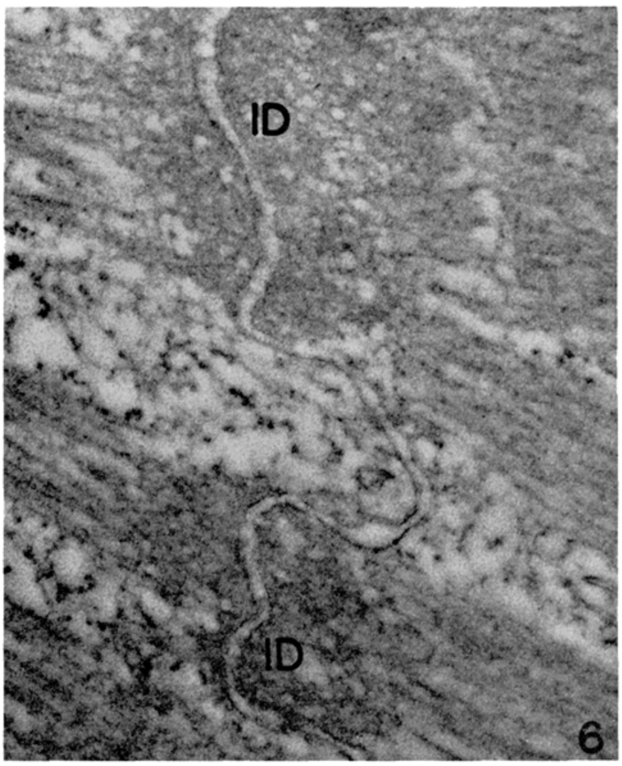
Intercellular abutments are of three types, all of which have been observed in higher vertebrate forms. The most common is the simple intercalated disc, interrupting the longitudinal patterns of single myofibrils (Fig. 1, 4, 5). These often occur in stepwise fashion, the steps being one sarcomere long (Fig. 4). Complex discs, interrupting the pattern of more than one fibril (Fig. 6) also occur, although less commonly. These discs are convoluted but not to the same degree as those of the mammalian cardiac muscle. Either type of intercalated disc may at times be continuous with Z band substance (Fig. 4) and indeed the Z band and disc substances appear to be similar. The similarity is not as striking in the fish as in the mammal, however, in that the Z bands are generally of little density, whereas the discs may be of considerable density and indeed of total width up to 1/3 μ. The disc itself comprises modified plasma membranes, vesicles, filaments, a disc substance, and an intercellular cement of medium density. Structures quite similar in their detail, but shorter, not normal to the long axis of the cells, and not always related to contractile filaments, have been observed associated with, but at times distant from, the intercalated discs. These are termed intercellular bars (Fig. 2–5). They seem to be always associated, at least in one of the abutting cells, with the Z band of a fibril. These intercellular bars correspond to, and include, structures previously referred to as mirror plaques[9], desmosomes[8], and specialized regions of the disc[6].



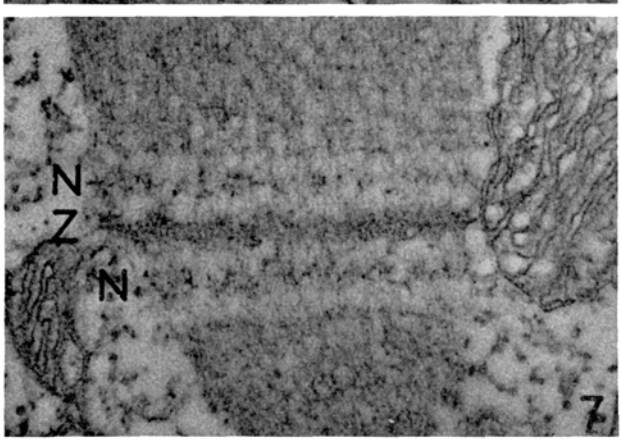


Fig. 6. — Complex intercalated disc (ID) in fish heart, interrupting the longitudinal pattern of several myofibrils. The complex disc of the fish is less convoluted than that of the mammal. × 60000

Fig. 7. — Detail of I region of a myofibril of the ventricular muscle fiber, showing *N* lines on either side of the fairly light *Z* band. × 60000

The results of the present study show that an intercalated disc, closely resembling that in mammalian cardiac muscle, exists in the heart of the fish. In addition, more examples of intercellular bars have been found, in close association with the intercalated disc[8], and the Z band, affording further evidence that these three structures are related to one another.

