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Orthotopic transplantation of the pig heart

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Orthotopic transplantation of the pig heart is described, with success in the last 12 of 23 studies, leading to survival until rejection in the last two. The pig heart was found to be easily damaged by cold. In contrast, organ viability was not appreciably affected when the warm ischaemic time was shortened by making the aortic anastomosis the initial manœuvre of implantation. The advantages of the pig as a model for the study of orthotopic cardiac allografts would appear to be its tolerance of long periods of cardiopulmonary bypass, immediate clotting after heparin reversal and ability to maintain the circulation without drug or pacemaker support. These factors have contributed to the relative simplicity of post-operative management.

Orthotopic cardiac implantation was first described in dogs by Lower and Shumway (1960) and has since continued to be reported by these workers and their associates with success (Shumway, Lower, Hurley, Dong, and Stofer, 1963; Shumway and Lower, 1964; Shumway, Angell, and Wuerflein, 1967). Their constancy has been achieved by few others in large animals (Willman, Cooper, and Hanlon, 1964; Cachera, Lacombe, Salamagne, Baumann, de la Fuente, and Dubost, 1966; Bos, Meeter, Israël, Stibbe, and van Rood, 1970). The pig, which has been used extensively in other areas of organ research, offers a possible alternative model for studies in this field.

METHODS

Inbred landrace all-white crossed pigs, weighing between 20 and 30 kg., were anaesthetized and maintained on a mixture of oxygen, nitrous oxide and halothane and ventilated through an extended armoured endotracheal cuffed tube, using a Starling pump at a minute volume adequate to sustain normal blood gas levels. Intermittent intravenous administration of pancuronium bromide during anaesthesia avoided halothane-induced hypotension.

Cardiopulmonary bypass was established after heparinization by extracardiac cannulation in a circuit primed with dextran 110 in 0·9% sodium chloride (Dextravan 110) and fresh citrated donor blood[1], using two cardiotomy reservoirs[2] in parallel, as described in detail by Clarke, Gorman, Orme, Howard, and Cullum (1970). Haematological and biochemical estimations (including blood gas analysis) were made before, during, and after bypass, and the acid-base balance was controlled by the addition of 8·4% bicarbonate.

The recipient heart was widely exposed through a left fourth intercostal approach which transected the sternum into the opposite interspace. The heart was isolated before excision by tying both azygos veins in continuity, tape-occluding the superior and inferior venae cavae, and cross-clamping the great vessels. The donor heart was similarly exposed but without the sternal transection and, after donor heparinization, was excised to include the great vessels, the atria and their appendages, and the sino-atrial node.

Twenty-three studies were divided into three groups of 3, 6, and 14 each, according to the degree of cooling of the donor organ immediately before implantation. The last 21 studies were consecutive. Groups I and II hearts were freshly harvested and immediately cooled, donor blood being collected from the chest cavity subsequently. In the first two hearts of group I, cooling was effected by perfusion with Ringer's lactate at 4° C. The remaining group I heart was excised and cooled in three washes of heparinized normal saline at 4° C. during a 5- to 10-minute interval before transference. All group II hearts were similarly cooled but with saline at 18° C. All group III hearts were harvested after aortic arch stab for 500 ml. of donor blood, and washed once in heparinized normal saline at ambient temperature.

Atrial closure preceded great vessel anastomosis in groups I and II, with late restoration of aortic

[1]Fenwall Double Blood Pack JD-2, Baxter Laboratories Ltd., Thetford, Norfolk

[2]Queen Elizabeth Hospital, Birmingham Pattern, HL-058, Polystan Ltd., Hitchin, Herts

TABLE I

HAEMATOLOGICAL, BIOCHEMICAL, AND BYPASS DATA

Group	Hb (g. %)	Na (mEq/l.)	K (mEq/l.)	Po_2 (mm. Hg)	Prime		Flow	MSAP
					Dextran	Blood		
I	10·6 7·1 6·5	135 141 149	6·0 6·0 6·6	136 106 —	18	7	55	73
II	8·3 8·3 7·7	143 148 149	7·0 6·6 8·2	137 85 —	15	22	61	64
III	9·5 8·7 8·8	143 144 144	6·1 5·6 5·1	200 81 255	14	17	52	79

