EXPERIMENTAL MODIFICATION OF THE SEXUAL CYCLE OF THE GREENFINCH

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(With Ten Text-figures)

INTRODUCTION

It is generally assumed that in birds of the northern hemisphere with a brief breeding season sexual maturity is directly related to the seasons. Consequently many attempts have been made to determine the factors which influence the gonads of these birds in spring, and to elucidate the mechanism involved.

Though it has been known in the poultry world for over a century that light has a great influence on reproduction—(artificial light has a stimulating effect on the egg production of hens (Baker & Ranson, 1932) so that an extended length of illumination in autumn protracts the laying period)—Rowan (1926, 1929, 1932, 1938) was the first investigator to demonstrate experimentally the great effect of light on reproductive activities. His experiments were designed to ascertain the relationship between the strong recrudescence of the gonads of migratory birds in spring and the impulse that induces birds to travel north at that time of the year. In this respect Rowan failed, for it has been shown by different authors, e.g. Putzig (1937), Hann (1939) and even by Rowan himself (1932, p. 645) that castrated migrants are able to migrate over long distances.

Starting from the fact that the increase in size of the gonads of migratory birds, the increase in daylight in spring and the northward migration of these birds run parallel, Rowan (1929) submitted juncos (Junco hyemalis, a North-American migratory song-bird, belonging to the finches) during midwinter, when their gonads are minute, to increasing amounts of light by means of artificial illumination after sunset. Consequently, the length of day for his experimental birds was increasing in the autumn instead of shortening. Thus, Rowan was able to induce in his birds a rate of gonad recrudescence, exactly comparable to that taking place in spring in control birds. Besides this, his male birds, thus brought into breeding condition, were constantly singing at the end of December, at temperatures between −20° and −45°C.; low temperatures have, therefore, no influence on the display of singing.

Post-mortem investigations revealed that the testes of the male juncos had increased enormously in size and that spermatogenesis was at its height. The ovaries of the female juncos had enlarged also under these experimental conditions, but by no means as much as the testes; full-grown follicles did not develop, the diameter of the largest ones measuring only 1.4 mm.
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Since Rowan's first detailed publication (1929) his work has been repeated and extended by himself and by many others, among whom Bissonnette and his co-workers especially must be mentioned here. Numerous species of birds and mammals were subjected to increasing dosage of daily illumination, of light of different wavelengths and of different intensities, and thus brought into oestrus or breeding condition.

Benoit (1935), Ivanova (1935), Ringoen & Kirschbaum (1937) and others experimented with birds which were completely draped in light-proof material or wore silk caps over their heads or had silk caps with slits admitting light freely to the eyes; moreover Benoit (1937) used ducks whose optic nerves were severed or whose eyes were blinded or even removed completely. As in all these cases the testes were activated, it is generally accepted that at least in birds light is the primary factor, causing gonad recrudescence in spring and that the pituitary functions as the intermediary organ between the light impulse and the activation of the gonads. The light stimulus reaches the hypophysis by way of the hypothalamus and activates this endocrine gland to secrete its gonadotrophic hormone, under the influence of which the gonads increase in size and, in case of the testis, sperm formation starts.

At one time in Holland enormous numbers of song-birds were caught in autumn by a traditional method which is sometimes used to-day by local bird-catchers. The song-birds are caught by the use of live male birds as decoys. As it is of primary importance that these decoys should be in full song in autumn, they are gradually put in the dark at the beginning of May and little by little exposed to light again in August, with the result that during September and October they are in full song and then are exceptionally suitable as decoys. This treatment of birds is called to 'muiten' birds; in the course of May the decoy birds are put into the 'muit' and in August they come out of it. This whole process of ‘mewing’ was elaborately described by Hoos (1937). Moreover, in Japan, the practice of ‘yogai’ has been in vogue, which consists in exposing cage-birds to artificial illumination towards the close of the year in order to bring them into singing condition in January.

In connexion with the work of Rowan and others we studied the process of mewing under laboratory conditions, investigation of the state of the gonads during and after mewing being our main concern. In addition we tried to bring these mewed birds again into spring condition by administering additional light during the winter months.

