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Studies on the Epidermal Structures of Birds

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V—Studies on the Epidermal Structures of Birds

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I—THE DEVELOPMENT OF FEATHERS

I—Introduction

Studies on the development of feathers have as yet provided only inadequate answers to many fundamental problems. The evolution of feathers themselves and their relation to reptilian scales are still matters of hypothesis, and will remain so until more crucial information is forthcoming. Again, the phylogenetic sequence of events relating embryonic to definitive feathers is inconclusive, and it cannot be said with certainty whether the embryonic down feather is a secondary modification of the ordinary contour feather, or whether the latter has been evolved from a primitive down-like primary feather.

Investigations upon the development of epidermal structures in general, and of feathers in particular, were favoured by workers in Germany during the last century, culminating in the classical work of Davies (1889) on the development of feathers in the pigeon. His views still form the accepted basis for work on plumage, but recent knowledge has necessitated a re-examination of them. (Lillie and Juhn, 1932.)

In 1902 Strong wrote an admirable account of the definitive feather in *Sterna hirundo*, which agrees fundamentally with the conclusions reached by Davies, although reference is made only to the latter for the development of the embryo feather.

Cossar Ewart's work on the nestling feathers of the Mallard (1921) was followed by Lamont's study of the development of embryo feathers in the Indian Runner duckling (1925). As the former dealt with external characters, and the latter took stages at intervals of several days and rarely sectioned them, knowledge concerning the structure and development of the embryo feather has advanced little since Davies's time.

Modern work on plumage reactions in relation to internal secretions has emphasized the need for a reconsideration of feather development, if satisfactory theories are to be formulated to explain the accumulated experimental data.

In this paper the sequence of plumage has been observed in several types of birds, and the histological development has been followed from the embryo to the adult.

II—Material and Methods

(*a*) *Embryology of Nestling Feathers*

For this study, stages were taken at daily intervals during the incubation period of the Domestic Fowl and two varieties of duckling (Khaki Campbell and Aylesbury). The Chinese Gosling was also used, but owing to poor fertility only four stages were obtained. These, however, were at convenient intervals for comparison.

The embryos were fixed in 10% formalin, picro-nitric or Bouin's solution. The latter, proving the most successful fixative was the most generally used, and was always heated to incubator temperature before immersion of the embryo.

(*b*) *Definitive Feathers*

Two varieties of the domestic fowl (Rhode Island Red, and the offspring of a Black Leghorn X Light Sussex) were reared in a bachelor brooder and watched for changes in plumage. At the age of eight weeks they were transferred to folding units out of doors. Stages were taken at intervals of two weeks in the former, and weekly in the latter over a period of eighteen weeks.

In addition, a series of Khaki Campbell ducklings (from one to eight weeks old) and a series of nestling starlings were obtained for comparison.

The skins of these birds were fixed in Bouin's solution after representative feathers had been plucked for reference.

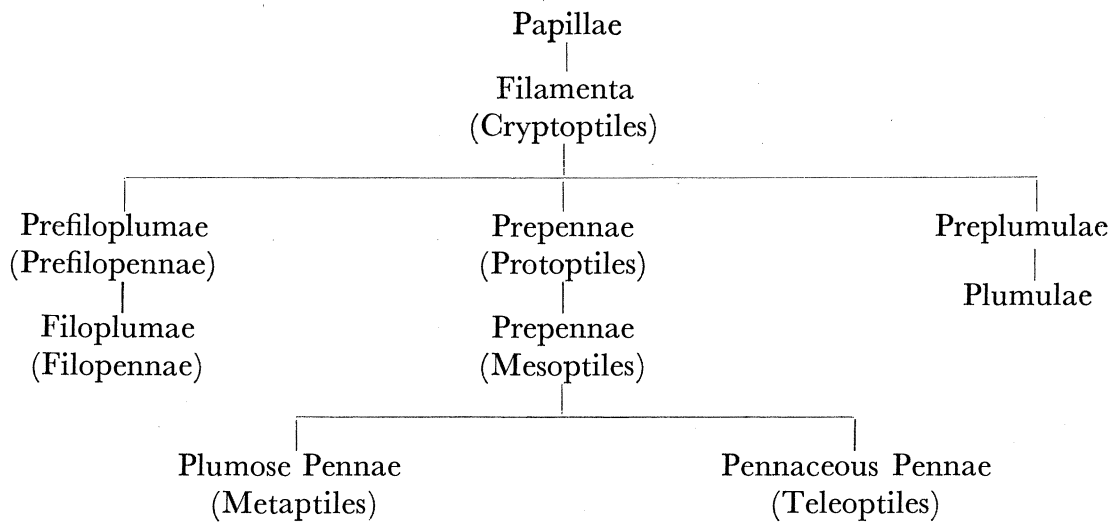
Portions of the skin were embedded in celloidin followed by paraffin wax. It was found necessary to embed the embryo feathers for two weeks in celloidin and the definitive feathers for at least three weeks, to get good infiltration. In the next stage, the small feathers were embedded rapidly in wax in the usual way, but larger feathers were left in a mixture of chloroform and wax for 24 hours at 40° C before embedding rapidly at a higher temperature.

Sections were cut from 6–8μ in thickness, and stained by the short method of iron haematoxylin (*i.e.*, only 15 minutes in both mordant and stain) otherwise it was impossible to differentiate the cornified parts successfully. Orange G, picro-indigo-carmine or picro-fuchsin were used as counter stains. In older feathers the last-named gave the most pleasing effect.

Whole feathers from the different pterylae were always mounted dry for comparison.

III—The Sequence of Plumage in Birds

It will be as well at the outset to ensure a correct interpretation of terms used in this work, by reference to the table compiled by Ewart (1921), showing the sequence of feathers in a hypothetical Avian type from embryo to adult.



Prefiloplumae, preplumulae and prepennae are typical constituents of nestling down, and will therefore be collectively spoken of as *Neossoptiles*; and instead of restricting the term *Teleoptiles* to Pennaceous Pennae as in the above table, it will be used for all types of adult feathers, as originally used by Gadow (1907), Pycraft (1907) and Newton (1896).

Hence the above table can be modified to give a clearer representation of what is usually found in feather sequence from the embryo to the adult bird.

Papillae

Filamenta

Neossoptiles (nestling feathers, consisting of prepennae, prefiloplumae and preplumulae).

Two generations are known :

(*a*) Protoptiles—prepennae, prefiloplumae and preplumulae.

(*b*) Mesoptiles—as yet known to be derived from prepennae only, or to arise directly.

Teleoptiles (adult feathers).

Three types exist :

(*a*) Plumulae—*i.e.*, down feathers.

(*b*) Filoplumae.

(*c*) Pennae—*i.e.*, quill feathers, whether plumose or pennaceous.

Of all the types of fledglings examined in the present investigation, only in the duck are three types of neossoptiles developed. Prepennae and preplumulae are present in the goose, and prepennae only in the fowl, on hatching. In birds with nidicolous young (*e.g.*, starlings) prepennae are present, but sparse and restricted in area.

Fowls—For reasons to be discussed later (*see* p. 169), it is assumed that the coat worn by the newly-hatched chick consists of protoptiles, the mesoptiles having been suppressed.

While it must be remembered that great variability exists between members of a flock, the order of appearance of feathers in the different pterylae is invariably constant, the difference being one of time only. On hatching, all except the most proximal primary and secondary flight feathers can usually be seen, together with the tectrices majores. These teleoptiles all bear protoptiles at their tips, which are shed during the first week after hatching. The remaining wing feathers (tectrices media and minores) and some body feathers in each area usually appear during the first month. The latter develop in the following order :—thigh, breast and neck, head, back, abdomen. In the Rhode Island Red fowl, tail teleoptiles are found in females during the second week, but the tail of the male is retarded in development. At 6 weeks, some teleoptiles are present in each pteryla, and the wings are completely fledged. Few bear protoptiles although many are still present between the teleoptiles, the latter being replaced completely by the end of the eighth week.

From the age of eight weeks to four months, the feathers are gradually shed from all pterylae and replaced by typical adult feathers with mature sexual characteristics.

After the assumption of this final type of plumage, which can only be changed by an improperly balanced endocrine condition (Torrey and Horning 1925), the gradual moulting ceases, and the feathers replacing those shed in the annual moult are similar in every respect to their predecessors, except under pathological conditions.

Ducklings—The main difference between the method of adoption of the teleoptile coat in the fowl and the duck appears to be correlated with differences in the habits of the birds. In the former, where the wings are of greater use than the tail, the first generation of feathers is replaced by the teleoptiles at an early age. In ducklings, however, where the wings are of little use, they are greatly retarded in development, and teleoptiles do not appear until the fifth week after hatching. The tail, which is of some advantage in swimming, shows enormous elongation of the calamus of the protoptiles during the first and second weeks after hatching. These are borne upon the tips of the teleoptiles, which appear during the fourth week, until the sixth week. On the rest of the body teleoptiles appear on the back, legs, and head during the third week, and in other pterylae about the sixth week. By the seventh week they are widespread, and most protoptiles have been shed.

The remiges, after their retarded start, grow very rapidly and during the eighth week surpass the greater wing coverts in size. No trace of the mesoptiles found in the ninth pair of rectrices of the Mallard duck (Ewart 1921) have so far been found in the Khaki Campbell duckling.

Thus the neossoptiles present in all nidifugous birds (*i.e.*, young completely clothed with nestling down) and to a lesser extent in nidicolous young, are replaced by teleoptiles during the nestling period. The order of formation of the adult

feathers differs in birds of different habits, *e.g.*, Galliformes and Passeriformes show precocious development of remiges and Anseriformes of rectrices. On the rest of the body, teleoptiles appear in the following order : thigh, breast and neck, head, back, abdomen.

IV—Nestling Feathers

(*a*) *A Comparison of Neossoptiles and Plumulae*

The development of the neossoptiles in the duck, goose, and fowl is more easily understood after a comparison of the external features of the various types of down.

Feathers first appear externally as minute opaque spots on the otherwise transparent skin of the spinal and femoral tracts of the embryo on the seventh day in the chick, the ninth day in the duckling, and about the twelfth day in the gosling. It was impossible to gauge the exact age of the Passerine embryos studied, but the protoptiles appear in the different pterylae in the same order as in the nidifugous types. The feather papillae are arranged in diagonal rows, in such a manner that some three days later, the backwardly growing rudiments overlap, as do reptilian scales. Feathers appear on the rest of the body a few days after their appearance on the back, but their development on the wings is delayed until the tenth and thirteenth days in the chick and duckling respectively.

At about this time, small papillae may be seen forming a circle round the base of each larger papilla in the duckling and gosling, but absent in the chick. These are the preplumulae from whose follicles the plumules of the next generation of feathers will be developed. The larger feather filaments are the prepennae. In the duckling the preplumulae may be secondarily divided on account of their size. The smaller papillae, which appear last, are the predecessors of the filoplumes.

