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Transplantation

ARTICLES

CANINE AND HUMAN LIVER PRESERVATION FOR 6 TO 18 HR BY COLD INFUSION¹

JOSEPH BENICHOV, CHARLES G. HALGRIMSON, RICHARD WEIL, III,
LAWRENCE J. KOEP, AND THOMAS E. STARZL

*Department of Surgery, Denver Veterans Administration Hospital and University of Colorado
Medical Center, Denver, Colorado, 80262*

SUMMARY

Forty-one dog livers were preserved with cold, lactated Ringer's, plasma, or intracellular (Collins) solutions. Consistent survival was obtained with all three solutions for 9 hr. After 18 hr, the plasma and Collins solutions permitted survival, with the Collins solution having a slight overall advantage. The method using Collins solution has been used to preserve seven human livers in Los Angeles, to transport the organs to Denver, and to transport them as orthotopic grafts from 6 hr, 45 min to 10 hr later.

MATERIALS AND METHODS

It has been well known for a decade that the preservation of whole animal and human livers is possible for many hr, as proved by subsequent life-sustaining function of the hepatic grafts in recipients (1-4, 7, 8, 10). The preservation methods can be divided into the more complicated ones involving continuous or intermittent perfusion (1-4), and those using simple intravascular infusion of cold solutions (7, 11, 13) followed by cold storage.

In this investigation, infusion solutions have been compared in cold storage experiments to determine the extent, if any, that the infusion fluid composition affects the outcome. The results have been used to design a successful clinical trial of liver graft preservation and transport over long distances.

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Canine studies. With preservation times of 9 and 18 hr, three solution types were compared. After removal, the livers were washed free of blood with 500 to 800 ml of lactated Ringer's solution at 4 C. The fluid was delivered through the portal vein at 100 cm H₂O pressure. This was followed with 400 to 600 ml of the test solution at 4 C. After placing it in a plastic bag, the organ had more test solution poured over it. The plastic bag was kept cold by packing it in ice, as is standard practice with kidney preservation (5, 6).

There were 41 experiments that were divided into 7 groups depending upon the solutions used and the time of preservation (Table 1). As the graft vessels were being anastomosed to the recipient structures, the preservation fluid was washed out with a slow final infusion of cold, lactated Ringer's solution. The transplantation technique was the one originally described in our laboratories (11), as slightly modified later (10). No immunosuppression was given postoperatively.

Human studies. The seven livers were removed in six different Los Angeles hospitals

under conditions of heart-beating brain death. Preservation was with Collins solution, using the same technique as described in dogs but with adjustments of infusing volume according to the size of the organ. The donors were 10 months to 43 years old. These livers were initially flushed with 500 to 1,300 ml of lactated Ringer's solution and subsequently with 400 to 800 ml of Collins solution (Table 2). The final flushing of the organ with lactated Ringer's solution during implantation was the same as that with the dog. Transplantation was by previously reported techniques (10-12).

RESULTS

Animal Studies

There was minimal acute ischemic damage of the grafts in control animals that had imme-

mediate transplantation of cooled livers (Fig. 1). Rejection supervened within a few days. Five of the control animals lived for at least 7 days.

9 hr. With preservation for 9 hr, the survival and behavior of the animals were similar to those of the controls. Furthermore, there were no differences by the criteria of postoperative behavior and mortality with lactated Ringer's, Collins, or plasma solutions. Nevertheless, biochemical evidence of ischemic damage was less with Collins solution than with lactated Ringer's or plasma solutions (Fig. 1).

One animal survived with no immune-suppression and is alive 3 months later.

18 hr. Recipients of three livers preserved with lactated Ringer's solution died within 1 day. With plasma and Collins solutions, about one-half of the recipients survived the immediate postoperative period, in spite of the fact that moderate to severe ischemic injury was always present (Fig. 2). In terms of both survival and liver function, there seemed to be a slight advantage with Collins solution compared with the plasma (Fig. 2).