Values in the first four columns refer above downwards in each group to pre-bypass, bypass, and post-bypass samples. Total circuit prime in ml./kg. body weight is made up by no-blood prime and blood prime. The flow in ml./kg. body weight and mean systemic arterial pressure (MSAP) in mm. Hg are average values.

continuity and coronary perfusion. In group III, initial aortic closure allowed early perfusion of the donor organ during cardiopulmonary bypass. Teflon felt strips on either side of the aortic suture line and on the recipient aspect of the right atrial suture line ensured surgical haemostasis. A vent emptying into the chest cavity was placed through the left atrial appendage and mitral valve to provide left ventricular decompression after atrial closure and AC defibrillation, until its subsequent removal at the end of bypass.

Support procedures were instituted when the mean systemic pressure arterial blood pressure fell below 60 mm. Hg after completion of implantation: by partial bypass, isoprenaline drip, or by pacing. When bypass support was no longer required, protamine was given and wound closure was performed with single chest drainage into a plastic bag.[3] Drug or pacemaker support was discontinued when appropriate. Extubation was delayed for half an hour following adequate reversal with atropine and neostigmine for confirmation of satisfactory cardiac and respiratory status by blood gas analysis. Oxygen administration was discontinued after about six hours, by which time surviving animals were able to 'turn' themselves from one side to the other.

RESULTS

The pertinent haematological, biochemical, and bypass data of the three groups are shown (Table I). The difference between the three can be accounted for by the differences in the methods of priming, as less plasma substitute was used during the course of the investigation. Also the reduced average priming volume of 25 ml./kg. body weight in group I was due to the exclusion of the heat exchanger in the bypass circuit in the first two of these three studies only. The total priming volume of this circuit of about 750 ml. includes a final reduced value of 10 ml./kg. of dextran with apparent improvement in clotting and haemostasis, which accounts for the higher post-bypass haemoglobin values and lower sodium values in later studies. Total bypass time averaged 108 minutes in the last 12 studies (75–146). Not more than 2 litres of blood were used during any one complete study.

Group I studies, with hearts cooled with perfusate or saline at 4° C. and with an average ischaemic period of 70 minutes, were unable to support the circulation without excessive drug or bypass support (Table II). Initially the hearts appeared pink and contracted well, but later they became hard with loss of compliance. Reduction of support procedures after implantation was completed resulted in patchy cyanosis, weakening contraction and further loss of compliance, with the development of rhythm irregularities, including irreversible third-degree heart block in one study. After one hour with continuous isoprenaline, bypass support was approaching total bypass conditions and the studies were terminated.

Group II studies, with hearts cooled in saline at 18° C. and with an average ischaemic period of 67 minutes, had similar findings. Discontinuation of bypass support was possible in one study

TABLE II

SURVIVAL AFTER DONOR ORGAN PRESERVATION

Group	No.	CIT	TIT	Support		Survival
				Bypass	Drug	
I	3	4	70	60	60	60
II	6	18	67	30	45	45
III	10	A	16	15	35	5 hours
	2	A	15	0	0	5 days

CIT = Cold Ischaemic Temperature in degrees centigrade; TIT = Total Ischaemic Time in minutes; A = ambient temperature; Other times refer to post-implant periods in minutes unless otherwise indicated.

[3]Aldon urine bag (Aldington Laboratories, Mersham, Ashford, Kent

but the heart failed to maintain the circulation after 45 minutes, requiring excessive isoprenaline support.

