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## THE EMBRYONIC DEVELOPMENT OF COCHLIDION LIMACODES HUFN.

(FAM. COCHLIDIDAE, LEPIDOPTERA)

A STUDY ON LIVING DATED EGGS

BY

PAUL J. HOLST CHRISTENSEN



København i kommission hos Ejnar Munksgaard 1953

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## A. Introduction.

Although the development of the eggs of butterflies is one of the most interesting subjects within insect embryology, we still lack many investigations which may shed light on problems as yet unsolved. This somewhat unfriendly treatment of that field of research is no doubt in great part due to the very considerable technical difficulties besetting such studies. For by far the majority of the eggs of the *Macrolepidoptera* are inclosed in an, as a rule thick, porcelain-like, and intransparent shell (the *chorion*) which renders impossible any direct observation of the living contents inside. If, therefore, the eggs are to be subjected to a histological-embryological investigation the shells must be removed prior to sectioning—a most difficult process, which it is understandable that many investigators have avoided. To this must be added that the eggs, owing to the copious yolk, are among the most difficult to microtomise; further, the form of the egg is often unfavourable to exact orientation in paraffin, especially in the case of globular eggs.

In contrast with the often fairly good-sized eggs of the *Macrolepidoptera*, those of the *Microlepidoptera* present certain unquestionable advantages for embryological studies: (1) The eggs are as a rule small, which reduces the expenses of the investigation (slides and cover-slips); (2) the moths are easily bred in cultures; (3) the eggs are often transparent; and (4) they are sometimes flat, which must be a great advantage in direct observation under the microscope. And as a matter of fact several valuable investigations on the embryology of butterflies have been made—more especially recently—precisely on *Microlepidoptera*, e. g. HUIE's work (1918) on *Eudemis nævana* (HB.), that of SEHL (1931) on *Ephestia kuehniella* ZELL., and MÜLLER's study (1940) of *Plodia interpunctella* HÜBN.

The ideal subject of investigation in the study of butterfly embryology must, therefore, according to the above, be a flattened, thin-shelled, transparent butterfly egg, preferably not very large. Where shall we be able to find such an egg? The fact that conditions are so favourable within the *Microlepidoptera* group will, I think, warrant the inference that we might with some probability expect to find something similar among the *Macrolepidoptera* placed lowest in the system, i. e. among the transitional forms between the *Micro-* and *Macrolepidoptera*. On a closer analysis this hypothesis has indeed proved correct. The moth *Cochlidion limacodes* 

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HUFN., belonging to the characteristic family of the *Cochlididae* but somewhat rare in this country (Denmark), has eggs of the desired type. I have indeed already discussed this question in two previous publications (HOLST CHRISTENSEN 1943 and 1950), where the embryology of this form was partly treated. Since I have moreover in the latter paper thoroughly described the anatomy of the *limacodes* egg I shall only include in the present work what may be considered of importance for the exposition to follow (cf. p. 5–6).

Before proceeding I should like in this place to express my thanks to Farmer ERLING PEDERSEN, Bogo, for his kind help in procuring eggs, larvae, and fertilised butterfly females. Without his ready assistance it would hardly have been possible to carry through the investigation. I am likewise indebted to Mr. E. PYNDT cand. pharm. for sending me some fertilised females. Further, my acknowledgments are due to the draughtswoman, Miss K. SVANE, for her beautifully and accurately executed drawings. Finally it is my pleasant duty to tender grateful thanks to the CARLSBERG FOUNDATION for financial support which has enabled me to carry out the present investigation, and to the RASK-ØRSTED FOUNDATION, which has defrayed the expenses of the translation done by Miss ANNIE I. FAUSBØLL, M.A.

## B. Material and Methods.

Having described at length how the material was procured in my paper of 1950 (cf. pp. 3—4) I shall not go further into this subject here but merely say that the fertilised females were confined singly in small rectangular glass cages specially constructed for the purpose. These were made from ordinary slides,  $26 \times 76$  mm, and some of a somewhat larger size,  $38 \times 88$  mm. The glass plates were fastened together lengthwise by means of black adhesive tape ("U-Form Feucht-Klebstreifen"). The cages could stand up or be laid down as it was most convenient, and in the latter case they were closed by a slide pressed against each end while in the former case they were placed on the top of a slide or a cover slip, and on the top of the cage again was laid a loose slide. Some of the erect cages were also placed on cellophane paper; afterwards the eggs could be used either for photographing, hatching, or fixing.

The females as a rule proved very willing to oviposit—particularly in the evening and the night—both on the glass plates (cf. HOLST CHRISTENSEN 1950, pp. 4 and 6) and on the cellophane paper. Gradually as the eggs were laid they could then easily be marked in ink with a number or a ring, and thus one could keep track of their age and appearance during the embryonic development. So as to secure a suitable degree of moisture for the eggs to develop in, the slides from the cages broken up after oviposition were placed in cardboard boxes containing damp cotton wool; now and again water was also actually dripped on the eggs from a small pipette. At a tempera-

ture of about  $20-24^{\circ}$  C. the eggs would generally hatch out on the 9th day after oviposition.

The eggs which were to be used for taking photomicrographs were laid in distilled water with a cover slip over, supported by two pieces of cut-off brass pins about 1 mm thick and almost the same width as the cover slip. The eggs were photographed under EDINGER's projection apparatus at an enlargement of 70 times and with the use of a green filter. For the most part PERUTZ's fine-grained "Silbereosin Platten" were used.

## C. The Embryonic Development of Living Dated Eggs.

We shall now proceed to describe in detail the embryonic development of the unfixed living *limacodes* egg. So far as it has been possible the investigation was made on eggs accurately dated, but since oviposition—as previously mentioned—chiefly takes place in the evening or at night I have not always been able to ascertain the quite exact age of the eggs; in such cases I have then been obliged to be content with approximate figures (cf. Table III, text figures and explanation of the plates). In this connection it must also be kept in mind that two eggs of the same age may very well be at quite different stages of development, since the temperature is of decisive importance for the embryonic development as for other metabolic processes. For the same reason the temperature was measured as far as it was practicable. For comparative purposes it would of course have been best if all the experiments had been made under constant temperature conditions, but that was unfortunately impossible, since a great many of the investigations were carried out in my home in the summer holidays under very primitive laboratory conditions. However—as already pointed out in my thesis for the doctorate (1942, p. 5)—much valuable embryological work on insects has been carried out under similar natural experimental conditions to those of my experiments, so I think that it may be of interest to give an account of my results. Finally I may just mention that of course it would be impracticable to review the variations in the embryonic development of all the numerous eggs examined. For the sake of clarity, therefore, only a few of the particularly interesting egg types have been discussed and for convenience they will in the sequel be denoted by capital letters: A, B, C, etc.

## I. The New-Laid Egg.

As already stated in two previous papers (HOLST CHRISTENSEN 1943 and 1950), the egg of *Cochlidion limacodes* HUFN. (cp. also 1943, Tafel IV, Abb. 7 and 1950, Tafel I, Abb. 2) is as a rule flat, scale-like, and oval, and measures about 0.90 mm in length and about 0.65 mm in breadth. The new-laid egg is yellowish in colour but gradually as it develops it assumes a more milky hue. Outermost it is surrounded

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by a thin egg shell (the *chorion*) shining with a mother-of-pearl lustre, especially when the larva has emerged from the egg. A microscopic examination of the chorion further reveals that the latter is provided with a characteristic sculpturing reminiscent of the meshes of wire-netting. At one end of the egg-often the pointed end-is observed the micropyle apparatus consisting of two concentric circles of wedge-shaped "leaves", the inner ones of which delimit a centrally placed area with about 6-8micropyle canals intended for the passage of the sperms. Owing to the shape of the eggs which, as already described, resembles that of a flat hill, the outermost part of the egg is quite flat and also transparent, consisting of the egg shell alone in this place. This transparent zone may be about 0.12 mm broad, but it varies a good deal as it is the broader the more flatly the egg adheres to the substratum. Inside the clear zone comes the living content itself, the composition of which corresponds closely to what we know from the typical *centrolecithal* egg occurring among the insects: Outermost a very fine contour which must no doubt be interpreted as a vitelline membrane (membrana vitellina); inside this a mixture of so-called formative yolk (cytoplasm) and nutritional yolk (*deutoplasm*). The cytoplasm is especially found as an extremely thin peripheral layer (periplasm) just under the vitelline membrane, and we must then imagine that it branches through the whole mass of the egg as an exceedingly fine sponge, so that in this way central cytoplasmic strands are organically connected with peripheral ones, and finally with the periplasm. The deutoplasm is embedded in the cytoplasm and occurs as a very complete emulsion of minute fatty droplets or of a fatty substance, staining vividly with 0s04. In the new-laid egg the fatty drops are of a very small order of magnitude (about  $5-8 \mu$ ), but gradually as the development progresses the fat accumulates in larger and larger globules (cf. the figures on Pls. I and II). Here it is a characteristic worth noting that the nutritional volk does not, as in the Orqua antiqua egg, consist of proteins (cf. HOLST CHRISTENSEN 1937, 1942, and 1943), but of fatty substances, for which reason microscopic sections through the *limacodes* egg bear some resemblance to a Swiss Emmenthaler cheese, the fat having been extracted during the preparation.

## II. The Egg when about I Day old.

#### 1. The Formation of the Blastoderm.

If *limacodes* eggs laid in the night at about  $20-25^{\circ}$  C. are examined the next morning they will as a rule be in the blastoderm stage. This is the case, for instance, with the egg (**A**) shown on Pl. I, fig. 1 which was laid on the evening of the 12th July, 1945, at a temperature of  $25^{\circ}$  C (Mean developmental temperature  $27^{\circ}$  C, cf. Table III). At 11 a.m. on the  $^{13}/_{7}$ , the following histological-embryological details could be observed:

Outermost an egg shell (Ch) with the characteristic chorion structure (Chs); above at the pointed end of the egg the micropyle apparatus (Mi) consisting of the two concentric rosettes and of the central area with 6 distinct micropyle canals (Mik).

Inside the outer zone formed by the chorion (Yz) occurs the actual living substance of the egg, composed of the cytoplasm and the deutoplasm. The former is only seen distinctly outermost where it extends as a fine zone, the periplasm (Pe), along the periphery of the whole egg. The outer contour has a tongue-shaped appearance, the blastoderm stage being under formation. Each "tongue" is the incipient primordium of a blastoderm cell (Bl) which is well marked off on the outside and towards the neighbouring cell, but is still without any sharp contour inwards towards the volk. The periplasm contains hardly any yolk mass, so this zone appears practically entirely devoid of granules and clear, whereas the area inside is filled with numerous small shining globules (F) consisting of fat or a fatty substance. The formation of blastoderm cells seems to take place at last at the posterior blunt end of the egg and at first the cells are rather large. Subsequently smaller cells develop, the first ones dividing and new ones being added. We can therefore distinguish between a large-celled and a small-celled blastoderm stage, according as the outer contour appears as large vigorously marked tongues or as smaller, more obliterated tongues. As an example of this we will choose 2 other eggs (**B** and **G**) of somewhat similar age, shown on Pl. I.

Fig. 2 shows the **B**-egg<sup>1</sup>, laid on the evening of the 12th July, 1945, at a temperature of  $18^{\circ}$  C (Mean developmental temperature  $24.3^{\circ}$  C), mostly in the large-celled blastoderm stage with very conspicuous large, strongly marked blastoderm cells (Bl)especially at the plump posterior end of the egg. Each cell contains a lightcoloured roundish spot (Ke) which must be interpreted as the nucleus. In the pointed front part of the egg there are also fairly large cells, and these as well as the posterior blastoderm cells form the cells which I have previously called the "serosagenic cells" (cf. HOLST CHRISTENSEN 1942, pp. 88-89), because they give rise to the outermost embryonic envelope, the serosa (see p. 8). The  $\mathbf{G}$ -egg depicted in fig. 3, on the other hand, is apparently in a somewhat earlier stage, the blastoderm cells being distinctly observable along the periphery of the whole egg. This egg approximately represents a transitional stage to the small-celled blastoderm stage, many of the blastoderm cells being not yet radially halved, and new cleavage cells from the interior of the egg may still be expected to arrive in the periplasm. As is well known, two kinds of cells are present in the egg contents: cleavage cells and vitellophags. Both are derivates of the fertilised nucleus of the egg, which at first is surrounded by a small island of cytoplasm formed by the union of the cytoplasmic areas of the male and female pronuclei. Later the zygote divides and by the further division of the offspring of the daughter cells thus formed, a large number of cleavage nuclei arise inside the egg. Of these some remain in the egg as vitellophags, intended for the liquefying of the yolk, others migrate centrifugally to unite at length with the periplasm. Absorbed herein the cytoplasm bulges like a tongue in front of each nucleus and the area now develops into a blastoderm cell which is marked off from the neighbouring cell and finally also from the yolk inside, by which procedure the blastoderm gradually arises.

 $^1$  Unfortunately the tables of the development of the  ${\bf B}\text{-}$  and  ${\bf D}\text{-}\text{eggs}$  have been left out to reduce the printing expenses.