As experimental bird the greenfinch (Chloris chloris) was chosen, a hardy species, which remained in good condition throughout the duration of the experiments.

This investigation was suggested by Prof. G. J. van Oordt and carried out under his direction; a preliminary communication about it has already appeared (van Oordt

* Prof. A. E. H. Swaen has informed me that the English equivalent for the Dutch ‘muiten’ is to ‘mew’, i.e. to put a bird into the mew, which was a cage used for hawks, particularly while they were molting. The word ‘mew’ will be used in this paper to describe the process of bringing encaged birds gradually into the dark, keeping them in the dark for some time and gradually exposing them to daylight again.
P. H. Damsté & Damsté, 1939). As a thesis (in Dutch) for the degree of Ph.D. it has been presented in detail to the Faculty of Science of the University of Utrecht.

MATERIAL AND METHODS

Preliminary investigations were begun in the autumn of 1937, when twenty-four greenfinches (13 ♂♂ and 11 ♀♀) were kept in a large aviary, standing in a laboratory room facing south. About half of these birds, which acted as controls, were sacrificed during the winter and spring months in order to investigate their gonads histologically. In the middle of May 1938 six male and six female greenfinches were confined, each in a cage of 28 x 23 x 19 cm. These cages were put into a cupboard, provided with two doors. Tests with a photographic plate showed that when the doors were closed no light entered. Ventilation was arranged through a bent funnel, placed on top of the cupboard.

Before the encaged birds were mewed, they were left in the cupboard with open doors for about a week. During this period they accustomed themselves completely to perches, food-tray and water-bottle. Then mewing was started: the doors were gradually closed, and on 25 May the birds were in complete darkness. Once a week every cage was cleaned and new food and fresh water were given. On 30 July the doors of the cupboard were gradually opened again and on 12 August the birds were once more exposed to full daylight. At regular periods both controls and experimental birds were sacrificed; their gonads were fixed in Bouin’s fluid, cut in sections of 5 or 10 μ and stained with haemalum-eosin.

The preliminary investigations had a positive result. When the experimental birds were in the dark their gonads were small and they did not sing, but after they had been exposed to light again they were found to be in breeding condition once more. The experiments were therefore carried out on a larger scale, and as it appeared that for comparative investigations the oval testes were much better suited than the irregularly shaped ovaries, only male birds were used.

In the summers of 1939 and 1940 instead of the cupboard I used a room which could be darkened by means of shutters.

In 1939 the light was decreased from 19 May onwards and on 31 May the birds were in darkness. On 14 August the shutters were reopened and the birds were gradually exposed to daylight; on 21 August they were in full daylight again.

In 1940 fourteen male birds were mewed from 21 May onwards; they were in complete darkness between 30 May and 10 August; on 24 August mewing was finished.

In these three years forty-four male birds in all were mewed.

The quantity of light to which the birds were subjected was measured by means of a luxmeter kindly placed at our disposal by the Physical Laboratory of the University of Utrecht. On 20 May 1939, just before mewing, the quantity of light, measured at a distance of 150 cm. from the window of the room facing north in which the birds were kept, amounted to 900 Lux.

Generally the birds withstood mewing well, but there were some casualties at the beginning of our experiments. In 1939 and 1940 we therefore resorted to the
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practice of the bird-catchers who give their birds a very small amount of light for
1 hr. per day: we subjected them to a quantity of only 0.006 Lux.

Some of the birds which had been mewed were subjected to further experiments: from
the beginning of December four of them received increasing daily amounts of
light by means of artificial illumination. The birds were put in a large cage in a room
at the south-west side of our laboratory, which was darkened every day by means of
curtains from 4.30 p.m. till 8 a.m. (M.E.T.). When the weather was dull, a daylight
lamp of 145 W. was kept burning just over the cage; in bright weather the birds
received only daylight through a large window from 8 a.m. till 4.30 p.m. From
3 December until 7 January the birds were subjected every day to an increased
amount of artificial light, thus having their day lengthened instead of shortened.
Owing to technical difficulties this extra illumination was given only in the evening.
For instance the birds received:

- On 3 December 1940, additional light from 4.30 till 6 p.m.
- On 4 December 1940, additional light from 4.30 till 6.10 p.m.
- On 5 December 1940, additional light from 4.30 till 6.20 p.m.
- On 7 January 1941, additional light from 4.30 till 11 p.m.