On hatching, the young birds are clothed in long backwardly projecting filaments, all of which have reached the same state of development in spite of their different times of appearance. In a few hours the filaments are dry, and then if friction, however slight, is applied, the sheath splits and the barbs splay out forming a dense covering of soft, warm down.

In the duckling, various types of down may be distinguished, differing in size and texture, but having the same general plan. According to Newton (1896) the neossoptiles of Anseres have a " feeble rhachis " bearing " all the biserially radiated rami, forming feathers, which clearly resemble the down of mature birds, and are devoid of an aftershaft". In reality, the prepennae of a duckling consist of both shaft and aftershaft, which are comparatively well developed, fig. 28, Plate 19. These unite near the base forming a definite calamus. Barbs, or rami, are arranged biserially as Newton says, but along both rhachis and hyporhachis, the former ending in two long barbs, which are characteristic of the duckling. The prepennae of the head are small, and with no caps in the short calamus. On the breast and back they are much longer, and usually have at least one cap on hatching ; while the tail prepennae are very long and stiff, with bristly barbs and a long calamus

containing several caps. The barbules are also arranged biserially along each barb of both shaft and aftershaft, but a superficial spiral arrangement is obtained by the twisting of the proximal cell of each barbule.

The adult down of a duck is fundamentally similar to the nestling down. Numerous slender barbs, only slightly longer than the barbs of the prepennae but much longer than those of the preplumulae, join forming a long rhachis in the shaft; while the aftershaft consists of fewer barbs, which are equal in length to those of the main shaft, and which join to form a definite hyporhachis. The chief structural difference lies in the restriction of cilia to the distal part of the barbule, while the next two or three cells bear pyramidal swellings, and the proximal cells are uniformly elongated with no visible swellings at the nodes, fig. 29*e*, Plate 20.

The neossoptiles of a newly-hatched gosling are essentially similar to those of a duckling. The rhachis is shorter in the gosling, ending in two very long, slender barbs, and the cilia from the nodes of the barbules are much longer than in the duckling, especially the distal cilia, fig. 29*c*, Plate 20.

The adult down of the goose consists of a veritable forest of barbs, but structurally differs from the nestling down only in the presence of pyramidal swellings between the elongated basal cells of the barbule and the ciliated distal cells, fig. 29*f*, Plate 20. This condition is very similar to that of the duckling, fig. 29*e*, Plate 20.

The neossoptiles of the chick, fig. 27, Plate 19, present a totally different appearance from those of the duckling and gosling. The rhachis is here so short that from a cursory glance the feather appears umbelliform. Most writers mention one barb being longer and stronger than the rest, and consequently regard it as a rhachis. But all the barbs are approximately of equal length, and the short rhachis is formed by the fusion of two of these, while others join lower down. The aftershaft is well developed, as in ducklings, consisting of barbs almost as long as the shaft; but the calamus is very short and contains no caps. The barbules are not twisted, but the basal cell is very broad and each subsequent cell is swollen at the node in receiving the narrow proximal end of the next cell; and no cilia are present, fig. 29*a*, Plate 20.

Unlike the duckling, there is no distinction except in size between neossoptiles from any part of the body, the tail feathers being as soft and slight as those on the breast.

A few days before hatching, the filaments of the protoptiles of the remiges are pushed out from their follicles by the rapidly growing teleoptiles, fig. 27, Plate 19. This, as will be discussed later, is due to continuity between a few barbs of the teleoptile and the calamus of the protoptile. The teleoptile consists of a very definite rhachis, which branches alternately at close intervals. The branch barbs are progressively shorter from the base of the feather upwards, so that they all end at the same level. Near the base about four pairs of shorter barbs representing the aftershaft, with no definite rhachis, join the shaft, forming a short calamus. Each barb bears biserially arranged barbules extending along its whole length, thus no long slender filaments form the termination of the barb as in protoptiles. The single barbule of a teleoptile represented in fig. 29*b*, Plate 20, shows great specialization

compared with the protoptile barbule in fig. 29*a*. The basal cell is still swollen, but its distal end bears a curved projection. This is emphasized in the next two or three cells, where the projection forms a typical barbicel. Thus a slight amount of interlocking is possible in the tip of the teleoptile.

The adult down of the fowl differs from the nestling down in having a relatively longer rhachis, so that the feather loses the umbelliform appearance of its predecessor.

The plumule of the fowl is really comparable to the preplumule, which is not developed in the chick, but as already shown for the duckling, the preplumule is only a minute edition of the prepenna. It is not comparable to the teleoptile described on page 149, which is here classed among nestling feathers, as it is developed during the incubation period, but the structure of which forms the fundamental basis of all teleoptiles, whether contour or quill feathers.

(*b*) *The Development of Protoptiles*

In early stages of development, the epidermis consists of two layers, an upper epitrichium with elongated nuclei, and a lower or inner stratum germinativum (stratum Malpighii or rete mucosum) consisting of cells with rounded nuclei. The latter layer is by far the most important and ultimately gives rise to all the layers present in the adult skin, and to the derivatives of the epidermis such as scales or feathers.

By continued proliferation, the stratum Malpighii gives rise to several layers, the lowermost of which has typically cylindrical nuclei and hence bears the name of stratum cylindricum. Above this are the numerous rounded cells of the stratum intermedium which effectually cut off the epitrichium from nourishment, so that it atrophies, becoming replaced by the stratum corneum.

All the layers of the epidermis of the adult bird therefore are directly derived from the lower of the two layers found in the embryo.

(i) Ducklings

The development of protoptiles in the duck has been studied by comparing stages during the incubation period at intervals of one day, starting from the eighth day.

8 *days*—The site of the feather embryo is visible microscopically in the spinal and femoral tracts. At intervals in transverse sections of the skin from these parts, a few of the rounded nuclei of the Malpighian layer appear slightly elongated, with their long axes vertical, *i.e.*, at right angles to the axes of the nuclei of the epitrichial layer.

9 *days*—The feather germs may be seen macroscopically as small white patches in the spinal and femoral tracts. The elongation of cells of the stratum Malpighii is even more apparent, and certain cells of the epitrichial layer have divided, the daughter cells passing down to the position of the future intermediate cells.

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This migration of epitrichial cells is interesting, as according to Davies (1889), the epitrichium takes but a passive part in feather development, remaining as a very thin covering external to the sheath, and soon being stretched to the breaking point.

10 *days*—The feather germs are visible macroscopically over the whole body. In an Aylesbury duckling of the tenth day, the whole of the feather germ is slightly depressed, fig. 1, Plate 16, as in early stages of hair development, but in the Khaki Campbell duckling it is correspondingly elevated. The epitrichial layer no longer contains a single row of nuclei, but two less regular rows, which are distinct from the nuclei of the intermediate cells (formed by the rapid division of the Malpighian layer cells) in the direction of their long axes.

11 *days*—The feather germ is distinctly elevated, and has increased to more than twice the size on the previous day, as may be seen by comparing figs. 1 and 3. The epitrichial layer is less compact than formerly, and is obviously dividing less rapidly than the Malpighian layer, which is now many cells wide. On one side of the feather germ, the intermediate cells are much less compact, and at that side the slope is more gradual, fig. 2, Plate 16. Davies (1889) attributes this to more active division at the steep sloped (*i.e.*, the anterior) side of the feather, resulting in a bending backwards.

A clear space between the dermis and Malpighian layer, which persists throughout the development of the feather, is possibly analogous with the " basal membrane " noted by Strong (1902*a*). High power microscopic examination reveals this to be composed of the bases of cells of the stratum Malpighii whose nuclei are uniformly withdrawn toward the periphery, and so agrees with the observation of Jeffries (1883).

Below the concentrated dermis of the feather germ are always two or more blood vessels. A distinct sheath envelops the feather before the next day of incubation, which according to Lamont (1925) is one cell deep and composed of flattened cells " derived from the epitrichium or outer layer of the epidermis". In both Khaki Campbell and Aylesbury ducklings, however, the sheath may be three or more layers wide, consisting of the epitrichial layer and its derivatives together with some intermediate cells, which have become secondarily elongated in the same direction as the cells of the epitrichial layer. This agrees with Davies's observations on the pigeon (1889).

The innermost layer of intermediate cells has now definite elongated nuclei, and unlike Davies's description of the pigeon, this shape is more or less retained until the final stages of development.

12—13 *days*—A tendency for the intermediate cells to become arranged in groups is apparent, at least in the Aylesbury duckling, although this process is delayed until the following day in the Khaki Campbell duckling.

14 *days*—Longitudinal sections on this day show that the base of the feather germ is slightly sunken below the general level of the skin. This is due to a downgrowth of the epidermis of the feather papilla and the surrounding skin, forming the feather follicle. The pulp in this region is very concentrated, and as the follicle becomes

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deeper, this dense pulp moves downwards so that it is always situated near the base of the feather, *i.e.*, the growing region. The blood supply consists of a central vessel surrounded by a circle of smaller vessels, but this is more clearly defined in later stages of development.

The grouping of intermediate cells, forming the so-called ridges seen in transverse sections on the 12th and 13th days of incubation, is complete. The stratum cylindricum has passed outwards between the ridges, eventually meeting the sheath cells, and thus continuing to form a boundary between the pulp and intermediate cells. According to DAVIES, the cylinder cells may extend between the sheath and intermediate cells of each ridge, but this certainly does not occur in the duck, goose or fowl.

The ridges are of unequal size, the larger ones tending to be concentrated to one side of the feather embryo, fig. 6, Plate 16, and in the largest of these in the Khaki Campbell duckling, pigment cells are present. Fig. 5, Plate 16, shows the ridges between the lines *a* and *b* in fig. 6 highly magnified, and the detailed structure of the pigment cells is clear.

15 *days*—During the late 14th and 15th days of incubation, the intermediate cells of each ridge become separated into three groups, the so-called "median" and "lateral plates". The median plate consists of small, rounded cells, which remain scattered about the central part of each ridge, figs. 2 and 8, Plates 16 and 17, until the 16–17th days, when they withdraw towards the apex of the ridge, forming a dense mass—the future barb, fig. 9, Plate 17. The lateral plates undergo a more rapid development. During the 14th day, in transverse section they appear to consist of a string of flattened cells on either side of the median plate, but each cell is really quite separate from its neighbour. This becomes emphasized as development proceeds, until it is clear that each is a cross-section of a developing barbule. Fig. 29*d*, Plate 20, shows the single barbule from a newly hatched duckling, and the longitudinal section in fig. 8 illustrates how the fundamental structure of the barbule (a single row of cells placed end to end) is laid down as early as the 15th day of incubation.

