Regeneration in Haploid Tissue in *Rana pipiens* Embryos

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Summary. It is shown that the haploid condition in *Rana pipiens* embryos does not, in any way, impede their ability to regenerate a tail tip. The implication is that the absence of the paternal genome leaves intact very complex physiological and developmental propensities.

The investigation of the effects of ultraviolet (UV) irradiation has largely been limited to either unicellular organisms, or to cell cultures. Yet the ellucidation of such UV-induced damages are of considerable interest when tested on higher animals. A convenient chordate system is the frog *Rana pipiens* in which the exposure of sperm to UV yields, upon a subsequent fertilization, temporarily viable haploid embryos[1-3]. Hence the study of haploidy in *Rana pipiens* is revealing not only because it represents a unique nucleocytoplasmic relationship but also because it affords an opportunity to assess the effects of UV treatment on such embryonic development. In spite of recent information regarding cytological observations[2] observable in haploid tadpoles, little is known about their ability to perform certain designated functions. For instance impairment in yolk utilization has been previously noted[4-6] in relation to their aberrant growth pattern.

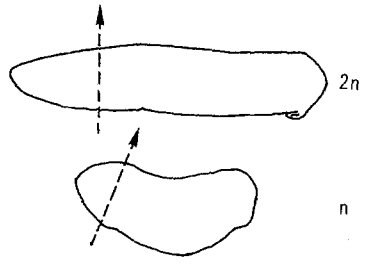

Fig. 1

In this investigation, we present evidence that the development of the haploid syndrome is not concurrent with a total functional debilitation: haploid embryos regenerate their tail tip as well as normal diploid tadpoles obtained from the same batch (Figure 1).

Figures 2–4 show representative phases during the regeneration of tails. Although haploid tissue is visibly different from control tails, particularly with respect to myotomal organization, it is clear that all the normal ($2n$) steps of tail regeneration are accurately mirrored in haploid embryos. These can be briefly described as follows. Shortly after amputation and healing a conspicuous apical tuft forms (Figure 3). This is followed by some internal reorganization as examplified by the formation of a notochordal cap (Figure 2) and myotomal breakdown (Figure 3). The regeneration of the tail tip seems to be initiated by the elongation of a notochordal funnel and an enlarged neural tube end (Figure 4). Finally, some myotomal reorganization can also be seen in the latter stages of regeneration.

Apparently, a number of highly complex physiological traits are retained in toto by the haploid condition. This being the case, we wonder about the possibility that, even during haploidy, certain basic and important physio-

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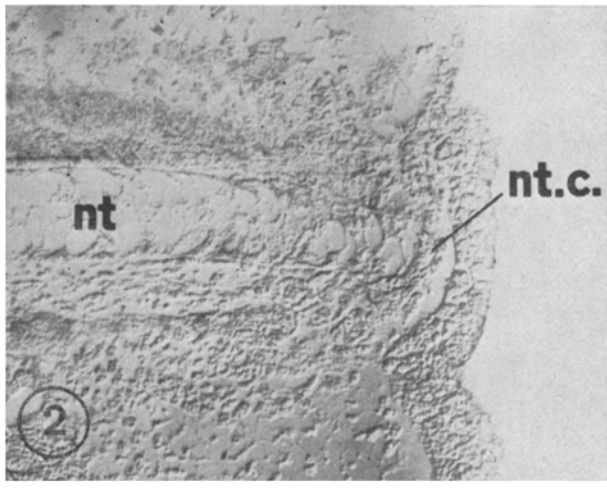

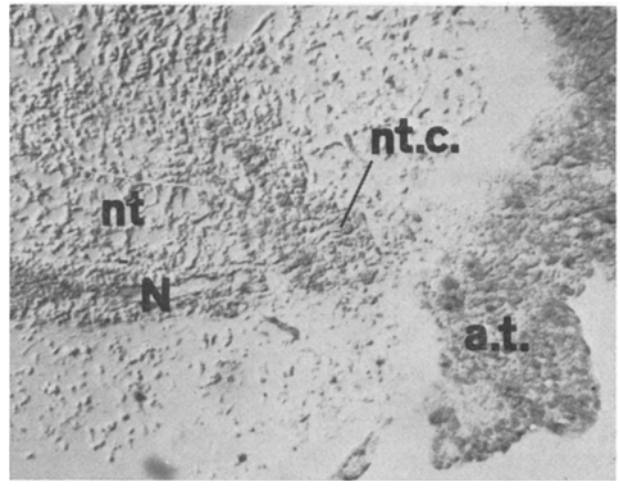

Figs. 2–4. All embryos were amputated as shown in Figure 1. After fixation and embedding, the sections were deparaffinized in xylene and immediately covered with Permount. The unstained material was then photographed on Plus X-Plan using a Zeiss, Nomarski interference contrast optical system. a. t., apical tuft; mb, myoblast-like cells; my, myotome; N, neural tube; nt, notochord; nt.c., notochordal cap. Left $2n$; right n.

Fig. 2. Wound healing is accompanied by the formation of a prominent apical tuft (day 1).

