## Radiation-Induced Castration in the Female Teleostean Fish, Heteropneustes fossilis Bloch

In fishes, castration is frequently difficult to accomplish without high mortality. The use of X-rays has not yielded clear-cut results because the heavy doses necessary to destroy the interstitial tissue as well as the germinal cells bring about deleterious side effects. In the present communication we record our preliminary observations on radiocastration of a common Indian catfish, Heteropneustes fossilis Bloch.

Female fish were collected in February, when the ovary contains oocytes in the perinucleolar stage of development. 40 fish, each weighing  $13 \pm 2$  g, were injected i.p. with 5  $\mu$ c of Co<sup>60</sup>. The controls received stable CoCl<sub>2</sub> in distilled water equivalent to the same dose. Co<sup>60</sup> was received from the Atomic Energy Establishment, Trombay (India) in the form of CoCl<sub>2</sub>. The fish were fed normally during the experiment. There was no mortality and the fish recovered very well from the slight ill effects of radiation within a fortnight. This observation is based on the studies on the gastrointestinal tract and hematology taken up in this laboratory. In April, i.e. 2 months after the first injection, the fish were given another dose of 5  $\mu$ c of Co<sup>60</sup>. The fish were sacrificed in June, 4 months after

the first injection. The weight of the ovary of the irradiated fish was  $0.13\pm0.02$  g while, in the controls it weighed  $1.5\pm0.2$  g, the difference between the 2 being highly significant.

Histologically, the control ovary contained mature oocytes packed with yolk-granules, while in the irradiated ovary all the oocytes were still in the perinucleolar stage (Figures 1-4). Similar results have been obtained by us on Lebistes reticulatus after maintaining them in water containing Co<sup>60</sup> (unpublished). The significant result of irradiation was complete castration, which was brought about by the cessation of growth and further development of the oocytes. All the oocytes in the irradiated ovary showed degenerating histopathological features such as vacuolation of the cytoplasm and the nucleus, chromatolysis, disintegration of the nuclear membrane in most oocytes resulting in an endomixis of nuclear and cytoplasmic content, knocking off of the nuclei, change from the eosinophilic to basophilic nature of the nuclear content, and appearance of irregularly distributed yellow pigments in the ovary. We feel that total castration could be achieved even if 1 µc of Co60 were injected in-

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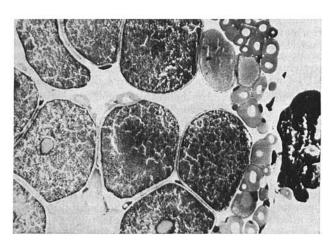


Fig. 1. Photomicrograph of transverse section (TS) of ovary (control) showing occytes in the yolk-granule stage. × 28.

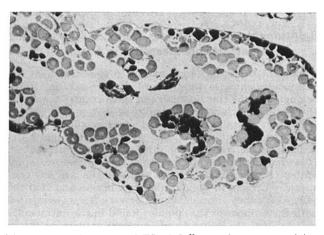


Fig. 3. Photomicrograph of TS of Co<sup>60</sup>-treated ovary containing occytes in the perinucleolar stage.  $\times$  28. (Compare with Figure 1.)

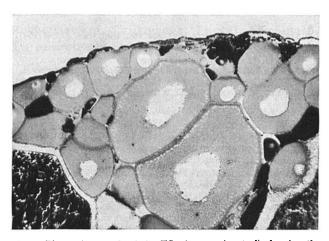


Fig. 2. Photomicrograph of the TS of ovary (control) showing the oocytes in the perinucleolar and yolk-vesicle stages. × 80.

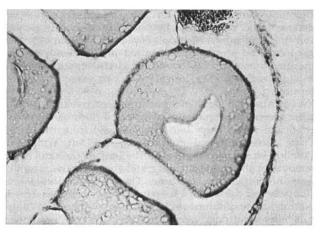


Fig. 4. Photomicrograph of TS of Co<sup>60</sup>-treated ovary with perinucleolar stage oocytes showing vacuolation of the cytoplasm and chromatolysis of the nucleus. × 320. (Compare with Figure 2.)

stead of 5  $\mu$ c. If necessary 3 injections could be given at regular intervals instead of 2.