16 *days*—The feather follicles are comparatively deep, fig. 7, Plate 17, and horizontal sections show that each is at the "knot of a mesh of muscle network", which LAMONT (1925) also noticed in a 17 days embryo Indian Runner duckling. At the distal end of a feather embryo about this time, signs of approaching cornification are to be seen in the shrinkage of cylinder cells from between adjacent ridges. This is foreshadowed in Aylesbury ducklings of the 15th day, in which as fig. 8 shows, the separation of barbules from barbs and of cylinder cells from barbules has commenced.

17 *days*—During the 16th and 17th days the barbs are formed by cells of the median plate in each ridge moving inwards towards the apex. Hence many newly formed barbs are pear-shaped in cross-section, with the pointed end consisting of traces of the last cells incorporated into the barb. Many authors mention "residual cells", which occupy spaces in the ridges, but these are very rare in the Khaki Campbell and Aylesbury ducklings.

18 *days*—The pulp of the distal end is now composed of very scattered nuclei connected by cytoplasmic strands, with a few blood vessels, and bounded by the cylinder cell layer which differs greatly from its appearance in earlier stages, fig. 9. The once distinct layer of compact cylindrical cells now forms a nucleated string with no definitely shaped cells, which is, as STRONG suggests, probably due to the great longitudinal growth of barbs and barbules. The barbs are differentiated into a central medulla, consisting of large vacuolated cells, and a thick-walled cortex of quadrangular cells with densely-staining properties. One barb is relatively large, with progressively smaller barbs on either side and very small barbs opposite. As these very small barbs are absent from the tip of the feather, they must represent the distal ends of shorter barbs. All pigment cells proper have disappeared before the barbs are formed, *i.e.*, the pigment granules have been distributed to the outermost cells of the cortex and all the barbule cells. Presumably the amoeboid processes of the original pigment cells are withdrawn, when their function is completed, and the cell which remains behaves as, and is indistinguishable from, an ordinary intermediate cell. The barbules are attached to the barbs near their apices, by cells elongated radially instead of lengthwise as are the barbule cells distally. This results in the flattened bases of the barbules seen in fig. 29, Plate 20.

The sheath is partly cornified, having the characteristic " layered " appearance, and no distinct cells being visible, but very occasional nuclei. The outermost epitrichial layers are now quite indistinct, being broken at intervals as early as the 15th day of incubation, and with long spaces between nuclei.

19 *days*—Preparations for cornification have proceeded still further, in that the cylinder cell layer has withdrawn from between ridges, and now forms a more or less circular boundary of the pulp, in transverse section. On this day, too, are the first signs of branching of the largest barb seen. This strongest barb is regarded as the rhachis.

20 *days*—The central cells of the rhachis appear greatly enlarged and have begun to become vacuolated, the comparatively small nuclei remaining attached by cytoplasm to the cell wall for some time. The smaller barbs become medullated soon after the rhachis and in a similar manner. The rhachis frequently branches, as more rarely do some of the small barbs. This " branching " is really fusion of barbs, for, as the feather grows from the base, the tips of the barbs are formed first. The cortical cells nearest the periphery are the first to fuse, the rest of the barb remaining quite distinct for some time, and after complete fusion, the cortical cells remain separating the medulla into two parts for a considerable distance downwards. From this method of fusion it would thus be impossible for a feather to have a spiral arrangement of barbs ; and the method of attachment of the barbules permits of no other than lateral arrangement of barbules, although spiral arrangement is superficially obtained in the nestling and adult down of Anseriformes by the twisting of the basal cells, figs. 29*c–f*, Plate 20.

21 *days*—Fusion of barbs occurs very rapidly in later stages of development, culminating in the calamus about the 24th day. Transverse sections near the base

of an embryo feather on the 21st day show the outer cortical cells of the rhachis extending almost completely round the pulp, which is very dense in this region, and which is separated from the intermediate cells by clearly defined cylindrical cells. The intermediate cells are arranged in two groups on the inside of the thickest part of the rhachis. This is seen in succeeding sections to be due to the recent fusion of two branch barbs. The whole of the tip of the feather is cornified, while only the peripheral cortical cells near the base show signs of approaching cornification. This process always commences by the coalescence of cells radially, so that the elongated nuclei seem to lie in a " layered " material. In later stages, the layered appearance is seen to precede the complete coalescence of cells, so that the nuclei now lie in a uniform matrix. Meanwhile the cell walls have become remarkably thick, literally squeezing the nuclei out of existence, so that their position is represented only by a short line (probably a minute cavity) when cornification is complete. In the vacuolated medullary cells this cavity is relatively large, as perhaps owing to vacuolation taking place comparatively early in development, the cell walls are only slightly thickened; but no line of demarcation separates them from the cortical cells. LAMONT's definition of cornification as " loss of cellular structure " is thus singularly applicable.

Meanwhile the feather follicle has become gradually deeper, owing to the continued downgrowth of the epidermis. Continuity is clearly seen between the stratum Malpighii of the follicle and of the base of the feather; but the epitrichial layers of both feather and follicle are indistinct. This is due to the proliferation of the latter layers giving rise to a continuous sheath round the base of the feather, but higher up, the narrow follicle sheath is separate from the broader feather sheath. As the epidermis of the follicle does not divide into intermediate cells, and yet a definite sheath is formed, this seems to be evidence in favour of the theory that the epitrichium is largely responsible for that structure in embryo feathers.

Transverse sections show the deepest part of the follicle to be in the form of a double crescent of epidermal cells, with the convexity nearest the surface of the skin, including the combined sheaths of the feather and its follicle. Below this, the characteristic whorls of dermal nuclei with very small blood vessels are seen, and the dermis surrounding the whole of the follicle has a similar whorled appearance.

22–24 *days*—Sections from the base of the feather show the gradual fusion of large barbs with the rhachis on the upper side of the feather germ (*i.e.*, the side nearest the upper surface of the skin, when the feather is in its follicle). The rapid fusion of the smaller barbs on the opposite side forming the short hyporhachis, terminates in the lateral fusion of the shaft and aftershaft, giving rise to a short calamus, fig. 19, Plate 18. Cornification takes place very slowly at points of fusion, fig. 20, Plate 18, for there the cortex is always deeply stained instead of clear as when completely cornified.

Figs. 17–21, Plate 18, are transverse sections taken at different levels of a feather of a 24 days Khaki Campbell duckling. Fig. 17 shows the crescentic base, with whorls of dermal cells and the characteristic blood supply; fig. 18 is in the region

of the calamus; fig. 19 shows the fusion of the rhachis with the hyporhachis; fig. 20 shows the hyporhachis to be much shorter than the rhachis, as the former has almost disappeared from the section while the latter is still very large; and fig. 21 is near the tip of the feather where the aftershaft is represented only by the tips of its barbs, and the rhachis has branched repeatedly, but without any great reduction in size. The withdrawal of the pulp and cylinder cell layer can also be traced in this series. Figs. 22–26, Plate 18, represent sections taken from corresponding levels in a preplumule of the same age. It will be noticed that the structure is exactly the same as in the prepenna, except for the reduction in size and in the number of barbs. The minute feather germs of the duckling, which probably are prefiloplumes, also have the same structure as the prepennae, but even smaller in size than the preplumulae.

The feather is now structurally complete, except for the full length of the calamus but this is obtained by cornification of the intermediate cells below the short calamus found on the 24th day. Before hatching, when the feather sheath splits and the barbs splay out, the pulp is withdrawn to the base of the feather. This process is very gradual, commencing about the 15th day, when as fig. 8 shows, the pulp near the tip of the feather is relatively less dense, and the cylinder cells have begun to withdraw from between adjacent ridges. When the tip begins to cornify, the pulp (always bounded by cylinder cells) shrinks towards the centre, but generally remains attached by the cylinder cells, in one or two points, to the sheath between two barbs. It remains in this condition at the tip of the feather for several days, while the preliminary stages of withdrawal are taking place lower down, as cornification proceeds downwards. The crowded barbs and barbules push inwards to occupy the space vacated by the pulp, losing their regular arrangement. About three days before hatching, the pulp is entirely withdrawn from the tip, but is still present at the top of the calamus, where the first "cap" is formed. The formation of feather caps is discussed in the development of definitive feathers (p. 163).

28*th day*—On hatching the feather filaments dry, but friction, however slight, is necessary for the sheath to split. When once the dried sheath is slightly torn, the crowded barbs and barbules within push outwards, thus tearing away the sheath to the base and presenting the appearance seen in the protoptile of fig. 28, Plate 19.

(ii) Goslings

Only four stages of development of the goose were obtainable, aged 11, 14, 25, and 27 days respectively. The last was dead when opened on the 28th day, but the material taken from it did not appear to have suffered by the delayed fixation.

The development of protoptiles in a goose is essentially similar to that of a duck, differing only in very small points.

11 *days*—Although the 11 days embryo showed no signs of feather papillae in any part of the body (skin being examined from the spinal tract, tail, and wing), development must have been relatively more rapid than in the duck, where feathers

first show in sections of 8 days embryos. On hatching on the 28th day, the feather filaments of the gosling are fully formed and much larger and stronger than those of the duckling.

14 *days*—The embryo feather corresponds to a 12 days duckling or a 10 days chick feather. It is elevated above the general level of the skin, and with a steeper slope on one side. This is presumably before the backward growth has commenced, and correlated with it is the absence of a follicle. Near the tip of the feather, groups of intermediate cells are seen, merging lower down into a common band. The cylinder cell layer is indistinguishable in this region ; apparently it does not appear until the intermediate cells have become arranged in groups. This supports the theory that it is the intermediate cells, which actually group themselves together, instead of being cut into ridges by the ingrowth of the cylinder cells.

Pigment cells containing very coarse granules, unlike the fine-grained pigment of the duck, are present near the apices of the ridges, and among the outermost intermediate cells near the base of the feather.

The stratum Malpighii is separated from the pulp by the bases of the cells forming a clear space (the " basal membrane ") through the withdrawal of nuclei towards the periphery. A definite sheath, several rows in thickness, is present at this stage, and sections through the tip of the feather show this to join above the termination of the ridges.

The pulp is concentrated only at the base, and especially on the steeper side of the feather papilla. Near the tip of the feather it consists of very scattered nuclei connected by cytoplasmic threads, which indicates a comparatively earlier withdrawal of pulp than in the duck protoptile.

