

it would appear that adequate amounts of heparin are effective in preventing the accumulation of platelet masses on the injured surfaces of veins.

The opinion has been wide-spread, however, probably largely as a result of a publication of Shionoya, that while heparin prevents the clotting process it does not influence the deposition of platelets. Rowntree and Shionoya² studied the accumulation of platelets in an "extracorporeal loop." Using this technique, Shionoya³ reported that heparin had no influence upon the rate of formation of white thrombi.

We have repeated the experiments of Shionoya and find that in dogs when the observations are made over a two- or three-hour period, there is almost invariably formation of white thrombi in the glass cannulae and the collodion or Cellophane tube used to connect the arterial with the venous cannula. Masses of white thrombi, large enough to produce partial or complete obstruction of the tubes, have been observed in 15 of the 16 experiments in which no heparin was used. In experiments, however, in which one intravenous injection of a solution of purified heparin (450 units per kilogram) was made, we have never observed any obstruction of blood flow or the accumulation of any platelet masses.

Microscopic examination of the obstructing masses in the experiments in which heparin was not used revealed, in every case, the typical structure of the white thrombus. In all except two of the experiments in which heparin was used microscopic examination of the Cellophane loop revealed only a few red and white blood cells. In two cases *very minute patches* of material which appeared to be composed of platelets were seen. It may be stated, however, that in no case was any mass which had the characteristic structure of a thrombus observed in the experiments in which heparin was administered.

The results of these studies demonstrate that purified heparin is very effective in preventing the formation of white thrombi under the conditions of our experiments.

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LIGHT AND THE SEXUAL CYCLE OF GAME BIRDS

SINCE Rowan's¹ original observations that lengthening the period of light in winter by means of artificial

illumination caused stimulation of normally quiescent gonads in juncos, similar results have been found in other species of birds and some mammals.

As a result of night lighting of upland game birds, Petty² secured gonad stimulation and eggs in quail during the non-breeding season in 1934, Martin³ announced mid-winter production of eggs in pheasants (*Phasianus colchicus torquatus*) in 1935, Clark, Leonard and Bump⁴ secured gonad stimulation in pheasants, quail and grouse (*Bonasa umbellus*) and Bissonnette and Csech⁵ secured early eggs from pheasants and quail by night lighting.

We wish to report briefly some of our experiments concerning the factors regulating sexual activity in the pheasant and grouse.

(1) The absence of light inhibits the onset of sexual activity in the grouse and pheasant. Two pair of grouse were continuously in the dark, except for a low illumination of 0.02 foot candles over the feeder from February 10 to June 20, 1936. No sexual activity was manifested during this time, although the experiment extended over the period of sexual activity of the control birds. Two pair of pheasants under the same experimental conditions also failed to come into breeding.

(2) Continuous illumination during the winter months can stimulate grouse into sexual activity and egg laying but does not prevent the cessation of egg laying. A pair of grouse were illuminated continuously, starting on February 10, by two 150 watt lamps. No daylight was admitted and the light intensity was maintained constantly at 22 foot candles. Egg laying began on March 6 and continued to April 3, yielding 14 eggs, the normal number for grouse. None of the control grouse laid before April 10. The experimental birds ceased laying in spite of the continued light. We were able to secure egg production prematurely in pheasants and quail with night light, but did not carry the experiment to the end of the normal breeding season.

(3) The cessation of egg laying is probably due to a failure of the hypophysis to furnish the necessary gonad stimulating hormones rather than to an exhaustion of the gonads. On June 23, 1936, two pair of grouse which had completed their reproductive cycle were autopsied, and it was found that the sex organs had regressed to approximately the mid-winter condition. The testes of the males weighed 41.4 and 41.6

² *American Field*, August 11, 1934.

³ L. E. Martin, *Game Breeder and Sportsman*, 39: 95, April, 1935.

⁴ L. B. Clark, S. L. Leonard and G. Bump, *SCIENCE*, 83: 2150, 268, March 13, 1936.

⁵ T. H. Bissonnette and A. G. Csech, *SCIENCE*, 83: 2156, 392, April 24, 1936.

² L. G. Rowntree and T. Shionoya, *Jour. Exper. Med.*, 46: 7, 1927.

