7, and 15.

A statistical analysis was performed using SPSS software version 14 by IBMTM. The estimate of patient survival was obtained using the Kaplan-Meier method. The non-parametric Mann-Whitney test was used to compare the liver enzymes between groups, whereas the χ² or Fisher exact tests were used for categorical and logistic regression to linear variables.

RESULTS

A total of 178 patients received transplants, with IGL-1 preservation solution being used in 113 patients (63.5%) and HTK preservation solution in 65 patients (36.5%). Donor and graft baseline characteristics according to each group are presented in Table 1. There were no differences in terms of age, skin color, weight, and height. The donor risk index, duration of intensive care unit stay, use of vasopressors drugs including noradrenaline concentration, cardiac arrest, infections, and incidence of steatosis of any degree in donor are featured in both study groups. Donor gender was of different incidence between groups. In the IGL-1 group there were 75 (66.4%) males and 38 (33.6%) females, whereas in the HTK group the proportion was inverse with 31 (47.7%) males and 34 (52.3%) females (P = .02).

The features of the recipients are shown in Table 2. There was no statistically significant difference between recipients in both study groups regarding age, skin color, gender, weight, height, and MELD score. Acute rejection, cholestasis, and reperfusion syndrome (RS) incidences were also similar. Other causes of recipients’ terminal complicated cirrhosis were autoimmune hepatitis, primary sclerosing cholangitis, and primary biliary cirrhosis.

The causes of the liver donor death were similar between groups. In the IGL-1 group there was stroke in 75 (66.4%), major encephalic trauma in 34 (30.1%), and anoxia in 4 (3.6%). In the HTK group, there was stroke in 48 (73.8%), major encephalic trauma in 16 (24.6%), and anoxia in 1 (1.5%; P a = .35).

The causes of liver recipient sudden death (before POD 30) were sepsis (2 cases: 1.8% in the IGL-1 vs 5 cases: 7.8% in the HTK group), perioperative cardiac arrest event (1 case: 0.9% in the IGL-1 vs 1 case: 1.6% in the HTK group), PNF (2 cases: 1.8% in the IGL-1 vs 2 cases: 3.4% in the HTK group) or portal mesenteric venous rethrombosis (5 cases: 4.4% in the IGL-1 vs 3 cases: 4.7% in the HTK group). Other causes of death in the IGL-1 group were severe acute necrohemorrhagic pancreatitis (0.9%) and perioperative hemorrhagic shock (2.6%).

Three patients were submitted to retransplantation in the IGL-1 group for PNF, arterial thrombosis, and venous thrombosis. In the HTK group, 4 patients were retransplanted, 2 for PNF and 2 more for arterial thrombosis (P a = .674). Only 2 patients with PNF, 1 from each group, recovered successfully.

Variation in liver enzyme levels during the first 2 weeks after transplantation is shown in Fig 1. The graphs show a statistically significant increase in AST and ALT levels at POD 1 in the HTK group. The ALT and AST serum levels on POD 1 were 707 and 1185 mg/dL, respectively, with the IGL-1 group and 1298 and 2291 mg/dL, respectively, with the HTK group. (P = .001 and P = .016, respectively). ALT serum levels on POD 7 were different between groups (P = .022), but similar for AST serum levels (P = .33). There was no difference among liver enzyme variation on POD 15 (P = .88). The incidence of cholestasis with transitory increase of serum bilirubin levels at the first month was similar between groups (31 cases: 29.2% in the IGL-1 vs 17 cases: 27.0% in the HTK group; P = .50).

The incidence of acute cellular rejection episodes during the first month after transplantation, confirmed by biopsy, was similar among the IGL-1 and HTK groups.

Vasopressor use was also similar between both groups and the standard drug used was noradrenaline. The hemodynamic parameters of donors regarding medium arterial pressure, use or not of vasopressors, and noradrenaline concentration were similar between groups (Table 1). The presence and duration of RS was the same among the IGL-1 and HTK groups (Table 2).

The actuarial surveillance was similar between groups. The mortality that occurred during the first 30 days was 11.5% with IGL-1 and 13.8% with HTK. The mortality after POD 30 was 12.4% with IGL-1 and 18.5% with HTK, as
shown in Table 3. The incidence of DGF and PNF is also shown in Table 3.

The Kaplan-Meier curve of patient survival is shown in Fig 2. The 2-year patient survival rate was 86% in the IGL-1 group and 82% in the HTK group ($P = .66$). An estimated 5-year patient survival rate in the HTK group was 75%; this cannot be calculated for the IGL-1 group because of a shorter follow-up period.