Group III studies (of which the first two are excluded as being necessary to examine the feasibility of initial aortic closure) with hearts washed in saline at ambient temperature during initial dissection and with an average ischaemic period of 16 minutes, supported the circulation without bypass, drug or pacemaker support within one hour of completion of implantation. There was little evidence of macroscopic change or loss of compliance. The average survival time of the first 10 of these 12 studies was more than five hours. Six of these animals died from technical faults (bleeding (4), premature donor death (1) and ventricular fibrillation (1)) without being extubated, although requiring anaesthesia to prevent spontaneous respiratory movements. Four survived for an average of 10 hours, breathing spontaneously for more than half of this time, and dying from secondary haemorrhage, drug overdose, ventricular fibrillation or accidental arterial catheter bleed. In each study the heart rate averaged between 110 and 150 beats per minute, with the maintenance of an average mean systemic arterial pressure of more than 80 mm. Hg and with a central venous pressure of less than 10 cm. of saline until death. In the last two studies no support procedures were required following implantation; extubation was performed within 90 minutes of bypass and both animals survived until the fifth and sixth days, showing a typical electrocardiographic pattern of rejection (Figure).

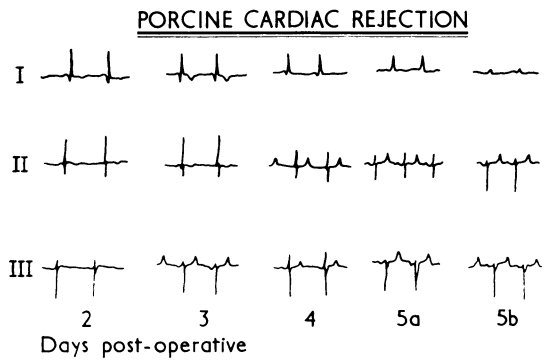

FIGURE. *Leads I, II, and III on days 2 to 5 (a and b both taken a few hours before death), showing progressive decline in voltage, especially well seen by R-wave diminution in leads I and II.*

DISCUSSION

Orthotopic transplantation of the heart is associated with donor organ ischaemia of about one hour while atrial anastomosis is completed and until the aorta is joined. Protection of cardiac muscle during this period is possible by preservation, either by cold immersion at 4° C., as reported by Lower and Shumway (1960), or at 20° C., as reported by Willman *et al.* (1964), or else by cold perfusion with a bypass circuit, as described by Barnard (1967). Cooley, Bloodwell, Hallman, and Nora (1968), while harvesting fresh material, found no need for donor organ protection with an anoxic interval of about 45 minutes in four patients.

Our results show that the pig heart cannot maintain the circulation after cold preservation, using techniques found to be successful in dogs by Shumway and his associates (Shumway *et al.*, 1963; Shumway and Lower, 1964; Shumway *et al.*, 1967), and Willman *et al.* (1964), even though similar periods of non-perfusion were observed. This suggests that the pig heart is more susceptible to ischaemia, especially cold ischaemia, than to other factors. Comparison of the haematological, biochemical, and bypass data (Table I) supports this view, as little difference is shown between the three groups presented, the last of which was successful (Table II). The failed groups I and II implants required increasing amounts of drug and bypass support after transplantation as progressive macroscopic damage and loss of compliance became evident. The successful group III implants maintained the circulation without support techniques within a short while of surgery. This contrasts with the experience reported in dogs by Shumway, Willman, and their associates, where drug or pacemaker support may be required for several hours or days. The difference is due to making aortic closure the initial manœuvre of implantation and reducing the ischaemic period to 16 minutes.

The pig has certain advantages as a model for the investigation of orthotopic cardiac allografts. Periods of total bypass of up to two hours, using simple bubble oxygenator techniques, have been tolerated, as shown by early spontaneous respiration together with normal blood gas values in air within six hours of surgery (Clarke *et al.*, 1970), and with subsequent survival. The low total priming volume necessary for the bypass circuit and immediate clotting after heparin reversal has reduced blood requirements to four units in any one study. Provided that a short warm ischaemic

period is observed these factors, together with the ability of the heart to maintain the circulation without drug or pacemaker support, contribute to the relative simplicity of the post-operative management in these animals.

There have been no other reports in the literature of successful orthotopic transplantation of the pig heart with survival.

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