Experimental Irradiated Nerve Heterografts*

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Over the years innumerable methods for repairing large defects in peripheral nerves have been tried with only minimal success. Huber\(^4\) reviewed the literature in 1895 and described the following methods: a) nerve stretching (Schüller); b) heterografts; c) tubular structures; d) nerve flaps; e) cross grafting from an adjacent nerve; f) resection grafting of bones (Löbker).

The number of methods has increased considerably since then as various attempts have been made to solve this problem.

Results of end-to-end suture were reviewed by Highet and Holmes\(^8\) who found that when the joints were placed in acute flexion, traction injuries occurred and poor results were obtained. The ability to use nerve grafts would furnish an ideal solution to the problem of the large nerve defect. There is no doubt that if heterografts were successful they would be the operation of choice.\(^28\)

The history of heterografts began in 1880 when Gluck\(^7\) implanted a fresh heterograft using a 3.5 cm. section of the sciatic nerve of a rabbit to bridge a 3 cm. defect in the sciatic nerve of a hen. He reported return of function in 11 days which according to modern knowledge is not possible. His experiment, however, is of historical interest. Johnson\(^14\) repeated the work in 1882 and found that the heterograft united with the proximal and distal nerve but that there was no sign of innervation after 23 to 34 days. Assaky\(^2\) in 1886 reported 4 transplants with 1 successful 3 cm. sciatic graft from a turkey to a rabbit with return of function in 35 days. Huber\(^10\) in 1895 reported 5 satisfactory transplants of sciatic nerve of a cat to the ulnar nerve of the dog with return of function within 190 days. He also reported 14 transplants in humans, 3 successful and 7 improved. One of the successful cases occurred within 16 days. Merzbacher\(^20\) in 1905 transplanted nerves from various animal species to other species and reported that they did not degenerate but that necrosis of the graft occurred.

Attempts were made also to prepare heterografts to make them more successful. Durous\(^5\) in 1911 placed a cat’s nerve which had been kept on ice for 24 hours into a defect in a dog’s sciatic nerve, but the result compared unfavorably with even a fresh heterograft. Ingebritsien\(^12\) in 1915 reported that heterografts are unsuitable for bridging nerve defects. In 1916 he reviewed the cases published to date\(^13\) and concluded that only 1 heterograft could be considered as successful. Nageotte\(^21\) in 1917 conceived the idea of using a heterograft that was fixed in alcohol and kept in 50 per cent alcohol until 24 hours before use when it was transferred to Ringer’s solution. Although he claimed functional recovery, no details were given in his reports. Huber\(^41\) in 1919 reported that grafts of this type were innervated in the rabbit. Nageotte\(^21\) in 1917, using alcohol-fixed calf heterografts in the sciatic nerve of the dog, observed return of electrical excitability in 138 days. Sencert\(^25\) in 1918 performed 15 heterografts of alcohol-fixed calf nerves to humans but he did not follow the cases long enough.

Policard and Leriche\(^22\) in 1922 published the results of a 2-year follow-up on a transplant of the sciatic nerve of a calf to a human. The nerve elements had grown into the implant from below, but the upper end was blocked with fibrous tissue. Vargas Salcado\(^29\) in 1925 took up the work of Nageotte in Chile and reported 5 successful cases of alcohol-fixed calf nerve transplanted to the human. Sweet\(^26\) in 1929 reviewed the literature on nerve regeneration and reported the work of Nageotte and Sencert. He utilized alcohol-fixed fetal calf nerve to bridge 1.5 to 3 cm. gaps in the sciatic nerve of the dog. A significant amount of scar tissue developed at both suture lines hampering the growth of the axons and the percentage of success was very small. Sanders and Young\(^23\) in 1942 re-
ported their work on heterografts using both fresh and alcohol fixed dog and rat nerves transplanted to rabbits. The grafts were attacked by white blood cells which at 25 days had almost destroyed the graft. Weiss and Taylor29 in 1943 stated that most devitalized heterografts behave like foreign bodies. Heterografts subsequently fell into disrepute and little has been added to the literature in the past 2 decades. Seddon24 stated in 1963, "heterogenous grafts behave as foreign bodies and are completely useless."

