Cephalic exchange transplantation in the monkey

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Short-term vascular transfer of the brain has been accomplished in the laboratory animal,[5] but its significance with reference to human cerebral transplantation is meaningless in the highest functional sense because the human adult central nervous system displays no capability of regeneration, thereby denying the brain any opportunity for afferent input or efferent expression.

Cephalic transplantation, on the other hand, would obviate this major problem by providing external environmental contact with information gathering and expression through the preservation of the cranial nerves.

While Demikhov[1] has accomplished prolonged vascular association between the upper portion of the canine body, including the head, and an intact recipient animal, the isolated cephalon until recently had not been transplanted.[6] Classically, the major problem in the design of this biological model has been the difficulty of maintaining the brain in a high state of performance after neurogenic transection of the spinal cord. The solution to this problem and the successful transplantation of the isolated primate cephalon to the cervical vasculature of an intact monkey has been the subject of a recent publication.[7]

This report deals with our initial experience in the vascular transfer of the isolated cephalon to the isolated body of a recipient with the persistence of normal cerebral function.

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PREPARATION AND INSTRUMENTATION

Eight small rhesus monkeys (weighing six to seven pounds), initially under intravenous pentobarbital (20 mg. per kilogram of body weight), tracheotomized and mechanically respired, were cervically transected at the level of the fourth to the fifth cerebral vertebrae with preservation of the carotid-jugular circulations. Each cephalon was instrumented for cortical electroencephalography (EEG), carotid pressure, and arteriovenous blood sampling; each recipient body was instrumented for electrocardiogram (ECG), arterial and venous pressure, discontinuously for blood gases, hematocrit, electrolytes, and glucose.

Procedural outline for cephalosomatic separation. With the satisfactory induction of anesthesia and instrumentation, surgical isolation was accomplished in the following manner:

1. Circumferential soft tissue and muscle were divided around the entire surface of the cervical vertebra with ligation and transec-

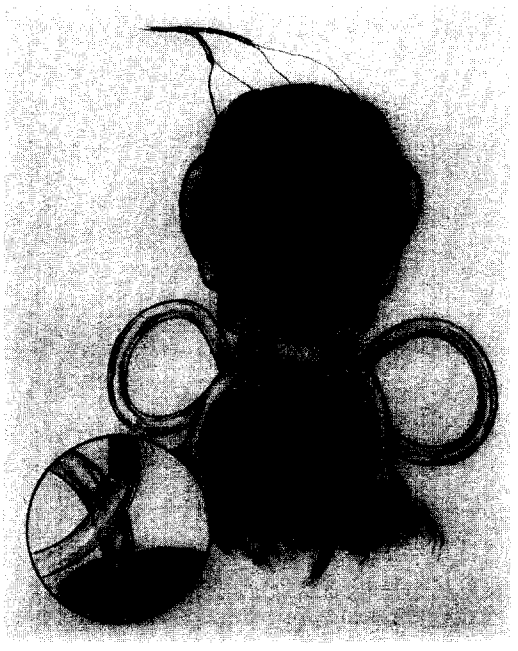

Fig. 1. Schematic drawing demonstrating the cannula loops associating the carotid-jugular circulation of the transplanted cephalon to the isolated monkey body.

tion of the trachea and esophagus following appropriate intubation.

2. Cervical laminectomy was performed at the level of the fourth to the sixth cerebral vertebrae with ligation and division of the spinal cord and its vasculature at levels 5 to 6. Following spinal cord division, an infusion of catecholamine (Levophed, 0.064 mg. per cubic centimeter) was begun to counteract the hypotension of ensuing spinal shock with the maintenance of mean arterial pressure between 80 and 100 mm. Hg. Mechanical pulmonary support by means of a Bird respirator was commenced and continued throughout the experimental run.

3. The vertebral sinus was obliterated with judicious use of cautery and intravascular injection of fast-setting Celloidin.

4. Intraosseous destruction of the vertebral arteries was carried out.

5. The vertebral body or interspace was transected.

With the completion of these five steps the head and body were completely separated save for the two neurovascular bundles.

Previous work[1] has demonstrated the capability of a single carotid artery and jugular vein to adequately sustain a rhesus monkey's brain when all other circulatory support has been eliminated. Consequently, each carotid artery and jugular vein in turn was divided and reconnected by means of suitable sized tubing arranged in loops during constant EEG surveillance. Prior to cannulation, the preparation was heparinized (2 mg. per kilogram of body weight) and the vagi were sectioned under ECG monitoring.

