Intra-abdominal Heterotopic Transplantation of the Canine Heart

Rodman Taber
Nobuatsu Kouno
Jorge Simon
Intra-abdominal Heterotopic Transplantation of the Canine Heart

Rodman Taber, M.D.,** Nobuatsu Kouno, M.D.*** Jorge Simon, M.D.***

Heterotopic transplantation of the dog heart to an abdominal location is a satisfactory experimental model for the study of immunosuppression. Rejection signs are readily detected by palpation and electrocardiographic monitoring.

Experimental cardiac transplantation was initiated in this country by Alexis Carrel in 1905. He was able to report a seven-hour survival of a grafted heart placed in a dog's neck. Both experimental orthotopic transplantation and heterotopic sites have been used subsequently to study the rejection process. Orthotopic heart transplantation requires the use of a pump oxygenator and is associated with a high mortality. Transplantation to an intra-abdominal location, which is a modification of the procedure originally described by Mann in 1935, may be done without cardiopulmonary by-pass and with a high rate of immediate success.

This report describes a series of abdominal heterotopic cardiac transplantations in which a variety of immunosuppressive regimes were employed to modify the rejection reaction.

Materials and Methods

Eighty experimental transplants were performed. Donor animals, weighing approximately ten kilograms, were heparinized before removal of the heart under general anesthesia. Ligation and division of the vena cavae and pulmonary veins was followed by transection of the aorta and pulmonary artery. The heart was then immersed in iced saline and the major vessels prepared for the anastomoses. A generous atrial septal defect was created through the orifice of one of the vena cavae.

Recipient dogs weighing between 20 and 25 kilograms were anesthetized with penthrane. The abdominal aorta and inferior vena cava were isolated through a midline transperitoneal incision. The ascending aorta of the donor heart was then anastomosed end-to-side to the recipient's abdominal aorta and the pulmonary artery to the host inferior vena cava (Fig 1). All trapped air was removed from the cardiac chambers by aspiration prior to releasing the vascular clamps. Ventricular fibrillation was commonly noted. This converted to sinus rhythm spontaneously within a few minutes in most of the animals. An anoxic period of 40 minutes was usually required to trim the donor vessels and perform the anastomoses. After restoring the circu-
Diagrammatic representation of experimental heterotopic transplantation of the heart to an abdominal position. The donor pulmonary artery is joined to the vena cava and the donor aorta to the abdominal aorta.

The transplanted heart was monitored for signs of rejection with an electrocardiogram every other day. Standard limb leads were used for the electrocardiogram as well as leads placed over the abdominal heart transplant site. Here details of the donor heart electrocardiogram were more readily seen (Fig 2). There were frequent blood counts, serum enzyme determinations (SGOT, LDH) and protein electrophoresis. The experiment was terminated whenever the donor heart stopped beating as determined by palpation and confirmed by the electrocardiogram. The donor heart was re-
Intra-Abdominal Heterotopic Transplantation of the Canine Heart

moved for pathological study when signs of rejection were complete.

Nine transplants served as controls and made up Group I. The remaining animals were divided into groups in which various immunosuppressive regimes were employed. In Group II, seven donors and recipients were matched according to red blood cell groups and crossmatching. In addition, they received azathioprine and prednisone (2-4 mgm/kg/day). In Group III, 45 dogs, splenectomy was added to the previous protocol. In Group IV, thymectomy was performed in 10 dogs several days prior to the transplantations. Group V consisted of nine animals receiving each of the previous measures plus irradiation (800-2400 rads) of the donor heart.

Rejection episodes were treated by temporarily increasing the dose of azathioprine and prednisone (Fig 3).

Results

In the entire series of 80 heterotopic heart transplants, four animals died of technical complications on the day of operation. Intra-abdominal hemorrhage and anesthetic mismanagement were usually at fault.

**Experimental Abdominal Heart Homotransplant**

![Figure 2](image)

Electrocardiogram (leads placed on the abdomen) of the donor and recipient hearts.
Group I (controls): The nine transplants in the untreated control group survived an average of 6.5 days (Table I). The rejected hearts showed typical signs of acute rejection. They were swollen, red and edematous, exhibiting numerous subendocardial and intramyocardial hemorrhages. Microscopically the myocardium was edematous and infiltrated with red blood cells and inflammatory cells. Myofibrillar necrosis and perivascular infiltrates were also present. Intracavitary thrombosis was rarely seen. Postmortem coronary arteriograms on several of the rejected hearts showed the major vessels to be patent.

Group II: In these seven transplants, azathioprine and prednisone were administered in addition to red blood cell matching. Survival averaged 30 days. Two hearts are still beating as of July, 1971, having survived more than 50 days. Rejection signs were minimal in these hearts.