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Résumé

L'examen au microscope électronique de coupes minces du cœur de cyprin doré révèle la présence de disques intercalaires de forme soit simple, soit complexe, semblables aux disques des vertébrés supérieurs. De plus, les barres intercellulaires sont présentes. La structure et les rapports des disques et des barres avec la bande Z de la myofibrille suggèrent leur proche similarité.

The Importance of Autoplastically Transplanted Pituitaries for Survival and for Regeneration of Adult *Triturus*

The essential role of the pituitary for the initiation of regenerative processes having been discovered in 1926[1] and definitively established in a series of other researches[2–4], it became imperious to investigate firstly, whether heterotopically transplanted pituitaries were able to support the vital functions indispensable for survival, and secondly, whether grafted pituitaries away from their normal nervous connections, were still capable of providing the required stimuli for regeneration. Autoplastic transpantations were performed on 163 adult *Triturus viridescens* (kept at 20° ± 1° C) into the anterior *eye-chamber*, the *manus* or the *tarsus*. Direct observation under the binocular permitted to establish vascular connections between the eye and the pituitary within 5 to 10 days; for the manus and the tarsus it was established by histological examinations that vascularization occurred somewhat later in most, but not in all the cases.

I. *Survival patterns after autoplastic pituitary transplantations.* Hypophyseoprivic controls begin to die fifteen days after the operation and only 11% live as long as fifty

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[3] A. B. HALL and O. E. SCHOTTÉ, J. exp. Zool. *118*, 368 (1951)

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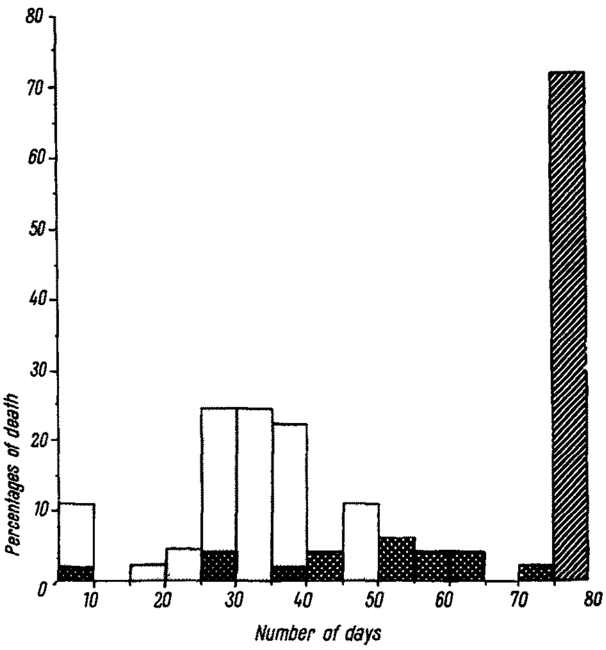
days. Newts with heterotopic transplants shed their epidermis regularly and keep their pigmentation as do normal animals. The data condensed on the histiogram (Fig.) show that hypophysectomized animals may be kept alive for an indefinite length of time; we carry at present numerous cases having survived for over two years.



Histiogram of death patterns of hypophysectomized newts (dotted colums) and of hypophysectomized newts bearing autoplastically transplanted pituitaries (checkerboard columns). The column with diagonal lines on the right represents the percentage of graft-bearing *Triturus* having lived longer than 75 days. (This column, then, refers to survival, rather than to death patterns.) The first double column (from 1 to 10 days) represents early post-operatory deaths within the two experimental series.