25 *days*—The feather follicle is well developed and the space between feather and follicle is occupied by a common sheath about six rows of cells in width. Cornification is in full swing in the upper part of the feather, the cylinder cells having withdrawn from between adjacent ridges, and now enclosing a small central pulp. Two large medullated and cornified barbs are present in the upper part of the feather filament, with progressively smaller barbs on either side. Lower down, these two large barbs fuse, forming the rhachis with which one or two of the neighbouring smaller barbs also fuse. In this way the feather consists near the base of a very broad rhachis, while the ventral part is still divided into smaller barbs. These rapidly fuse, resulting in the narrow hyporhachis or aftershaft. One side of this fuses with the rhachis, while a few barbs of the aftershaft are still free on the other side. This one-sided fusion seems characteristic of geese, although it occasionally occurs in the fowl. The lower region of the feather germ is still uncornified, and consequently the fusion of barbs can be clearly seen.

Each barb is circular in section, like the barbs of the duckling protoptile, and consists of elongated cells nearest the periphery, and more rounded cells towards the apex. During fusion the elongated cells join the circles of intermediate cells, which are not distinguishable except in their slightly greater width from the sheath cells. No barbules are present in this region. The calamus is succeeded by a broad

band of intermediate cells in the base of the feather, and unlike the condition on the 14th day, the cylinder cell layer is here quite distinct.

The musculature between the feather follicles is very definite, strands running from the top of one follicle to the base of the next. Traces seem to pass between both preplumulae and prepennae, but never so distinctly as between two prepennae.

28 *days*—The dead specimen of a Chinese gosling opened on the 28th day is covered with fully formed feathers, similar in structure to those described for the 25th day, but completely cornified (except for points of fusion), and with a longer calamus. The latter has the same crescentic base in transverse section as a duckling protoptile, but with the convexity facing the side, instead of towards the surface of the skin. This may be associated with the fact that the follicle of a goose protoptile is much deeper and correspondingly more oblique than that of a fowl. Pulp is still present, although withdrawn towards the centre in the tip of the feather.

The most striking feature of this gosling is the clearly defined blood system ; the course of the single central blood vessel surrounded by a circle of anastomosing capillaries is easily followed. In the definitive feather, the central vessel is clearly seen to be an arteriole, and the anastomosing vessels in the periphery are venules.

(iii) Chickens

6 *days*—Feather papillae appear on the 6th day of incubation in the chick, when sections of the skin of the spinal and femoral regions show similar concentrations of the dermis, and slight elongations of the Malpighian cells as on the 8th day in the duck and about the 12th day in the gosling. The developmental stages are similar to those described for the duckling, although always more precocious (correlated with earlier hatching).

10 *days*—The downgrowth of the epidermis which culminates in the feather follicle begins ; and the following day sees the formation of ridges. In the duckling and gosling these processes are reversed and the follicle is formed about two days after the ridges.

11 *days*—In sections of an embryo on this day of incubation, pigment is found scattered about the epidermis surrounding the follicle, the walls of the follicle and the intermediate cells of the feather germ. Thus pigment must be of endogenous origin, as it is found in the epithelium with no previous traces in the dermis.

12–16 *days*—The regrouping of intermediate cells to form barbules and barbs occurs on the 12th day, and by the 16th day the barbs are well developed and medullated, while the barbules are very crowded. The shape of the barbs is characteristic, being oval in cross-section, fig. 11, Plate 17, unlike the rounded barbs of duck and goose, fig. 9, Plate 17. But, as in the duck in early stages of development of the barbs, a point may be seen projecting from the side opposite the apex of the ridge, which is attached to cytoplasmic strands, as though intermediate cells have lately been drawn into the barb. There are very few medullary cells in these barbs, the central spaces rarely being divided.

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The chief difference in the development of protoptiles in ducks and fowls lies in the fact that the former have a well developed rhachis and hyporhachis, while the latter have a very short rhachis, and therefore fusion of barbs does not occur until late stages of development (about the 19th day). Consequently, for a considerable distance from the tip of the feather, the uniform barbs (of which only about three in shaft and aftershaft are slightly larger than the rest) are crowded indiscriminately into the space vacated by the pulp just before hatching. The calamus is very short and no caps are formed during the incubation period.

21 *days*—Some time after hatching, the protoptiles are freed from their dried sheaths, and the barbs spread out, forming a dense covering of soft, warm down.

(*c*) *The Development of Teleoptiles in the Wings of the Chick*

Feathers first appear microscopically on the wings of the chick about the 10th day of incubation, but their development is relatively more rapid than the development of the protoptiles on the rest of the body. About the 13th day, a second generation of feathers makes its appearance below the prepennae of the remiges. These are the teleoptiles, which on hatching are equal in length to their predecessors which they still bear on their tips, fig. 27, Plate 19.

The first signs of teleoptiles are seen in longitudinal sections of the wing when the follicle of the protoptile becomes greatly elongated, extending to the position of the differentiating cartilage. The sheath, the intermediate cell layer below the short calamus of the protoptile and the cylinder cell layer are also continued downwards, retaining their connexion with the sheath and Malpighian layer of the follicle. The dermis becomes greatly concentrated in this region, and meets the dense pulp at the base of the protoptile.

The calamus of the protoptile is continued for a short distance further on the upper side of the feather, resulting in a crescentic shape, when seen in transverse section. On the opposite side the intermediate cells become aggregated together to form the ridges of the teleoptile, inside the narrow inferior umbilicus of the protoptile. Thus transverse sections at a later stage show a few barbs of the teleoptile in the same section, but on the opposite side of the feather germ to the barbs of the protoptile, fig. 16, Plate 17. Formation of ridges continues, and certain of these on the dorsal side fuse at intervals, giving rise to the rhachis. As may be seen in fig. 12, Plate 17, barbs from each side of the rhachis fuse with it almost simultaneously.

Pigmentation and differentiation of barbules and barbs take place as in the protoptiles, but the resultant shape of the barbs is much more elongated than in the protoptile. This is probably due to the presence of a greater number of barbs inside a sheath of only slightly greater diameter than that of the protoptiles. Correlated with this is the relatively thin cortex of the barbs, only the apex and opposite end retaining their characteristic thickness. The proximal barbule cells are comparatively long in transverse section, fig. 13, Plate 17, resulting in the broad flattened base of the fully formed barbules seen in fig. 29*b*, Plate 20. The hamuli,

which make their first appearance in the teleoptiles are possibly due to the crowding of barbules in very early developmental stages, resulting in the curvature of the more distal cells.

Towards the tip of the teleoptile, the ridges decrease slightly in size on either side of the rhachis. No ridges are present on the ventral side of the feather germ, but they appear lower down, fusing near the base to form the short aftershaft. Fig. 10, Plate 17 shows a transverse section of this region, the smaller barbs of the left hand side representing the aftershaft; fig. 14, Plate 17, shows the tips of the barbs of the aftershaft; fig. 15, Plate 17, is a section showing the main shaft persisting to the tip of the feather, and fig. 16, Plate 17, illustrates the manner in which the barbs of the teleoptile are present at the same time as barbs of the protoptile.

It has been suggested that there is direct continuity between a few barbs of the teleoptile and the calamus of the protoptile. Although this is certainly true of the duckling, in the chick, however, by following serial transverse sections from base to tip of the feather it is seen that the barbs of the former have disappeared, though the distal ends of the barbules are still present when the barbs of the protoptile appear. It seems equally probable that the manner in which protoptiles are carried for some days on the tips of the teleoptiles after the latter have broken from their sheaths, is due to the narrow base of the protoptile constricting the developing barbs of the teleoptile. At any rate, the barbs of the teleoptile show a distinct curve distally when they first spring from the calamus of their predecessor, fig. 27, Plate 19.

Cornification takes place as in the protoptile but it has not reached the base of the teleoptile papilla when the chick is hatched.

The development of protoptiles is therefore fundamentally similar in the duck, goose, and fowl, minor differences being due to relative lengths of incubation periods, resulting in differences in times of appearance of the constituent parts of a feather, and to the differences existing between the specific forms of the protoptile on hatching. Bearing in mind, that the feather is continuously growing from the base, the stages in development may thus be summarized as follows:—

(1) The concentration of dermal nuclei and the elongation of epidermal cells above this concentration.

(2) Proliferation of the stratum Malpighii, resulting in the formation of intermediate and cylinder cell layers.

(3) The backward elongation of the papilla thus formed and the formation of the sheath by the division of epitrichial and intermediate cells.

(4) The aggregation of intermediate cells forming ridges, followed by the binding cylinder cell layer; the appearance of pigment cells, formed in situ in the epithelium; the formation of the feather follicle by the downgrowth of the epidermis.

(5) The division of intermediate cells forming one median and two lateral plates in each ridge, the former giving rise to the barb and the latter to the barbules.

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(6) The differentiation of barb cells into medulla and cortex.
(7) The fusion of barbs dorsally, forming the rhachis, and later the fusion of barbs ventrally, forming the hyporhachis.
(8) The lateral fusion of the rhachis and hyporhachis, forming the calamus.
(9) The withdrawal of the pulp and cylinder cell layer, and the cornification of the feather from the tip downwards.
(10) The formation of the feather caps, and the elongation of the calamus by cornification of undifferentiated intermediate cells at the base of the papilla.

The teleoptile in the wing of the newly hatched chick develops from the same papilla as its protoptile predecessor, and remains attached to it by continuity between a few barbs of the former and the calamus of the latter.

V—Definitive Feathers

The histology of definitive feathers is fundamentally similar in feathers from the different pterylae (*i.e.*, head, neck, breast, thigh, tail, wing, and back), and therefore will not be considered separately.

As might be expected from the study of nestling feathers, the epidermal layers giving rise to the definitive feather are all derivatives of the stratum Malpighii. The stratum corneum forms the sheath of the feather and its follicle ; the stratum intermedium forms the appendicular parts of the feather, and the stratum cylindricum forms the feather caps. The dermal component, *i.e.*, the pulp, is, of course, merely a transitory structure.

The development of the definitive feather will therefore be considered under the following headings : (*a*) stratum corneum ; (*b*) stratum intermedium ; (*c*) stratum cylindricum ; and (*d*) pulp, in both the domestic fowl and the starling.

(*a*) *The Development of Definitive Feathers*

(i) Fowls

(*a*) *Stratum corneum*

This uppermost layer of the epidermis may be distinguished in transverse section passing down from the general surface of the skin into the follicle, and at the base, of this, its continuity with the sheath of the feather is obvious. Contrary to the statement made by Poulton (1894) the feather sheath consists of not one, but sometimes as many as ten or more layers in thickness (*e.g.*, tail feathers). Cornification takes place simultaneously in the sheaths of both feather and follicle, and passes gradually down towards the base. It is practically impossible to distinguish one from the other, until this process is almost complete. Then, however, the slight amount of basal growth, which still takes place, causes the feather sheath to be pulled outwards and away from the follicle sheath. The close connexion between the sheaths of the feather and its follicle may be seen by the irregularity of the break

between them, layers adhering at one point to the follicle sheath, and the same layers adhering to the feather sheath at other points, fig. 30, Plate 21.