#### 2. The Formation of the Germ Band and the Embryonic Envelopes.

When the blastoderm has been formed the more detailed differentiation of the blastoderm cells begins in the equatorial region of the egg, giving rise to the germ band or germ disk, or embryonic rudiment, or ventral plate. In order to follow its formation in detail we select a new, specially suited, egg  $(\mathbf{D})$  which was laid in the evening of the  $^{17}/_6$ , 1948, at a temperature of about  $181/_2^{\circ}$  C (Mean developmental temperature 20.5° C). The next morning, at 10<sup>15</sup> a.m. (text fig. 1 a) we see that the embryonic rudiment (K) is beginning to appear, most plainly in the right side of the egg. While the cells here and at the top are quite small they seem still to be in the large-celled blastoderm stage (Bl) at the back (cp. Pl. I, fig. 2), being very large here (about 50  $\mu$  broad and 28  $\mu$  high) and clear on account of their slight content of fatty droplets and granules. The fatty droplets inside the yolk too (F) are still very small, being only of the order of magnitude of about 8  $\mu$ . At 10<sup>30</sup> a.m. (text fig. 1 b) the ventral plate (K) is distinctly observable and appears as a very wide, slightly curved band extending across the egg a good way on either side of the equator. In this way the germ band comes to occupy about  $\frac{1}{3}$  of the whole area of the ventral side, being about 444  $\mu$  high and about 789  $\mu$  broad<sup>1</sup>. The ventral plate, however, does not keep to the ventral side alone but also with its rounded ends extends some way on to the dorsal aspect of the egg (cp. Pl. I, fig. 4,  $K_1$  and  $K_2$ ). The fatty droplets (F) have now grown to a size of about 9–17  $\mu$ , and the large blastoderm cells (serosagenic cells) (Bl) can still be observed posteriorly. Coincidently with the rise of the germ rudiment the formation of the embryonic envelopes now sets in. Thus it can be observed how the overlapping edge formed of blastoderm cells  $(Se_1)$  has, as it were, slipped some way down over the germ disk, thus gradually giving rise to the outer embryonic envelope, the serosa. Almost at the same time the inner embryonic envelope, the amnion, is also formed by an intensive growth of cells from the edges of the germ band. We must, however, give up following its further development as it can only be satisfactorily studied in sections and not in the living egg.—Now too the gradual immersion of the embryonic rudiment into the yolk begins, a marked reduction of the height and breadth of the ventral plate taking place, a process which I have called the regression of the germ disk (cf. HOLST CHRISTENSEN 1942, p. 157). Thus the dotted and later stippled line in the left side of the egg shows how the lateral contour of the germ disk gradually moves inwards. Now and then deep dints are also observed in this place, but they quickly straighten out again. At  $10^{55}$  a.m. (text fig. 1c) the ventral plate (K) is about 300  $\mu$  high and 800  $\mu$  broad. A strip of serosa  $(Se_2)$  is now also plainly present at the lower end of the germ band and both strips have further approached each other in an equatorial direction, the distance between them being about 289  $\mu$ .<sup>2</sup> At 11 a.m. (text fig. 1d) the ventral plate (K) measured about 322  $\mu$  in height and about 793  $\mu$  in breadth, while the strips of serosa (Se<sub>1</sub> and Se<sub>2</sub>) were about 267  $\mu$  distant from each other; at 11<sup>06</sup> a.m. (text fig. 1e) the distance between them was reduced to about  $250 \,\mu$ ; at  $11^{14}$  a.m. (text

<sup>&</sup>lt;sup>1</sup> The breadth was only measured on the ventral side, not on the dorsal aspect.

<sup>&</sup>lt;sup>2</sup> The distance was measured between the apexes of the strips.



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fig. 1 f) it was only about 206  $\mu$ . Moreover, it is observed that the measurements of the germ disk (K) are practically unchanged, the height being still about 322  $\mu$  and the breadth about 800  $\mu$ , and in the left side of the egg, as in the preceding stage, the serosa strips are just on the point of joining each other and closing. At 11<sup>25</sup> a.m. (text fig. 2a) it has almost happened, and the lateral contour of the ventral plate is now already seen some way inside the yolk. At the immersion of the germ disk its breadth has been reduced to about 706  $\mu$ ; its height, on the other hand, constitutes about 333  $\mu$ . On the right side the two serosa strips (Se<sub>1</sub> and Se<sub>2</sub>) are about 156  $\mu$ distant from each other, and at 11<sup>30</sup> a.m. (text fig. 2b) the distance has been reduced to about 145  $\mu$ . The ventral plate (K) is now also immersed in the yolk on the right side and the breadth has therefore been reduced to 667  $\mu$ . The height is only about  $300 \ \mu$ . Above and below in the germ disk an indentation may moreover be traced, which is shown schematically by a dotted line. It is a characteristic feature worth noting that the joining of the serosa strips and the detachment of the ventral plate by immersion do not coincide. This again means that the formation of the amnion is not, probably, synchronous with that of the serosa but most likely takes place later and independently of the outer embryonic envelope. This curious fact I have already noted in two previous publications (HOLST CHRISTEN-SEN 1942, p. 168, and 1943, pp. 209-215) and described at length in the case of Orgyia antiqua L.

At  $11^{46}$  a.m. (text fig. 2c) the breadth of the ventral plate has been diminished a very little (656  $\mu$ ), whereas the height has increased somewhat (322  $\mu$ ); the distance between the two serosa strips now amounts to about  $89 \mu$ . At  $12^{08}$  p.m. (text fig. 2d) the breadth of the germ band (K) has again increased a little (678  $\mu$ ) at the expense of its height (305  $\mu$ ), which in fact means, as shown indeed in text figures 2b-2d, that only an oscillation around a state of equilibrium has been observed, the dimensions and form of the ventral plate not having altered very much since 11<sup>30</sup> a.m. At  $12^{08}$  p.m. the serosa strips are still about 89  $\mu$  distant from each other. At  $12^{11}$  p.m. (text fig. 2e) comes a turning point in the development of the ventral plate; now it begins decidedly to grow in the longitudinal direction of the egg at the same time as its breadth decreases visibly, the height being now about  $344 \mu$ , the breadth about  $644 \mu$ . The distance between the two strips of serosa has been reduced to some 78  $\mu$ , and at 12<sup>15</sup> p.m. (text fig. 2f) it has further diminished to about 56  $\mu$ , whereas the proportion of the dimensions of the germ disk has not altered in the 4 minutes that have elapsed. At  $12^{18}$  p.m. (text fig. 3 a) the serosa strips (Se1 and Se2) have very nearly joined (the dotted line), at 1229 they meet (the heavy line), and at  $12^{40}$  p.m. the union is complete. The closing and formation of the serosa in the right side of the egg has then exactly, at c.  $20^{\circ}$  C, taken some 130 minutes, from which it will be seen that this interesting process occurs fairly rapidly and therefore may easily escape notice. As a further illustration of the facts concerning the closing of the serosa I refer the reader partly to my previous publication (HOLST CHRISTENSEN 1943, pp. 211-213 and Tafeln IV-V), partly to fig. 4 on Pl. I.

The figure represents a photomicrograph of the above-mentioned B-egg which







Fig. 3. Diagrammatic representation of the development in an approximately dated egg (D) of *Cochlidion limacodes* HUEN, Mean developmental temperature 20<sup>1</sup>/<sub>2</sub>° C, ×50. Fig. 3 a about 15<sup>1</sup>/<sub>4</sub>—15<sup>1</sup>/<sub>2</sub> hours, 3 b about 15<sup>3</sup>/<sub>4</sub> hours, 3 c about 17<sup>1</sup>/<sub>4</sub> hours, 3 d about 17<sup>3</sup>/<sub>4</sub> hours, 3 e about 19 hours, and 3 f about 39 hours after oviposition (see also the text).

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has here been taken on the  ${}^{13}/_{7}$ , 1945, at 1<sup>45</sup> p.m., i. e. 165 minutes after the stage shown on Pl. I, fig. 2. The ventral plate (K) is seen as a wide curved band running obliquely across the egg with its rounded ends ( $K_1$  and  $K_2$ ) curving round on to the dorsal side. The embryonic disk which measures about 329  $\mu$  in height and about 686  $\mu$  in breadth is already partly immersed in the yolk, the lateral contours of the ventral plate having moved somewhat inwards. While the closing of the serosa has been accomplished in the left side of the egg, it is only just on the point of closing on the right side. The two serosa strips ( $Se_1$  and  $Se_2$ ) are indeed quite close together, but the outer contour has not as yet straightened out entirely as a sign of the completion of the process. The figure also conveys an excellent idea of the different dimensions of the fatty globules (F) in the central and peripheral areas of the egg, the diameter varying from 5—10  $\mu$ .

We now return to the D-egg previously mentioned to point out that at 12<sup>30</sup> p.m. (text fig. 3a) the secondary cleavage of the volk mass begins. For the time being it only happens in the left side of the egg, and the process consists in the yolk being divided by radial partition walls into large, well-marked territories (Dt). Later the cleavage proceeds in a centripetal direction by the formation of walls in other directions. Coincidently with the cleavage the yolk withdraws a little from the outer wall (Serosa, Se), thus leaving inside the latter a comparatively clear area (O) containing few small granules of yolk. In contrast with this the fatty globules inside the egg have grown to about 19  $\mu$ , and at 12<sup>43</sup> p.m. (text fig. 3b) the diameter of some of these is about 28  $\mu$ . The embryonic rudiment (K) which has a height of 361  $\mu$  and a breadth of 667  $\mu$ —at 12<sup>30</sup> p.m. the dimensions were respectively 350  $\mu$  and 639  $\mu$ —is now, it is plain, wholly immersed in the yolk, the serosa contour (Se) being also entirely straightened out on the right side  $(Se_3)$ . The secondary cleavage of the yolk (Dt) started on the left side has advanced further and the clear space (O) inside the serosa has increased. At 1<sup>13</sup> p.m. the first vigorous pulsations in the egg are observed, so that on the left side deep dints arise at certain intervals, which quickly straighten out again. At 210 p.m. (text fig. 3c) the secondary cleavage of the yolk mass1 is far advanced, though the central areas are still undivided. In the right side of the egg the volk territories (Dt) from both sides are pressing downward (cp. also here Pl. II, fig. 5) while at the same time the embryonic rudiment (K) curves more and more inward from side to side. The height now amounts to some 350  $\mu$  and the breadth to about 572  $\mu$ . The fatty globules (F<sub>1</sub>) in the peripheral area (O) are small, while inside the yolk territories they (F) have a diameter of about 17  $\mu$ . At 2<sup>45</sup> p.m. (text fig. 3d) not a few have attained a diameter of about 28  $\mu$ , and the yolk mass has now undergone a total secondary cleavage into numerous yolk territories (Dt). The ventral plate (K) which has a height of about 367  $\mu$  and a breadth of about 606  $\mu$ , has become further immersed in the yolk so that the yolk territories from above and below have reached each other  $(Dt'_1)$  on the right of the embryonic disk.—At 16 p.m. (text fig. 3e) the regression has reached its climax, the embryonic rudiment (K) being

 $<sup>^{1}</sup>$  For greater clarity only a part of the yolk territories is included in the diagrammatic drawings of the eggs.

highly incurved from all sides; thus it measures about 317  $\mu$  in height whereas it is only about 417  $\mu$  in breadth, which shows that it is especially in the breadth that the incurvation has taken place.

We will now for the time being leave this egg though it was followed till it hatched out  $({}^{26}/_{6}, 1948, \text{ at } 12^{03} \text{ p.m.})$ , so as to study another egg (**E**) where conditions in the subsequent developmental processes are still more instructive. The egg is a *limacodes* egg laid on the  ${}^{15}/_{6}$ , 1948, at 6 p.m. at a temperature of 24° C (Mean developmental temperature 21.15° C, see Table I). On the  ${}^{16}/_{6}$  at 11 a.m.—17 hours later—the following histological-embryological details (text fig. 4a) could be observed:

Outermost the chorion (Ch) with the micropyle apparatus  $(Mi)^1$  above, and inside this a clear finely granulated zone (O) delimited outwardly by the serosa (Se). Then follows the yolk mass, throughout divided into yolk territories (Dt) where the fatty droplets (F) have a diameter of about  $22-33 \mu$ . Innermost, deeply immersed in the yolk, is seen the obliquely placed oval ventral plate (K), which is about  $322 \mu$ high and some 589  $\mu$  broad. In other words, the egg is in a stage similar to that of the **B**-egg of which a photomicrograph was again taken on the  $^{13}/_{7}$ , 1945, at 4<sup>45</sup> p.m. (Pl. II, fig. 5). The tongue-shaped embryonic rudiment (K), some  $314 \mu$  high and 586  $\mu$  broad, extends obliquely across the egg, and on the right side the two strips  $(Dt_2' \text{ and } Dt_3')$  of the yolk mass have not yet reached each other. The fatty globules (F) have a diameter of about 22  $\mu$ , and as regards the secondary cleavage of the yolk mass the **B**-egg is still very backward. Further advanced in this respect is the C-egg<sup>2</sup> shown on Pl. II, fig. 6, which was photomicrographed on the  $^{12}/_{7}$ , 1945, at  $2^{15}$  p.m. i. e. exactly  $17^{1/2}$  hours old (see Table II), though the ventral plate (K) is only about  $300 \ \mu$  high and about  $494 \ \mu$  broad; many of the fatty droplets (F) have a diameter of about 29  $\mu$ .—A fine secondary cleavage of the yolk mass is further seen in the A-egg, which was again drawn on the  $^{13}/_{7}$ , 1945, at  $2^{30}$  p.m. (Pl. II, fig. 7). Inside the seros contour (Se) the clear area (O) can be observed and then follows the yolk mass divided into numerous territories (Dt). Deeply immersed in the latter we observe the somewhat obliquely placed embryonic rudiment (K), about 265  $\mu$  high and some 458  $\mu$  broad. Above this are seen some large fatty droplets (F) with a diameter of about 18 µ.

When the **E**-egg is  $17^{1/2}$  hours old  $({}^{16}/_{6}, 1948, 11^{30} \text{ a.m.})$  there is a distinct change in the form of the ventral plate (K) (text fig. 4b); having now an incurvation both above and below it has become doubly tongue-shaped. The maximum height and the maximum breadth are now about 378  $\mu$  and 556  $\mu$  respectively. A similar stage is seen on Pl. II, fig. 8, which shows a photomicrograph of another egg (**F**). The figure also exhibits a very fine secondary cleavage of the yolk mass, and the fatty droplets (F) in some of the yolk territories (Dt) have attained a size of about 30  $\mu$ . The maximum height and the maximum breadth of the double-tongued embryonic rudiment (K) are 357  $\mu$  and 557  $\mu$  respectively. Inside the serosa is seen the clear zone (O) with very small fatty droplets.

<sup>&</sup>lt;sup>1</sup> These constant structures will only exceptionally be mentioned in the description to follow.

<sup>&</sup>lt;sup>2</sup> Mean developmental temperature 25.6° C.



Fig. 4. Diagrammatic representation of the development of an exactly dated egg (E) of Cochlidion limacodes HUFN. Mean developmental temperature about  $21^{\circ}$  C.  $\times$  50. Fig. 4a 17 hours, 4b  $17^{1}_{2}$  hours, 4c 18 hours, and 4d  $18^{1}_{2}$  hours after oviposition (see also the text).



Fig. 5. Diagrammatic representation of the development of an exactly dated egg (E) of Cochlidion limacodes HUFN. Mean developmental temperature about  $21^{\circ}$  C.  $\times$  50. Fig. 5a 19 hours, 5b  $19^{1}_{2}$  hours, 5c 20 hours, and 5d  $20^{1}_{2}$  hours after oviposition (see also the text).