From 7 January till 13 February 1941, the same duration of light was maintained.

THE NORMAL REPRODUCTIVE CYCLE

(a) The testis

It is a well-known fact that the gonads of birds breeding in the temperate and arctic
zones are much larger in the breeding season than during the rest of the year.
According to Stieve (1921) the testis of the jackdaw (Coloeus monedula) in the
breeding condition has a volume which is about 1500 x the volume of the quiescent
testis; in the house-sparrow (Passer domesticus) this factor is about 1100. The
minimum size of the testis of the greenfinch is about 0.5 mm. in diameter.

Under laboratory conditions the diameter of the November-December testis of
the greenfinch is about 1 mm.; in the breeding season this diameter increases to
8 mm. and more. This considerable enlargement is due partly to the enormous
increase in diameter of the testis tubules and partly perhaps to an increased content
of fluid, whereas the intertubular spaces are much narrower in spring than in the
winter months.

It was found (Table 1) that under laboratory conditions testis activity starts in the
first half of February and that the first sperms are formed in the tubules towards the
end of March. Sperm formation continues till June; then testis regression begins
rather quickly till, in the second half of July, the stage is reached which I have
termed for the sake of convenience the quiescent stage and which lasts till the
first half of February.

In Table 1 the average diameter of twenty testis tubules is given. From this it
follows that the increase in diameter of these tubules coincides with the increase in
size of the testis. In Fig. 1 a graph is given, showing the size changes in testes
during the course of a year; its maximum occurs during the first half of May.
Table 1. Showing details of testes of control birds

<table>
<thead>
<tr>
<th>No.</th>
<th>Date of autopsy</th>
<th>Average diameter of testis in mm.</th>
<th>Average diameter of testis tubules in μ</th>
<th>Histological details of testes</th>
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<tbody>
<tr>
<td>C1</td>
<td>5 Jan. 1938</td>
<td>1.3</td>
<td>65</td>
<td>Quiescent stage</td>
</tr>
<tr>
<td>C2</td>
<td>11 Feb. 1939</td>
<td>1.2</td>
<td>110</td>
<td>First beginning of spermatogenesis</td>
</tr>
<tr>
<td>C3</td>
<td>11 Feb. 1939</td>
<td>0.8</td>
<td>60</td>
<td>Quiescent stage</td>
</tr>
<tr>
<td>C4</td>
<td>25 Feb. 1938</td>
<td>2.2</td>
<td>135</td>
<td>Progressive intermed. stage</td>
</tr>
<tr>
<td>C5</td>
<td>10 Mar. 1938</td>
<td>4.7</td>
<td>195</td>
<td>Progressive intermed. stage</td>
</tr>
<tr>
<td>C6</td>
<td>29 Mar. 1938</td>
<td>7.4</td>
<td>355</td>
<td>Complete breeding condition</td>
</tr>
<tr>
<td>C7</td>
<td>31 Mar. 1939</td>
<td>4.4</td>
<td>230</td>
<td>Almost complete breeding condition</td>
</tr>
<tr>
<td>C8</td>
<td>1 May 1939</td>
<td>8.2</td>
<td>340</td>
<td>Complete breeding condition</td>
</tr>
<tr>
<td>C9</td>
<td>19 May 1939</td>
<td>7.6</td>
<td>285</td>
<td>Complete breeding condition</td>
</tr>
<tr>
<td>C10</td>
<td>19 May 1939</td>
<td>6.3</td>
<td>345</td>
<td>Complete breeding condition</td>
</tr>
<tr>
<td>C11</td>
<td>2 June 1939</td>
<td>6.2</td>
<td>410</td>
<td>Complete breeding condition</td>
</tr>
<tr>
<td>C12</td>
<td>7 July 1939</td>
<td>7.1</td>
<td>245</td>
<td>Beginning of regression</td>
</tr>
<tr>
<td>C13</td>
<td>17 July 1939</td>
<td>1.3</td>
<td>70</td>
<td>Almost quiescent stage; much pigment in intertubular tissue</td>
</tr>
<tr>
<td>C14</td>
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<td>1.7</td>
<td>80</td>
<td>Almost quiescent stage; much pigment in intertubular tissue</td>
</tr>
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<td>28 July 1939</td>
<td>1.1</td>
<td>70</td>
<td>Quiescent stage; intertubular spaces still narrow</td>
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<tr>
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<td>1.1</td>
<td>60</td>
<td>Quiescent stage</td>
</tr>
<tr>
<td>C17</td>
<td>16 Aug. 1940</td>
<td>2.1</td>
<td>125</td>
<td>Regression stage; intertubular spaces rather narrow</td>
</tr>
<tr>
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<td>24 Aug. 1940</td>
<td>1.2</td>
<td>65</td>
<td>Quiescent stage</td>
</tr>
<tr>
<td>C19</td>
<td>19 Oct. 1939</td>
<td>0.5</td>
<td>45</td>
<td>Quiescent stage; much pigment in intertubular tissue</td>
</tr>
<tr>
<td>C20</td>
<td>10 Nov. 1937</td>
<td>1.0</td>
<td>35</td>
<td>Quiescent stage</td>
</tr>
<tr>
<td>C21</td>
<td>8 Dec. 1937</td>
<td>1.1</td>
<td>55</td>
<td>Quiescent stage</td>
</tr>
</tbody>
</table>