(*b*) *Stratum intermedium*

From this layer, the rhachis, hyporhachis, barbs and barbules arise, *i.e.*, all except the basal part of the developing feather (chiefly within the follicle) are formed from the stratum intermedium. The method of formation of these structures is fundamentally similar to that described for neossoptiles. Slight modifications, however, are present and are correlated with the different types of feathers produced (*i.e.*, from the thigh, back, wing, etc.), as distinct from their uniform downy predecessors. The structures arising from this layer will therefore be considered under separate headings : (1) ridges ; (2) rhachis and hyporhachis ; (3) barbs ; and (4) barbules.

(1) *Ridges*—The formation of ridges in a definitive feather proceeds as in the nestling down, although (and especially towards the end of development) the barbule plates may be differentiated before the ridges are cut off. In rapidly cornifying feathers (*e.g.*, ear bristles) this acceleration of formation of parts may be carried even further, and the barbs and barbules cornified without any definite ridge formation, *i.e.*, with the cylinder cells completely encircling the pulp.

(2) *Rhachis and Hyporhachis*—As in the development of protoptiles, the rhachis and hyporhachis of the definitive feather are formed by fusion of barbs, fig. 32, Plate 21. In bilaterally symmetrical feathers, the so-called ventral point at which the hyporhachis arises, lies diametrically opposite the dorsal point. This is well seen in back feathers, fig. 30, Plate 21, in which the aftershaft is well developed. Almost halfway between the dorsal and ventral points (*i.e.*, halfway between rhachis and hyporhachis in transverse section) lies the region of plasmatic growth, fig. 31, Plate 21, where barbs destined to form either shaft or aftershaft arise. What causes their growth in a dorsal direction towards the rhachis, or in the opposite direction towards the hyporhachis is uncertain, but it appears that the intermediate cells are divided primarily into four large ridges—one on either side of the dorsal point, and one on either side of the ventral point, fig. 30, Plate 21. These are then secondarily divided into the small ridges giving rise to the barbs and barbules and all the barbs in the large ridge on either side of the dorsal point fuse to form the rhachis, while all those in the large ridge on either side of the ventral point ultimately fuse to form the hyporhachis. In feathers with rudimentary aftershafts, this is less obvious but still apparent.

The remiges of the fowl are an instance of asymmetry due to the deflexion of the ventral point from its position diametrically opposite the dorsal point, fig. 53, Plate 28. This results in the vane being narrower on one side than on the other, and in a slight curvature of the rhachis. In remiges and retrices, the rhachis may be so disproportionately large as to occupy the greater part of the pulp cavity.

The actual structure of the rhachis varies in different types of feathers. In filoplumes and in down feathers it is solid, very rarely in the latter but never in the former, having traces of medullation.

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In pennaceous feathers of all descriptions, the rhachis is always medullated. The outer cortex, which in coloured feathers invariably receives the greatest amount of pigment, is uniformly cornified early in development, as in nestling down; while in the region nearest the pulp, as might be expected, cornification is retarded, and cell structure is still visible when barbs, barbules and the dorsal side of the rhachis are completely cornified.

In large feathers (*e.g.*, remiges and rectrices) and in the basal region of smaller ones, projections from the cortex extend into the medulla of the rhachis, presumably forming stiffening rods. These have no connexion in number or time of origin with the fusion of barbs.

The medulla consists, as in nestling down, of large vacuolated cells, fig. 32, Plate 21. In very rare cases, small deposits of pigment may be found within these cells, usually restricted to the region where the cytoplasm containing the nucleus withdrew at the commencement of vacuolation.

(3) *Barbs*—Barbs differ greatly, in transverse section, in size and shape in different types of feathers and according to their position. In down feathers or the downy region of contour feathers, they are rarely if ever medullated, and typically consist of barrel-shaped masses, becoming slightly elongated when nearing the ventral point.

In pennaceous feathers, the barbs are invariably medullated for the greater part of their length. The medulla, absent at the extreme tip, usually disappears again before fusion occurs with the rhachis. Continuity has not yet been observed between the medulla of barbs and rhachis.

Near the ventral point, barbs tend to be round or barrel-shaped, becoming progressively elongated in transverse section, in nearing the rhachis, fig. 30, Plate 21. In the rounded region, the medulla may be several cells wide; yet towards the base of the barb, always consisting of a single layer of cells, fig. 32, Plate 21. As in the rhachis, pigment is invariably more densely deposited on the outer side of the barbs.

It is almost certain that cells which are destined to form the barb are the pigment cells in early stages of development. It is invariably in that region of the ridge which ultimately gives rise to the barb, that the pigment cells are found, fig. 31, Plate 21, sending pigment granules by means of long pseudopodia to the barbule plates. The latter are formed before the barb plates, which usually arise after pigmentation is complete. Then, possibly through loss of pigment to the barbules, the pigment cells are much smaller and less dense than before, and these cells are incorporated into the barb plate, thus losing their identity. This possibility of pigment forming in situ in the cells of the barb itself, has been previously mentioned by Lillie and Juhn (1932).

The origin and fate of pigment cells which have been observed among the cylinder cells surrounding the rhachis, fig. 32, Plate 21, just before cornification of the latter, is uncertain.

(4) *Barbules*—These too differ in size and shape according to position and type of feather. In down feathers and the downy basal areas of contour feathers, they

are usually square in cross-section, and appear to be attached end to end. On cornification, this attachment is broken, and each barbule square becomes a separate unit.

In the lacey tips of feathers, barbules appear as narrow elongated cells in cross-section, but in the compact region of the vane, the hooked ends of the barbule cells form the hamuli. This differentiation of barbules has been claimed as a possible means of distinguishing the sex of the fowl by studying the germs of such feathers as the neck hackles, which have a broader lacey tip in the cock than in the hen.

The hooked barbules arise at a lower level on the side away from the rhachis than the straight barbules of the opposite side, fig. 33*a*, Plate 21. This is to be expected from the interlocking method of barbules, where the upper barbules of one barb hook on by means of their hamuli to the slight groove on the lower lying barbules of the adjacent barb.

Pigmentation of barbules takes place relatively early in development, and dense masses of granules may be seen encircling the nucleus. As the barbules elongate, the pigment invariably becomes aggregated at the end which is distal with reference to its attachment to the barb.

(*c*) *Stratum cylindricum*

During the early development of a feather, this layer proliferates and adds a few cells to the rapidly multiplying intermediate cells. But its later function is quite passive, as it merely acts as a binding layer, which follows the divisions of the intermediate cells into ridges, or in the region of the calamus, retains its circular nature.

Immediately following the onset of cornification, the cylinder cells play a more conspicuous though still passive part. Cornification and withdrawal of pulp proceed simultaneously whatever the type of feather, and they are definitely correlated. The conditions initiating keratinization are still obscure, though attempts have been made to explain it in hairs by means of a cystine gradient (King and Nicholls, 1932).

The cylinder cells at this time are still in an obviously healthy condition, and retain their normal cytoplasm and nuclei, although they have long since ceased to divide and may be stretched out of their typical shape. As the pulp withdraws, the cylinder cells remain attached to the still living material, and are pulled away from the cornified barbs and barbules. That a definite pull is exerted by the pulp is seen in the stretched and broken cytoplasmic strands connecting the cylinder cells with the intermediate cells in the early stages of withdrawal.

As the pulp passes down the feather, the cylinder cells become congested, and the outermost ones, being further from the nourishment contained in the abundant blood supply of the pulp, are overtaken by the process of cornification. This occurs at the sides as well as the tip of the pulp, fig. 37, Plate 22, so that a series of cones is formed, connected with each other.

In this way the feather caps arise, and, where protected by the calamus, persist, although distal to the superior umbilicus they soon break away from the feather.

DAVIES (1889) describes the method of cap formation from an entirely different point of view. He considers the cylinder cells to recommence dividing at certain stages during withdrawal, when the pulp is stationary. He does not explain what causes the pulp to remain at a certain level, and then suddenly to continue passing proximally after such a pause, nor why cells which have remained passive for so long should suddenly start dividing again when comparatively far from nourishment.

(*d*) *Pulp*

In a developing feather, the dermis is concentrated round the follicle, and also, and to a greater extent, within and below the base of the feather germ. In successive cross-sections through the base of the feather, this aggregation of whorls of dermal cells may be seen to extend for some distance below the actual base of the feather. This basal region is the growing part.

Nourishment is carried to the developing feather by means of a definite blood supply in the pulp. In a large definitive feather, *e.g.*, a remex or rectrix, the blood system differs from that of an embryonic down feather only in its greater complexity. The general plan of a central arteriole branching at the top of the pulp into numerous anastomosing venules still persists, but the arteriole of a definitive feather may be duplicated and occasionally branch, while the venules are more extensive.

This dermal component of a feather is transitory and as soon as its function of nourishing the developing barbs and barbules is finished, its withdrawal commences. As already stated, this occurs at the same time as cornification begins in the peripheral regions of the tip of the feather, starting at the sheath and passing inwards.

The circulatory system is entirely remodelled during the withdrawal of the pulp, tiny vessels fusing to form large blood spaces which often occupy the whole of the space beneath the developing feather cap, and may be left there when the pulp withdraws further.

This might be thought by adherents of DAVIES's theory of cap formation to be an incentive for cylinder cell division ; but these cells may form many congested layers, and cornification, having once started, seems to gain speed in passing downwards. Hence the accumulation of blood sometimes occurring at the top of the withdrawing pulp can have little effect on the outermost cylinder cells.

The withdrawn pulp is absorbed by the dermis, and no trace of its previous extension within the developing feather exists, except for a small papilla projecting within the inferior umbilicus. Immediately before the moulting of a feather, however, the quiescent pulp recommences growth and keeps pace with the developing intermediate cells up to the mouth of the follicle.

The pulp has been considered (LILLIE and JUHN, 1932) to be constantly growing at its base, and constantly dying off and being resorbed at its apex. From the fact that both in longitudinal and transverse sections the pulp may be seen to be actually torn away, first from the barbs as they move upwards, and secondly from the feather caps in process of formation, fig. 37, Plate 22, it would seem more probable that the pulp actually withdraws and is resorbed at the base, as earlier authors (DAVIES,

1889 and Strong, 1902*a*) have observed. This is confirmed by the fact that growth of the pulp is no longer necessary at this time, as when pulp withdrawal takes place, the whole of the feather (except the calamus) has been formed, and only cornification is to be completed.

(ii) Ducks

The development of definitive feathers in the Khaki Campbell duckling was studied for the first eight weeks after hatching. In general, this process is similar to that of the fowl, the chief differences being in the relative times of appearance of the second generation of feathers. In the duckling, the wing feathers are retarded in development and do not appear externally until the 5th week. The tail teleoptiles however develop during the second week at the base of the greatly elongated calamus of the protoptile. These are the only feathers which show major structural differences from the fowl feathers already described.