When the **E**-egg has attained an age of exactly 18 hours  $({}^{16}/_{6}, 1948, at 12 noon)$ the ventral plate (K) has again changed its form (text fig. 4 c), being now some 516  $\mu$ broad and about 406  $\mu$  high; it has, however, still the incurvation above and below. Some of the fatty globules (F) are seen to have attained a diameter of about 33  $\mu$ . Half an hour later (text fig. 4d) the germ disk (K) has become still shorter, being about 478  $\mu$  broad and about 400  $\mu$  high. The incurvation above and below is still retained and the diameter of some of the fatty droplets has increased to about 39  $\mu$ . When the egg is 19 hours old (text fig. 5a) the ventral plate (K) has almost lost its incurvation below, while it is still distinct above. The form of the germ disk is now more roundish, the height being increased to about 428  $\mu$ , whereas the breadth has now decreased to 462  $\mu$ . Above, on the right (K<sub>3</sub>), the growth of the ventral plate would seem to be particularly lively, and this is seen still more distinctly when the egg is  $19^{1/2}$  hours old (text fig. 5b,  $K_{3}$ ). The form is now more like that of a short thick pear (the finely dotted line), the maximum height of the embryonic rudiment (K) being about 456  $\mu$ , its maximum breadth about 462  $\mu$ . At the same time a blastokinesis takes place, the ventral plate turning to the left, i. e. anticlockwise. Some of the fatty droplets (F) have in this stage attained a diameter of about 44  $\mu$ .—Before continuing our description of the further development of the **E**-egg we would point out that quite similar changes in the germ disk may also be instructively studied in the photomicrographs reproduced on Pls. II-III.

Fig. 9 on Pl. III shows a picture of the C-egg of which a photomicrograph was again taken on the 12/7, 1945, at 415 p.m., i. e. at an age of exactly 191/2 hours. Within a clearer finely granulated zone (O) come the numerous yolk territories (Dt), some of which contain fatty globules (F) with a diameter of about 36  $\mu$ . Innermost is seen the germ disk (K) which is broader on the left side; its maximum breadth is about 449  $\mu$ , whereas the maximum height is about 336  $\mu$ . The incurvations above and below, on the other hand, are not very distinct. $-1^{1/2}$  hours later (at 5<sup>45</sup> p.m.) a photomicrograph of the same egg (Pl. III, fig. 10) shows that the "pear-shaped stage" previously mentioned has been reached (cp. text figs. 5b and 5c). The maximum height now amounts to about 350  $\mu$  and the maximum breadth to about 435  $\mu$ , and many of the fatty globules (F) in the volk territories (Dt) are fairly large, about  $36 \mu$ . Some fine "pear-shaped stages" are also seen on Pl. III, figs. 11 and 12, and on Pl. IV, fig. 13 where a photomicrograph of the  $\mathbf{F}$ -egg has again been taken. Fig. 11 shows a slight incurvation below in the germ disk (K); the maximum height is about 386  $\mu$ , the maximum breadth about 414  $\mu$ . The fatty droplets (F) are fairly large in many of the yolk territories (Dt), about 36  $\mu$ . The chorion (Ch) with the micropyle apparatus (Mi) as well as the finely granulated zone (O) are very plainly seen in all three figures. Fig. 12 shows a more elongate pear-shaped stage of the ventral plate (K), and it can be observed that the left side of it is beginning to show signs of increased growth above. The maximum height of the germ disk (K) is now about 407  $\mu$ , while its maximum breadth is only  $386 \mu$ . In fig. 13, Pl. IV this ratio between them is still more marked, the left side above having quite distinctly taken the lead. It can even now be observed how this rapidly growing area is differentiating itself into a "head 3

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part" ( $H_1$ ); the maximum breadth of the germ band (K) has now decreased to about 336  $\mu$ , while, on the other hand, its maximum height has increased to about 443  $\mu$ . When the germ disk has once reached this stage a rapid growth in length (and height) takes place, as can easily be verified under the microscope (see below).

While figs. 11 and 12 most nearly correspond to text figs. 5a and 5b, fig. 13 is considerably more advanced. This is indeed plainly seen on considering text fig. 5 c, which shows the  $\mathbf{E}$ -egg when it is exactly 20 hours old. On a comparison with text fig. 5 b it is noted that the germ band has extended very considerably, the maximum height being now about 488  $\mu$  and the maximum breadth about 362  $\mu$ . If we copy the contour of the germ disk in the 5c stage on to the 5b stage it will be seen that it is principally the "head part"  $(H_1)$  of the embryo which has grown. At the same time it may be observed, on comparing the ventral plates in stages 4 a-4 d and 5 a-5d, that a very conspicuous movement (blastokinesis) with a consequent new position of the embryonic rudiment has taken place, for it is now partly seen in side view, whereas it was previously seen from the surface. This fact, that the embryo turns sideways, must no doubt be connected with a more favourable utilisation of the space within the egg. And as a matter of fact it may be observed that a rapid growth in length now occurs, connected with a straightening out of the germ band. This we already find clearly displayed when we consider text fig. 5d, where the egg has attained an age of  $20^{1/2}$  hours. Here it is seen that the embryo (K) has not only increased in breadth above but the length also has been augmented by additional growth both above  $(H_1)$  and below  $(H_2)$ ; of this fact one also gets a vivid impression by following the development of the limacodes egg under the microscope. The maximum height of the embryo is now about 533  $\mu$  and the maximum breadth about 356  $\mu$ ; the clear area (O) has increased in volume and the fatty globules (F) in the yolk territories have a diameter of up to about  $42 \mu$ . In the succeeding stage (text fig. 6 a), where the  $\mathbf{E}$ -egg is 21 hours old, it is observed that the embryo (K) by a turn to the right (clockwise) has assumed a position more parallel to the longitudinal axis of the egg. The breadth of the germ band has decreased somewhat compared with stage 5d, being now about  $328 \mu$ , on the other hand the maximum height has distinctly increased, being now about 544  $\mu$ .

In the 6a stage many of the fatty globules have attained a diameter about 37  $\mu$ .

The above-mentioned straightening out of the embryo is also clearly displayed in the photomicrographs shown on Pl. IV. In fig. 14 it is seen that the germ disk (K) in the **F**-egg has become appreciably elongated, the maximum height now amounting to 472  $\mu$  and the maximum breadth to about 315  $\mu$ . The "head part" ( $H_1$ ) and the "tail part" ( $H_2$ ) of the embryo are now growing more and more distinct and the distance between gradually increases, amongst other things by a rapid downward growth of the tail part. In fig. 15—where a photomicrograph of another *limacodes* egg (**H**) is shown—these two areas are plainly seen at a considerable distance from each other, and both in fig. 13 and fig. 14 it will be noticed that the yolk territories (*Dt*) often contain a single large fatty droplet (*F*) which may have a size of up to



Fig. 6. Diagrammatic representation of the development of an exactly dated egg (E) of *Cochlidion limacodes* HUFN. Mean developmental temperature about  $21^{\circ}$  C.  $\times$  50. Fig. 6a 21 hours, 6b 38 hours, 6c 45 hours, and 6d 63 hours after oviposition (see further the text).

43  $\mu$ . By the further growth and elongating of the embryonic rudiment a rapid coiling up of the latter gradually takes place and we now approach the appearance which the embryo has when it is about 2 days old.

### III. The Egg when about 2 Days old.

As a typical example of the development in the egg when it is 2 days old we first select for discussion the **D**-egg (text fig. 3 f), which was drawn again on the  $^{19}/_{6}$ , 1948, at 12 noon. It will be seen that the embryo  $(Em)^1$  is markedly spiral in shape, coiled up at both ends and especially in the tail part  $(H_2)$  below. The maximum height<sup>2</sup> amounts to about 578  $\mu$ , the maximum breadth to about 389  $\mu$ , and across the "back" the embryo is about 78  $\mu$ . In the drawing is also seen the finely granular zone (0) and the fatty droplets (F), up to about 44  $\mu$  in size, within the yolk territories (Dt). On Pl. IV, fig. 16 a quite similar stage is shown, where a photomicrograph has been taken of the **B**-egg on the  $\frac{14}{7}$ , 1945, at 11 a.m. Inside the "fine-grained zone" (0) we have the yolk-mass with the yolk territories (Dt) and fatty globules (F), some of which measure about 29  $\mu$ . Innermost is seen the embryo (*Em*) closely coiled up in a spiral and measuring about 514  $\mu$  in length, about 372  $\mu$  in breadth and about 100  $\mu$  across the back. This, in addition, shows a distinct segmentation (Sq) in several places; above is observed a broader head part  $(H_1)$ , which I interpret as the primordium for the "cephalic lobes" (J).—A nearly similar stage is seen on Pl.V, fig. 17, where the C-egg is shown in a photomicrograph from the  $^{13}/_{7}$ , 1945, at 12<sup>45</sup> p.m. -i. e. at the exact age of 40 hours. In the egg is seen the fine-grained zone (O), and the yolk mass with the yolk territories (Dt) containing fatty droplets about 29  $\mu$  in size (F). Innermost is found the spirally coiled up embryo (Em) with the tightly rolled up tail part  $(H_2)$  below and the broad head part  $(H_1)$  above. The maximum height of this embryo is about 558  $\mu$ , the maximum breadth 472  $\mu$ , and the dorsal thickness is about 143  $\mu$ .—It is true that the segmentation (Sq) is not so marked as in the case of the **B**-egg, but on the other hand the two cephalic lobes of the head are more plainly to be seen in this embryo.—Practically the same conditions may still be observed about 4 hours later (Pl.V, fig. 18) when a photomicrograph was again taken of the C-egg on the  $^{13}/_{7}$ , 1945, at 5 p.m., only the thickness of the dorsum has increased somewhat (about 157  $\mu$ ).

A little further advanced in its development is the **E**-egg (text fig. 6b), which, as a matter of fact, started at a developmental temperature of  $24^{\circ}$  C.<sup>3</sup> It was drawn again on the  ${}^{17}/_{6}$ , 1948, at 8 a.m., i. e. exactly 38 hours after oviposition. The maximum height of the embryo (*Em*) is about 656  $\mu$ , its maximum breadth about 533  $\mu$ , and the dorsal thickness is about 106  $\mu$ . The finely coiled up tail part ( $H_2$ ) and the head ( $H_1$ ) furnished with cephalic lobes (*J*) are plainly seen; some of the largest

<sup>&</sup>lt;sup>1</sup> From now on the embryonic rudiment will be designated by this term.

<sup>&</sup>lt;sup>2</sup> The coiling up has not been taken into account in the measurement.

<sup>&</sup>lt;sup>3</sup> From the  $^{17}/_{6}$ , 1948, the temperature was about  $20^{1}/_{2}^{\circ}$  C (see Table I).

fatty globules (F) are 50  $\mu$  in diameter. In the case of this **E**-egg the development seems to take place with the greatest rapidity, for already the same day (the <sup>17</sup>/<sub>6</sub>, 1948, at 3 p.m.) at an age of exactly 45 hours (text fig. 6c) it has attained a stage almost corresponding to that of the **B**-egg which is about 3 days old (cp. Pl. V, fig. 20). The maximum height, maximum breadth, and dorsal thickness of the embryo (*Em*) have increased to about  $660 \times 533 \times 222 \,\mu$  respectively. The head (*H*<sub>1</sub>) is furnished with 2 broad cephalic lobes, of which especially the left one (*J*) can be distinctly seen, and the tail (*H*<sub>2</sub>) is closely coiled up in a spiral and provided with an outgrowth (*T*) at the tip. Down the middle of the dorsum (*R*) and in the left cephalic lobe are seen areas of yolk with fatty drops in them; some of these (*F*) have a diameter of about 33  $\mu$  outside the animal; in several places along the dorsal side of the embryo a distinct segmentation (*Sq*) is moreover observed. Further, most of

the secondary yolk areas have become fused and the area (O) inside the serosa is extremely fine-grained. At this stage pulsations often occur in the egg along the dorsal side of the embryo, which results in more or less marked embryonic movements.

The furthest advanced in development of the eggs 2 days old is beyond doubt the A-egg (Pl.V, fig.19), which was drawn again on the  $^{14}/_{7}$ , 1945, at 10 a.m. The zone with the small yolk bodies (O) is seen and then comes the yolk, the yolk territories of which seem to have disappeared; the fatty droplets average about 24  $\mu$  in size. Innermost is the embryo itself (Em) tightly coiled up; on it is seen both a head part (H<sub>1</sub>) and a tail part (H<sub>2</sub>). The maximum height, maximum breadth, and dorsal thickness are about  $543 \times 416 \times 193 \mu$  respectively.

### IV. The Egg when about 3 Days old.

Here we shall begin by discussing the **E**-egg (text fig. 6d) which was drawn again on the  ${}^{18}/_{6}$ , 1948, at 90<sup>5</sup> a.m.—i. e. exactly 63 hours after it had been laid. By comparing it with the **E**-egg which is about  $2^{1}/_{2}$  days old (text fig. 6c) it is easily seen that the embryo (*Em*) has grown much and at the same time altered its shape. For the maximum height, maximum breadth, and thickness of the dorsum now amount to about  $650 \times 550 \times 278 \ \mu$  respectively; the cephalic lobes (*J*) are no longer so distinct and the dorsum has grown appreciably broader over a long part. The tail part (*H*<sub>2</sub>) with the outgrowth (*T*) has also increased in size and the segmentation (*Sg*) may be observed a good way along it. In the head (*H*<sub>1</sub>) as well as along the middle of the dorsum (*R*) there is a good deal of yolk which at this stage exhibits fatty globules (*F*) with a diameter of about 50  $\mu$ .—On the other hand, the **B**-egg (Pl.V, fig. 20), which was again photomicrographed on the  ${}^{15}/_{7}$ , 1945, at 3 p.m., does not seem to have progressed quite so far in its development as the **E**-egg, the head lobes (*J*) being more marked, the plate-shaped outgrowth (*T*) and the tail (*H*<sub>2</sub>) somewhat smaller, and finally the thickness of the dorsum does not appear to be so great either.

The C-egg, on the other hand, is somewhat further advanced in its development (Pl. VI, fig. 21), as was indeed to be expected from the particulars given above; it Dan. Biol. Skr. 6, no. 9.

was again photomicrographed on the  $^{14}/_7$ , 1945, at  $10^{45}$  a.m.—i. e. at the exact age of 62 hours. The maximum height, maximum breadth, and thickness of dorsum were about  $650 \times 643 \times 221 \ \mu$  respectively. The broad head part  $(H_1)$  with the cephalic lobes (J) strike the eye at once, likewise the tip of the tail  $(H_2)$  with the plate-shaped outgrowth (T). In the head and along the trunk (R) there is a good deal of yolk which, incidentally, contains fatty droplets (F) of about 29  $\mu$  in diameter, whereas outside the animal they reach a size of about 50  $\mu$ .

