

9. The parietal eye is easily removed, and complete removal can be determined by visual inspection under a dissecting microscope. Complete removal of the pineal organ was confirmed histologically in five of the nine pinealectomized lizards. One lizard showed some pineal tissue remaining after pinealectomy. Three of the lizards died before the brains could be taken for histology. The paraphysis was often removed along with the pineal organ, and damage to adjacent nervous tissue was sometimes evident. All operations were performed on lizards immobilized by cooling and embedded in crushed ice.
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Reinnervated Eye Muscles Do Not Respond to Impulses in Foreign Nerves

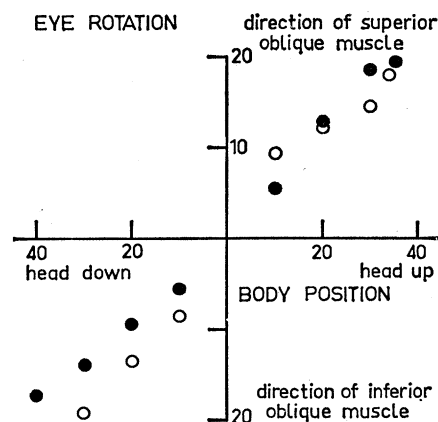
Abstract. Normal movements return to carp eyes after section and regeneration of the IIIrd and IVth nerve trunks. Two months after reinnervation, records of impulses in the inferior oblique nerve during tilting of the body show activity of the normal motoneurons to that muscle, together with discharge patterns characteristic of the antagonistic superior oblique and some of the rectus muscles. These axons must have found their way into the inferior oblique trunk during sprouting at the lesion and must be maintained after reinnervation. Impulses from foreign axons are without detectable effect on eye movement and therefore must be blocked at their termination in the muscle. Previous study of cross-innervated and doubly innervated fish eye muscles revealed only structurally normal neuromuscular junctions. Transmission from foreign junctions in multiply innervated muscle is blocked by competitive molecular recognition and control mechanisms that do not cause degeneration.

Regrowth of cut motor nerves in some lower vertebrates restores full coordinated movement to the reinnervated muscles (1). When the superior oblique muscle of the carp eye is innervated first by the nerve to the antagonist muscle and then superinnervated by the correct nerve, contractions caused by the antagonist innervation cease as soon as the response to the original nerve appears. Foreign nerves of proven ability stop working when the original nerve grows back. No ultrastructural signs of degeneration of neuromuscular junctions accompanies the repression of the first innervation. Either foreign motoneurons no longer bring impulses to the muscle or neuromuscular transmission is blocked (2). We have distinguished between these two possibilities by the following experiments.

In two carp the IIIrd and IVth nerves to one eye were cut where they run together in the cranium. The eye was paralyzed except for flick movements from the posterior rectus muscle which is innervated by the VIth nerve. Nineteen days later other movements began to return, weakly at first but ap-

propriate in direction and timing as indicated by the conjugate behavior of the intact eye. After two more months the movements of both eyes were coordinated and equal. Figure 1 shows the rotation of both eyes in response to tilting the whole body up and down. The difference between the curves is no greater than normal (3).

The fish were then anesthetized with MS 222 and paralyzed with gallamine. The eyes and extraocular muscles were removed, leaving the nerve stumps free.



The animals were mounted in an apparatus which could be tilted up or down and rotated to the left or right. An electrode was placed successively on the cut ends of the inferior oblique nerves of the normal and the reinnervated sides. Figure 2 shows the responses to tilting. Normally the inferior oblique neurons discharge when the head is down and stop firing if the head moves up. Neurons of the superior oblique nerve discharge for movements in the opposite direction. On the reinnervated side there were fibers in the inferior oblique trunk which increased their discharge when the head was down and others which discharged when the head was up—reflex behavior which usually identifies motoneurons of the superior oblique. Other maneuvers revealed the discharge patterns of motoneurons from the rectus muscles in the same nerve.

Therefore, sprouting and regrowth of the IIIrd and IVth nerves must have been haphazard, and axons from different kinds of motoneurons must have entered channels which eventually led them out into the inferior oblique branch of the IIIrd nerve in the orbit. Two months later they were still there and they showed normal reflex responses. Nevertheless, the movements of the eye produced by the inferior oblique muscle corresponded not to the totality of the impulses in the trunk but only to those delivered by fibers of the original inferior oblique nerve. Thus, when the head is held up, superior oblique neurons should discharge to pull the eye around toward the horizontal while the inferior oblique motoneurons should stop. The neurogram from the operated side (Fig. 2) shows a strong burst of superior oblique neurons in the inferior oblique nerve. If this burst had been transmitted to the muscle, coinervation of the two

Fig. 1. Graph of the wheel-like rotation of the carp eye about the optic axes when the body of the fish is tilted up and down from the normal horizontal position. The reflex movements, mainly of labyrinthine origin, tend to keep the eye horizontal. Rotation when the head is up is almost entirely due to contraction of the superior oblique muscle; rotation when the head is down is due to the inferior oblique muscle [see (5) for further details]. Left operated eye, filled circles; right control eye, open circles. Measurements were made 2 months after reinnervation of the left eye muscles. Both eyes show normal reflex movements, and the difference between them is no greater than that in normal fish. Axes in degrees from horizontal.