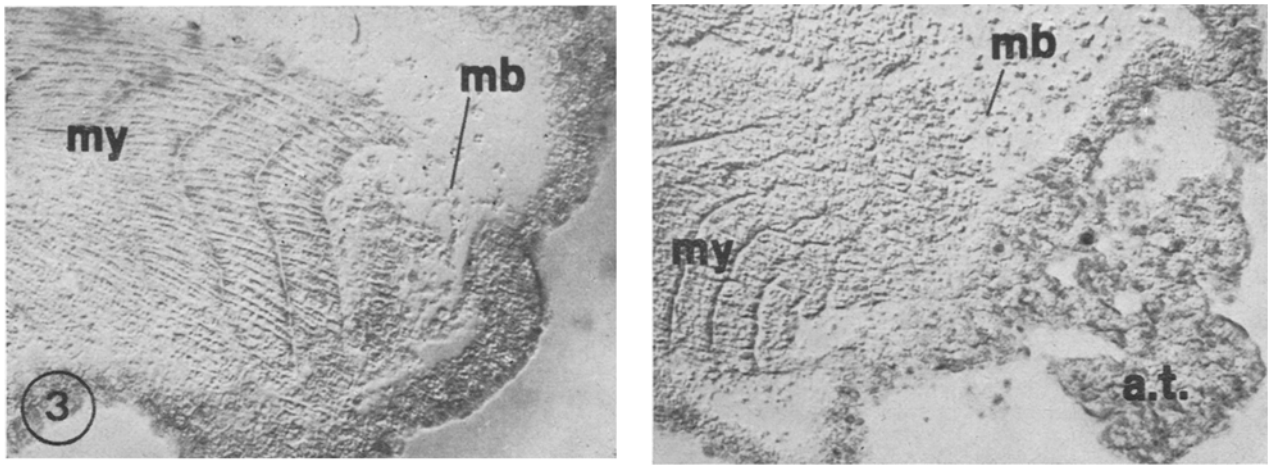

Fig. 3. Internal healing is also seen in the form of notochordal cap for instance (day 3).

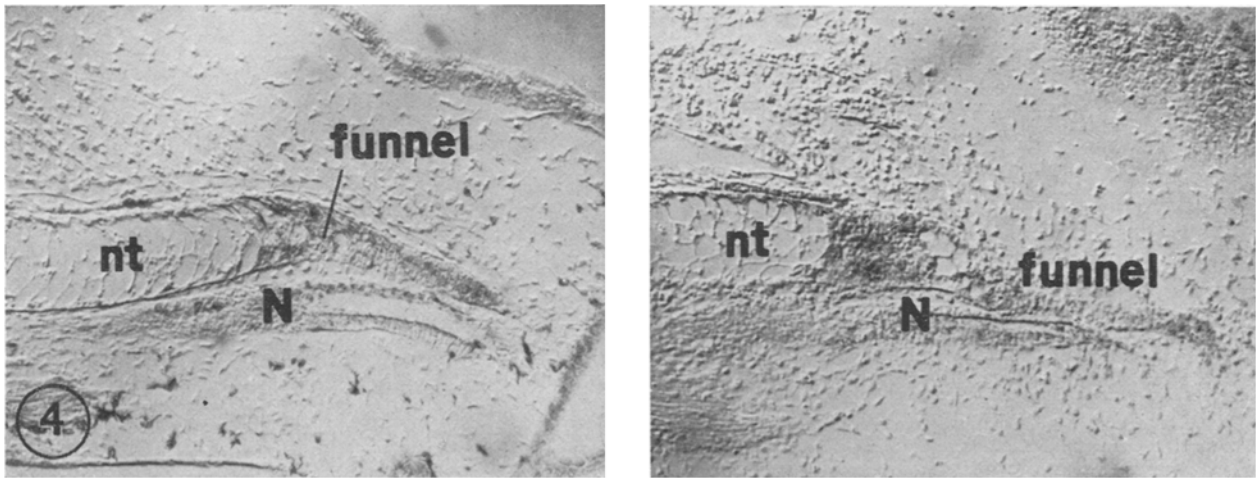

Fig. 4. Even myotomes disorganize into loose masses of myoblast-like cells (day 5).

logical functions are perhaps sequestered and left unaffected by the elimination of the paternal genome. One of these must then be a general propensity to grow and regenerate. The apparent normal regeneration in haploid tadpoles can be related to the significant finding that certain organelles (mitochondria and cilia) have retained the same dimensions in *n* as in *2n* cells of *Xenopus* larvae[7].

Admittedly, it is not possible to follow regeneration of haploid tails to its completion because of the precocious death of the tadpoles. But it is clear that much information regarding nucleocytoplasmic interactions in general and haploidy specifically will be gained by a continual appraisal of haploid function and dysfunction. In this manner it should become possible to catalogue and separate those functions which are affected by the haploid condition from those which are not as has already been done with respect to water regulation during haploidy[8].

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9,10-Dihydroergotamine: Production of Antibodies and Radioimmunoassay

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Summary. Antibodies against 9,10-dihydroergotamine (DHE) were produced by immunizing rabbits with a conjugate of 6-nor-6-carboxymethyl-9,10-dihydroergotamine and bovine serum albumin. A highly specific and sensitive radioimmunoassay for DHE has been developed.

Complete separation of antibody-bound hapten from free hapten is a prerequisite for an efficient radioimmunoassay. The successful use of the coated charcoal technique[2] allowed us to realize a simple but highly sensitive and specific test for an ergot alkaloid. Owing to the low plasma concentration of 9,10-dihydroergotamine (DHE) in the blood after an oral dose of 2.5 mg DHE mesylate, it is not possible to determine the concentration of the active compound in plasma samples by physico-chemical methods. For this reason, a radioimmunoassay has been