For vascular transference of the cephalon to the new isolated body, the individual cannulas were occluded and withdrawn from the parent body carotid arteries and jugular veins (in sequence, allowing for continuous cerebral perfusion from one set of cannulas during exchange) and replaced into the appropriate somatic vessel under EEG observation. To date we have attempted to salvage only one exchanged preparation, although both rejoined pairs should be usable (Fig. 1).

In one preparation, following successful cannula-vascular transfer, direct suture anastomosis of the carotid arteries and jugular veins was undertaken, with the use of No. 6-0 and 7-0 silk, respectively (Fig. 2), and the operating microscope. This permitted discontinuance of purposeful anticoagulation.

Freshly drawn monkey blood was available for infusion if significant losses were encountered during acute transplantation under prolonged heparinization. Both systemic (penicillin and chloromycetin) and topical (neosporin) antibiotics were generously administered.

Just prior to the conclusion of each experiment, Evans Blue dye was injected to define grossly the status of the blood-brain barrier.[2] At the completion of each run, the brain was removed, sectioned coronally, examined, and placed in formalin. After fixation, representative sections of brain were prepared, stained with hematoxylin and eosin, and examined under the light microscope.

RESULTS

All four of the cephalic exchange transplantation preparations survived; their

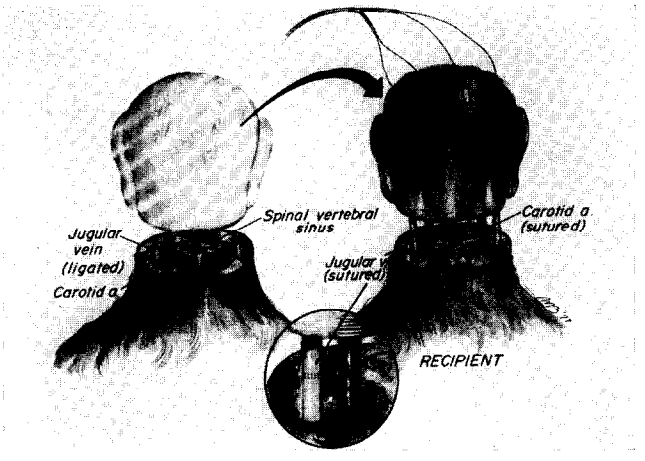

Fig. 2. Schematic drawing of the isolated primate caphalon transplanted to the isolated monkey body employing direct suture of the carotid and jugular vessels.

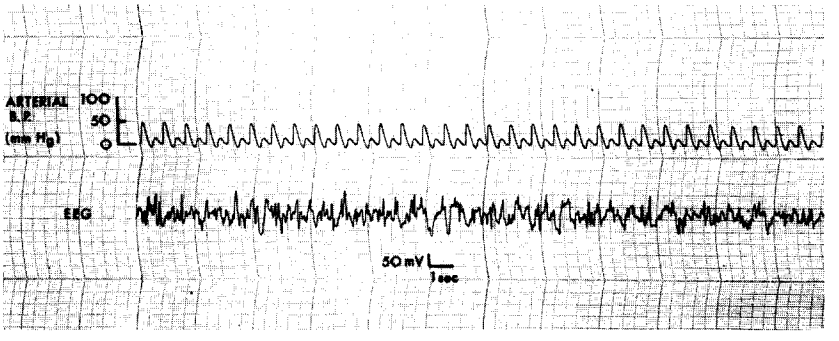

Fig. 3. Graph of the arterial blood pressure of the isolated body and a single channel recording of the EEG activity (electrodes in the extradural space) of the associated isolated cephalon. Note the persistence of excellent electrical activity of brain in the face of significant hypotension.

periods of viability ranged from 6 to 36 hours. In three to four hours, each cephalon gave evidence of its awareness of the external environment by accepting and attempting to chew or swallow food placed in its mouth. The eyes tracked the movement of individuals and objects brought into their visual fields and the cephalons remained basically pugnacious in their attitudes, as demonstrated by their biting if orally stimulated.

Throughout the long periods of observation, their EEG records reflected the establishment of a characteristic awake pattern. If the arterial pressure was allowed to fall below 40 mm. Hg. the cephalon became less responsive; however, the EEG activity, while demonstrating a reduction in amplitude and some slowing, remained quite respectable (Fig. 3). Tremendous seizure activity, as reflected in the EEG tracing and the muscle movements about the head and face, was easily elicited with the infusion of metrazol even hours after transfer.