Sections through the tip of a tail protoptile show the rhachis to be indistinguishable from neighbouring barbs, but more proximally, a one-sided fusion takes place rapidly followed by a second fusion on the opposite side. The median barb may therefore be regarded as the rhachis, and after this preliminary fusion, it is always characterized by its greater size. At the proximal end of the protoptile, the rhachis may occupy one half of the transverse section and form one side of the circular calamus. It then becomes much narrower, leaving the calamus of uniform width. This marks the distal limit of the elongated calamus peculiar to the rectrices.

The rectrices of the duckling are of particular interest in that there is no definite line of demarcation between protoptile and teleoptile. The barbs of the former fuse to form rhachis and hyporhachis, which in turn fuse to form the calamus. The calamus then splits up into barbs, fig. 28, Plate 19, which fuse to form the rhachis and hyporhachis of the teleoptile. A slight constriction is usually present at the base of the calamus of the protoptile, probably marking the site of the inferior umbilicus, and below this the calamus splits into the barbs of the succeeding definitive feather.

It is possible that this peculiar state of affairs is due to the rapid growth of the teleoptile. Usually a quiescent period sets in between the formation of the two generations of feathers, but the feather papilla of the rectrix of the duckling is obviously in a very active state throughout the nestling period.

This offers striking proof of the facts disputed by previous authors that (*a*) the second generation of feathers is formed in the same follicles as their predecessors, and (*b*) that the feather papilla resumes activity prior to the shedding of the old feather.

(iii) Starlings

The first coat worn by the starling consists of a very rudimentary nestling down similar fundamentally to that of the chick. It was unfortunately impossible to obtain sufficiently early stages to study the development of these protoptiles.

The youngest stage obtained had feathers visible macroscopically on the head and round the eyes and ears ; on the neck, ventrally two rows of feathers converging below the head and extending backwards almost to the cloaca, and dorsally, extending from the head to the tail in the middle line ; on the thighs and wings, with a few on the legs.

None of these feathers had emerged from its follicle, but could be seen by means of the pigment contained, or by the raised lumps which they formed on the surface. All follicles, however, had downy protoptiles protruding.

The largest specimen obtained had primaries measuring 4 cm, with other feathers proportionately long, and all having burst from their sheaths for the distal quarter of their length.

(*a*) *Stratum corneum*

This layer forms a thick sheath to the feather and a thinner sheath to the follicle, but they are much more widely separated than in the fowl.

(*b*) *Stratum intermedium*

Residual cells are more definite and in larger numbers than in the fowl. In longitudinal sections of primaries the barbule plates of each ridge are seen bounded by cylinder cells and with a central layer of residual cells lying between them, in appearance very similar to the true cylinder cells. These join up a triangular colony of cells near the sheath to the developing barb. Presumably the residual cells are either cornified with the sheath, or atrophy in the centre of the ridge.

As in the fowl, the shape of barbs and barbules differs according to the position with regard to the rhachis, its level in the feather and the position of the feather on the body.

In wing and tail feathers, the barbule plates nearest the rhachis consist of very long, deeply pigmented cells, with blunt inner edges and narrow outer ones. On the far side of the barb from the rhachis, the barbules are narrower, less pigmented and hooked, and develop later than do their neighbours.

In thigh feathers, the barbules are wedge-shaped and do not bear hooks.

(*c*) *Stratum cylindricum*

This layer typically consists of cells with rounded nuclei containing two to three nucleoli. Between the ridges, the rounded shape may be lost and the cylinder cells represented only by a double narrow, nucleated string. This is undoubtedly due to the close proximity of the ridges.

(*d*) *Pulp*

The formation of feather caps and the withdrawal of the pulp take place as in the fowl. Fig. 37, Plate 22, shows the pulp actually pulling away from the developing cap, in a tail feather of a starling.

Thus development of definitive and nestling feathers differs only in that the latter arise from the general epidermis of the body, while the former already have the

dermal papilla sunk within a follicle. Differentiation of the epidermal covering of this papilla takes place in the same way as differentiation of analogous structures in the embryo, with modifications resulting in the different types of feathers found in the adult.

(*b*) *Moulting and Replacement of Feathers*

The development of regenerating feathers does not appear to have received much attention. As Lillie and Juhn (1932) have stated regarding the papilla, " it has not been known whether its activity is resumed prior to actual moulting . . . or whether the shedding of the feather is the actual stimulus to renewed activity ".

The usual method employed in the study of plumage variations is to pluck the feathers from a given area before injecting or feeding an endocrine substance, and then to compare the regenerated feathers with normal ones.

For these reasons it has been thought advisable to study the feather germ (*a*) under normal conditions (*i.e.*, after moulting) and (*b*) after deliberate plucking (Part III). In this section, normal regeneration is considered.

Sections were taken of young feathers developing in the follicles of feathers which were about to be moulted, or which had recently been shed. Such stages are easily obtained in the fowl up to the assumption of the adult plumage, as no definite moult occurs between the different coats worn by the young animal, since a gradual shedding of old feathers and replacement by others of a slightly different type takes place.

On emerging from the follicle, the new feather is still covered by a sheath, which is crowned distally by a small hook-like projection. Serial transverse sections through this region show the hook-like projections to consist entirely of sheath and a layer of cornified cells similar to a feather cap, and therefore presumably consisting of cylinder cells. Lower down, the tips of the barbs and barbules appear on one side only (*i.e.*, the dorsal side) between approximately the 3rd and 4th layers of cornified cells from the centre. More proximally, pulp is seen beneath a definite feather cap, and below this region, the feather is formed in the usual way.

A longitudinal section through such a feather may show as many as five caps between the tip of the feather and the pulp, and the narrow cornified layers going up to the cap are continuous with the stratum cylindricum and the stratum intermedium. The actual tip is sheath only, and therefore represents the stratum corneum, fig. 34, Plate 22.

Figs. 35 and 36, Plate 22, show the method of development of this tip. The calamus of the old feather becomes much narrower and slightly constricted in nearing the base, and into this constricted region (the inferior umbilicus) the papilla projects. The epidermis covering this is complete in all its layers. The cylinder cells are continuous with the cylinder cells of the follicle walls and are quite separate from those (now cornified) of the last feather cap. The intermediate cells are similarly continuous, and already show signs of ridge formation. Finally, the stratum corneum

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of the papilla is continuous with the sheath cells of the follicle and with the extreme base of the old feather.

The manner in which the complete epidermal covering of the tip arises may be followed from serial transverse sections. Near the base of the feather the calamus becomes narrower, owing to cornification of the outer intermediate cells only. The inner layers become slightly vacuolated, so that a new sheath is formed within the calamus. This is, naturally, continuous at the base with the sheaths of the old feather and its follicle, although the former is very narrow and scarcely distinguishable from the calamus.

The order of layers from the follicle sheath inwards now consists of feather sheath, calamus, sheath of new feather, intermediate cells, cylinder cells and pulp. As the pulp withdraws and the last feather cap is formed, the intermediate cells within the calamus are drawn over the tip of the pulp together with the cylinder cells above which they lie. This then forms the new papilla, the old calamus breaking away from the new sheath cells laterally.

It is evident, therefore, that the feather papilla resumes activity prior to the shedding of the old feather, and this is most conspicuously demonstrated by the protoptiles borne on the tips of the teleoptiles in the fledgling.

VI—Discussion

These investigations deal with three main groups of problems associated with (*a*) sequence of plumage, (*b*) histological development of feathers, and (*c*) the actual structure of a feather.

(*a*) *Sequence of Plumage*

The sequence of plumage in birds has been the subject of discussion at various times. In 1907, Lynds Jones pointed out that " the first down and its succeeding definitive feather are produced by one continuous growth, and therefore cannot be regarded as two distinct feathers. The first down is the plumulaceous tip of the definitive feather."

At a cursory glance this might appear true, as invariably the protoptile is borne on the tip of its successor for some time ; but, except in areas where the development of the second coat is accelerated (as in the remiges of the fowl), the fact that a definite calamus exists at the base of the protoptile, into which the distal barbs of the second generation feather project as in the fowl, or with which they fuse as in the duck, shows the protoptile and its successor to be distinct feathers produced consecutively by the same papilla.

The sequence of plumage in the Brown Leghorn fowl has recently been studied by Domm, Gustavson and Juhn (1932) who describe four stages, down, chick, juvenile, and adult ; but " because their development is a gradual piece-meal affair, succeeding stages are invariably intermingled before the attainment of the adult plumage ",

The chick plumage differs from the down, according to these authors, by the precocious development of remiges and rectrices, together with a few feathers along sides of breast and belly. Now in the Rhode Island Red and the Black Leghorn X Light Sussex, and according to Finn (1919) in all Galli, the remiges are developed on hatching, and breast and abdominal feathers appear later. Thus the "down" stage may be considered as relegated to the incubation period. In some breeds, *e.g.*, Rhode Island Red, the rectrices do not usually appear until after the fourth week in the male birds, although present in the female during the second week. The difficulty of dividing the plumage into definite stages is thus obvious.

Clarke as early as 1906 announced the discovery of two coats of nestling feathers in Ringed and Gentoo penguins, and in 1907 Pycraft assumed that most common birds have lost their protoptile coat, while in Ratitae the mesoptiles may persist in the adult. Ingram (1920) suggests that it is the second coat which is lost in most birds, and Cossar Ewart's work on the Mallard duckling (1921) corroborates this.

As already noticed, the neossoptile coat exists in members of Anseriformes, Galliformes, and Passeriformes, but while Ewart (1921) has figured three definite coats in the first of these groups, it has so far been impossible to find them all in the others.

The mesoptile is usually portrayed (*e.g.*, Ewart's duck) as a structure intermediate in type between the down feather and the pennaceous form. In the chick it was at first thought that the wing feathers present on hatching represented mesoptiles, but their later development showed them to be similar to the adult type of feather in structure. Differences of course exist, but since these consist of shape of vane or pigmentation—which vary considerably in the adult—they can hardly be called characters defining generations of feathers.

The question then arises as to whether the nestling down of the chick consists of protoptiles or mesoptiles. Newton (1896) defines a typical protoptile as consisting of :—

(*a*) a very short calamus ;
(*b*) an insignificant or ill-defined rhachis, if there be one at all ;
(*c*) the almost universal absence of cilia, and
(*d*) the absence of an aftershaft, except in *Dromaeus*.

Now according to Ewart (1921) the nestling down of the Mallard duck consists of feathers characterized by :—

(*a*) a well-developed calamus which may have as many as 20 cones,
(*b*) a well-developed rhachis, .
(*c*) well-developed, sometimes hook-like cilia,
(*d*) short, stiff as well as long, slender barbs, and
(*e*) a well-developed aftershaft, the barbules of which bear cilia.