It is reasonable to infer that irradiation disturbs the physiological activity of the pituitary gland, and decrease in the secretion of FSH would result in the cessation of the growth and development of the oocytes. A similar view has also been expressed by Egami et al.2 on the basis of their experiments on Oryzias latipes. VIVIEN<sup>8-5</sup> has been able to sterilize Xiphophorus and Lebistes by maintaining them in water containing P32, and has concluded that radioactive phosphorus first acted on the hypothalamic nerve centres and thus inhibited the pituitary. Golgi bodies and mitochondria are believed to be responsible for the origin of the yolk vesicles and yolk globules. It is probable that radiochemical changes in these organelles impair their activity, thereby inhibiting the development of the oocytes in the irradiated fish?.

Zusammenfassung. Indischen Katzenwels-Weibchen (Heteropneustes fossilis Bloch) wurden im Februar und

April 5  $\mu$ c Co<sup>60</sup> injiziert, was zur vollständigen Unfruchtbarkeit führte. Alle Fische erholten sich aber innerhalb von 14 Tagen auffallend gut von den Injektionsschäden.

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Radiation Biology Laboratory, University of Rajasthan, Jaipur (India), September 14, 1966.

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- Thanks are due to the Rockefeller Foundation and the Ford Foundation (USA) for financial assistance, to the Indian Council of Medical Research for the award of a research fellowship to one of us (S.K.R.), and to Prof. L. S. RAMASWAMI for his keen interest in the work.

## Relationship of Gasserian Cells to Extraocular Muscle Proprioception in Lambs

Afferent discharges in response to the stretch of the extraocular muscles were recorded from nervous fibres belonging to the third, fourth and sixth cranial nerves 1-4. It was claimed that the cell bodies of such proprioceptive fibres were contained either in the mesencephalic nucleus of the trigeminus nerve 5-7 or in the small ganglia described on the trunk of the same oculomotor nerves 8-10. However, arguments against both these views were presented. It was objected that such ganglia are not consistently present in the 3 oculomotor nerves and that the cells which they contain should be very few as compared with the number of the spindles found in the extraocular muscles. On the other hand, a clear-cut demonstration was given that the mesencephalic nuclei of the fifth cranial nerve are concerned with jaw muscle proprioception both in mammals and in birds 11-14. However, another possibility could be taken into account. Ocular afferents were described in the trigeminal nerve by STIBBE 10 and connections between ocular muscle nerves and the trigeminus were observed in Ungulata 15. The results of the present investigation provide evidence for the first time that cells contained in the semilunar ganglion of lambs are concerned with eye muscle proprioception.

21 lambs were employed in the present research. Under ether anaesthesia the left extrinsic eye muscles were isolated and the left eye ball was removed from the orbit. Then, under surgical anaesthesia, the left semilunar ganglion was gently exposed at the base of the skull after removal of the cerebral cortex and the operative wounds were infiltrated with procaine. The ether anesthesia was stopped, and the lambs were paralysed with Intocostrin T Squibb and maintained under artificial respiration. Tungsten microelectrodes were introduced into the semilunar ganglion by means of a microcontrol. The microelectrodes were connected through a Grass Hip 5A high-impedance probe with a Grass P5CR preamplifier. The upper beam

of a Tektronix 502 A oscilloscope and a loud-speaker monitored the inputs. The lower beam recorded the stretching of the eye muscles by means of a Basile MDI 4 microdynamometer. Films were taken by means of a Cossor type 1431 kymograph camera. The exact position of the recording microelectrode tip was ascertained in all the experiments by histological control. The following criteria, proposed by Darian-Smith, Mutton and Proctor 16, were followed in order to ensure that the microelectrode tip was located in the region of the pericaria of the semilunar ganglion: (1) negative polarity of the units, (2) the unitary activity could be recorded while advancing the microelectrode tip up to 250 microns, (3) the histological control showing that the electrolysis of the recorded site was within a cellular pool.

A cellular pool was found in the medial-dorsolateral part of the semilunar ganglion of all 21 lambs at a depth of about 1-1.5 mm, which contained units responding to

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