With the advent of cathode irradiation to sterilize homografts3,18,29 and decrease their inflammatory response in the host,5,17,19 it was decided to investigate the value of irradiation in nerve heterografts. The grafts utilized in the following experiments were obtained as follows:

The grafts were removed from various species of animals under unsterile but clean conditions and immediately packaged in heat-sealed polyethylene bags and frozen to $-12^\circ$F. The grafts were maintained in a frozen state and irradiated with 2,000,000 r.e.p. by a Van de Graaff generator which sterilizes the graft within the bag and alters its rejection by the host. The grafts were then stored in a freezer at $-12^\circ$F until used 4 to 8 weeks later.

**Experiment 1**

This experiment was set up to determine the effect of cathode irradiation on peripheral nerve heterografts. Irradiated and nonirradiated implants in animals were to be compared.

**Method.** Eleven rats of different strains and size were utilized for this study. Previously prepared frozen nonirradiated and irradiated peripheral nerves from dogs were cut into 1 cm. sections to serve as grafts. The rats were anesthetized with ether and a subcutaneous incision made in each thigh. The skin flap was dissected to allow the implant to be placed away from the skin incision. An irradiated heterograft was placed in the right thigh and a nonirradiated heterograft in the left thigh. Three rats were killed after one week, 3 after 2 weeks, 3 after 3 weeks, and 2 after 6 weeks. Gross examination and microscopic study were performed to determine the inflammatory response produced.

**Results.** Examination of 3 nonirradiated implants after 1 week revealed a moderate amount of swelling and inflammation about the heterograft. The microscopic sections were stained with hematoxylin and eosin. There were inflammatory cells about the implant with some areas of axis cylinder destruction.

Gross examination of the irradiated heterograft after 1 week showed very little reaction about 2 of the implants and swelling about the third, but very little inflammation. The microscopic sections revealed severe inflammatory reaction in 2 and a slight reaction in the third.

Two weeks after implantation the nonirradiated nerve revealed considerable swelling in all 3 implants. Microscopically there was a tremendous inflammatory response with invasion and destruction of the graft.

The irradiated nerve after 2 weeks revealed a small amount of swelling about one nerve and some slight scarring about the other 2, with little edema. Histologically there was a marked inflammatory response but less destruction of the nerve as compared to that in the nonirradiated nerve.

After the third week the gross appearance of the nonirradiated heterograft showed adhesions, edema, and a moderate inflammatory response. Microscopic sections revealed tremendous destruction with complete invasion of the graft by inflammatory cells.

The irradiated grafts after 3 weeks showed minimal adhesions, no edema and no inflammation. Microscopically the grafts revealed a severe inflammatory response about 2 implants and a minimal response about the third.

Six weeks after implantation, biopsy of the nonirradiated graft showed the implants to be bound down by adhesions. Microscopic sections revealed inflammatory cells in the graft and invasion of the periphery with a few cylinders remaining (Fig. 1 (a)).

The irradiated grafts revealed some edema with adhesions about the graft. Histologically the axis cylinders were still present and one rat showed very little scar tissue (Fig. 1 (b)).

**Discussion.** It was apparent from this study that irradiation had a slight depressive action on the inflammatory response produced by a nerve heterograft in a rat. Nonirradiated implants were all severely affected in contrast to some of the irradiated heterografts which were still satisfactory, with open axis cylinders.

Although some decrease in the inflammatory response was produced by this method, the degree of response varied considerably.

**Experiment 2**

The second experiment was set up to study the results of heterografts in a small laboratory animal. The facial nerve of the rat was selected as the peripheral nerve to be used in the study due to its accessibility and the fact that there are 2 main branches which innervate the whisker