From the present investigation, the nestling down of the Khaki Campbell and Aylesbury ducklings and the Chinese gosling is in these respects similar to the Mallard duckling, and hence probably to all Anseriformes.

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The chief difference between the nestling down of the chick and the duckling lies in the ring-like swellings at the nodes of the barbules in the former, and the presence of cilia (*i.e.*, projections from the nodes) in the latter.

It may thus be concluded that EWART's description of a typical protoptile is more accurate than the earlier one of NEWTON.

On this similarity of all essentials in the first coat of the chick and the duckling, we may assume the nestling down of the chick to consist of protoptiles, and therefore the mesoptiles have been suppressed.

In the Mallard duckling and the Chinese gosling, EWART claims to have found all three types of feather in continuity with each other. The proximal and youngest feather represents the teleoptile ; the median feather, which is definitely less highly specialized than its successor, being the mesoptile, and bearing on its tip the first nestling down or protoptile. No such intermediate form has been found in the chick.

Not only is the mesoptile coat totally suppressed in the chick, but in the region of quickest growth of feathers (*i.e.*, primaries and secondaries) the protoptile coat is less highly developed than elsewhere. This refers only to the completion of the calamus, which usually grows to contain several caps, but the precocious formation of the teleoptiles of the remiges prevents more than the beginning of the calamus from being formed.

(*b*) *The Histological Development of Feathers*

The attention of earlier writers in this field was mainly concentrated on such points as ridge formation and pigmentation, while modern work stresses the importance of the method of formation of the rhachis. Hence these three problems will be considered separately.

(i) Ridge Formation

The problem of ridge formation has resolved itself into the question whether the grouping of intermediate cells to form ridges is due to an invasion of the intermediate cells by the cylinder cells, or whether the intermediate cells themselves first form groups, the cylinder cells merely retaining their limiting position. Both DAVIES (1889) and STRONG (1902 *a*) consider the last condition most probable, as the intermediate cells may be seen to change their position during the early stages of barbule formation. Again MAURER (1892) and STRONG (1902 *a*) agree that the rapid growth of intermediate cells causing increased pressure on the pulp, may be a factor in ridge formation. In later stages of development of protoptiles, ridges may be formed before the cylinder cell layer is differentiated from the intermediate cells ; and cylinder cells are not present between ridges when they are fusing to form the rhachis.

In the development of teleoptiles, barbs and barbules may be formed from the intermediate cells before any ridges exist, and in some feathers, cornification proceeds so rapidly that only attempts at ridge formation are possible. In thigh and back feathers with very large aftershafts, the ridges are crowded into such a confined space that there is little room for cylinder cells to extend from the apex

of the ridge to the sheath, figs. 30 and 52, Plates 21 and 28. They may be traced for a certain length and then become indefinite, and it is doubtful whether the ridges are ever quite separated from each other near the periphery.

It is evident, therefore, that the intermediate cells take the initiative in forming ridges, and the cylinder cells later pass between them.

(ii) Pigmentation

The origin of pigment has been the subject of various theories but its distribution in the duck, goose, and fowl supports the view advocated by Demiéville (1884), Mertsching (1889), Jarisch (1891 and 1892), Rabl (1894), Post (1894), Rosenstadt (1897), Loeb (1898), Prowazek (1900), and Strong (1902 *a*). These authors believe it to be the result of metabolic activity of either the nucleus or cytoplasm of epithelial cells.

Strong, working on the definitive feathers of *Sterna hirundo*, found pigment first appearing in the intermediate cells before the formation of ridges, and no pigment in the pulp. These results were based on sections cut at different levels of a fully formed feather, and consequently the absence of pigment in the pulp might be due to its migration outwards between the cylinder cells and so to the intermediate cells, as writers in favour of the exogenous (*i.e.*, dermal) origin of pigment believe. However, in the sections of embryonic and definitive feathers studied in the present enquiry pigment first appears in the intermediate cells after the formation of ridges, and no traces are found in the pulp. Further, in the goose and fowl, pigment may be present in the epidermis surrounding the follicle, but not in the dermis. In hairs, pigment has also been found appearing first in the Malpighian layer of the papilla (Spencer and Sweet, 1899).

Pigment appears concentrated towards the outermost intermediate cells nearest the sheath, in the earliest stages at which it is present in embryonic feathers, and in successive days of incubation, wanders towards the apex of each ridge, from there sending out amoeboid processes along which pigment granules flow, fig. 5, Plate 16. The word " flow " is used since different concentrations of pigment are visible in different parts of the processes. This tendency of pigment cells to move inwards from their place of origin, accounts for isolated pigment cells which have been found on extremely rare occasions in the pulp of duckling and gosling feathers.

The fact that in definitive feathers pigment cells may be incorporated into the barb as normal cells, also supports the view that in feathers pigment is formed within the epithelial cells.

(iii) Rhachis

The study of the development of both nestling and definitive feathers confirms the theory advanced by Davies (1889) and Strong (1902 *a*) that the rhachis is formed by fusion of barbs. This method of formation of a feather is illustrated in fig. 56. The first ridges formed give rise to barbs of approximately equal size, and at this point (*i.e.*, the future tip of the feather) it is impossible to distinguish the primordium of the rhachis, fig. 56A. This condition extends for a very short distance,

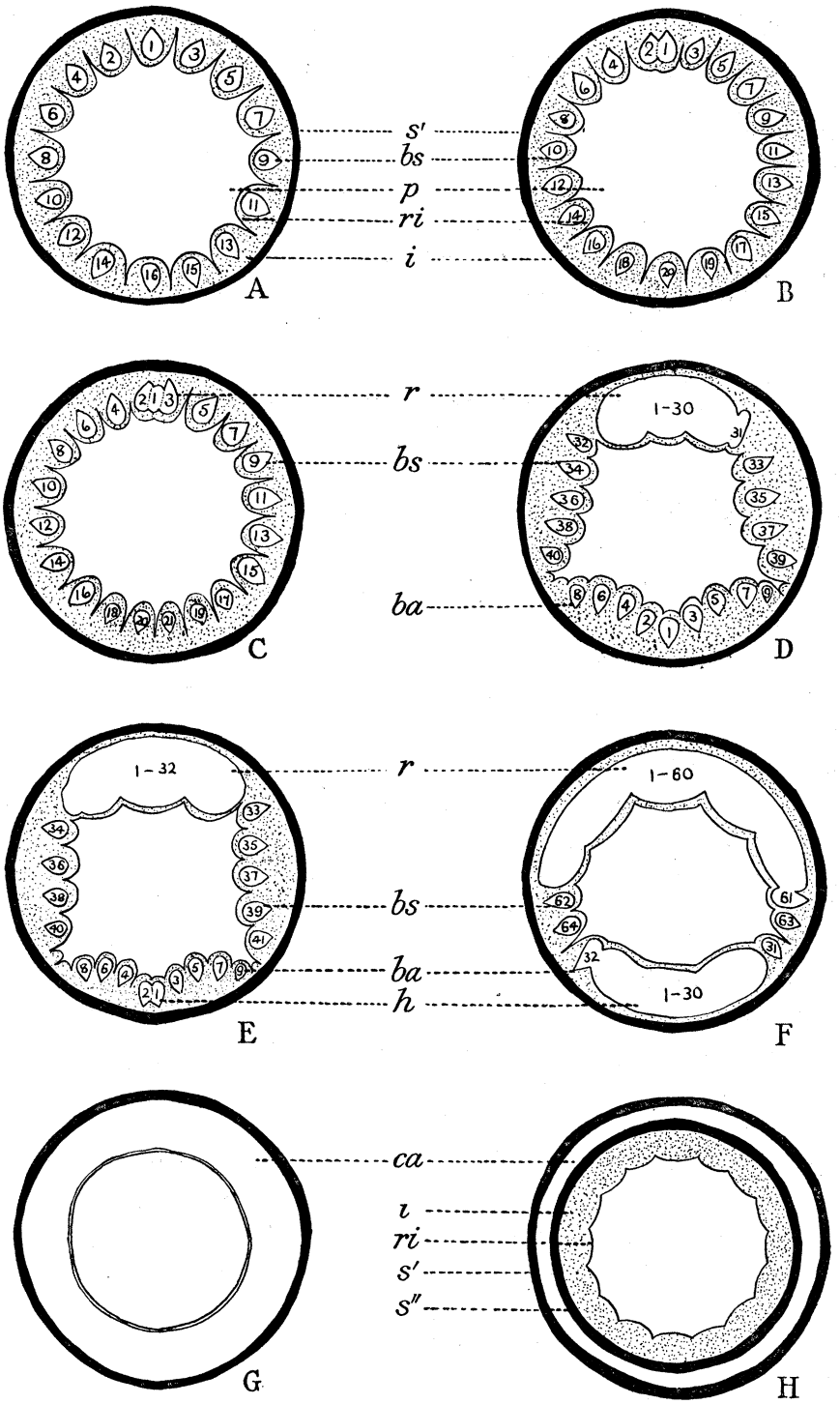

FIG. 56—Diagrams of cross-sections of a feather to show the dorsal fusion of barbs to form the rhachis (A-F); the fusion of barbs ventrally to form the hyporhachis (D-F); the lateral fusion of rhachis and hyporhachis to form the calamus (F); the structure of the calamus (G) and the tip of the new feather forming within the base of the calamus of its predecessor (H). *ba*, barb of aftershaft; *bs*, barb of shaft; *ca*, calamus; *h*, hyporhachis; *i*, intermediate cells (*i.e.*, collar); *p*, pulp; *ri*, ridge; *s'*, sheath of old feather; *s''* sheath of new feather.

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the length varying in different types of feathers, and the fusion of two of these identical barbs marks the beginning of the rhachis, fig. 56B. By repeated fusion, the rhachis becomes conspicuously large and may occupy the dorsal half of the feather papilla. Near the base (the distance again varying according to the type of feather) barbs arise on the ventral surface, which, like the first formed barbs on the dorsal side, show little difference in size, fig. 56D. Fusion of these barbs gives rise to the hyporhachis, fig. 56 E. The superior umbilicus is formed by the lateral fusion of the rhachis and hyporhachis, fig. 56 F, and proximally the fused structures form the calamus, fig. 56 G. Fig. 56 H shows the tip of the succeeding feather within the calamus of its predecessor.

(*c*) *The Actual Structure of a Feather*

Until quite recently, ornithologists have been uncertain as to the true composition of nestling down, and the generally accepted view was that no aftershaft is present (Headley, 1895). Ewart (1921) summarizes the reasons for regarding the aftershaft as an accessory and secondarily acquired structure, as follows :—

(*a*) That the aftershaft is developed from a forward elongation of the calamus (according to Gadow) ;

(*b*) that the tip of the aftershaft is never attached to the calamus of the feather about to be shed.