Fig. 1. Graph, showing size changes in control testes in the course of a year. Crosses indicate testes from 1937-38; black dots testes from 1939 and circles testes from 1940.

Approximately the same graph results when the testis tubule diameter is taken into consideration (Fig. 2); its maximum occurs during May.

It is not necessary to give here a full histological description of the gonads of our birds during the course of a year, as an account of the spermatogenesis and a detailed histology of the testis of the greenfinch and of other song-birds has been given.
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elsewhere (e.g. Rowan (1929) for the junco, Bissonnette (1930) and Bissonnette & Chapnick (1930) for the starling). Therefore, only schematic drawings, showing at the same magnification sections of testes during the quiescent stage, the intermediate and the reproductive stage, will be given here.

In Fig. 3 a section of a winter testis of a greenfinch, killed 5 January 1938, is drawn. The section in Fig. 4 represents a testis in the progressive intermediate stage of a greenfinch, killed on 10 March 1938. And finally a cross-section of some tubules of a mature testis belonging to a bird killed 2 June 1939, is shown in Fig. 5.

In these figures the relative decrease in size of the intertubular spaces and the enormous increase in diameter of the testis tubules are distinct. Later in the season the quiescent testis stage is approached quickly, in 2 or 3 weeks.

(b) Song

The male greenfinch has a pretty song, a warbling twitter, in which its characteristic note, a long drawn ‘dwee’ is often repeated. The song is mostly uttered from a perch, and, in nature, also in flight. In confinement the call note, a short twitter, is regularly heard throughout the year; in early spring the typical ‘dwee’ is heard first and is soon followed, in March and later, by the true song.
In the laboratory the male control birds, which had started singing from the beginning of March, stopped singing in June.

Fig. 4. Contours of testis tubules of a normal greenfinch in the progressive stage. C5, killed 10 March 1938. ×45.

Fig. 5. Contours of testis tubules of a normal greenfinch in breeding condition. C11, killed 2 June 1939. ×45.

(c) Moult

The adult greenfinch undergoes a complete moult in nature between the end of July and the beginning of September. In spring there is no moult; but by abrasion of the tips of the feathers the upper parts of the male bird become greener and its under parts brighter yellowish green, while its greater wing coverts and inner
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secondary flight feathers become greyer and less brown by the same process. The same changes in respect of moult, colour and time of the year were noted under laboratory conditions.

THE EXPERIMENTS

As already mentioned greenfinches were in complete darkness in 1938 from 25 May till 30 July, in 1939 from 31 May till 4 August and in 1940 from 30 May till 10 August. Table 2 gives details of the testes of these experimental birds.

