growth which becomes more marked in subsequent generations. The results are, however, suggestive and the problem deserves further investigations.

Somatic Maturity. Minor variations in the time of first appearance of hair and of opening of eyes and ears occurred in both operated and control groups, however the differences between the averages of the 2 groups were insignificant.

Sexual Maturity. As criterion of sexual maturity we used the ability of the males to beget and of the females to produce, young. No significant difference in this respect between the 2 groups was observed. The greatest difference in the dates of birth of the first litter in each group in the fifth generation was only 17 days. This would seem to disprove any retardation or acceleration of sexual function.

9526 P

Experimental Studies on Eggs and Miracidia of *Renifer aniarum* (Leidy, 1891) and *Dasymetra villicaeca* (Byrd, 1935).

J. HENRY WALKER. (Introduced by Ernest Carroll Faust.)

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Previous investigators of trematodes of the Subfamily Reniferinæ, to which the 2 species studied by the writer belong, have found the eggs fully developed when oviposited. In the present studies 2 groups of parasite-free *Physa halei* Lea (the snail host) were inoculated October 1 and October 6, 1936, respectively with freshly oviposited eggs of *Renifer aniarum* (Leidy, 1891), which no doubt had been naturally incubated in the uteri of the trematodes while in the mouth of the snake, *Natrix cyclopion* (Duméril and Bibron), and shed cercarial larvae of this fluke 36 days later. During the 30 days previous to the dates of inoculation the mean air temperature was 28°C. in the laboratory where the snakes were kept. Subsequently 3 groups of parasite-free *P. halei* were inoculated with eggs of *R. aniarum* November 4, 11, and 20, 1936, respectively, and one group of parasite-free *P. halei* was inoculated November 20, 1936, with eggs of *Dasymetra villicaeca* Byrd, 1935. Individuals from these 4 groups were later fixed at various time intervals after the dates of inoculation and were found on careful examination to be uninfected. Daily attempts were then made to hatch freshly ovi-

posited eggs in the digestive tract of the appropriate snail host, with negative results. During October, 1936, the mean air temperature was 22.5°C. and during November, 1936, 15.7°C.

The eggs of both species, embryonated 1-30 days at room temperature (17-20° C.) failed to mature. Experiments demonstrated that during the colder months it was necessary to incubate artificially the freshly oviposited eggs for a period of 6 days at a temperature of 25° to 30°C. to produce mature and usually viable eggs so that, after the ingestion of these eggs by the appropriate snail host, hatching would occur. Artificial incubation of eggs of both species was unnecessary after April 15, 1937.

Experiments showed that the daily average number of eggs extruded per worm was 89.21 and that the total capacity of the uterus was 2669 eggs; thus previous to oviposition the age of those eggs at the extreme distal end of the uterus must have been 29.9 days. Since 1868 eggs were found to be extruded during the first oviposition, the age of eggs at the distal end of the uterus on the second day must have been 8.97 days.

The eggs of trematodes with known life histories either (1) hatch in water and their miracidia actively penetrate the first intermediate host (usually a Gastropod) or (2) the eggs are ingested by the snail and hatching occurs in the digestive tract of the snail. *Fasciola hepatica*[1] is the classical example of the former method. A survey of the literature showed that probably 20 known species among the Families Opisthorchidæ, Heterophyidæ, Dicrocoeliidæ, Azygiidæ and Plagiorchidæ employ the latter method. All attempts to hatch the eggs of *R. aniarum* and those of *D. villicaeca* outside the digestive tract of the appropriate snail host were consistently negative. While many workers have seen living miracidia attempting penetration of the gut of a dissected snail there is only one record of a miracidium having been observed in a sectioned gut wall of an infected snail.[2]

In the present studies a large number of the appropriate snail hosts were killed and fixed at regular intervals after the ingestion of fully developed eggs of *R. aniarum* and *D. villicaeca.* A complete examination of stained serial sections has shown (1) hatched eggs in the crop-gizzard, stomach and mid-intestine of the snail; (2) free-swimming miracidia in the stomach and mid-intestine; (3) miracidia attempting to penetrate and depressing the epithelium of the stomach and intestine; (4) miracidia at various levels within the epithelium of the stomach and intestine; (5) miracidia between the epithelium

[1] Thomas, A. P., *Quart. J. Micros. Sc.*, 1883, **23**, 99.

[2] Ingles, L. G., *Univ. Cal. Pub. Zool.*, 1933, **19**, 143.

and the surrounding connective tissue of the stomach and intestine; (6) the transformation of the miracidia into the mother sporocysts; and (7) all stages up to the emergence of the cercariæ.

The consistent evidence, stage by stage, of the hatching, penetration and metamorphosis phenomena within the snail indicates for the first time the exact locations and methods by which the snail becomes infected with these *Reniferine trematodes*. Information is now available indicating the method by which those families of trematodes whose eggs do not hatch *in vitro* are able to infect appropriate snails and to proceed with development within those molluscs.

9527

Choline-Esterase Content of Normal and Denervated Submaxillary Gland of the Cat.*

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The heightened sensitivity of the cat's submaxillary gland to sympathetic stimulation, following degenerative section of the chorda tympani, appears to be due to some spontaneous activity of the decentralized parasympathetic supply, the phenomenon thus being analogous to the well-known "augmented secretion".[1] Histological examination of the unstimulated paralytic gland, indeed, reveals changes characteristic of activity in the demilune cells,[2] which are normally controlled by the sympathetic.[3] An attractive hypothesis is that the latter elements are sensitized by acetylcholine which is liberated elsewhere in the gland through the spontaneous activity of the postganglionic neurones, and which reaches the demilune cells by diffusion. After degenerative section of the chorda tympani, how-

* During the preparation of this paper for publication, a paper by F. Th. v. Brücke has appeared (*J. Physiol.*, 1937, **89**, 429) in which it is shown that section of the cervical sympathetic nerve in the cat greatly diminishes the choline-esterase content of the superior cervical ganglion. This case is clearly not analogous to that of the submaxillary gland, in which doubtless only a small part of the total esterase content has a special relation to the terminations of the (preganglionic) fibres which are cut.

1 MacIntosh, F. C., and Rawlinson, H. E., *Quart. J. Exp. Physiol.*, 1935, **25**, 199.

2 Rawlinson, H. E., *J. Anat.*, 1935, **70**, 143.

3 Rawlinson, H. E., *Anat. Rec.*, 1933, **57**, 289.