Human Studies

Six of the seven cadaveric livers functioned well after 6 hr, 45 min to 10 hr of preservation, with prompt relief of jaundice and generation of good clotting factors. The serum glutamic-oxaloacetic transaminase always rose (Fig. 3), but the increase was to above 1,000 in only two of the six patients, and in these recipients there was a return to normal within 3 to 10 days. Five of these six patients are still alive from 3 weeks to 8 months post-transplant. The patient who died had normal liver function but developed lethal cardiopulmonary complications.

The seventh patient was 30 years old. At

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TABLE 1. Canine experimental groups

| Group | No. of dogs | Preservation time (hr) | Infusion fluid |
|----------------|-------------|------------------------|---------------------------------|
| A | 8 | 1 | Lactated Ringer's |
| B ₁ | 6 | 9 | Collins I ^a |
| C ₁ | 6 | 9 | Plasma solution II ^b |
| D ₁ | 3 | 9 | Lactated Ringer's |
| B ₂ | 7 | 18 | Collins |
| C ₂ | 8 | 18 | Plasma solution |
| D ₂ | 3 | 18 | Lactated Ringer's |

^a Commercially available solution (code 5A 7810) which, after the recommended additives, contains for 1 liter: potassium, 115 mEq; sulfate, 60 mEq; magnesium, 60 mEq; chloride, 15 mEq; sodium, 10 mEq; bicarbonate, 10 mEq; and phosphate, 100 mEq.

^b Shalm's solution (8) as modified by Calne et al. (4) contains: dog plasma, 1 liter; heparin, 2,000 IU; hydrocortisone, 250 mg; ampicillin, 500 mg; dextrose, 250 mg; magnesium sulfate, 5 ml of a 10% solution; and potassium phosphate, 15 mEq.

TABLE 2. Data of the 7 human liver transplantations with preservation procedure

| OT no. | Age of | | Recipient diagnosis | Total ischemia | Solution amounts (ml) | |
|--------|-----------|-----------|------------------------------------|----------------|-----------------------|---------|
| | Donor | Recipient | | | Lactated Ringer's | Collins |
| OT113 | 22 months | 3 years | Biliary atresia | 6 hr, 45 min | 500 | 400 |
| OT 116 | 4 years | 7 years | Neonatal hepatitis | 7 hr, 30 min | 600 | 500 |
| OT 118 | 43 years | 24 years | α_1 -antitrypsin deficiency | 8 hr, 52 min | 1300 | 800 |
| OT 125 | 10 months | 10 months | Biliary atresia | 6 hr, 34 min | 600 | 400 |
| OT 126 | 7 years | 11 years | Biliary atresia | 7 hr, 20 min | 1000 | 650 |
| OT 127 | 3½ years | 5½ years | α_1 -antitrypsin deficiency | 9 hr, 30 min | 1000 | 600 |
| OT 128 | 17 months | 3½ years | Biliary atresia | 10 hr | 1000 | 500 |