Finally there is the A-egg (Pl.VI, fig. 22), which seems to be at a slightly earlier stage of development than the E-egg previously mentioned. The A-egg was drawn again on the  ${}^{15}/_{7}$ , 1945, at  $2^{30}$  p.m. After the fine-grained zone (O) comes the yolk where the territories (Dt) have become almost entirely fused and where some of the fatty globules (F) may attain a diameter of 28  $\mu$ . Innermost is seen the embryo (Em), the tail part (H<sub>2</sub>) of which especially is closely coiled up and ends in a broad plateshaped expansion (T). The maximum height, maximum breadth, and thickness of the dorsum are 596 × 488 × 205  $\mu$  respectively. Observation of the exact shape of the embryo is, however, rendered extremely difficult by the yolk, part of which is found in the head part (H<sub>1</sub>) and along the trunk of the embryo (R).

### V. The Egg when about 4 Days old.

Here again we start with a description of the **E**-egg (text fig. 7a) which was drawn again on the  ${}^{19}/_{6}$ , 1948, at 12 noon—i. e. exactly 90 hours after oviposition. It is at once noted that the shape and size of the embryo have changed very considerably; the maximum height, maximum breadth, and thickness of the dorsum now amount to about  $672 \times 550 \times 283 \ \mu$  respectively. The thin "tail" with the characteristic "tail-plate" (cp. text figs. 6 b - 6 d) has disappeared, and instead we find an almost straightly cut off tail area ( $H_2$ ) with 2 peculiar "humps" ( $P_1$  and  $P_2$ ) towards the ventral side of the animal. The latter, incidentally, is strongly curved, "maggot-like", and along the dorsal side of the embryo a very pronounced segmentation (Sg) is observed. Inside the yolk, some yolk territories (Dt) are still seen to have been preserved and they may contain a fatty droplet (F) up to 28  $\mu$  in diameter.

Somewhat similar developmental conditions are found in the A-egg (Pl.VI, fig. 23), which was drawn again on the  ${}^{16}/_{7}$ , 1945, at 10 a.m. Inside the serosa (Se) the fine-grained zone (O) is seen, and inside this again there is yolk with some few fatty globules (F) 30  $\mu$  in size, and with the yolk territories (Dt) partly preserved, especially in the upper part of the egg. Innermost appears the "maggot-like" embryo (Em) itself, measuring about 596  $\mu$  in height, about 512  $\mu$  in breadth, and with a dorsal thickness of about 223  $\mu$ . The strongly curved position of the embryo will be noticed, besides the marked segmentation (Sg) of the dorsum and the characteristic humps (P<sub>1</sub> and P<sub>2</sub>) of the tail part (H<sub>2</sub>).—Almost similar developmental conditions may be observed in the **B**-egg (Pl.VI, fig. 24), of which a photomicrograph was again taken on the  ${}^{16}/_{7}$ , 1945, at 11 a.m. The tail part (H<sub>2</sub>), cut off almost straight, will be noted,



Fig. 7. Diagrammatic representation of the development of an exactly dated egg (E) of Cochlidion limacodes HUFN. Mean developmental temperature about  $21^{\circ}$  C.  $\times$  50. Fig. 7a 90 hours, 7b  $112^{1}_{2}$  hours, 7c 136 hours, and 7d  $146^{3}_{4}$  hours after oviposition (see also the text).

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and in addition the 2 humps ( $P_1$  and  $P_2$ ) at the top and the bottom of it. Segmentation (Sg) is present but not equally distinct everywhere.—This, on the other hand, is especially the case in the C-egg (Pl.VII, fig. 25), of which another photomicrograph was taken on the  $^{15}/_{7}$ , 1945, at  $2^{45}$  p.m., that is to say, at the exact age of 90 hours. Further, the difference in the size of the embryo in the **B**-egg and the C-egg will be noticed. The maximum height, maximum breadth, and thickness of dorsum are in the former case about  $500 \times 400 \times 257 \mu$  respectively, in the latter about  $615 \times 528 \times 329 \mu$ .

## VI. The Egg when about 5 Days old.

When the **E**-egg is 5 days old (text fig. 7 b) the embryo has again become much changed in size and shape. It is characteristic of this egg, which was drawn again on the  ${}^{20}/_{6}$ , 1948, at  $10^{30}$  a.m.—i. e. exactly  $112^{1}/_{2}$  hours after the moment of oviposition—that a re-orientation, a blastokinesis II, of the embryo (*Em*) has again taken place. The maximum height, maximum breadth, and thickness of the dorsum now amount to about  $672 \times 566 \times 278 \mu$  respectively. The head ( $H_1$ ) of the embryo no longer points downwards (cp. text fig. 7 a), but approximately sideways. An eye rudiment (*Ey*) is now for the first time plainly visible, and the segmentation (*Sg*) is very marked—in the head part (*Sg*<sub>1</sub>) too. The tail part ( $H_2$ ) is no longer seen under the head but has in an apparently mysterious way been moved so that it lies along the dorsal aspect of the animal. The mid-intestine ( $T_1$ ) filled with yolk will also be noticed, and more posteriorly the rectum (*Et*) with the dilated part ( $T_2$ ) at the extremity. Some of the fatty droplets (*F*) outside the animal may have a diameter of about 33  $\mu$ .

In order to understand clearly how the above-mentioned blastokinesis, takes place it will be necessary to study another *limacodes* egg  $(\mathbf{I})$ , where conditions are particularly instructive (text figs. 9a-9d). But first we must again recall certain anatomical details concerning the peculiar development of the tail part in the previously mentioned, 90-hour-old, E-egg (text fig. 7a). The almost straightly cut off "tail"  $(H_2)$  was, as we know, provided with 2 distinct humps,  $P_1$  and  $P_2$ . During the blastokinesis the very peculiar fact is observed that the area  $P_2$  is now everted and continues its growth up along the whole length of the dorsal aspect of the embryo. The first stage in the process of eversion is shown in text fig. 9a, which represents the I-egg drawn on the 21/6, 1948, at 140 p.m. A distinct segmentation (Sg) of the embryo (Em) is observed on the dorsal aspect; in addition the downwardbent head part  $(H_1)$  is seen. The tail area  $(H_2)$  with the rectum (Et) and its dilated part  $(T_2)$  is noticed below, and it is observed that the former has already pushed its way in under the embryo and some distance up along the dorsum. Already at 400 p.m.—140 minutes later—(text fig. 9b) conditions have chonged so that the tip of the abdomen  $(H_2)$  has reached almost halfway up along the back of the animal. The mid-intestine  $(T_1)$  filled with fatty globules (F) is seen, as well as the rectum

(*Et*) with the dilated part ( $T_2$ ) posteriorly and the distinct segmentation (*Sg*), and some of the spiracles (stigmata, *St*). In addition, the head ( $H_1$ ) has changed its position, being no longer turned so much downwards but more lifted up. At 5<sup>00</sup> p.m.—200 minutes later—(text fig. 9c) the tail part ( $H_2$ ) with the rectum (*Et*) has advanced still further upwards and the head now points sideways, as can indeed be seen by the distinct eye rudiment (*Ey*). At 9<sup>10</sup> p.m.—450 minutes later—(text fig. 9d) the blastokinesis is approaching its completion, the tip of the tail part ( $H_2$ ) now lying on a level with one eye rudiment (*Ey*). The head ( $H_1$ ) with the two eye rudiments points directly sideways, but it will not, probably, be long before the final position of the abdomen has been reached and the head thus turned into its place fronting the micropyle (cp. text figs. 7b and 7c). The part of the blastokinesis described here has then in this case taken some 7<sup>1</sup>/<sub>2</sub> hours at a developmental temperature of  $23^{1}/_{2}^{\circ}$  C.

That the blastokinesis takes place precisely in the egg that is about 5 days old will indeed appear very plainly from a consideration of the C-egg (Pl.VII, fig. 26), of which a photomicrograph was again taken on the <sup>16</sup>/<sub>7</sub>, 1945, at 10<sup>45</sup> a.m.—i. e. at the exact age of 110 hours. The embryo (Em) whose maximum height, maximum breadth, and dorsal thickness amount to about  $578 \times 543 \times 257 \mu$ , is here slightly ∫-shaped. This is owing to the fact that the embryois undergoing a blastokinesis, the aim of which—as previously stated—is partly to get its head (H1) turned towards the micropyle (cp. Pl. VII, fig. 25), partly to make room for the rapidly growing animal and obtain the most ideal utilisation of the space within the egg. Further the distinct segmentation (Sq) on the left side of the abdomen appears very plainly, as well as the rectum (Et) with the posterior dilatation  $(T_2)$ . In addition the mid-intestine  $(T_1)$  filled with volk is observed more anteriorly, and the anterior  $\delta$ -shaped area of the embryo and the head which—in spite of its orientation towards the micropyle (Mi)—has not yet reached its final position, because the caudal area  $(H_2)$  has not been pushed into place. In other words, the last phase of the blastokinesis is fixed in this photomicrograph. Finally we may also mention that many of the yolk territories (Dt) contain several fatty droplets (F), some of which attain a diameter of about 36 µ. The proportion of the embryo to the yolk mass within the egg now seems to be as 1:1.

As might be expected from what has previously been stated, the A-egg and the **B**-egg have progressed further in their development on account of the start they had in temperature, though they too are only about 5 days old. This is observed at once on inspecting the A-egg (Pl.VII, fig. 27), of which a drawing was again made on the 17/7, 1945, at 10 a.m. Inside the fine-grained zone (O) we note the yolk in which the yolk territories (Dt) can be distinguished and in which some of the fatty globules (F) have a diameter of about 28  $\mu$ . Innermost may be noted the embryo (Em) itself, the height<sup>1</sup> (= length) now amounting to about 603  $\mu$ , while the breadth is about 530  $\mu$ , and the thickness of the dorsum about 289  $\mu$ . Unlike the 4-day-old A-egg (cp. Pl.VI, fig. 23) the form has now changed again from the "maggot-stage" to a more

<sup>1</sup> As usual the height was measured without taking into account the coiling of the embryo. Thus the measurement merely denotes the height of the coiled up embryo, not its absolute height = length.

caterpillar-looking stage. The animal is, however, still closely coiled up, though, indeed, in another way, namely from side to side, so that the posterior part of the body lies parallel with and close up against the anterior part. Moreover, a typical change has taken place in the position of the head  $(H_1)$  which is now no longer turned a way from the micropyle but towards it; owing to this fact the posterior extremity of the animal  $(H_2)$  and the tip of the head are almost on a level with each other, though the contours of the anterior extremity cannot be quite distinctly seen because the head is deeply buried in the yolk mass. In other words, the blastokinesis previously mentioned has been accomplished in this case. Very characteristic of the 5-day-old **A**-egg is likewise the presence of 2 distinct eye rudiments (Ey) in the form of 1 oval pigment spot on either side of the head. The segmentation (Sg), so distinct before  $(r_1)$  containing yolk is present.

Similar developmental conditions are seen in the 5-day-old **B**-egg (Pl.VII, fig. 28) of which a photomicrograph was again taken on the  ${}^{17}/_{7}$ , 1945, at 11 a.m. The eye rudiments (*Ey*) are seen, the mid-intestine (*T*<sub>1</sub>), and part of the segmentation (*Sg*), which is particularly distinct in the right half of the abdomen; finally, posteriorly part of the rectum (*Et*) with a dilatation (*T*<sub>2</sub>) at the end may also just be discerned. The maximum height, maximum breadth, and dorsal thickness of this embryo are about  $543 \times 436 \times 222 \ \mu$  respectively.

## VII. The Egg when about 6 Days old.

When the  $\mathbf{E}$ -egg (text fig. 7c) is about 6 days, or exactly 136 hours old, it is, owing to the somewhat lower developmental temperature  $(18^{1/2} \, ^{\circ} \text{C})$ , in a similar phase of development as has already been mentioned for the egg about 5 days old (cp. Pl.VII, figs. 27 and 28). Thus if we study text figure 7c of the E-egg, of which another drawing was made on the <sup>21</sup>/<sub>6</sub>, 1948, at 10 a.m., it will be seen that the position of the embryo (Em) corresponds very closely to that observed for the **B**-egg when about 5 days old (cp. Pl.VII, fig. 28); for as in that stage, the tip of the abdomen  $(H_2)$  only projects a little way past the anterior margin of the head, and the head itself  $(H_1)$  is deeply immersed in the yolk. The maximum height, maximum breadth, and dorsal thickness of the embryo amount to about  $678 \times 623 \times 278 \ \mu$  respectively. On the anterior part of the animal are observed 2 distinct light-brown eye spots (Ey)besides a more or less distinct esophagus (Sp). Further back may be noticed the mid-intestine  $(T_1)$ , and then the rectum (Et) with the dilated part  $(T_2)$ . The segmentation (Sg) is less distinct, most conspicuous where the two sides of the animal lie against each other. Along the dorsal aspect of the animal there is a series of pigment spots mainly light-brown in colour. The numerous yolk territories (Dt) contain large fatty droplets which, however, as a rule seem to be smaller than in the preceding stages. Outermost the volk is as usual surrounded by a narrow granular zone (O).

When, on the other hand, the **E**-egg approaches the exact age of 6 days (= 144

hours), its developmental stage is much more like that of the B-, C-, and A-eggs, which will be discussed presently (cp. Pl.VIII, figs. 29, 30, and 31 respectively). This appears plainly in text fig. 7 d, representing the E-egg drawn again on the  $^{21}/_{6}$ , 1948, at  $8^{45}$  p.m.—i. e. exactly  $146^{3}/_{4}$  hours after oviposition. In the course of about 11 hours the embryo (Em) has grown considerably in size, the maximum height, maximum breadth, and dorsal thickness now amounting to about  $728 \times 638 \times 334 \,\mu$  respectively. This is also manifested by the great reduction of the yolk mass surrounding it. The head  $(H_1)$  is much more differentiated, with distinct mouth-parts (Md) in front and well developed eyes (Ey) more posteriorly. Particularly conspicuous is the esoph gus  $(S_p)$  which adjoins the mid-intestine  $(T_1)$  filled with yolk; posteriorly it passes into the rectum (Et) with the characteristic dilatation ( $T_2$ ). The segmentation (Sq) has practically quite disappeared along the outer margin of the animal, but can still be seen along the two inner adjoining sides of the embryo. On the dorsal aspect are observed a series of large pigmented spots (P) mainly light-brown of colour. Surrounding the embryo is, as usual, seen the fine-grained zone (O) and the chorion (Ch) with chorion structure and micropyle apparatus (Mi).

