CONDITIONED ELECTRICAL DEFENSE REFLEX IN DOGS WITH AUTOTRANSPLANTED LIMBS

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(Received June 3, 1955. Presented by V. N. Chernigovsky, Member of the Acad. Med. Sci. USSR)

According to I. P. Pavlov, "restoration of locomotion is achieved in dogs with the aid of the cerebral hemispheres, i.e. with the aid of conditioned reflexes. If a dog which has learned to walk after an injury is deprived of its cerebral hemispheres, it will become an incurable cripple" [2, p. 480].

E. A. Asratyan and his collaborators [1] have for many years studied the role of the cortex of the cerebral hemispheres in restoring locomotor functions of animals. They have shown that the process of restoration of motor functions of deafferentated limbs is connected basically with the activities of the cerebral cortex. For this reason, in addition to the study of the unconditioned reflexes of an autotransplanted limb, we determined the rate of formation of conditioned reflexes, as well as their characteristics.

Only the conditioned reflex method is able to show how soon after autotransplantation of a limb establishment of its communications with the brain takes place. The solution of this problem is of both theoretical and practical interest, inasmuch as fully efficient voluntary movements of the transplanted limb, as well as the trophism of its tissues, are possible only after establishment of functional relations with the central nervous system.

Ordinarily, the development of a conditioned electronociceptive reflex in normal dogs presents no difficulty, owing to the rapid formation of conditioned links, which gain strength with each repetition of the experiment. In dogs which have undergone autotransplantation of the left hind limb, the possibility of forming a conditioned electronociceptive reflex involving the transplanted limb depends on the time required for the limb to establish itself, inasmuch as restoration of the functions of such a limb is a slow process. The locomotor function of the limb is restored sooner; the dog begins to walk on the 4th or 5th day after the operation, dragging the transplanted leg, but 3-4 weeks later the leg begins to take part in locomotion, at first supporting itself on the back of the paw, and then more and more frequently using the sole of the paw. The normal position of the paw is finally established 5-7 months after the operation but, even after this, deterioration of the motor function of the leg occurs in cold weather, or in the case of additional injury or generalized disease.

In the early, critical stage of the denervated limb and of the animal as a whole, its muscle tissue displays enhanced excitability to various stimuli. Because of the raised sensibility to electric stimulation of the muscles of autotransplanted limbs, it is possible, from the 5th to 7th day after operation, to determine the threshold of excitability of the muscles and their constitutional chronaxie. The motor effects, in the form of movements of the digits or the whole foot are evoked by weaker currents (4-13 v) than in the normal limb (9-30 v), which is evidence of the lowering of the threshold of motor excitability of the denervated muscles (Fig. 1); at the same time the chronaxie of these muscles amounted to 28-40 σ. Such a discrepancy between the values for rheobase and for chronaxie is characteristic of the denervated muscles of an autotransplanted limb, and is most pronounced during the first three months after the operation. After 3-4 months, evidence of the gradually increasing function of the regenerating motor nerve begins to appear, and only some time later does the possibility of determining subordinational chronaxie arise. The chronaxie values gradually approach the preoperational ones, normal levels being attained 8-12 months after operation, or even later.

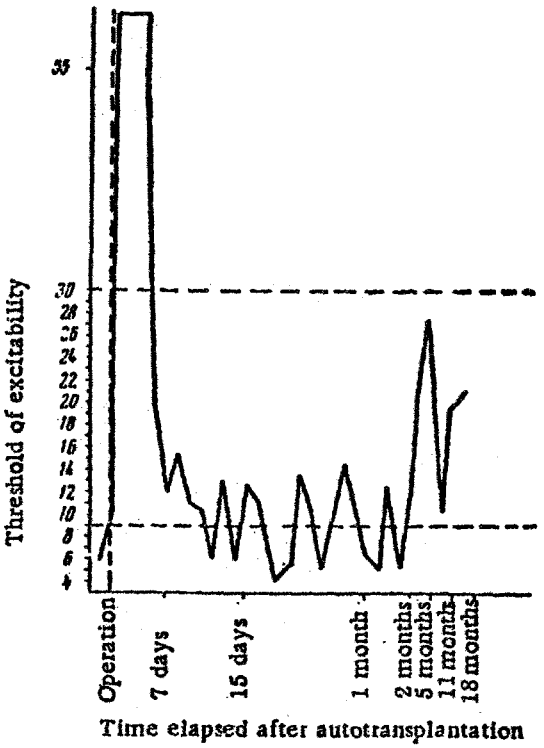

Fig. 1. Changes in the excitability threshold of the muscles of the transplanted leg.
- - - range of excitability of the muscles of the contralateral normal leg (9-30 v) after the operation.