**DISCUSSION**

Both solutions seem to be equally efficient for liver preservation in DDLT. Some advantages seem to favor effectiveness of IGL-1 solution.

Concerning costs in Brazil, the price per liter of both solutions is almost equivalent. IGL-1 is indicated when using only 4 L of solution, whereas HTK is preferable for the infusion of 10 to 6 L to achieve a homogeneous perfusion in all abdominal organs, increasing the cost per decease donor. This is a major issue considering the economic status of Latin America.

The remarkable increase of AST and ALT serum levels in the HTK group at POD 1 and the decrease of those levels through the first week after transplantation requires attention and further study [26,28,30,37].

![Fig 1. (A) Aspartate aminotransferase (AST) and (B) alanine aminotransferase (ALT) serum levels post-operative curves.](image)

<table>
<thead>
<tr>
<th>Table 3. Outcomes of Liver Transplantation</th>
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<td></td>
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<td>-------------------------------------------</td>
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<tr>
<td>Outcomes</td>
</tr>
<tr>
<td>DGF</td>
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<tr>
<td>PNF</td>
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<td>PNF death</td>
</tr>
<tr>
<td>Retransplantion</td>
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<tr>
<td>Death within 30th day</td>
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<td>Death after 30th day</td>
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</tbody>
</table>

Abbreviations: IGL-1, Institut Georges Lopez – 1 solution; HTK, histidine-tryptophan-ketoglutarate solution; DGF, delayed graft function; PNF, primary nonfunction.
The relationship between early liver graft viability and hepatic enzyme activities in effluent of preservation solution has already been established [8,26]. Serum levels and effluent levels greater than 10,000 mg/dL implicate a higher risk of PNF and, consequently, patient death. AST and ALT serum levels greater than 2,500 mg/dL are commonly observed in patients with DGF during the first week after transplantation. Most of these patients will recover after that, but long-term liver graft outcome is still unknown. Will the graft surveillance be shorter or will the main disease relapse precociously and suddenly?

IGL-1 and HTK solutions contain low concentration of sodium and potassium and a lower index of viscosity, but these have different substrates to anaerobic glycolysis. HTK acts like an extracellular solution in the interstitial compartment. Histidine is a powerful substance to maintain acid basic balance during cold ischemia time (CIT) with tryptophan and alfa-ketoglutarate for cellular feeding [1,6]. IGL-1 cellular protection is due to oncotic agent polyethylene glycol, glutathione anti-oxidant properties, raffinose, and lactobionate glycolysis substrates [22–24]. These solutions function through different ways in graft microcirculation to preserve hepatocytes and cuboids biliary cells [7,9]. Therefore, some unknown mechanisms could result in more hepatocellular ischemic and reperfusion damage with HTK than with IGL-1, translated by a greater hepatic enzyme level increase during the first postoperative days.

AST and ALT postoperative levels in the IGL-1 group encouraged our team to use IGL-1 in our center as a routine preservation solution. We also considered our excellent previous experience with IGL-1, a similar solution to the gold standard UW solution, from 1991 until 2010.

Despite the difference in donor gender among the groups, it does not seem to have an impact on major outcomes and can be controlled in multivariate analysis. Other variables of both donors and recipients were similar, as shown in Tables 1 and 2.

The incidence of DGF and PNF was statistically similar comparing both groups (4.5% and 2.7% in IGL-1 group vs 4.6% and 3.1% in HTK group, respectively). These similarities have probably occurred due to our policy of low use of extended criteria grafts and a CIT average of approximately 8 hours and a warm ischemia time average of approximately 30 minutes. Incidence of PNF was similar to the literature (2% to 23%) [10,16,21].

We did not evaluate the incidence of biliary complication on this trial. A long-term follow-up study is needed to diagnose and treat biliary stenosis. More than 50% of our IGL-1 cases were transplanted in the last 2 years, which makes it difficult to compare this condition with HTK cases that have already completed 5-years follow-up. Even so, there was no case of biliary fistula and no death related to biliary complication in IGL-1 group.

Considering that the CIT average was similar in both groups and that we do not transplant livers with more than 12 hours of CIT, we did not evaluate stratified and prolonged cold ischemia periods. Whether IGL-1 is better than HTK solution for liver preservation when the cold ischemia period is prolonged (>12 hours) is still to be determined. A larger stratified sample as well as experimental models should address this question in the future.

Important prospective trials must be developed to evaluate each preservation solution particularities. A major follow-up study is necessary to diagnose biliary complications and also a considerable number of patients are needed to be able to stratify steatosis degrees.

The preservation solution availability in each transplantation center must guide its use at the present moment.

**REFERENCES**