(a) The testis

The data of Table 2 show that under mewing conditions the testes have involuted completely in 2½–3 weeks (experimental birds E5 and E7) and that they are quiescent during the second half of June and during July. Consequently, the mewed testis is in every way comparable to the winter testis. On the other hand the recrudescence of the testes, induced by light after mewing, is almost as fast as the experimental regression; in experimental birds, which were gradually brought into light in the beginning of August, spermatogenesis is distinct after 1½–2 weeks (birds E24, E25 and E26), whereas sperm formation is complete after 3 weeks (E30 and E32). In this condition the birds remained till about the beginning of October. Then, either owing to the gradually decreasing daylight in autumn or to other inhibiting factors which cause the natural regression of the testis, the gonads diminish in size and towards the end of November or the beginning of December the testes of the experimental birds have reached their winter minimum once more.

Moreover, we have tried to induce a renewed testis recrudescence in these mewed birds. As already mentioned (p. 23) four of our previously mewed birds were subjected in the course of December 1940 and January 1941 to regularly increasing amounts of artificial light, with the result that by the end of December 1940 and in January 1941 the testes of these birds were in full breeding condition, i.e. they had again increased enormously in size and contained large quantities of sperm.

These experiments were concluded at the end of February. It may be accepted that from that time onwards the testes of these birds have shown a renewed regression, possibly due to the relatively shorter days or to some inhibitory influence of the hypophysis. As a matter of fact, at the end of February 1941, the testes of one bird, which had been submitted from 7 January 1941 onwards to artificial light from 4.30 till 11 p.m. and to daylight from about 7.15 a.m. until 4.30 p.m., still showed breeding condition, but at the end of March when only one experimental bird was available, this bird (E44) possessed small testes which were in distinct regression. On that date the bird was exposed to light for about 16 hr.

In Fig. 6 a graph is given, showing the size-changes in the testes of our experimental birds. For the sake of clearness the graph of Fig. 1 is repeated in Fig. 6. In full darkness the testes of the experimental birds are at their minimum, but they enlarge enormously under the influence of daylight during August; in September and the beginning of October they are at their maximum. In November a second minimum is reached, but towards the end of December the testes regain their maximum size under the influence of a newly administered, daily increasing, amount of artificial light.
<table>
<thead>
<tr>
<th>No.</th>
<th>Date of autopsy</th>
<th>No. of days after beginning of light decrease</th>
<th>No. of days in total darkness</th>
<th>No. of days since beginning of light increase</th>
<th>No. of days since end of mewing</th>
<th>Average diameter of testes in mm.</th>
<th>Average diameter of 20 testis tubules in μ</th>
<th>Histological details</th>
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<tr>
<td>E1</td>
<td>19 May 1939</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1-3</td>
<td>260</td>
<td>Breeding condition</td>
</tr>
<tr>
<td>E2</td>
<td>21 May 1940</td>
<td>7</td>
<td>5</td>
<td>7</td>
<td>2</td>
<td>1-3</td>
<td>260</td>
<td>Breeding condition</td>
</tr>
<tr>
<td>E3</td>
<td>26 May 1939</td>
<td>14</td>
<td>2</td>
<td>14</td>
<td>2</td>
<td>6-2</td>
<td>260</td>
<td>Breeding condition</td>
</tr>
<tr>
<td>E4</td>
<td>2 June 1939</td>
<td>21</td>
<td>9</td>
<td>21</td>
<td>9</td>
<td>6-2</td>
<td>260</td>
<td>Breeding condition</td>
</tr>
<tr>
<td>E5</td>
<td>4 June 1940</td>
<td>14</td>
<td>5</td>
<td>14</td>
<td>5</td>
<td>5-1</td>
<td>440</td>
<td>Beginning of regression</td>
</tr>
<tr>
<td>E6</td>
<td>9 June 1939</td>
<td>14</td>
<td>2</td>
<td>14</td>
<td>2</td>
<td>5-2</td>
<td>355</td>
<td>Breeding condition</td>
</tr>
<tr>
<td>E7</td>
<td>15 June 1938</td>
<td>28</td>
<td>16</td>
<td>28</td>
<td>16</td>
<td>6-6</td>
<td>440</td>
<td>Quiescent stage</td>
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<td>E8</td>
<td>21 June 1939</td>
<td>29</td>
<td>17</td>
<td>29</td>
<td>17</td>
<td>5-5</td>
<td>290</td>
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<td>29</td>
<td>17</td>
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<td>17</td>
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<td>E10</td>
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<td>28</td>
<td>28</td>
<td>28</td>
<td>28</td>
<td>1-9</td>
<td>100</td>
<td>Almost quiescent stage</td>
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<td>28 June 1939</td>
<td>28</td>
<td>28</td>
<td>28</td>
<td>28</td>
<td>2-4</td>
<td>140</td>
<td>Almost quiescent stage</td>
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<td>28 June 1939</td>
<td>28</td>
<td>28</td>
<td>28</td>
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<td>33</td>
<td>45</td>
<td>33</td>
<td>2-2</td>
<td>230</td>
<td>Strong regression</td>
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<td>37</td>
<td>49</td>
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<td>57</td>
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<td>51</td>
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<td>61</td>
<td>1-2</td>
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<td>Strong regression</td>
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<td>69</td>
<td>81</td>
<td>69</td>
<td>1-2</td>
<td>60</td>
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<td>105</td>
<td>Almost quiescent stage</td>
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<td>12 Aug. 1938</td>
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<td>85</td>
<td>93</td>
<td>85</td>
<td>1-6</td>
<td>340</td>
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<td>85</td>
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<td>85</td>
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Experimental modification of the sexual cycle of the greenfinch

