females may keep the last inseminated egg in the uterus. Such an egg begins the embryonic development within the mother and is generally laid as an 'overaged' egg during the 7 mm period. (2) Our experience has shown that females are too much disturbed by changing the dishes every 3 mm and egg-laying activity decreases rapidly after about 1 h. With the alternative schedule egg collection can be extended for 6 h or more. If for a particular experiment the extreme short periods are not needed, regular changes every 5 min (or longer) are possible. Some tests showed that the collection period could not be reduced to less than 3 mm without a drastic reduction in the number of eggs deposited.

To keep the disturbance of the flies to a minimum, the change of the dishes is done as follows: the egg-laying apparatus stands on a solid, vibration-free table. The lid of the wooden box is carefully lowered. An automatic switch (14) turns off the room light at the same time in order to prevent attraction of the flies by light. Now, in the dark, the spring is removed and the dish is gently rotated back and forth during about 10 sec. This causes the flies (which do not fly in the dark) to walk off the surface of the dish. Thereby they cross the ring of plaster which absorbs any fluid on the animals' legs. Thus neither the blotting paper nor the bell-glass ever get wet. A new dish is now fixed to the plate. Upon closing the lid of the box, the light in the room is turned on automatically.

The special temperature and humidity conditions are based on preliminary measurements of the dew-point temperature on the surface of a medium on which females have been laying eggs. Using these conditions in the whole experimental room has 2 advantages: (1) a dish brought into the apparatus has already the optimal temperature and (2) eggs collected from the apparatus remain under constant conditions.

Factors Influencing Rates of Tail Regeneration in the Lizard *Anolis carolinensis*

In an investigation of the somatotropic effects of certain hormones in the lizard *Anolis carolinensis*, tail regeneration was studied as one of a number of physiological variables related to growth. In spite of a rigidly controlled experimental regime and use of only males of restricted age and size, considerable individual variation in tail regeneration was found. Although such variations have been reported, they have not been studied in detail. There are conflicting reports regarding certain possible regulatory factors in lacertilian tail regeneration, especially the role of the vertebral autotomy plane, and there have been speculations on largely uninvestigated factors such as epidermal involvement. We attempted to elucidate the basis for individual variation in the regenerative response in *A. carolinensis* by examining these and other factors, especially temperature.

**Materials and methods.** In early September, 60 adult male *A. carolinensis* (average snout-vent length 64.5 mm, body weight 5.0 g) were put at 32 ± 0.5°C with 6 h light daily. Some animals were injected with gonadotropins, or gonadotropin plus prolactin, but there were no significant differences in tail regeneration and the data were pooled for this analysis. Procedures for hand-feeding, assessing growth and autopsy are reported elsewhere.

The original tail (average length 124 mm, range 111–138) was amputated with a razor blade 18–21 mm behind the vent: amputated portions averaged 360 mg. None of the animals appeared to have had previously regenerated tails except at the very tip. The length of the regenerating tissue was measured weekly and after 6 weeks the newly regenerated portion was removed and weighed.

The epidermal condition at amputation and the position of amputation relative to the natural autotomy plane (Figure 1) was determined from histological preparations of the proximal 1.5 cm of the amputated portion; methods are described elsewhere.

In order to facilitate comparison between our results and those of previous workers who have used temperatures around 18–22°C, a second experiment at 21°C was conducted with 18 males in April. Ad libitum feeding maintained or increased the animals' weights. Severals were transferred to higher temperatures as described below. The average growth rate for the 10th to 42nd day after amputation was 0.98 mm/day. The mean length of the amputated tail at the end of 6 weeks averaged 28.5 mm, representing 28% replacement of amputated tissue. Prominent

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scales and greatly reduced growth rates during the 6th week suggested that the regenerates were approaching their final length.

The regenerated tails averaged 109 mg (range 45–180 mg) at autopsy, an average replacement of 30%. The final length and weights of the regenerates were highly correlated ($r = 0.8, p < 0.001$). Short tails tended to be proportionally lighter (3 mg/mm) than the long ones (4.3 mg/mm). In general, individual differences in these final values reflected consistent differences in regeneration rates throughout the experimental period.

Despite the relatively uniform rate of blastema formation, rates and final extent of tail regeneration differed markedly among the animals studied. The following factors were tested against final length and weight of the regenerates: initial and final body weight, change in body weight (all gained weight but this ranged from 1–25%), change in body (snout-vent) length, final weight of the liver and abdominal fat bodies, testicular weight and condition, and thyroid epithelial height at autopsy. None of these variables were significantly correlated ($p > 0.10$) with tail regeneration.

There was no correlation between tail regeneration and the position of amputation with respect to the autotomy plane nor the original epidermal condition (Figure 3).

**Tail regeneration at 21°C.** At 21°C, the time for the first external signs of blastema formation to become evident averaged 36 days, after 35 days in 8 individuals and on the 28th, 39th, 42nd and 45th day in 4 others. This contrasts with an average of about 8 days at 32°C. There was only negligible tail growth during the 2 weeks following the appearance of the blastema. The regenerates grew to 4–7 mm within the next month, averaging 0.15 mm/day. Four animals had regenerated tails of lengths: 3, 11, 14 and 20 mm respectively after 6 months and no increase occurred during a further 4 months.

Three lizards were transferred from 21–32°C 14 days after amputation. Blastema formation was observed 6 days after the transfer. Thus, there was apparently little progress toward blastema formation in the first 2 weeks at the lower temperature. 5 animals kept initially at 21°C were transferred to 32°C after blastema formation. In the