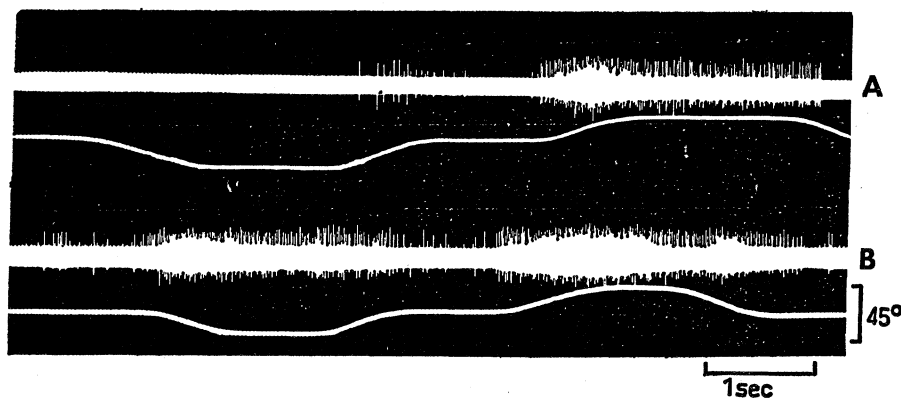


Fig. 2. Records of impulse traffic in the inferior oblique nerve trunk of the fish whose eye movements are illustrated in Fig. 1. (A) Normal right eye. Top trace, nerve recording; bottom trace, position of fish; head-up movements signaled by depression of the trace; head-down movements signaled by elevation of the trace. Note that the normal inferior oblique motoneurons only fire as the head is lowered and only discharge tonically when the fish is held head down. (B) Reinnervated left eye. The nerve record shows fibers that discharge both when the head is up and when it is down. Continued discharge when the head is up is the response pattern normally seen in records from the superior oblique nerve.

oblique muscles in the same eye would have reduced or abolished the normal counterrotation of the eye. Figure 1 shows that this was not so. When the head was up movements of the operated and control eyes were equal.

It is clear that nerves can grow into foreign muscles and retain their characteristic reflex behavior. The selectivity on which coordination depends is therefore peripheral, between nerve and muscle. Inappropriate nerves either do not form terminals, or those formed do not work. We favor the latter view because of the results of electron-microscopic studies of cross-innervated and doubly innervated eye muscles. Only morphologically normal nerve endings were seen when it was clear from the reflexes that nerves that once worked had just been suppressed (2). So far there seems to be no reason to think that foreign nerves establish themselves in these multiply innervated muscles except by the formation of the usual terminal synaptic apparatus. No unusual endings have been seen in the present muscles. However, whether or not multineuronal innervation is allowed, a subtle but strong selectivity must operate to block transmission of excitation from foreign nerves as long as the correctly matched nerves are present. Perhaps the developmental mechanisms for neuromuscular connection permit the formation of inappropriate synapses but regulate transmission precisely. The morphological and physiological correlates of suppressed transmission are not known.

The mechanism of coordination in reinnervated muscles recalls the reso-

nance theory of Weiss (4), who suggested that nerves broadcast messages for several muscles, each of which could decode the correct command and ignore all the others. After regeneration, nerves of one kind are no longer segregated in trunks and reinnervated muscles receive a mixture of commands for many muscles but, through peripheral filtering, respond only to the correct ones. Presumably they do so, not because they resonate to the right message, as Weiss thought in 1924, but

Harderian Gland: Influence on Pineal

Hydroxyindole-O-Methyltransferase Activity in Neonatal Rats

Abstract. *A circadian rhythm has been found in hydroxyindole-O-methyltransferase activity of the pineal gland of blinded 12-day-old rats. Five additional hours of lighting can partly prevent the nocturnal increase in pineal hydroxyindole-O-methyltransferase activity in such rats. Removal of the Harderian gland abolishes this response to light in 12-day-old blinded animals, giving further support to the suggestion that this gland may function as an extraretinal photosensitive organ influencing the pineal gland in blinded suckling rats.*

The concentration of serotonin in the pineal gland of the rat undergoes a rhythmic variation, reaching a maximum at about the midpoint of the light cycle, and falling rapidly after the onset of darkness. Extending the lighting period prevented the serotonin fall in intact, but not in blinded mature rats. In immature rats, however, extended lighting prevented the pineal serotonin fall in both intact and blinded animals (1).

In a previous study (2), we confirmed the persistence of the pineal

because some competitive process of chemical recognition has made ill-matched synapses ineffective.

If the regulatory mechanisms of neuronal growth tolerate and maintain the terminals of active motoneurons in a muscle, even when they are prevented from transmitting, we wonder whether a similar mutual repression of synapses might control interneuronal connections in the central nervous system. A repressive process that depended on the activity of a synapse would provide a learning mechanism based on the faculties of intercellular recognition that are important in brain development (5).

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serotonin rhythm in immature blinded rats and the effect of additional light on that rhythm. We also reported that the removal of the Harderian gland abolished the effect of light on the pineal serotonin levels of the blinded 12-day-old rat and suggested that the Harderian gland may act as an extraretinal photoreceptor influencing the pineal serotonin rhythm in immature rats.

Pineal hydroxyindole-O-methyltransferase (HIOMT) activity also undergoes a circadian rhythm. In intact