Group III: The 45 animals in this series had splenectomy added to the measures employed in Group II. The hearts survived an average of 39 days with four still alive more than 50 days

| Blood count and serum enzyme changes noted during treatment of rejection crises, 14 and 23 days following abdominal heterotopic transplantation. These studies were not as helpful as the electrocardiogram for indicating impending rejection. |

<table>
<thead>
<tr>
<th>Days</th>
<th>WBC (count)</th>
<th>Lymphocytes (%)</th>
<th>SGOT</th>
<th>Prednisone (mg)</th>
<th>Azathioprine (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>14</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>21</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>28</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 3
Intra-Abdominal Heterotopic Transplantation of the Canine Heart

as of July, 1971 and two continuing to beat beyond a year.

Group IV: The 10 recipient dogs in this group had thymectomy performed besides those measures employed in Group III. The average survival was 32 days.

Group V: Nine donor hearts received varying dosages of irradiation immediately prior to transplantation, in addition to the measures used in Group IV. Average survival was 34 days with three living beyond 50 days and one still alive as of July, 1971.

Discussion

Survival in the control group and the treated transplants was similar to that reported by Crosby.$^3$ In nine treated animals, the hearts continued to beat for periods in excess of 50 days and three are long-term survivors beyond one year. It is not possible, however, to assign any convincing benefit to a particular immunosuppressive regime. Prolonged survival seemed to be more the result of accidentally favorable matching of dogs. Paired animals in the treated groups were selected for matching red blood cell groups in addition to satisfactory crossmatching. This pairing seemed to result in only minimal improvement of the average survival period as compared to results previously reported$^4$ in which this selection was not performed. Although we were able to obtain a few transplants surviving longer than those reported by others, the total number of these animals was too small to be significant. Red blood cell antigens are known to be considerably weaker in the dog than man.$^5$ It is also known that white cell transplantation antigens in the dog exert a significant effect on organ transplantation results.$^6$ We were unable to identify any impressive effects resulting from splenectomy and thymectomy in this group of experiments. However, we regard this area of investigation as undecided until longer observation periods are recorded. If these procedures are beneficial, they would influence the cellular phase of

<table>
<thead>
<tr>
<th>Group</th>
<th>No.</th>
<th>Prednisone*</th>
<th>Azathiopine</th>
<th>rbc typing</th>
<th>Splenectomy</th>
<th>Thymectomy</th>
<th>Irradiation</th>
<th>Average Survival</th>
<th>Still Alive</th>
</tr>
</thead>
<tbody>
<tr>
<td>I (Controls)</td>
<td>9</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>6.5 dys.</td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>7</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>30 (2&gt;50d)</td>
<td></td>
</tr>
<tr>
<td>III</td>
<td>45</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>39 (4&gt;50d)</td>
<td>2</td>
</tr>
<tr>
<td>IV</td>
<td>10</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>32</td>
<td></td>
</tr>
<tr>
<td>V</td>
<td>9</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>34 (3&gt;50d)</td>
<td>1</td>
</tr>
</tbody>
</table>

* 3mgm/kg

Table I

Summary of results in 80 experimental cardiac heterotopic transplants.

215
the immunologic reaction. Thymectomy in adult mice has been shown to exert a favorable immunosuppressive effect. \textsuperscript{6} Irradiation of the donor heart just prior to transplantation did not produce any obvious additional immunosuppressive effect.

Rejection signs were readily monitored by daily palpation of the transplanted heart which exhibited weak contractions when rejection was imminent. Electrocardiographic confirmation was evident with the appearance of bradycardia and conduction disturbances such as prolonged QRS segments and reduced R wave potential. The serum enzyme determinations and white blood cell count were of little value in detecting early rejection.

We frequently found the signs of impending rejection difficult to reverse. When vigorous treatment was required, prolonged survival was rare as the increased doses of immunosuppressive agents led to death from secondary pneumonia in at least 15 animals. The transplanted hearts in most of these animals dying of infection showed no signs of rejection. Possibly, decreased doses of azathioprine and prednisone would have lowered the mortality from infection. Intracavitary thrombosis was seldom seen. This was probably due to the free intracardiac circulation resulting from creation of the interatrial septal defect.

Anti-dog lymphocyte serum was investigated during a portion of this experimental series. The results were not included, however, as the sera obtained from two different sources were of varying precipitin titers and had questionable immunosuppressive effects.

Summary

Eighty heterotopic transplants were performed. Nine controls survived an average of 6.5 days. The remaining 71 transplants were used to evaluate modification of the rejection reaction by chemotherapy with azathioprine and prednisone, red blood cell matching, splenectomy, thymectomy and irradiation of the donor organ. The average donor heart survival in treated animals was 34 days. Transplant survival beyond 50 days was obtained in nine animals and beyond one year in three. No improvement in the average survival time was noted over that obtained with administration of azathioprine and prednisone when various supplemental measures were added to the protocol.

REFERENCES