We shall now proceed to describe the **B**-egg (Pl.VIII, fig. 29) of which a photomicrograph was again taken on the  ${}^{18}/_{7}$ , 1945, at 11 a.m. The embryo (Em) is as usual closely bent together from side to side, and the tip of the abdomen ( $H_2$ ) protrudes a little way past the head. The animal is seen to have grown very much, the maximum height, maximum breadth, and dorsal thickness now amounting to about  $594 \times 507$  $\times 286 \mu$  respectively. The head ( $H_1$ ) is deeply immersed in the yolk, which owing to the growth of the embryo has shrunk considerably, particularly along the dorsal aspect of the animal. Surrounding the embryo is seen the yolk with the yolk territories (Dt), some of which contain fatty globules (F) with a diameter of up to about 29  $\mu$ .

Somewhat more advanced in development is the C-egg (Pl.VIII, fig. 30) of which another photomicrograph was taken on the  $^{17}/_{7}$ , 1945, at  $10^{45}$  a.m.—i. e. at the exact age of 134 hours. In this egg too, the tips of the abdomen and the head are no longer on a level, that of the abdomen  $(H_2)$  lying a little way past the anterior margin of the head  $(H_1)$ . The maximum height, maximum breadth, and thickness of the dorsum are, in the case of this embryo, about  $686 \times 594 \times 286 \mu$  respectively. Further, the 2 distinct eye rudiments (Ey) will be noticed, as well as the mid-intestine  $(T_1)$ and the rectum (Et) with the posterior dilatation  $(T_2)$ ; anteriorly the esophagus (Sp)and mandibles (Md) are just visible. In addition the segmentation (Sg) is distinctly seen, particularly on the left side of the abdomen, as well as the numerous, wellmarked yolk territories (Dt), the main part of which contains several fatty droplets (F), which are therefore as a rule not very large, about  $20 \mu$ . As an extremely narrow border may be observed the fine-grained zone (O) outside.

Furthest advanced in development, however, is the A-egg (Pl.VIII, fig. 31), which was drawn again on the  $^{18}/_{7}$ , 1945, at  $2^{30}$  p.m. Inside the fine-grained zone (O) we observe the yolk, which is divided into yolk territories (Dt), mostly containing several fatty globules (F) of small size (about 18  $\mu$ ). Innermost appears the embryo (Em) itself, bent strongly sideways; its maximum height, maximum breadth, and dorsal

thickness are about  $783 \times 614 \times 343 \mu$  respectively. That this embryo is older than those previously mentioned is seen amongst other things from the fact that the tip of the abdomen  $(H_2)$  lies in front of the anterior margin of the head  $(H_1)$ . On the front part of the head one notices the mandibles (Md) and on the sides the eye rudiments (Ey); further back on the animal the mid-intestine filled with yolk  $(T_1)$  is observed, and above that a series of pigmented brownish spots (P) which also appeared in the photomicrographs of the **B**- and **C**-eggs (cp. Pl. VIII, figs. 29 and 30). The segmentation (Sg) is now less distinct but is still observed on the right side of the front part of the animal.

## VIII. The Egg when about 7 Days old.

Here, too, we begin with the  $\mathbf{E}$ -egg (text fig. 8a) of which a drawing was again made on the  $^{22}/_{6}$ , 1948, at 2 p.m. At this juncture the egg is about 7 days old or exactly 164 hours. It will soon be seen that with respect to its developmental stage the embryo (Em) corresponds approximately to that described for the **B**- and **C**-eggs when about 6 days old (cp. Pl.VIII, figs. 29 and 30); for here too the tip of the abdomen  $(H_2)$  protrudes a little in front of the anterior margin of the head  $(H_1)$ , and a great deal of the yolk mass is still present, particularly in front of the head. The position of the embryo is also the same, it is closely bent together from side to side. The maximum height, maximum breadth, and dorsal thickness amount to about  $823 \times 689$  $imes 350 \ \mu$  respectively. On the anterior part of the animal are seen the head with the head capsule (Hk), the eve rudiments (Eq), and the mouth-parts (Md). In front of these may be noted 2 fatty globules  $(F_1)$  which the animal is about to swallow; at this period there is no doubt that an active consumption of food takes place. On a level with the eyes a little of the esophagus (Sp) is observed, further back we notice the mid-intestine  $(T_1)$  filled with yolk. Along the dorsal aspect are observed the previously mentioned brownish dorsal spots (P), and posteriorly the abdomen ends in a light-brown double-tongued contour. Immediately surrounding the embryo is seen, on the left side, an apparently "empty" light space (A), which should probably be interpreted as an amniotic cavity. Surrounding this again is the volk containing volk territories (Dt) often with one fatty droplet (F) in them; finally, outermost there is the fine-grained zone (O), besides of course the chorion (Ch) with the micropyle apparatus (Mi).

As was to be expected, the **B**-egg (Pl.VIII, fig. 32), of which a photomicrograph was again taken on the  $^{19}/_7$ , 1945, at 11 a.m., is much further advanced in development. The phase corresponds approximately to that described for the **A**-egg when about 6 days old (cp. Pl.VIII, fig. 31). This in itself is not surprising, considering that the caterpillar already emerged from the latter egg on the 8th day, i. e. on the  $^{20}/_7$ , 1945, at 9<sup>30</sup> a.m. (cf. later, p. 31 and Table III). As in this egg, the embryo (*Em*) occupies a large part of the space within the egg, and much of the yolk outside has been consumed, particularly along the convex side of the animal. The maximum height, maximum breadth, and dorsal thickness amount to about 643 × 586 × 300  $\mu$ 



Fig. 8. Diagrammatic representation of the development of an exactly dated egg (E) of Cochlidion limacodes HUFN. Mean developmental temperature about  $21^{\circ}$  C.  $\times$  50. Fig. 8a 164 hours, 8b  $183^{1/2}$  hours, 8c  $207^{1/2}$  hours, and 8d  $207^{3/4}$  hours after oviposition (see also the text).



Fig. 9. Diagrammatic representation of the second blastokinesis of the embryo in an egg (I) of *Cochlidion limacodes* HUFN.  $\times$  50. Mean developmental temperature  $23^{1/2}$ ° C. 4 different phases in the development of the blastokinesis (II) showing how the tail part of the embryo is gradually everted backward and up along the dorsal aspect, so that the downward-pointing head is gradually turned towards the micropyle. Fig. 9a on the  $^{21}/_{6}$ , at 1<sup>40</sup> p.m.; 9b 140 minutes later, 9c 200 minutes later, and 9d 450 minutes after phase 9a (see further the text).

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respectively. On the front part of the head  $(H_1)$  the mandibles (Md) can be discerned and on the sides the eye rudiments (Ey); further back are seen numerous fine silvery tracheal trunks (Tr), some of which can be traced far back along the side of the abdomen. Further the mid-intestine  $(T_1)$  is also seen, and the pigmented dorsal spots (P) previously mentioned. The partition of the yolk territories (Dt) containing fatty globules (F) may likewise be observed, whereas the fine-grained zone seems to have totally disappeared.

Still further advanced in development is the C-egg (Pl. IX, fig. 33), of which a photomicrograph was again taken at 10<sup>45</sup> a.m. on the <sup>18</sup>/<sub>7</sub>, 1945<sup>1</sup>,—i. e. at the exact age of 158 hours. Amongst other things it may be observed that the tip of the abdomen  $(H_2)$  lies a good way past the anterior extremity of the head  $(H_1)$ , and a large part of the yolk, particularly along the convex side of the animal, has been consumed. The embryo (Em) itself has grown rapidly, its maximum height, maximum breadth, and dorsal thickness now amounting to about  $800 \times 686 \times 343 \,\mu$  respectively. The segmentation (Sq) is no longer very marked, and the form and appearance of the animal are more like those of a coleopterous or a hymenopterous than of a lepidopterous larva. Some of the mouth-parts are seen anteriorly on the head, especially the mandibles, (Md), also the esophagus (Sp); and on the sides the eye rudiments (Ey) are visible. In addition numerous tracheae (Tr) are observed in the head, and further back, too, a good many are present. Inside the animal the mid-intestine  $(T_1)$  can just be made out, and along the dorsal aspect accumulations  $(Dt_3)$  of absorbed volk are seen in several places. Surrounding the larva there is still a little volk, and in front of the head several yolk territories (Dt) are observed containing distinct fatty droplets.

Furthest advanced in development of all the 7-day-old eggs, is, however, the A-egg (Pl. IX, fig. 34), which was again drawn on the 19/7, 1945, at 10 a.m. Outermost is seen the chorion (Ch) with shell structure (Chs), whereas the micropyle apparatus can no longer be distinctly made out. The fine-grained zone as well as the embryonic envelopes and the yolk mass have completely disappeared, the larva having consumed them all. The embryo (Em) which is strongly bent over sideways fills up almost the whole space within the egg shell, and from exigencies of space the tip of the abdomen  $(H_2)$  projects far past the head  $(H_1)$ , and even actually overlaps it. The maximum height, maximum breadth, and dorsal thickness of the larva now amount to about  $982 \times 728 \times 337 \ \mu$ . On the front part of the head are seen the mandibles (Md), on the sides the eyes (Ey); the head capsule (Hk) can also be distinguished. Further back on the animal the mid-intestine  $(T_1)$  is faintly visible, and in addition one observes numerous absorbed fatty globules  $(F_2)$  derived from the yolk mass. The segmentation (Sq) can be seen in several places, though most distinctly along the dorsal and ventral sides of the abdomen, and along the dorsal aspect appear the series of brown pigmented spots (P) previously mentioned. In other words, the embryo is so far advanced in its development that hatching is imminent; it did in fact occur the next day, the 20/7, 1945, at 930 a.m.-i. e., as previously stated, 8 days after oviposition.

 $^1$  The next day at  $9^{30}$  a.m., 8 days after oviposition, the caterpillar had emerged from this egg (cf. Table II).

## IX. The Egg when about 8 Days old.

Text fig. 8b shows the **E**-egg, which was drawn again on the  $\frac{23}{6}$ , 1948, at  $9^{30}$  a.m. At this juncture it is about 8 days old, or exactly  $183^{1/2}$  hours; the maximum height, maximum breadth, and thickness of dorsum are now about  $862 \times 750 \times 372 \ \mu$ respectively. The developmental stage of the embryo (Em) almost corresponds to that previously described for the  $\mathbf{B}$ -egg when it was about 7 days old (cp. Pl.VIII, fig. 32). Here too the closely doubled up embryo fills the greater part of the space within the egg, and only a very little amount of yolk is left along the dorsal aspect of the larva. On the front part of the head  $(H_1)$  the mandibles (Md) are noticed, on the sides the eye rudiments (Ey), and posteriorly the esophagus (Sp) containing particles of yolk  $(Dt_a)$  of which small portions are sucked in between the mandibles at intervals. The head further contains many tracheae  $(Tr_1)$ , several of which are very massive. Along the sides of the abdomen distinct stigmata (St) are observed in several places, in addition to a very large tracheal trunk  $(Tr_2)$ , from which smaller tracheae issue vertically. Finally the mid-intestine  $(T_1)$  can just be made out inside the animal, and along the dorsal aspect of the embryo there is a series of the pigmented spots (P) previously mentioned. Immediately surrounding the animal is seen an empty space (A), probably an amniotic cavity, and outside this again is observed the almost entirely consumed yolk mass which is divided into yolk territories (Dt)containing one  $(Dt_1)$ , two  $(Dt_2)$  or several fatty droplets (F), some of which attain a size of about 22  $\mu$ . Outermost is seen a zone (O) with very small particles of volk (= the fine-grained zone).

From our previous statements (cf. p. 28) it was indeed to be expected that the B-egg when about 8 days old (Pl. IX, fig. 35), of which a photomicrograph was again taken on the  $^{20}/_{7}$ , 1945, at 11 a.m., would be considerably more advanced in development. This is at once seen from the fact that the entire yolk mass has been consumed in addition to the embryonic envelopes. The maximum height, maximum breadth, and dorsal thickness of the still very closely doubled up embryo (Em) amount to about  $714 \times 672 \times 350 \ \mu$  respectively. Above, near the micropyle area, the head  $(H_1)$ is distinctly seen surrounded by its head capsule (Hk). Anteriorly the mandibles (Md)can be discerned, on the sides the eves (Ey), and inside the head the numerous tracheae (Tr). Near the head the tip of the abdomen  $(H_2)$  is also distinctly visible, and in several places there are indications of a segmentation (Sq), particularly along the dorsal aspect of the abdomen. The larva is now so far advanced in development that hatching is imminent. Hence it is seen that it makes lively cutting movements with the mandibles, while now and then "peristaltic waves" pass through it. At last the caterpillar manages to pierce the thin hyaline egg shell (Ch) and it then works its way out, apparently with great effort, and with the most vigorous peristaltic movements of its whole body. At 12 noon, on the  $\frac{20}{7}$ , 1945, it had emerged from the egg shell after a total embryonic period of about 8 days, at a mean developmental temperature of 24.3° C.

## X. The Egg when about 9 Days old.

Since the greater part of the yolk mass has already been consumed in the **E**-egg when it is about 8 days old, (cp. text fig. 8b), it is no wonder that the larva in this egg, when it is about 9 days old (text fig. 8c), is very near to hatching out. A drawing was again made of the egg in question on the  $^{24}/_{6}$ , 1948, at 9<sup>30</sup> a.m. and it was then exactly  $207^{1/2}$  hours old. The maximum height, maximum breadth, and dorsal thickness of the animal (Em) are now  $978 \times 906 \times 478 \,\mu$  respectively. Anteriorly, to the right of the micropyle of the egg, the head  $(H_1)$  is to be seen, and on its sides the eves (Ey); near the head the tip of the abdomen  $(H_2)$  is observed, and along the back of the animal some characteristic protuberances are visible (H). These are the peculiar hairs of the caterpillar, which are gradually everted by means of the blood pressure. Most frequently eversion does not occur till after emergence, but in this case, we see, it has partly preceded the hatching. The larval hairs, which in the fully everted state are provided with a small lateral branch, were closely described and figured in my previous publication on the post-embryonic development of Cochlidion limacodes HUFN. (HOLST CHRISTENSEN 1950, see Tafel II, figs. 6, 7, and 8, and pp. 12 -13). Already at 940 a.m. the caterpillar, about 9 days old (text fig. 8d), now measuring about  $1133 \times 918 \times 512 \ \mu$  in maximum height, maximum breadth, and dorsal thickness respectively, is working its way out of the egg shell, and at 945 a.m.—i. e. at the age of  $207^{3}/_{4}$  hours after oviposition—at a developmental temperature of  $18^{1}/_{2}$ — 24° C (mean temperature about 21° C), the hatching occurred.