³ T. Shionoya, *Jour. Exper. Med.*, 46: 19, 1927.

¹ W. Rowan, *Nature* (London), 115: 494-495, 1925.

mgs, as compared to 27.3 mg for the testes of a male autopsied in January of the same year. The ovaries weighed 126.8 and 110.3 mgs, as compared to 56.7 mg for the ovaries of a bird killed in mid-winter. The injections of 5 to 10 grams equivalent of an alkaline hypophyseal⁶ extract in 3 male grouse for 10 days resulted in testes weighing 238.2, 206.4 and 179.4 mgs, an increase of approximately five times.

Similar results were found in the pheasants. The testes of 3 male pheasants autopsied on August 3, 1936, three weeks after the close of the breeding season, weighed 367.8, 617.1 and 612.5 mgs. The testes of birds in the winter weighed from 69.7 to 164 mgs, while in full sexual activity they weighed over 6 grams. Three male pheasants, each receiving a pyridine extract of sheep hypophysis equivalent to 5 grams of dried glands for a period of 10 days, yielded testes weighing 1,232, 2,209 and 3,001 mgs, as compared to 907 and 1,052 mgs for two uninjected controls. The difference in weight of the testes of the controls and those autopsied August 3 is due to the fact that they belong to different age groups. The average increase was 119 per cent.

In both grouse and pheasant, histological examination of the testes confirmed the gross observations. In all injected birds, the testes showed mitotic activity, an increase in the number of cells in the germinal epithelium and an enlargement of the tubules. Normal sperm were found in the largest testes of the injected pheasant; none were found in the controls. The control testes of both pheasant and grouse gave evidence of degenerating to the resting condition, the grouse testis having regressed further.

(4) The gonads of immature pheasants will respond to adequate hypophyseal stimulation. A group of 8-week-old male pheasants were given a similar amount of the sheep hypophyseal extract for the same period as the adult pheasants. The testes of the young injected birds weighed 197.0, 143.3 and 93.2 mgs, and the testes of the controls weighed 61.2, 45.2 and 67.8 mgs, an average increase of 148 per cent. Microscopically, the stimulated testes were easily distinguished by the enlargement of the tubules and the increased number of division figures in the germinal epithelium.

SUMMARY

The results of these experiments has led us to conclude that light is a primary factor in inducing sexual activity in pheasants, quail and grouse, through the mediation of the hypophysis. Once this hypophyseal-gonad reaction is started it can not continue indefinitely with adequate light but only until the hypophysis falters in the production of the gonad-stimulating hormone through causes unknown. The immature pheasant resembles the adult at the close of the breeding season in that the gonads are capable of responding to adequate hypophyseal stimulation.⁷

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

THE PHOTOELECTRIC DETERMINATION OF PHOSPHORUS IN ESTUARINE WATERS

In estuarine waters there is much variation in salinity and heavy metal content, both of which factors may interfere with phosphorus determination, when the Denigès method is followed. Errors due to such factors must be taken into consideration. Also, the blue color of the reduced phospho-molybdate, as produced in the Denigès method, may be of improper magnitude (intensity and quality) for comparison with a standard blue solution by means of an ordinary comparison colorimeter. Accordingly, a photoelectric colorimeter was devised to study the color production.

A Weston photometer measuring light intensity in foot candles, a ruby glass filter, a pyrex tube 30 cm long closed at both ends to hold the solution and a

Spencer microscope lamp served as the photoelectric colorimeter. The solution to be tested, which was blue in color after being treated according to the Denigès method, was put into the 30 cm pyrex tube, in the ends of which plane glass pyrex windows were fused. An opening left on the side of the tube served for filling and emptying. By placing such a cylinder, filled with the blue solution to be tested, in the path of the beam of light, the absorption of the light by the solution expressed in terms of change in light intensity would be measured by the galvanometer of a light-sensitive instrument, such as the Weston photometer, at the other end. The ruby glass filter which transmitted only a narrow band of red (an average wavelength of 6,500 Å) was used because in the region of the band mentioned there was a maximum change in

⁶ Acknowledgment is made to Parke Davis Company for their kindness in furnishing the hypophyseal glands.

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