II. *The influence of autoplastic pituitary transplants upon forelimb regeneration* may be correlated, particularly in the eye-chamber cases, with their status of vascularization (Table, 3rd horizontal column, but details on vascular connections are not specified): among 47 survivors, 36 had displayed within five to ten days visible vascular loops; subsequently, of these, 34 regenerated normally and 2 did not; conversely, among the 11 cases where no vascularization was observable only 3 did regenerate their limbs. Other differences in regard to regeneration were observed between 'young' and 'old' transplants.

(a) '*Young' transplants* are cases where amputation of the forelimbs was performed simultaneously with the pituitary transplantation. The data of the Table show that 82 (or 80·2%) of these animals exhibited normal regeneration, while 20 (or 19·6%) failed to regenerate. In control experiments with hypophyseoprivic newts 6 (15%) did regenerate and 34 (85%) did not. Histological studies have, in addition, revealed a relation between the site of transplantation and successful regeneration: better 'takes' are obtained from pituitaries placed within the larger tarsus than when transplanted into the much smaller manus of newts; they also showed that, when total resorption of the graft was diagnosed on sections no regeneration took place.

(b) In *'old' transplants* forelimb regeneration was tested 7 to 8 months after transplantation (Table). Results from these animals differed from the previous group since, in spite of healthy appearance and normal pigmentation, 27 among 38 cases showed appreciable delays in the initiation of regenerative processes.

Table

Site of autoplastic transplantations of pituitaries and their effects upon regeneration in forelimbs of *Triturus viridescens*. (A control series dealing with the effects of hypophysectomies and amputation without concomitant heterotopic transplantations is included.)

Site of autoplastic transplantation	Total number of operations	Number of cases observed	Observations at 30 days regeneration		
			Normal	Absent	Delayed
Carpal region of forelimb	35	32	24	8	—
Tarsal region of hind limb	30	23	21	2	—
Anterior eye chamber (a) young transplants	48	47	37	10	—
(b) old transplants .	50	38	10	1	27
Hypophysectomy and amputation . . . . (without transplantation)	45	40	6*	34	

* Numerous previous investigations performed at this laboratory had shown, that no matter how carefully performed the hypophysectomies are, there are always some cases (appr. 15%) in which small remnants of pituitaries (discovered upon histological examination of the cranial cavity in supposedly completely hypophysectomized newts) account for regeneration in such cases (HALL and SCHOTTÉ[8]). This explains the occurrence of positive regeneration in the hypophysectomized newts of the control series.

Conclusions. – These experiments show: that (1) autoplastically transplanted anterior pituitaries may become properly vascularized within a matter of days and that they may remain 'alive' for many months within the eye-chamber and the carpal or tarsal regions; (2) heterotopic pituitaries maintain essential life functions well beyond the normal life expectancy in hypophysectomized newts; (3) after amputation, these transplants are also efficient to support regeneration, but this ability depends on adequate vascular supply and after months of sojourn in heterotopic position a slowing down in the initiatory phases of regenerative processes becomes noticeable. It is therefore concluded that a transplanted pituitary is capable of maintaining in its new site a level of activity sufficient to induce the adrenal end of the pituitary-adrenal axis (shown at this laboratory[5–7], to be essential for regeneration) to react positively to the stimulus of even much delayed amputations, in spite of severance of the normal vascular and nervous connections of the pituitary with the hypothalamus.

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Department of Biology, Amherst College, Amherst (Mass.), July 29, 1959.

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Résumé

Une hypophyse greffée autoplastiquement dans la chambre antérieure de l'œil, dans le *manus* ou dans le *tarsus* s'est avérée capable de remplacer l'hypophyse *in situ* dans son action sur la régénération et la survie chez le Triton, à condition que s'établisse une vascularisation satisfaisante du greffon et que la durée de l'expérience ne dépasse pas plusieurs mois.

Does the Ectodermal Proctodaeum Participate in the Development of the Pronephric Duct in Urodele Amphibia?