# D. Summary.

While previously, on following the embryonic development of butterflies, investigators have had to resort almost exclusively to the study of whole mounts or sections of dead, fixed eggs, the present investigation breaks new ground. Its main purpose has been to supply an accurate description of the progressive development of exactly dated living moth eggs at known temperatures, and under developmental conditions as nearly as possible like the natural conditions. This task, extremely difficult in a technical respect, I have tried to solve by using the oval, flat, and transparent egg, about 1 mm long, of the moth *Cochlidion limacodes* HUFN., somewhat rare in Denmark and occurring mainly in the southern parts of the country (cf. Holst Christensen 1950, p. 4). In order to procure sufficient material it was therefore indispensable to undertake hatching on a large scale, besides collecting larvae and imagines. The fertilised  $\Im$  were confined in cages, specially constructed for the purpose, of slides or cellophane paper, on which they very readily laid their eggs, particularly in the night. The glass or cellophane plates were then placed directly under the microscope, by the aid of which they could be studied and 5

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photomicrographed. When the eggs were kept suitably moist they developed quite normally on this foreign substratum, in the course of 8–10 days according to the temperature.—Since I have become increasingly convinced that *Cochlidion limacodes* is an even extremely well-suited "laboratory animal" for many purposes, I have thought that it might be of the greatest interest to later investigators—not least in the field of experimental embryology—to have a schematic view of the course of development similar to Keibel's "Normentafeln", so at the end of the present section I give 3 summaries in tabular form (Tables I—III)<sup>1</sup> of the development of 3 different *limacodes* eggs (**E**, **C** and **A**) at different temperatures  $(18^{1}/_{2} \degree \text{C} - 29^{1}/_{2} \degree \text{C})$ . Of these eggs **C** and **E** are dated with absolute exactitude, whereas only an approximate dating of the **A**-egg and the other investigated **B**- and **D**-eggs has been possible.

If we select as a paradigm a *limacodes* egg laid at night and evolving at a temperature of about  $20^{\circ}$  C, the embryonic development will, in broad features, take the following course:

On the first day in the life of the egg the zygotic nucleus produced by the fertilisation will give rise to the formation of numerous cleavage nuclei as well as vitellophags. The former gradually migrate through the yolk mass, which mainly consists of fatty globules embedded in a fine network of cytoplasm, to the peripheral layer of cytoplasm, the periplasm, in which they are absorbed and develop into blastoderm cells. The vitellophags, on the other hand, remain in the egg to contribute to the liquefying of the yolk.

On the second day in the life of the egg (when it is about one day old), the formation of the blastoderm will be completed, so that gradually there arises a broad band-like embryonic rudiment or ventral plate, with rounded ends; the latter extend on to the dorsal aspect of the egg, while the rest of the ventral plate occupies about  $\frac{1}{3}$  of the ventral aspect. Such are the conditions about 14 hours after oviposition, and when some 17 hours have passed the ventral plate is tongue-shaped and considerably shorter. A so-called "regression" has occurred, by which both the height and especially the breadth have been reduced. Coincidently with the shortening, a deeper and deeper immersion into the yolk takes place, and the ends of the ventral plate bend more and more towards each other. Synchronously with the regression and immersion the two protective envelopes of the embryo are formed, the amnion innermost, and the serosa outermost. The latter arises in the course of a few hours by an overlapping process of special blastoderm cells, the so-called "serosagenic cells", whereas the former arises as an independent formation along the edge of the ventral plate. After about 18 hours the ventral plate has the form of a double tongue, after about 19 hours it is roundish, after about 20 hours distinctly pear-shaped. The height of the embryonic rudiment is now rapidly increasing, and about 21 hours after oviposition the form has become elongate oval. From considerations of space the embryo must thereupon perform a rotation, blastokinesis I, in the egg, by which it settles on its side. When this has occurred a vigorous growth in length takes place, particularly a gradual growth of the lower part, which is thus by degrees removed

 $^{1}$  I refer the reader to the footnote p. 7.

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from the "head part". We can now speak of a distinct embryo formed like a C, with a "head part" and a "tail part"; the yolk has on that occasion undergone a secondary cleavage into large polygonal yolk territories.

On the third day in the life of the egg (when it is about 2 days old), about 38 hours after oviposition, the embryo is worm-like and coiled up in a close spiral, particularly below; above, two lateral growths, the so-called head- or cephalic lobes, are evolving. About 45 hours after oviposition the embryo has become appreciably thicker and has stouter head lobes; at the same time a "tail-plate" has formed, and the segmentation has begun to be distinct. The fatty globules in the yolk territories now attain a diameter of about 44  $\mu$ .

On the fourth day in the life of the egg (when it is about 3 days old), about 63 hours after oviposition, the embryo is still spiral-shaped but more clumsy. The cephalic lobes are no longer so dominant, whereas the "tail-plate" is large.

On the fifth day in the life of the egg (when it is about 4 days old), some 90 hours after oviposition, the embryo has become thick and maggot-like. A distinct segmentation is present, but the "tail-plate" has disappeared; instead there are two vigorous caudal "humps" on the tail which is almost straightly cut off.

On the sixth day in the life of the egg (when it is about 5 days old), about 112 hours after oviposition, the embryo again changes its position, blastokinesis II is going on, partly for the purpose of turning the head towards the micropyle, partly to obtain a better utilisation of the space within the egg. The process is initiated by the tail part being, as it were, twisted backward and upward along the dorsum, where it continues its growth till it is on a level with the head; at the same time the latter is gradually lifted up so that finally it points towards the micropyle when the blastokinesis has been accomplished. The shape of the embryo during the process therefore recalls an  $\int$ , and the blastokinesis must be estimated to have lasted some 8—9 hours at about 20° C. Distinct eye-rudiments are usually present in this phase.

On the seventh day in the life of the egg (when it is about 6 days old) some 136 hours after oviposition, the embryo, after the completed blastokinesis, is closely bent together from side to side and is more like a caterpillar. The tip of the abdomen lies either on a level with or a little way past the anterior edge of the head. The eyes, esophagus, rectum, and mid-intestine, the latter containing many small fatty droplets, are now distinctly visible; the yolk mass has shrunk considerably. Along the dorsal side of the animal a row of pigment spots, mostly of a light brown colour, are observed.

On the eighth day in the life of the egg (when it is about 7 days old), some 164 hours after oviposition, the embryo is still more markedly bent sideways, and the large dorsal spots have become more brownish black. The embryo now fills a large space in the egg, and it can be observed that an active consumption of food takes place, so that the yolk mass rapidly shrinks. The amnion must consequently be assumed to have been ruptured at the tip of the head, which is now much more distinctly seen. There may further be observed eyes, mouth-parts, head capsule, esophagus, and the mid-intestine, which contains yolk that has undergone secondary cleavage. On the ninth day in the life of the egg (when it is about 8 days old), some 183 hours after oviposition, the embryo has grown considerably, so that the greater part of the yolk mass also has been consumed. The animal is still bent strongly sideways, and the tip of the abdomen is seen a good way past the anterior margin of the head. The eyes, mouth-parts, head capsule, esophagus, and mid-intestine, which now and then makes vigorous peristaltic movements, can be distinctly observed; further the tracheae and spiracles are visibles.

On the tenth day in the life of the egg (when it is about 9 days old), some 205 hours after oviposition, the larva is fully developed and fills the greater part of the egg. The embryonic envelopes as well as the yolk mass have been consumed, and the caterpillar, which may have partly everted dorsal hairs, is now very restless. By means of the mandibles it pierces the egg shell and with violent convulsive movements gradually works its way out of the shell. Hatching usually occurs some 207 hours after oviposition.

The Institute of General Zoology, University of Copenhagen.

# TABLES

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# Tabular View of the Course of Development of an exactly dated Egg of Egg ( $\mathbf{E}$ ) laid on the $^{15}/_{6}$ , 1948,

Date of examination	Develop- mental	Age		Illustration	Appearance of the embryo	
	ture	Hours	Day(s)			
<sup>16</sup> / <sub>6</sub> , 1948, 11 a.m	$24^\circ$ C	17	c. 1	Text fig. 4 a	Germ band tongue-shaped.	
<sup>16</sup> / <sub>6</sub> , 1948, 11 <sup>30</sup> a.m	$24^{\circ}$ C	$17^{1}/_{2}$	c. 1	Text fig. 4b	Germ band double-tongued.	
<sup>16</sup> / <sub>6</sub> , 1948, 12 noon	$24^{\circ}$ C	18	c. 1	Text fig. 4 c	,, ,,	
<sup>16</sup> / <sub>6</sub> , 1948, 12 <sup>30</sup> p.m	$24^{\circ}$ C	$18^{1}/_{2}$	c. 1	Text fig. 4 d	,, ,,	
<sup>16</sup> / <sub>6</sub> , 1948, 1 p.m	$24^{\circ}$ C	19	c. 1	Text fig. 5 a	Germ band slightly double-tongued,	
<sup>16</sup> / <sub>6</sub> , 1948, 1 <sup>30</sup> p.m	24° C	$19^1/_2$	c. 1	Text fig. 5 b	Germ band pear-shaped.	
<sup>16</sup> / <sub>6</sub> , 1948, 2 p.m	$24^{\circ}$ C	20	c. 1	Text fig. 5 c	Germ band more pear-shaped.	
<sup>16</sup> / <sub>6</sub> , 1948, 2 <sup>30</sup> p.m	$24^{\circ}$ C	$20^{1}/_{2}$	c. 1	Text fig. 5d	Germ band elongate oval.	
<sup>16</sup> / <sub>6</sub> , 1948, 3 p.m	$24^{\circ}$ C	21	c. 1	Text fig. 6a	" "	
<sup>17</sup> / <sub>6</sub> , 1948, 8 a.m	$20^1/_2{}^\circ~\mathrm{C}$	38	c. 2	Text fig. 6b	Embryo worm-like and closely coiled	
<sup>17</sup> / <sub>6</sub> , 1948, 3 p.m	$20^1/_2^\circ$ C	45	c. 2	Text fig. 6 c	Embryo perceptibly thicker, spiral-	
<sup>18</sup> / <sub>6</sub> , 1948, 9 <sup>05</sup> a.m	$20^{\circ}$ C	63	c. 3	Text fig. 6d	Embryo still spiral-shaped but more	
<sup>19</sup> / <sub>6</sub> , 1948, 12 noon	20° C	90	c. 4	Text fig. 7 a	Embryo maggot-like, incurved and	
$^{20}/_{6}$ , 1948, 10 <sup>30</sup> a.m	20° C	$112^{1}/_{2}$	c. 5	Text fig. 7b	Embryo slightly ∫-shaped with distinct eye rudiments. No longer maggot-like.	
<sup>21</sup> / <sub>6</sub> , 1948, 10 a.m	18 <sup>1</sup> / <sub>2</sub> ° C	136	c. 6	Text fig. 7c	Embryo bent together from side to side and more caterpillar-like. Apex of ab- domen projecting a little beyond front	
$^{21}/_{6}$ , 1948, 8 <sup>45</sup> p.m	221/2° C	$146^{3}/_{4}$	с. б	Text fig. 7 d	edge of head. Embryo closely bent together from side to side. Along the dorsal aspect	
<sup>22</sup> / <sub>6</sub> , 1948, 2 p.m	$22^{1/2}^{\circ}~\mathrm{C}$	164	c. 7	Text fig. 8 a	Embryo now occupying a large space within the egg. Active consumption of	
<sup>23</sup> / <sub>6</sub> , 1948, 9 <sup>30</sup> a.m	$20^1/_2^\circ~{\rm C}$	$183^{1}/_{2}$	c. 8	Text fig. 8b	Embryo bent very much over side- ways; apex of abdomen a good way	
<sup>24</sup> / <sub>6</sub> , 1948, 9 <sup>30</sup> a.m	$21^{1/2}^{\circ}~{\rm C}$	$207^{1}/_{2}$	c. 9	Text fig. 8 c	Larva fully developed, filling greater part of egg.	
<sup>24</sup> / <sub>6</sub> , 1948, 9 <sup>40</sup> a.m	$21^{1}/_{2}^{\circ}~\mathrm{C}$	$207^{3}/_{4}$	с. 9	Text fig.8d	Larval hairs observed in several places in the much contracted animal.	

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at	18	$3^{1}/_{2}$	-2	$4^{\circ}$ C	(Me	ean	temp	perati	ire :	21.	$15^{\circ}$	C)
Ce	och	lid	ion	lima	code	es H	UFN.					
at	6	p.	m.	(ten	np.	$24^{\circ}$	C).					

Maximum dimensions of the embryo			Embryonic	Yolk		Comments		
Height = length	Breadth	''Dorsal thickness''	envelopes					
$322 \ \mu$	589 $\mu$		Amnion and serosa present.	The yolk mass h gone secondary	as under- cleavage; her small	Germ band immersed in yolk; the rounded ends bent inward.		
$378~\mu$	556 $\mu$		"	<i>,,</i>	<i>,,</i>	The inward-bent rounded ends give the germ band the shape of a ''double-tongue''.		
$406 \ \mu$	516 $\mu$		,,	Fatty globules larger.	a little	Germ band deeply immersed in yolk.		
$400~\mu$	478 $\mu$		,,	Fatty droplets la few in each yolk	arge, and territory.	Ends of germ band bent much inward; tongue- shape disappearing.		
$428~\mu$	$462\;\mu$		,,	,,	,,	Breadth of germ band much reduced; rapid growth of one lateral half above.		
$456~\mu$	$462~\mu$		,,	,,	"	Continued one-sided growth above. Blasto- kinesis I started.		
$488~\mu$	$362 \ \mu$		,,	,,	"	Blastokinesis in full progress.		
533 $\mu$	356 $\mu$		"	"	"	The germ band, which has increased greatly in length, has turned over on one side.		
544 $\mu$	328 $\mu$		,,	,,	,,	Longitudinal axis of germ band almost par- allel with that of the egg.		
$656~\mu$	533 $\mu$	106 $\mu$	"	Very large fatty	globules.	The germ band changed into a spiral-shaped		
$660~\mu$	533 $\mu$	$222~\mu$	"	"	"	"Tail-plate" developing; incipient segmenta- tion		
$650~\mu$	$550~\mu$	$278 \ \mu$	"	"	"	Head lobes no longer dominant; "tail-plate"		
$672~\mu$	$550~\mu$	$283~\mu$	"	"	"	"Tail-plate" gone; distinct segmentation pre-		
$672 \ \mu$	566 $\mu$	$278~\mu$	,,	"	"	Blastokinesis II in progress, the tail part growing up along the dorsum and the head gradually turning towards the micropyle.		
$678 \ \mu$	$623 \ \mu$	$278 \ \mu$	,,	Few and rath fatty droplets yolk territory.	ner large in each	Blastokinesis completed, the head being now turned towards the micropyle. Eyes, eso- phagus and rectum plainly visible.		
728 $\mu$	$638 \ \mu$	$334 \ \mu$	"	Yolk mass much	reduced.	Eyes, mouth-parts, esophagus, and rectum very plainly visible; front part of head now more distinct.		
823 $\mu$	$689 \ \mu$	$350~\mu$	Amnion, am- niotic cavity,	Yolk mass still duced.	more re-	Eyes, mouth-parts, head capsule, and mid- intestine plainly visible. Apex of abdomen double-tangued		
862 $\mu$	750 $\mu$	$372 \ \mu$	,, ,,	Greater part of consumed.	yolk mass	Eyes, mouth-parts, head capsule, esophagus, and mid-intestine plainly visible; tracheae		
978 $\mu$	906 $\mu$	478 $\mu$	The embryonic envelopes have disappeared	Yolk mass enti sumed.	rely con-	Larva very restless within egg. Partly everted hairs of the caterpillar now also observed, besides the organs already mentioned.		
1133 $\mu$	918 $\mu$	$512 \mu$	33	"	,,	Hatching in progress with violent convulsive contractions of the animal. The head is already, by the aid of the mandibles, partly outside the egg shell.		