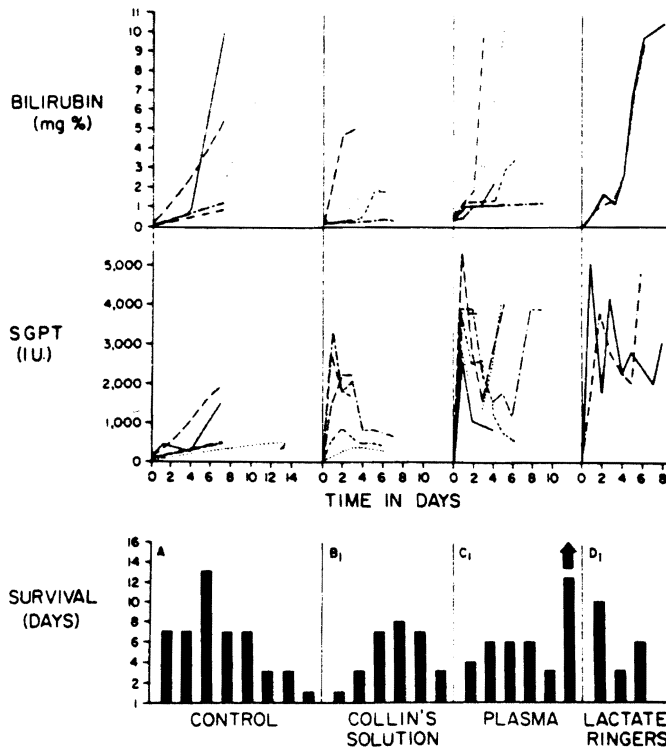


FIGURE 1. The effect of preservation for 9 hr upon orthotopic liver grafts in dogs. In the control experiments, transplantation was carried out immediately after flushing with cold, lactated Ringer's solution and after preservation for 1 hr or less. Although immunosuppression was not used, one animal in group C₁ (arrow) is still alive after 80 days.

eration, the liver could not be completely vascularized because of thrombosis of the recipient's portal vein. The graft functioned poorly, and there was an increase of serum glutamic-oxaloacetic transaminase to more than 3,000 IU (normal <50).

Eleven days later, another donor became available. Retransplantation was performed with the second homograft that was transplanted with the usual cooling (10, 11), without prolonged preservation, behaved the same as the first and the patient died 2 days later. The histopathological changes were much the same in the first and second organs, including widespread ischemic changes.

DISCUSSION

The intravascular infusion of cold electrolyte solution was the first effective technique of liver preservation (11), and it was not surprising that the same general method immediately became the standard for the quick cooling of the kidney (9) and other organs.

However, in the early experience with canine

liver transplantation, it was found that delay of more than 2 hr after this kind of cooling was not compatible with survival. From our present results, it is obvious that the time of 2 hr was a gross underestimate of what can be expected after simple infusion, providing other aspects of the operation and care are of high caliber. Even after 9 hr the results with lactated Ringer's solution alone were almost equal to those achieved with the complex solutions. Thus, the most important ingredient of success, no matter what the solution, was organ hypothermia.

In our experiments, the extra value of special solutions became clear only with longer preservation times. At 18 hr, both the plasma solution of Shalm et al. (8) modified by Wall et al. (13) and the Collins solution yielded better results than lactated Ringer's. Lambotte et al. (7) have previously shown the supplementary value of Collins solution for liver preservation.

If Collins solution is used, or for that matter the plasma solution preferred by Calne et al. (3), the safe time for liver preservation is

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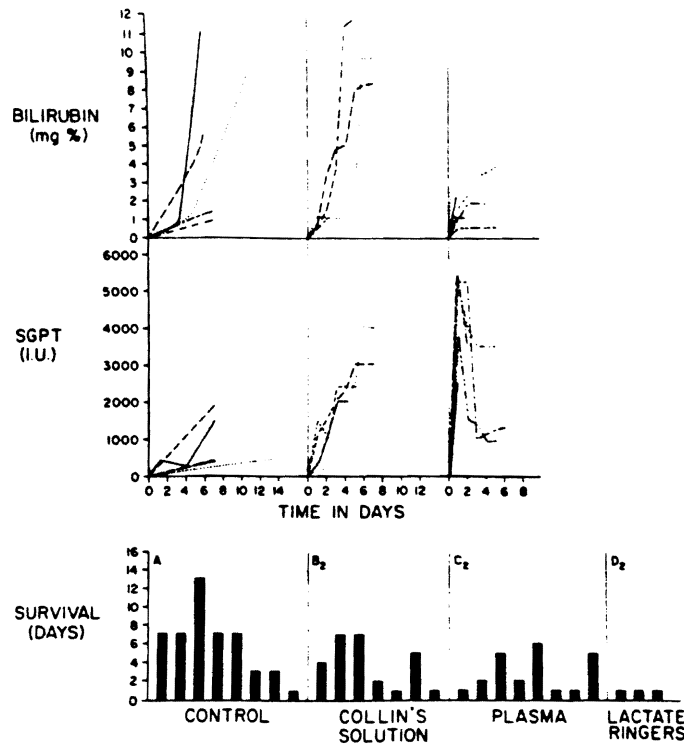


FIGURE 2. The effect of preservation for 18 hr upon orthotopic liver grafts in dogs. The experimental and control conditions were the same as for Figure 1.