In the experimental birds the testis tubule diameter runs parallel to the total testis size, which is clearly perceptible in Fig. 7, showing a graph of the different testis tubule diameters throughout the experimental period.

As it may be accepted that these birds were also in breeding condition in the spring of the same year, it is obvious that the experimental birds were mature three times within a year: first, in April and May before they were mewed; secondly in August and September after having been mewed; and thirdly, in December and January when their testes underwent a third recrudescence under the influence of administered artificial light.

I think it superfluous to reproduce here many sections of the testes of the experimental birds with histological details, as the latter are exactly the same as in the
control testes. The experimental testes are normal in all respects, except as to dates. Fig. 8 shows a part of a section of a testis in regression; it belonged to a greenfinch which had been in total darkness for only 5 days. Fig. 9 represents a drawing of the section of a testis of a greenfinch in the quiescent stage during mewing. In Fig. 10 histological details of the same testis are shown.

Fig. 8. Contours of regressive testis tubules of an experimental greenfinch during mewing. E 5, killed 4 June 1940. × 45.

Fig. 9. Contours of testis tubules of an experimental greenfinch during mewing; quiescent stage. E 7, killed 15 June 1938. × 45.

(b) Song

Very soon after the birds were exposed to a decreased quantity of light they ceased singing; only the characteristic 'dwee' and an occasional twitter were heard, but as soon as the birds were kept in continual darkness they were quite silent. With the regression of the testes the song had come to an end.
Fig. 10. Histological details of part of the section of the testis shown in Fig. 9. E 7, killed 15 June 1938. × 600.
When at the end of the mewing period light was once again administered, the
twitter was heard again, soon followed by the ‘dwee’-call and at the end of August
and in September the experimental birds were in full song. The same thing was
observed in birds brought into breeding condition by artificial light in December.
Therefore recrudescence of the testes coincides with initiation of singing; when the
testes are in breeding condition the song is also at its height.

(c) Moult

It was observed that during mewing the birds were not only moulting, but also
that moulting and refeathering started much earlier than ordinarily. As mentioned
on p. 26 the summer moult and refeathering take place under normal conditions
in August. In our experimental birds, on the contrary, moulting and refeathering
started as early as the first half of June when they were in utter darkness. Then
many coverts were shed and shortly after not only the flight- and tail-feathers but
also the down followed. In the experimental birds moulting was at its maximum
at the end of June. Then the feathers reappeared gradually and by the end of July
refeathering was completed in most of the birds. Therefore, it is possible that a
relation exists between the effect of darkness and the processes of moulting and
refeathering. However, from our observations, it could not be ascertained whether
this precocious moult might not be due to some other cause, e.g. the state of the
gonads or the state of the thyroids, which, as one knows, play an important part in
the process of moulting. We shall consider this subject in the discussion (p. 34).