Table I.

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# Tabular View of the Course of Development of an exactly dated Egg of Egg (C) laid on the $^{11}/_7$ , 1945,

Date of examination	Develop- mental	Age		Illustration	Appearance of the embryo		
	ture	Hours	Day(s)				
$\frac{12}{7}$ , 1945, 2 <sup>15</sup> p.m	$25^{\circ} \mathrm{C}$	$17^{1}/_{2}$	c. 1	Pl. II, fig. 6	Germ band tongue-shaped.		
$^{12}/_{7}$ , 1945, 4 <sup>15</sup> p.m	25° C	$19^{1}/_{2}$	c. 1	Pl. III, fig. 9	Germ band still tongue-shaped, but shorter and higher.		
$^{12}/_{7}$ , 1945, 5 <sup>45</sup> p.m	$25^{\circ}$ C	21	c. 1	Pl. III, fig. 10	Germ band pear-shaped; left upper		
$^{13}/_{7}$ , 1945, 12 <sup>45</sup> p.m	$25^1/_2{}^\circ~{\rm C}$	40	c. 2	Pl.V, fig.17	area shows vigorous growth. Germ band transformed into a worm- like embryo, closely coiled up in a spiral.		
<sup>13</sup> / <sub>7</sub> , 1945, 5 p.m	$25^1/_2^\circ~{\rm C}$	$44^{1}/_{4}$	c. 2	Pl.V, fig.18	») »)		
$^{14}/_{7}$ , 1945, 10 <sup>45</sup> a.m	$25^1/_2^\circ$ C	62	c. 3	Pl.VI, fig. 21	Embryo perceptibly thicker, spiral- shaped, with large head lobes.		
<sup>15</sup> / <sub>7</sub> , 1945, 2 <sup>45</sup> p.m	$26^{\circ}$ C	90	c. 4	Pl.VII, fig. 25	Embryo maggot-like, incurved, and		
<sup>16</sup> / <sub>7</sub> , 1945, 10 <sup>45</sup> a.m	27° C	110	c. 5	Pl.VII, fig. 26	Embryo no longer maggot-like, but slightly ∫-shaped.		
$^{17}/_{7}$ , 1945, 10 <sup>45</sup> a.m	26° C	134	c. 6	Pl.VIII, fig. 30	Embryo bent together from side to side and more caterpillar-like. Apex of ab- domen a little way beyond anterior edge of head Large light-brown spots		
<sup>18</sup> / <sub>7</sub> , 1945, 10 <sup>45</sup> a.m	25° C	158	c. 7	Pl. IX, fig. 33	along the dorsal aspect. Embryo now filling a large space within the egg; active food consumption takes place. Apex of abdomen projecting far boyond outerior edge of head		
<sup>19</sup> / <sub>7</sub> , 1945, 9 <sup>30</sup> a.m	25° C	$180^{3}/_{4}$	c. 8	•••			

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# at 25—27° C (mean temperature 25.6° C) Cochlidion limacodes HUFN. at 8<sup>45</sup> p. m. (temp. 25° C).

Table II.

Maximum dimensions of the embryo			Embryonic	Yolk	Comments		
Height = length	Breadth	"Dorsal thickness"	envelopes				
$300~\mu$	$494~\mu$		Amnion and serosa present.	Yolk mass shows second- ary cleavage; diameter of fatty droplets c 29 u	Germ band immersed in yolk; left end broader than right.		
$336 \ \mu$	$449 \ \mu$		"	Yolk mass shows second- ary cleavage; fatty glob- ules in yolk territories up to c. $36 \mu$ in diameter.	Germ band deeper immersed in yolk; left end markedly broader than right.		
$350~\mu$	435 $\mu$	••	,,	,,	Germ band shorter and higher and still deeper immersed in volk.		
558 $\mu$	$472~\mu$	$143 \mu$	,,	Fatty droplets in yolk territories c. 29 $\mu$ in diameter.	A head- and tail part distinctly visible, the former with well-developed head lobes.		
558 $\mu$	$472~\mu$	157 $\mu$	"	,,	"		
$650~\mu$	$643~\mu$	221 $\mu$	"	,,	"Tail-plate" well developed and distinct seg- mentation present. Some yolk present in head lobes and along trunk.		
$615~\mu$	528 $\mu$	$329~\mu$	,,	,,	The "tail-plate" has disappeared, and seg- mentation is very distinct in many places.		
$578~\mu$	543 $\mu$	$257 \ \mu$	"	In some of the yolk terri- tories the fatty droplets attain a size of c. $36 \mu$ in diameter.	Blastokinesis II is taking place, the tail part growing up along the dorsum, and the head gradually turning towards the micropyle. Final phase of this process fixed in photograph.		
$686 \ \mu$	$594 \ \mu$	$286~\mu$	,,	More fatty globules in each yolk territory, so they are not very large. Diameter c. $20 \mu$ .	Blastokinesis II completed, the head being now turned towards the micropyle. Mandibles, esophagus, eyes, mid-intestine, and rectum with posterior dilatation plainly visible.		
$800 \ \mu$	$686 \ \mu$	$343 \mu$	The amnion has disappeared, but the serosa is still present.	Yolk mass as above, but much reduced in volume.	Mandibles, esophagus, eyes, mid-intestine, rectum, and rectal dilatation, as well as nu- merous tracheae plainly visible.		
					The caterpillar has emerged from the egg shell.		

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# Tabular View of the Course of Development of an approximately dated Egg of Egg (A) laid on $^{12}/_{7}$ , 1945,

Date of examination	Develop- mental	Age		Illustration	Appearance of the embryo		
	ture	Hours	Day(s)				
<sup>13</sup> / <sub>7</sub> , 1945, 11 a.m	$25^{1/2}$ °C	c. 13 <sup>1</sup> / <sub>2</sub>	c. 1	Pl. I, fig. 1	Blastoderm formed but not yet the		
<sup>13</sup> / <sub>7</sub> , 1945, 2 <sup>30</sup> p.m	$25^{1}/_{2}^{\circ} \mathrm{C}$	c. 17	c. 1	Pl. II, fig. 7	Germ band deeply immersed in yolk; form slightly double-tongued.		
<sup>14</sup> / <sub>7</sub> , 1945, 10 a.m	$26^{1}/_{2}^{\circ}~\mathrm{C}$	c. 36 <sup>1</sup> / <sub>2</sub>	c. 2	Pl.V, fig.19	Ventral plate transformed into worm- like embryo, coiled in close spiral.		
$^{15}/_{7}$ , 1945, 2 <sup>30</sup> a.m	$28^1/_2{}^\circ~{\rm C}$	c. 65	с. 3	Pl.VI, fig. 22	Embryo perceptibly thicker, spiral- shaped and with large head lobes.		
<sup>16</sup> / <sub>7</sub> , 1945, 10 a.m	$29^1/_2{}^\circ~\mathrm{C}$	c. $84^{1}/_{2}$	c. 4	Pl.VI, fig. 23	Embryo maggot-like, incurved and with 2 distinct caudal "humps".		
<sup>17</sup> / <sub>7</sub> , 1945, 10 a.m	26° C	c. $108^{1}/_{2}$	с. 5	Pl.VII, fig. 27	Embryo bent together from side to side and more caterpillar-like. Tip of ab- domen projects slightly beyond an-		
<sup>18</sup> / <sub>7</sub> , 1945, 2 <sup>30</sup> p.m	27° C	c. 137	с. б	Pl.VIII, fig. 31	terior edge of head. Embryo greatly bent together from side to side. Large brownish spots along the dorsal aspect. Apex of abdomen well beyond anterior edge of head.		
<sup>19</sup> / <sub>7</sub> , 1945, 10 a.m	28° C	c. 156 <sup>1</sup> / <sub>2</sub>	c. 7	Pl. IX, fig. 34	Larva full-grown, filling greater part of egg shell. Dorsal spots very distinct; apex of abdomen far beyond anterior edge of head		
<sup>20</sup> / <sub>7</sub> , 1945, 9 <sup>30</sup> a.m	27° C	c. 180	c. 8		···		

at	25 - 25	$9^{1/2}$	C (m	ean tem	perature	$27^{\circ}$ (	C)
Co	chlidio.	n lii	nacod	es Hufn			
at	about	930	p. m.	(temp.	25° C).		

Table III.

Maximum dimensions of the embryo			Embryonic	Volk	Germande		
Height = length	Breadth	"Dorsal thickness"	envelopes	TOIK	Comments		
				Size of fatty globules	Blastoderm cells distinctly larger in the front		
$265 \ \mu$	458 $\mu$		Amnion and serosa present.	Yolk mass shows plain secondary cleavage; fatty droplets c. $18 \mu$ in diameter.	Rounded and somewhat expanded ends of ventral plate curved deeply into yolk.		
543 $\mu$	416 $\mu$	$193 \mu$	,,	Average size of fatty globules c. $24 \mu$ in dia- meter	A distinct head- and tail part now formed, the former with well-developed head lobes.		
596 $\mu$	$488~\mu$	$205 \ \mu$	"	The fatty droplets in yolk territories attain c. $28 \mu$ in diameter	"Tail-plate" very well developed and distinct segmentation present. Some yolk in the head		
596 $\mu$	$512 \ \mu$	$223 \ \mu$	"	A few fatty globules at- tain a size of c. $30 \mu$ in diameter	The "tail-plate" has disappeared, segmenta- tion very distinct in many places.		
$603 \ \mu$	$530 \ \mu$	$289 \ \mu$	"	The diameter of some fatty droplets attains a size of c. $28 \mu$ .	Blastokinesis II accomplished, the head being now turned towards the micropyle. Eyes, eso- phagus, mid-intestine, and dorsal spots plainly seen. Segmentation no longer a complement		
783 µ	$614 \ \mu$	$343 \ \mu$	,,	Yolk mass much reduced in extent; in the yolk territories several small fatty globules c. 18 $\mu$ in size	Mandibles, eyes, head capsule, and mid- intestine plainly present, whereas the seg- mentation is more vague.		
982 μ	728 $\mu$	337 µ	The amnion and serosa have dis- appeared.	Yolk mass entirely con- sumed.	Mandibles, eyes, head capsule, esophagus, mid-intestine and tracheae distinctly visible. Larva very restless, hatching imminent.		
					Caterpillar out of egg shell.		

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## LITERATURE

A more detailed list is found in the author's previous works (cf. HOLST CHRISTENSEN 1942, 1943 & 1950).

- CHRISTENSEN, P. J. HOLST (1932), Om Sporsansen hos en Natsommerfugle-Han (Orgyia antiqua Linné). Naturens Verden, Vol. 16.
- (1937), Zur Histologie und Embryologie der überwinterten Eier von Orgyia antiqua LINNÉ. Zool. Jb. Anat., Vol. 62, 1936—37 (Heft 4, 1937).
- (1942), Embryologische und zytologische Studien über die erste und frühe Eientwicklung bei Orgyia antiqua LINNÉ (Fam. Lymantriidae, Lepidoptera). Reprint from Vidensk. Medd. fra Dansk naturh. Foren., Vol. 106. Thesis.
- (1943), Serosa- und Amnionbildung der *Lepidopteren*. Reprint from Entom. Medd. XXIII (Jubilæumsbind), 1943.
- (1950), Studien über die postembryonale Entwicklung bei *Cochlidion limacodes* HUFN. (Fam. *Cochlididae*, *Lepidoptera*). Dan. Biol. Skr., Vol. 7, no. 2. 1950.
- EASTHAM, L. E. S. (1927), A Contribution to the Embryology of *Pieris rapae*. Quarterly J. microsc. Sci., New Ser., Vol. 71, 1928 (No. 283, December 1927).
- (1930), The Formation of Germ Layers in Insects. Biol. Rev. Cambridge, Vol. 5.
- (1930), The Embryology of *Pieris rapae*—Organogeny. Phil. Trans. R. Soc. London, Ser. B, Vol. 219, 1931 (462, 1930).

HUIE, L. H. (1918), The Formation of the Germ-Band in the Egg of the Holly Tortrix Moth, *Eudemis nævana* (HB.). Proc. R. Soc. Edinb., Vol. 38, 1919 (Part II, No. 15. 1918).
IMMS, A. D. (1930), A General Textbook of Entomology. 2. Ed. London.

JOHANNSEN, O. A. (1929), Some Phases in the Embryonic Development of *Diacrisia virginica* FABR. (*Lepidoptera*). J. Morph., Vol. 48, No. 2, December.

KLÖCKER, A. (1917), Sommerfugle V. Natsommerfugle IV. Del. "Danmarks Fauna", Vol. 21. Müller, K. (1938), Histologische Untersuchungen über den Entwicklungsbeginn bei einem

Kleinschmetterling (*Plodia interpunctella*). Zeitschr. f. wiss. Zool., Vol. 151. 1938. SEHL, A. (1931), Furchung und Bildung der Keimanlage bei der Mehlmotte *Ephestia Kuehniella* 

Zell. Z. Morph. Ökol., Vol. 20.

SOUTH, R. (1919), The Moths of the British Isles. Ser. 2. London.