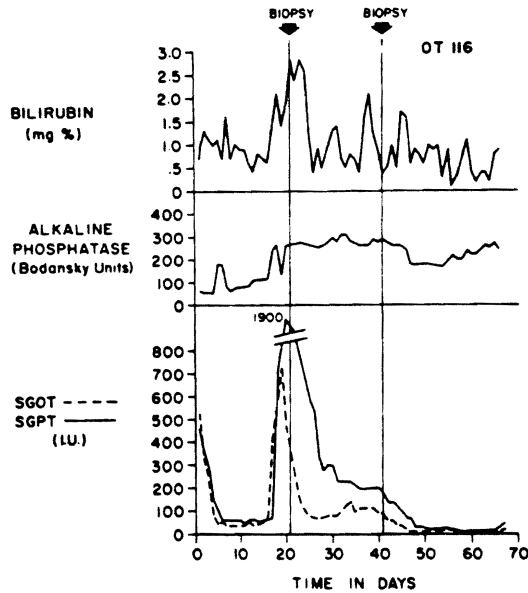


FIGURE 3. The course of a 7-year-old child who received a liver that was removed in Los Angeles, preserved for 7½ hr, flown 1,000 miles to Denver by a commercial airliner, and transplanted. Initial function was excellent. A biopsy taken 2 ½ weeks

extended to at least 9 hr and probably considerably beyond this. With this much time, the shipping of livers between most major American cities has become a real possibility. The implications of such a development were emphasized by Calne et al. (3) in their efforts at liver procurement in Europe.

The precaution of final flushing with lactated Ringer's solution should be scrupulously taken if the Collins solution is used. In one of our patients, this step was omitted. When the organ was revascularized, the serum potassium abruptly rose to 6.5 mEq/p 1,000 ml, followed by cardiac arrest from which resuscitation fortunately was possible. The sudden efflux of the potassium-rich intrahepatic fluid with revascularization of the unflushed graft was undoubtedly responsible.

Acknowledgments. The organ procurement was organized and carried out by Shawney Fine, Natalie

later, when homograft function deteriorated, was interpreted as hepatitis. The recipient is well 6 months postoperatively.

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Franklin, Barbara Schulman, and Roger Smith, coordinators of the Terasaki Organ Procurement Program. Paul D. Taylor, Senior Instructor in Surgery, at the University of Colorado Medical Center arranged and carried out the organ transfer to Denver, Colorado.

LITERATURE CITED

- Belzer FO, May R, Berry MN, et al: 1970 J Surg Res 10: 55.
- Brettschneider L, Daloze PM, Huget C, et al: 1968 Surg Gynecol Obstet 126: 263
- Calne RY, Dunn DC, Herbertson BM, et al: 1972 Br Med J 4: 142
- Calne RY, Dunn DC, Herbertson BM, et al: 1974 Transplant Proc 6: 289
- Collins GM, Shuggerman MB, Terasaki PI: 1969 Lancet 2: 1219
6. Fine SE, Clark RN, Fine RN, et al: 1975 Transplant Proc 7: 565
7. Lambotte L, Pontegnie-Istace S, Otte GD, et al: 1974 Transplant Proc 6: 301
8. Shalm SW, Terpsin JL, Popesen DT, et al: 1975 Surgery 78: 1637
9. Starzl TE: 1964 *Experience in renal transplantation*. WB Saunders Co, Philadelphia
10. Starzl TE: 1969 *Experience in hepatic transplantation*. WB Saunders Co, Philadelphia
11. Starzl TE, Kaupp HA, Brock DP, et al: 1960 Surg Gynecol Obstet 111: 733
12. Starzl TE, Porter KA, Putnam CW, et al: 1976 Surg Gynecol Obstet 142: 487
13. Wall VJ, Calne RY, Herbertson BM, et al: 1977 Transplantation 23: 210

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