DISCUSSION

From the above it follows that as a consequence of mewing the testes of the experi-
mental birds were minute and that after mewing the birds developed testes with
large quantities of sperm and were in full song. Moreover, some of these birds,
having been exposed to increasing quantities of (electric) light towards the close of
the year develop fertile testes and at the same time come again into full song.
So it was possible by means of mewing and ‘extra lighting’, within the course of
a year, to bring greenfinches twice into breeding condition after the normal breeding
season in April and May.

In the light of Rowan’s and Bissonnette’s investigations it is easy to understand
that after an involution stage, the result of bringing the birds into darkness, their
testes enlarge rapidly till, under the influence of the increasing daylight, they are
fully mature after some weeks of increasing spermatogenesis. Then, in August, the
birds are in full song. So it is likely that song coincides with sexual maturity and is
induced by the male sex hormone, this being in agreement with the experiments of
Leonard (1939) and of Frederiks (1941) who were able to induce singing by means
of androgen administration in female canaries, and with the fact that castrated male
birds do not sing.

Most probably, due to the shortening days in October, testis regression starts
again in the course of this month, followed by a stage of sexual inactivity, which
coincides with a non-singing period. Exposure of these birds to increasing quantities
of (electric) light from the beginning of December till the beginning of February again induces full spermatogenesis and full song.

According to Rowan (vide Rowan, 1938, for a summary) the influence of light on the testis, which results in sperm formation, is not direct but indirect, as the light merely permits the birds to remain awake, and this wakefulness causes an increased physical activity, which is the primary sexually activating stimulus. Rowan’s hypothesis has not been confirmed by the experiments of many other investigators (Bissonnette, 1931, 1933, 1936; Riley, 1937; Kendeigh, 1941) who found that enforced exercise in darkness did not induce testicular activation in starlings or house-sparrows. Neither are the experiments of Benoit (1937) who tied up his ducks nor those of Miyazaki (1934, 1935) who observed that during ‘yogai’ (cf. p. 21) his birds (Zosterops palpebrosa japonica) were sleepy and inactive, in favour of Rowan’s opinion. Moreover, my own experiments do not support his view; my greenfinches were placed in small cages, in which they could only move a little; nevertheless, when they were exposed to light again, their testes were activated very quickly.

Rowan (1929) and Miyazaki (1934) also succeeded in bringing birds into the breeding condition more than once within the course of a year. Rowan treated juncos with extra light during the autumn, then to shortened days in January and finally to the increasing daylight of February and March. By the end of May these birds were found to be in full breeding condition once more. Miyazaki practised ‘yogai’ with his birds during December, and got singing birds in January; then a second ‘yogai’ was initiated during September and stopped at the end of October when the birds were again in full song; and after a third ‘yogai’, started at the beginning of December, the birds reached breeding condition again towards the end of the year.

As to the time of year in which song-birds may be brought into breeding condition, the opinions of the authors differ. Riley (1937) was not able to activate the testes of house-sparrows in September, except in juvenile birds; the same negative result was obtained by Schildmacher (1938, 1939) with redstarts (Phoenicurus phoenicurus), robins (Erithacus rubecula) and blackcaps (Sylvia atricapilla) in the early months of autumn. My own experiments showed, however, that testis recrudescence may occur already in August after the birds have been mewed.

Some investigators (e.g. Benoit (1937) and Bissonnette (1938)) have found that after a period of extra lighting the enlarged testes regress spontaneously. This phenomenon was also observed during my experiments in a bird (E44), which had been in breeding condition three times within 12 months and in which testis regression occurred, although it was subjected to increasing lengths of daylight during March. The origin of this regression is not clear; internal factors may play an important part.