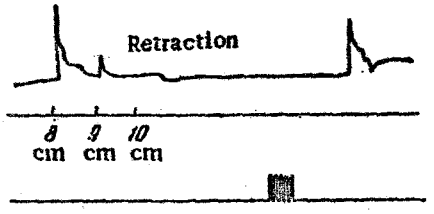

Fig. 2. Determination of threshold of excitability to application of an induction current to the dog Beshka. The conditioned nocicept-reflex appeared 20 seconds after the conditioning stimulus (skin stimulation).
Explanation of curves (from above down): flexion of foot, stimulation signal (the figures refer to the distances between the induction coils).

The receptor function of the autotransplanted limb (coarse pain and touch sensibility) reappears 4-5 months after operation; the time required for normalization depends on the rate of re-establishment of nervous continuity between the limb and the rest of the organism.

Thus, during the first three months, before establishment of nervous connections with the organism, the limb takes only a passive part in locomotion; it is atonic, hyperthermic, and edematous, and has a raised sensibility to stimulation, with a prolonged latent unconditioned reflex period.

After 3-4 months, the limb actively participates in locomotion. The atonic state disappears, and is gradually replaced by heightened tonus of the flexor muscles. The heightened sensitivity gradually diminishes, and reactions to pain, heat, and touch reappear.

With restoration of unconditioned reflex activity of the transplanted limb, 5-6 months after the operation, the possibility of forming conditioned electronociceptive reflexes arises. These were developed in the usual way, by coupling conditional stimulants (a skin stimulator or bell) with an electric shock. The electrodes were usually fastened to the dorsal surface of the foot, and the skin stimulator to the skin of the thigh of the operated leg, 3-5 cm below the suture.

Altogether, 205 experiments were performed on 7 dogs with autotransplanted left hind legs (the operations were performed by N. P. Petrova).

Before each experiment we determined the excitability threshold by means of an induction coil, and we used currents slightly higher than threshold for evoking a nociceptive reaction.

The conditioning stimulus usually preceded the electric shock by 2 seconds, and the two stimuli acted together during the subsequent 3 seconds, after which there was an interval of $1\frac{1}{2}$ -2 minutes before the next set of stimuli. The conditioned and unconditioned stimuli were recorded in ink on a kymograph.

Elaboration of the conditioned reflex was always started soon after the operation (2-3 months), but, in spite of the large number of repetitions of combined stimuli (66 or more), we were not successful in developing a conditioned reflex from the autotransplanted leg earlier than 5-6 months after the operation. The reflex then obtained was not at first stable or of constant strength, and during its formation a number of characteristic features were observed, such as its weak motor expression and its prolonged latent period, which we found to persist for 25 seconds or longer (Fig. 2).

Fig. 2 shows a prolonged latent period of a conditioned reflex to skin stimulation, with normal reaction times to unconditional stimulus (induction current), for the dog Beshka.

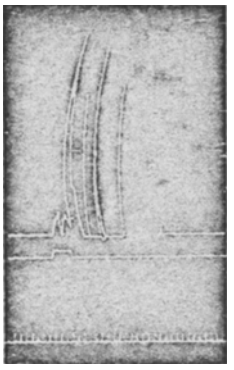

Fig. 3. Conditioned electronocieeptive reflex to the sound of a bell, from an autotransplanted limb of the dog Silva.
Explanation of curves (from above down): movement of limb, stimulating signal (bell), time marker (1 second).

Similar delayed motor reactions were found for other dogs, in experiments performed before the conditioned reflex appeared. Thus, the conditioned reflex was formed in the dog Seryi over the period $2^1/_2$ to $4^1/_2$ months after transplantation of the limb. The reflex was achieved rapidly in determining the threshold of excitability for movement of the foot, by means of an induction current, but when the sound of a bell was associated with the current it was retarded in 13 cases out of 22 by 15-35 seconds. After associating current with a skin stimulator, a latent period of 5-22 seconds was observed in 8 cases out of 20. Similar effects were found for the dogs Kisa, Orion, and Druzhok, for which elaboration of the conditioned reflex was begun 2-5 months after healing of the wound.

Formation of the conditioned reflex was started later for the remaining dogs, and the latent period was of normal duration for both conditioned and unconditioned reflexes, i.e. the protective reaction, in the form of flexion and drawing up of the limb, corresponded in time with the action of the conditioned and unconditioned reflex stimuli.

Reflexes elaborated a long time (9-10 months) after transplantation are rapidly established, and have high stability.

Fig. 3 shows a kymogram of an experiment with the dog Silva, on May 27, 1952; this shows the presence of an electronocieeptive reflex from a transplanted limb in response to the sound of a bell.

A distinctive feature of the electronocieeptive reflex formed in dogs 5-6 months after the operation is the extreme difficulty in inducing discrimination between stimuli which closely resemble each other.

This instability of the reflexes is confirmed by their ready extinction.

LITERATURE CITED

[1] E. A. Asratyan, Uspekhi Sovremennoi Biol. 5, 804-824 (1936).

[2] I. P. Pavlov, Collected Works* (Moscow, 1951), pp. 478-480.

*In Russian.