Strкöm, V. (1891), Danmarks større Sommerfugle (*Macrolepidoptera*) systematisk beskrevne. Lehmann & Stages Forlag, København.

WEBER, H. (1933), Lehrbuch der Entomologie. Jena.

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# EXPLANATION OF THE PLATES

Both the text figures and the figures on the plates have all been produced by means of ABBE's drawing apparatus, the former being 90 times enlarged, the latter 83 times. The photomicrographs on Plates I—IX I have taken with EDINGER's projection apparatus (cf. p. 5) at an enlargement of 70 times. The enlargements mentioned in the text, however, all have reference to the degree of enlargement after reproduction.

## Abbreviations occurring in the text:

A	amniotic cavity
Bl	blastoderm cell
Ch	egg shell, chorion
Chs	chorion structure
Dt, $Dt_1$ and $Dt_2$	yolk territory
$Dt_1'$ and $Dt_2'$	upper and lower yolk strip
Dt <sub>3</sub>	yolk absorbed in the embryo
Em	embryo
Et	rectum
Еу	eye rudiment; eye
F	fatty globule
F <sub>1</sub>	fatty droplets in the "fine-grained zone" or before the mouth
F <sub>2</sub>	fatty droplet absorbed in the embryo
Fo	fold in egg shell
Н	evertible larval hair
$H_1 \dots \dots \dots$	head part or head of the embryonic rudiment
$H_2 \dots \dots$	tail part of the embryonic rudiment
Hk	head capsule
J	head- or cephalic lobe
К	germ band or germ disk, or embryonic rudiment, or ventral plate
$K_1$ and $K_2$	overlapping ends of germ band
K <sub>3</sub>	the upper rapidly growing part of the germ band
Ke	nucleus of blastoderm cell
Md	mouth-parts; mandibles
Mi	micropyle
Mik	micropyle canal
0	outer, fine-grained zone
P	pigmented dorsal spot
$P_1$ and $P_2$	upper and lower caudal "humps"
Pe	peripheral cytoplasmatic layer, periplasm
R	yolk mass along the dorsum, or along the side of the embryo

Se and Se<sub>3</sub>..... outer embryonic envelope, serosa Se<sub>1</sub> and Se<sub>2</sub>..... upper and lower serosa strip Sg ..... segment or segmentation Sg<sub>1</sub>..... segmentation in the head part of the embryo Sk.... butterfly scale Sp ..... esophagus St .... spiracle, stigma T.... caudal outgrowth or "tail-plate"  $T_1$  .... mid-gut, mid-intestine  $T_2$  .... dilated part of rectum Tr .... trachea Tr<sub>1</sub> and Tr<sub>2</sub>.... trachea in head and trunk Yz .... outer zone

> Indleveret til selskabet den 18. september 1951. Færdig fra trykkeriet den 30. januar 1953.

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PLATES

## Plate I.

- Fig. 1. Egg (A) of *Cochlidion limacodes* HUFN. about 1 day old—approximately about  $13^{1/2}$  hours. × 70. Blastoderm stage. Note the numerous fatty droplets of which the yolk mass chiefly consists, as well as the peripheral cytoplasmatic layer, the periplasm, with the numerous blastoderm cells only partly marked off.
- Fig. 2. Egg (B) of Cochlidion limacodes HUFN. about 1 day old—roughly about 11<sup>1</sup>/<sub>4</sub> hours. ×70. "Large-celled blastoderm stage". Note on either side the small blastoderm cells of which the germ band consists, the larger cells above, and the very large ones below, which together give rise to the outermost embryonic envelope, the serosa.
- Fig. 3. Egg (G) of Cochlidion limacodes HUFN. about 1 day old.  $\times$  70. Blastoderm stage.
- Fig. 4. Egg (**B**) of *Cochlidion limacodes* HUFN. about 1 day old—estimated to be about 14 hours old.  $\times$  70. The broad, band-shaped germ band across has been formed. The rounded ends are seen to overlap the dorsal aspect of the egg. On the right the serosa is about to close, the 2 serosa strips lying quite close to each other.

## Plate I





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## Plate II.

- Fig. 5. Egg (**B**) of *Cochlidion limacodes* HUFN, about 1 day old—age estimated at about 17 hours.  $\times$  70. The sunken, tongue-shaped germ band lying across is plainly visible. To the right of it the 2 yolk strips have not yet reached each other.
- Fig. 6. Egg (C) of *Cochlidion limacodes* HUFN, about 1 day old—exactly  $17^{1/2}$  hours.  $\times$  70. The somewhat shortened tongue-shaped germ band is deeper immersed in the yolk which has undergone secondary cleavage into numerous yolk territories.
- Fig. 7. Egg (A) of *Cochlidion limacodes* HUFN, about 1 day old—estimated age some 17 hours. × 70. The germ band, which is deeply immersed in the yolk, is vaguely double-tongued in shape. The secondary cleavage of the yolk as well as "the outer fine-grained zone" are plainly visible.
- Fig. 8. Egg (F) of *Cochlidion limacodes* HUFN. about 1 day old.  $\times$  70. The deeply immersed germ band has here distinctly the shape of a double tongue, owing to the rounded, expanded ends. The yolk territories and the outer fine-grained zone are also very conspicuous in this egg.





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#### Plate III.

- Fig. 9. Egg (C) of *Cochlidion limacodes* HUFN, about 1 day old—exactly  $191/_2$  hours.  $\times$  70. The germ band, deeply immersed in the yolk, has become still shorter. The yolk territories are very distinct.
- Fig. 10. Egg (C) of *Cochlidion limacodes* HUFN, about 1 day old—exactly 21 hours.  $\times$  70. The deeply immersed germ band has now assumed the shape of a pear. Here too the yolk territories are very conspicuous.
- Fig. 11. Egg (F) of *Cochlidion limacodes* HUFN, about 1 day old,  $\times$  70. The much shortened germ band is pear-shaped with an indentation below.
- Fig. 12. Egg (F) of *Cochlidion limacodes* HUFN, about 1 day old.  $\times$  70. The germ band shows a further growth in height by which it has become more slenderly pear-shaped. Note in this as well as the previous figure the very distinct yolk territories, which often contain few very large fatty droplets.





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## Plate IV.

- Fig. 13. Egg (**F**) of *Cochlidion limacodes* HUFN, about 1 day old.  $\times$  70. The pear-shaped germ band is now opening above, a head part becoming distinctly visible to the left.
- Fig. 14. Egg ( $\mathbf{F}$ ) of *Cochlidion limacodes* HUFN, about 1 day old.  $\times$  70. The embryonic rudiment, which has grown much, is now elongate oval and has settled on its side. A head- and tail part are in course of differentiation, the former with an indication of head lobes. In this as well as the previous figure it is seen that many of the fatty globules in the yolk territories have fused into one large drop; the fine-grained zone too is very distinct in both cases.
- Fig. 15. Egg (**H**) of *Cochlidion limacodes* HUFN. about 1 day old.  $\times$  70. The embryonic rudiment which shows further elongation, has now assumed the form of a hoop, and a "head", a "trunk", and a spiral "tail" can be plainly distinguished.
- Fig. 16. Egg (**B**) of *Cochlidion limacodes* HUFN, about 2 days old—estimated at  $35^{1}_{/4}$  hours,  $\times$  70. The embryo is worm-like and closely coiled up in a spiral, particularly the tail part. The left head lobe, the fine-grained zone, and the segmentation along the dorsum are distinctly visible.







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#### Plate V.

- Fig. 17. Egg (C) of *Cochlidion limacodes* HUFN. about 2 days old—exactly 40 hours.  $\times$  70. The embryo is worm-like and closely coiled up in a spiral, especially the tail part. The 2 head lobes and the segmentation are very plain.
- Fig. 18. Egg (C) of *Cochlidion limacodes* HUFN, about 2 days old—exactly  $44^{1/4}$  hours old.  $\times$  70. The shape of the embryo as above, but with a somewhat stouter trunk. The yolk territories with the large fatty globules and the fine-grained zone are distinctly visible.
- Fig. 19. Egg (A) of *Cochlidion limacodes* HUFN, about 2 days old—estimated age  $361/_2$  hours.  $\times$  70. The embryo is worm-like and closely coiled up in a spiral, especially the tail. The left head lobe as well as the fine-grained zone can be plainly observed.
- Fig. 20. Egg (**B**) of *Cochlidion limacodes* HUFN, about 3 days old—estimated age  $63^{1/4}$  hours.  $\times$  70. The embryo is worm-like and closely coiled up in a spiral, especially the caudal part, which has developed a distinct "tail-plate". Both the right and the left head lobe are plainly present; this is also the case with the segmentation and the fine-grained zone.



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### Plate VI.

- Fig. 21. Egg (**C**) of *Cochlidion limacodes* HUFN, about 3 days old—exactly 62 hours.  $\times$  70. The embryo is worm-like and closely coiled up in a spiral, expecially the caudal part, which has developed a distinct "tail-plate". Note further the segmentation; also the yolk along the dorsum of the embryo, and finally the right head lobe, which is very conspicuous.
- Fig. 22. Egg (**A**) of *Cochlidion limacodes* HUFN. about 3 days old—estimated age about 65 hours.  $\times$  70. The embryo is worm-like and coiled up in a close spiral, especially the caudal part, which has developed a distinct "tail-plate". Some of the yolk can be observed in the head as well as along the dorsum. At the bottom of the egg, especially, the fine-grained zone is very distinct.
- Fig. 23. Egg (A) of *Cochlidion limacodes* HUFN, about 4 days old—estimated age about  $84^{1/2}$  hours.  $\times$  70. The previously spiral-shaped embryo has now become maggot-like. The tail part is very sharply marked off and provided with a caudal "hump" above and below. Both the segmentation and the fine-grained zone are very distinct.
- Fig. 24. Egg (**B**) of *Cochlidion limacodes* HUFN, about 4 days old—estimated age about  $83^{1}/_{4}$  hours.  $\times$  70. The embryo is maggot-like with distinct segmentation along the dorsal aspect.



Mi--------- Mik ---- Ch - - Yz Yz -Ch ----*Sk* -Chs Sg-- R F - Em - Em H1-- $H_{1}$ P1--Dt  $H_2 - H_2 - Sg = F - F$  $-H_2 = P_2 - F$  $P_7$ --Se Se - P<sub>2</sub> --0 --Dt 0---24

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#### Plate VII.

- Fig. 25. Egg (C) of *Cochlidion limacodes* HUFN. about 4 days old—exact age 90 hours.  $\times$  70. Note the maggot-like shape of the embryo and the distinct segmentation; the sharply marked off tail part with the 2 "humps" is also very conspicuous.
- Fig. 26. Egg (C) of *Cochlidion limacodes* HUFN, about 5 days old—exact age 110 hours.  $\times$  70. The embryo is accomplishing its second blastokinesis (II), by which the head, which is on the right side, gradually turns towards the micropyle. Note further the segmentation, the mid-intestine filled with yolk, and the rectum with the characteristic dilatation posteriorly.
- Fig. 27. Egg (A) of *Cochlidion limacodes* HUFN, about 5 days old—estimated age about  $108^{1/2}$  hours.  $\times$  70. The second blastokinesis of the embryo has been completed and the caterpillar-like animal is closely bent together from side to side. Note also the eyes, the esophagus, the mid-intestine and some characteristic spots along the dorsal aspect.
- Fig. 28. Egg (**B**) of *Cochlidion limacodes* HUFN, about 5 days old—estimated age about  $107^{1}/_{4}$  hours.  $\times$  70. The caterpillar-like embryo, bent strongly over to one side has the head deeply immersed in the yolk mass. Note the eyes, the esophagus, the mid-intestine, and the rectum with the dilatation posteriorly; further the segmentation and the dorsal spots.





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#### Plate VIII.

- Fig. 29. Egg (B) of Cochlidion limacodes HUFN, about 6 days old—estimated age about 131<sup>1</sup>/<sub>4</sub> hours. × 70. The embryo has increased considerably in size at the expense of the yolk, which does not even constitute half of the egg content now. The anterior end of the head is more distinctly discerned, and the eyes, esophagus, mid-intestine, rectum and the dorsal spots are plainly visible.
  Fig. 30. Egg (C) of Cochlidion limacodes HUFN, about 6 days old—exact age 134 hours. × 70. The embryo fills more than half of the egg and the posterior extremity now projects a little beyond the anterior margin of the head. The eyes, esophagus, mid-intestine, rectum with dilatation, as well as the dorsal spots are very plainly visible; the same is the case with the yolk territories which often
- Fig. 31. Egg (A) of *Cochlidion limacodes* HUFN, about 6 days old—estimated age 137 hours.  $\times$  70. The embryo fills about  $^2/_3$  of the egg, and the tip of the abdomen projects far beyond the anterior edge of the head. Note further the mandibles, the eyes, and the head capsule besides the mid-intestine and the dorsal spots; further the yolk territories with few and small fatty globules and the fine-grained zone.

contain several small fatty droplets.

Fig. 32. Egg (**B**) of *Cochlidion limacodes* HUFN, about 7 days old—estimated age about  $155^{1}/_{4}$  hours.  $\times$  70. The embryo constitutes about  $^{3}/_{4}$  of the egg content, and the yolk mass is chiefly present in the region in front of the animal's head. Besides the mandibles, the eyes, the mid-intestine, and the dorsal spots, the tracheae are now also plainly visible.





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## Plate IX.

- Fig. 33. Egg (C) of *Cochlidion limacodes* HUFN, about 7 days old—exact age 158 hours.  $\times$  70. The embryo has consumed the greater part of the yolk mass, and the tip of the abdomen projects a good way beyond the anterior edge of the head. Mandibles, eyes, tracheae, mid-intestine, and dorsal spots are plainly seen, the same applies to the remainder of the yolk territories.
- Fig. 34. Egg (A) of *Cochlidion limacodes* HUFN, about 7 days old—estimated age  $156^{1/2}$  hours.  $\times$  70. The larva is now fully developed and fills the greater part of the egg shell. The embryonic envelopes and the yolk have been consumed, and the animal is ready to emerge at any moment. The mandibles, eyes, head capsule, mid-intestine, and dorsal spots are plainly visible; in addition absorbed fatty droplets are seen in several places within the caterpillar.
- Fig. 35. Egg (**B**) of *Cochlidion limacodes* HUFN, about 8 days old—estimated age about  $179^{1/4}$  hours.  $\times$  70. The fully developed larva is emerging from the egg shell, which it has already pierced (the light, triangular hole on the right). The head, which is much retracted, is provided with distinct mandibles, eyes, and tracheae. Note also the head capsule, segmentation, and light-coloured tip of the abdomen.







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