As to the path along which the light stimulus is transmitted to the testes, it is generally accepted that it reaches the hypophysis by way of the eyes, optic nerves and hypothalamus, and activates this endocrine gland to secrete its gonadotrophic hormone, under the influence of which the gonad recrudescence and sperm production take place.
It would therefore have been important if it had been possible to detect in the anterior lobe of the hypophysis, simultaneously with the gonad recrudescence, an increase in number of the so-called basophile cells, to which the gonadotrophic function of the hypophysis is generally attributed. However, I was not able to establish a distinct difference between the acidophile (the α-) and the basophile (the β-) cells with the aid of the usual colour methods ('Azan'); even 'Duazor' (Ruyter, 1943) gave no results. Therefore it was not possible to discern a parallel between the gonad activity and an increased activity or a larger number of β-cells in the hypophysis. As far as I am aware the avian hypophysis stains very poorly, a fact which has also been established by other investigators in our laboratory; so it was impossible to make a qualitative or a quantitative cytological study of the anterior lobe of the hypophysis.

A precocious moult and refeathering were observed in the birds subjected to mewing. In nature the summer moult of the adult greenfinch takes place between the end of July and the beginning of September. Under the experimental conditions, described above, moulting and refeathering occur very much earlier; the reduction of light causes these phenomena to take place in the first half of June. Burger (1941) found in starlings that a reduction in length of day favours or speeds up moulting and refeathering. Moreover, Miyazaki (1934) observed a moult in his birds (Zosterops palpebrosa japonica) subjected to 'yogai', after which the birds received as a matter of course relatively less light than under 'yogai' conditions. Walton (1937) caused mallard drakes to take an eclipse plumage during February and March instead of during June and July by subjecting them to extra lighting.

Moulting and refeathering, being the expression of processes of metabolism, are undoubtedly related to the hormonal function of the thyroid; therefore it seems important to study the histology and cytology of this endocrine gland in order to establish a possible relation between the microscopical structure of the thyroid and the precocious moult. As the function of the thyroid is in turn regulated by the anterior lobe of the hypophysis it would also be important to study the anterior lobe, as the acidophile cells are possibly the source of the thyrotrophic hormone. Preliminary investigations on this subject have been carried out already, but will not be published till more experimental material is available.

My experiments as well as those of numerous other investigators have shown that in the male song-bird light is an essential factor in producing testis recrudescence in spring. Of course other factors, amongst which are the reciprocal stimulation of the males and the presence of the female, certainly have an important role in testis activation. In the female, however, these factors are more important and more numerous, as has been shown by the experiments of Polikarpova (1940) with female sparrows, where ovulation occurs only under special circumstances.

**SUMMARY**

1. 'Mewing' of song-birds is a traditional practice of Dutch bird-catchers who make use of it to produce singing decoys in the autumn. The birds are encaged early in May, the daylight is progressively reduced by means of curtains during May and
Experimental modification of the sexual cycle of the greenfinch

the birds are kept in darkness throughout June, July and the first half of August. From the middle of August onwards the curtains are gradually opened and by the end of the month the birds are in daylight and in full song once more.

2. This procedure was carried out with the greenfinch (*Chloris chloris*) under laboratory conditions in order to study the behaviour, song and moulting of the birds and the histology of their gonads before, during and after mewing.

3. In the early stages of mewing the testes decrease in size till in total darkness they have the same diameter as the inactive testis.

4. When the birds are exposed to normal daylight again the testes enlarge rapidly until after some weeks of increasing spermatogenesis they are fully mature. By this time the birds are in full song and it is likely that song coincides with sexual maturity and is induced by the male sex hormone.

5. The diameter of the testis tubules varies in the same way as the diameter of the gonads.

6. In the middle of October, after some weeks of sperm production, regression of the testis sets in. During the ensuing stage of sexual inactivity some birds were exposed to increasing quantities of (electric) light from the beginning of December until the beginning of February. These birds showed increasing spermatogenesis and developed their full song.

7. Thus by means of mewing and 'extra lighting' it was possible to bring birds into breeding condition three times within one year.

REFERENCES

[Literature quoted in the text but not mentioned in this list is to be found in Rowan (1938).]