With time, some blood loss was encountered from the muscles at the surfaces of surgical transection. This was attributed to chronic heparinization and represented a limiting factor in the prolongation of these experiments. The initial attempt to suture the vessels directly and thus eliminate the necessity of anticoagulation was only partially successful because of the constriction that developed in the jugular vein at the suture line, impeding venous return from the head.

Gross examination of each brain revealed no abnormal areas of uptake of infused Evans

Blue vital dye. Light microscopic review of multiple sections of each brain including cortex, white matter, basal ganglia, brain stem, and cerebellum gave no evidence of cellular changes compatible with a hyperrejection reaction in cerebral tissue.

DISCUSSION

These experiments demonstrated for the first time that it is possible to vascularly transplant the isolated cephalon to the isolated body at a primate level. Admittedly, the primary success has been accomplished with the use of cannula connections requiring continuous heparinization; there was only limited success in direct vascular suture. Nevertheless, direct anastomosis of the carotid and jugular circulations in this biological model is fully possible with sufficient practice and with the use of the operating microscope.

It must be acknowledged that the preparations described here are considerably in advance of those classical models of Demikhov[1] (and more recently Santo and associates[3]), since ours represent true isolated cephalons whereas theirs represented transections at the level of the thorax (including upper limbs) and therefore more correctly should be classified as "upper body grafts." Demikhov's transplantations were accomplished at a canine level and involved the transference of the graft to an intact animal. Ours utilize subhuman primates and achieve direct cephalosomatic association which obviously would be the only framework applicable to clinical transplantation.

Our major thrust to date in these experiments has been directed toward the solution of the technical and surgical problems that have attended the development of this biological model. Obviously, in the long-term management of these cephalic transplants, continuous respiratory support will be required. Whether exogenous support of blood pressure will likewise be necessitated is not known; however, after many hours of vascular linkage, catecholamine infusion has been needed. Certainly direct vascular suture will eliminate the long-term need for anticoagulation.

In the final analysis, the cephalosomatic transplant should reach a state of physiological equilibrium equitable with the individual who has suffered a cervical transection high enough to require respiratory support but who can be maintained in a viable, conscious state indefinitely.

SUMMARY

The surgical technique, instrumentation, and short-term performance of the primate cephalic exchange transplant model has been reviewed. For the first time, the subhuman primate cephalon has been vascularly transferred with the use of loop cannulas to the isolated body of the monkey with normalization of cerebral function.

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DISCUSSION

Dr. Bernard S. Linn (Miami, Fla.). I just rise to make a small point that is sort of jingoistic. I believe that in 1907 or 1908 in the *J. A. M. A.* the upper half of the trunk was reported to be transplanted by Carrel and Guthrie in a series of dogs.

Dr. Robert J. White. Yes, I am well aware of Dr. Guthrie's work. As some of you know, there has been serious question as to whether Dr. Guthrie should at least have shared the Nobel Prize that eventually went to Dr. Carrel.

However, I think Dr. Guthrie would be the first to admit that the time delays and the limitations of the surgical techniques in those days would mitigate seriously against the probability of the brain being preserved in a normal state. This is really the major message that I would like to get across—our efforts have been to transplant the brain and retain its capability of being and remaining normal. This is, of course, not like taking out the kidney, and so, therefore, our problems are a little bit different.

I fully agree that in terms of thinking out these problems Drs. Guthrie and Carrel were among the first. The difficulties of preserving the organ during the surgery and the period of transfer would obviously render the brain, in their preparations, suspect of being either dead or far from normal.

Chairman Starzl. Dr. White, there is a preparation called the *encephale isolé* that has been used extensively for neurophysiologic studies. Have you in your experience come across any difference in the behavior of the forebrains in your animals, compared to those that were studied by H. W. Magoun and others, with their *encephale isolé* preparations?

Dr. White. If the fifth nerve is preserved in our preparations, because of the great load of sensory information which is delivered over it, the electroencephalographic patterns which are taken off the cortex would appear to be awake and hard to distinguish from that of an intact animal. In contrast, the brains in Magoun's isolated experiments often exhibited a sleep state. We believe that in cutting the brain stem, and probably opening the basilar artery, Magoun's animals were really in a shock state. Our animals were not sleepy: They tracked; they ate; and they bit you if you brought your delicate hands near the mouth.

Chairman Starzl. Thank you, Dr. White. Actually, I have been bitten by *encephale isolé* monkeys, or cats, while working with Dr. Magoun a number of years ago. The features of your isolated head preparations as you just described them do not seem to me to be particularly different from the older observations of Dr. Magoun.