In 1926 and 1929, VOGT[1] demonstrated conclusively, by means of the vital staining method, that invagination of mesodermal material in urodele Amphibia continues after the neural groove has appeared and the blastopore has taken on a slit-like form. In 1929, I made the same observation and found that stained material of areas lying in the above-mentioned stage on both sides of the blastopore formed somites of the hindpart of the trunk after invagination, and – in some experiments – appeared as a cord-like structure extending along the ventral edge of the somites in the same part of the body. This 'cord' of cells shining through the ectoderm was seen turning off with its caudal extremity in the direction of the ectodermal proctodaeum (BIJTEL[2], 1929, Fig. 6, 1931, Fig. 4). After dissection of the living anaesthetized larvae, this structure was supposed to be the hindpart of the pronephric duct. Thus it would seem that the ectodermal proctodaeum had taken part in the development of the hindpart of the pronephric duct. Histological examination of the stained tissues was not possible at that time, as I did not succeed in preserving the vital stains for that purpose.

Staining experiments of O'CONNOR[3] in *Amblystoma tigrinum* and *Triton taeniatus*, and of VAN GEERTRUYDEN[4] in *Rana fusca*, have proved that the pronephric duct in larvae of these Amphibia is formed by the independent caudal growth of the pronephric rudiment. The caudal extremity of the developing duct joins with a small diverticulum formed by the ectodermal proctodaeum. By blocking up the way taken by the blastema of the pronephric duct, O'CONNOR[5] did not usually find a pronephric duct behind the obstructing transplant. In some experiments, however, at the caudal end of the trunk, a small tag of a duct-like structure was attached to the cloaca. This observation suggested the possibility that the cloaca should contribute to the development of the hindmost part of the pronephric duct. In this connection, O'CONNOR referred to my staining experiments mentioned above.

Results of more recent blocking experiments (BIJTEL[6]), like those of similar experiments of GIPOULOUX[7] however, have shown that the pronephric duct is completely missing only in the case when the obstruction is made in an early stage of development of the pronephric rudiment. If part of the blastema of the pronephric duct has passed the place where the obstacle is implanted, this part continues on its way, joins the small diverticulum pushed forward by the ectodermal proctodaeum and gives rise to the small duct-like structure seen by O'CONNOR. These experiments prove conclusively that the ectodermal proctodaeum does not contribute to the hindpart of the pronephric duct.

One may ask: what is the significance of the stained cord of cells extending along the ventral edge of the somites in the hindpart of the trunk in my experiments of 1929? In recent experiments, the same effect could be reproduced. Figure 1 gives an example of it. In the larva VK 24 of *Siredon mexicanum*, the surroundings of the ventral half of the blastopore have been stained in the early neural plate stage. In about stage 35 (HARRISON), on both sides of the hindpart of the trunk a stained cell cord was seen, extending along the ventral edge of the somites and turning off to the cloaca which was stained as well. As the vital stain could now be preserved, histological examination was possible. It appeared that the stained cord of cells was formed by cell groups of the mesonephros which accompany the unstained pronephric duct, as is shown in Figure 2. In none of these experiments was the pronephric duct stained together with the anlage of the mesonephros.

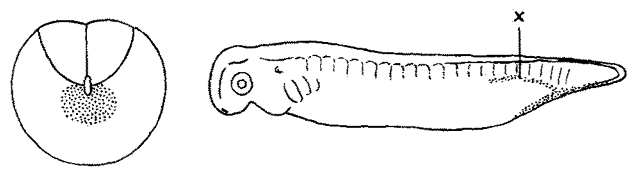

Fig. 1. – Siredon mexicanum VK 24. *a* larva in stage 13 with stained mark round the ventral half of the blastopore. *b* larva in stage 35, *x*: stained mark shining through the ectoderm.

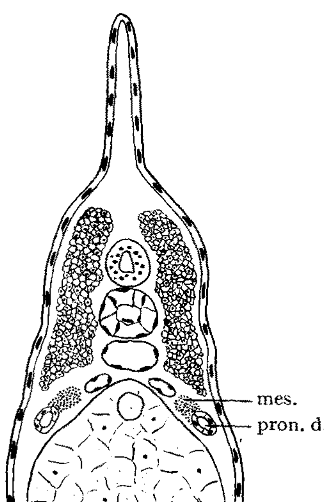

Fig. 2. – Transverse section of larva VK 24 in stage 40. *pron. d.*: pronephric duct; *mes.*: cell group of mesonephros, stained with the vital stain.

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