Ewart dispenses with the last reason by means of numerous convincing illustrations of the connexion between the aftershafts of two, and even three generations of feathers in birds of such widely separated families as Casuariformes, Sphenisciformes, Anseriformes, and Galliformes.

The first reason is seen to be equally unfounded from a study of the development of the aftershaft. As the feather grows from tip to base, the barbs of the aftershaft are formed at the same time as the barbs of the shaft, and it is the fusion of these two structures which forms the calamus.

Thus a complete feather must consist of a calamus, shaft and aftershaft, although the latter may appear vestigial owing to the delayed fusion of the barbs of the ventral side of the feather.

VII—Summary

The nestling coat of the duck consists of prepennae, preplumulae and prefiloplumae ; of the goose, prepennae and preplumulae, and of the chick and Passerines (except the House Sparrow) of prepennae.

These feathers represent protoptiles, the mesoptiles having been suppressed.

Nestling and definitive feathers arise in a similar manner from the stratum Malpighii of the skin or the papilla respectively, the stages consisting of :—

(*a*) Proliferation of intermediate cells.

(*b*) Grouping of intermediate cells to form ridges.

(*c*) Grouping of intermediate cells within ridges, forming one median and two lateral plates.
(*d*) Appearance of epidermal pigment cells.
(*e*) Formation of barbs and barbules from the plates, and their pigmentation.
(*f*) Fusion of barbs dorsally forming the rhachis, and ventrally forming the hyporhachis.
(*g*) Lateral fusion of rhachis and hyporhachis forming the calamus.
(*h*) Withdrawal of pulp from the apex of the feather, simultaneously with cornification, and followed by the formation of feather caps.

The feather sheath consists of the outermost layers of intermediate cells in both protoptiles and teleoptiles, in the former also supplemented by the epitrichium.

Prior to moulting, the feather papilla resumes activity and the tip of the new feather is formed inside the base of the calamus of its predecessor.

From its mode of development, a typical feather must consist of shaft, aftershaft, and calamus.

II—THEORIES OF FEATHER DEVELOPMENT

I—Feather Development

The theories of feather development propounded by Davies (1889) for the pigeon, and supported by Strong (1902*a*) for *Sterna hirundo* passed unquestioned until Lillie and Juhn (1932) put forward the concrescence theory. This is an attempt to explain variations in form and pattern of feathers, produced experimentally by the injection of female hormone or by thyroid medication, as due to different rates of growth of individual barbs, and the basis of this theory lies in their conception of the rhachis of a feather as formed by concrescence of the two halves of the "collar".

The "collar" is defined as the Malpighian layer before the commencement of ridge formation, and Lillie and Juhn further consider the part of the collar in which the bases of the primary ridges end as the primordium of the shaft, which thus has the form of a ring. The shaft itself according to these investigators, is formed by the concrescence of the halves of this ring in the mid-dorsal line (the dorsal point being fixed by the position of the rhachis) and the growth-energy is furnished by growth and multiplication of its cells directed dorsally from the mid-ventral part of the ring. The rate of growth of each barb is so regulated that it is completed by the time the base meets the mid-dorsal line. There is thus no time in development when the barb is not attached to the primordium of the shaft.

Domm, Gustavson and Juhn (1932) illustrate the concrescence theory by fig. 57.

The diagrams in fig. 57 offer no indication of the method of formation of the aftershaft—an essential part of the feather, however rudimentary, but it may be inferred that the aftershaft arises at the ventral point in the same way that the shaft arises at the dorsal point. A reversal of the "streaming movement" of cells somewhere between the dorsal and ventral points would be necessary to account

for this, giving the so-called " region of plasmatic growth " where barbs pass either dorsally or ventrally. The apparently double origin of the rhachis in these figures may possibly have been imagined from the longitudinal depression which is obvious macroscopically on the ventral side of the shaft. Lowe (1933) certainly regarded the longitudinal furrow in the middle of the ventral surface of Carinate feathers as

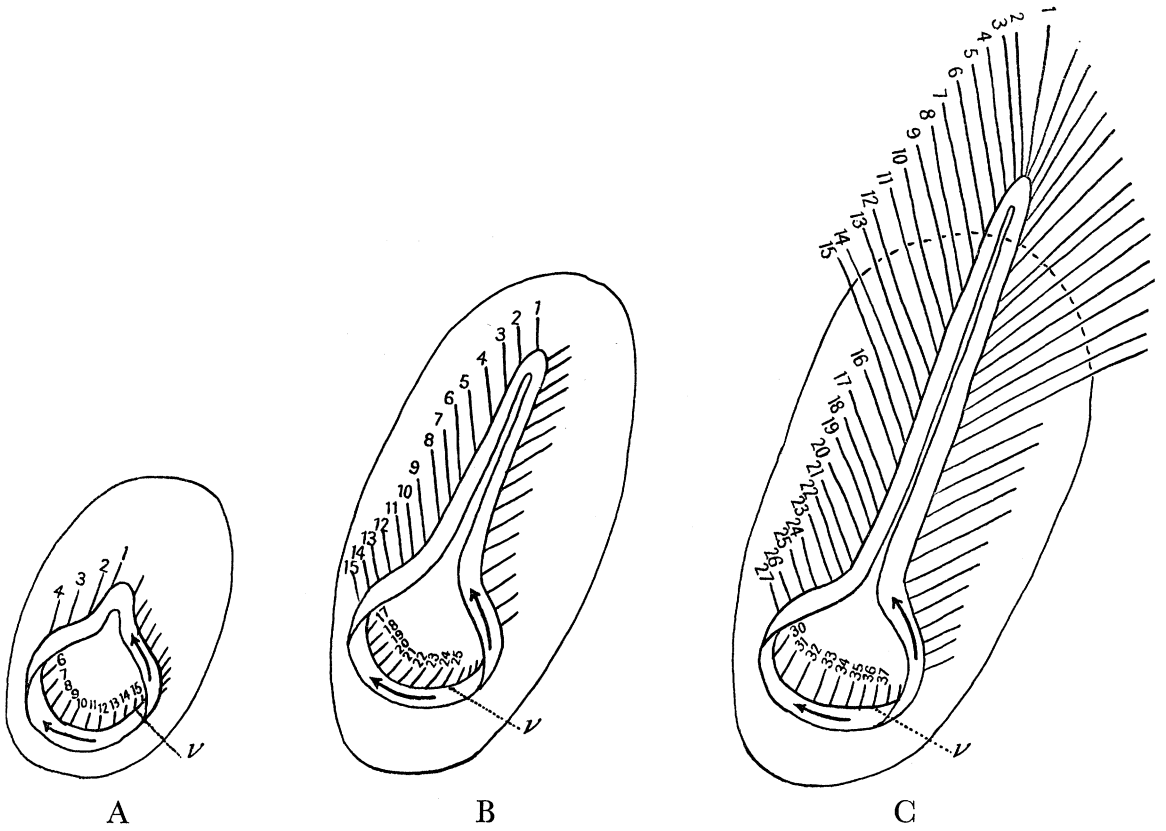

Fig. 57.—The diagrams illustrate the principle of concrescence in the origin of a feather. The two halves arise from a ring of embryonic cells, the " collar " surrounding the base of the feather germ ; the ridges or barbs (Nos. 1–15, fig. 57A) arise, each with a separate growth centre from the collar at right angles to it, thus parallel to the axis of the feather germ. The rhachis is formed by a process of concrescence of the continually growing right and left halves of the collar, the levels from apex to base being formed successively, fig. 57A. The forming barbs are carried along with the constantly streaming halves of the collar to their definitive positions at the sides of the shaft with consequent change of orientation. As the series of barbs move dorsally (Nos. 1–15, fig. 57A), new barbs, (Nos. 16–25, fig. 57B), take their origin in the space thus provided at the ventral surface of the collar. At any given time, then, the collar is beset with a series of forming barbs on each side which range in age from the dorsal to the ventral surface of the feather germ and in state of development from the completely formed barbs dorsally, (Nos. 1–25, fig. 57C), to mere apical rudiments ventrally, (Nos. 35, 36, 37, fig. 57C). The order of formation of the shaft is apico-basal and the order of age of the barbs is naturally the same. Similarly in each barb the apex at the margin of the feather is the first formed and the central end attached to the shaft last. Thus there are two time gradients in each feather, from apex to base along the shaft and from margin to centre along the barbs. (From Domm, Gustavson and Juhn, 1932, p. 638.)

" marking the point where the fusion of the two incurving lateral halves of the feather has not been quite complete" (p. 492). In this case, how would these authors explain the triplication of such depressions in feathers from certain regions which are so conspicuous microscopically, fig. 52, Plate 28.

A more exact representation of the sequence of events is given in fig. 58.

The barbs form simultaneously near the dorsal point at the tip of the feather germ, one of which may be slightly larger than its neighbours, or there may be no difference in size, fig. 58 A. The fusion of barbs to form the shaft, and the similar fusion to form the aftershaft are shown in fig. 58 B, C, and D. Meanwhile, growth has been continuing at the base, so that the whole feather germ has greatly increased in length, but the pulp is still keeping pace with the rest of the feather, fig. 58 E. Fig. 58 F shows that withdrawal of pulp and formation of feather caps has begun, and the bases of the rhachis and hyporhachis are rapidly thickening. This is followed by the lateral fusion of rhachis and hyporhachis, giving rise to the calamus, and marking the site of the superior umbilicus, fig. 58 G. At this time, too, the first formed barbs have broken from the sheath. The series of cross-sections in fig. 56, also illustrate this. The difference between the two theories of feather development is made clear by a comparison of figs. 57 and 59.

Lillie and Juhn summarily dismiss the theory that the rhachis is formed by fusion of barbs, saying " This is however, incorrect. . . . The two dorsalmost barbs are laid down and become pigmented and keratinized while still separate from one another. After that there can be no fusion except at their undifferentiated bases ; it is at their bases, as a matter of fact, that the shaft proper begins by a fusion of the two half-rings of the collar to which the bases of the first barbs, as well as those of all succeeding barbs have a primary attachment." (Pp. 142–143.)

If successive cross-sections of a feather are taken from tip to base, it will be seen that the two most apical barbs fuse to form the rhachis before cornification commences, and since the feather is growing only at the base, this fusion is really a combination of their growing basal regions. In fact, the process of cornification does not usually begin until several barbs have thus fused. When sections are taken through older feathers in which cornification has proceeded almost to the base, there might appear superficial grounds for such a statement. Actually, the tips of the barbs are keratinized before the bases, but it must be remembered that growth takes place in the reverse direction (*i.e.*, base to tip) and fusion of barbs occurs before cornification has begun. There is thus no justification for the first part of the above quotation.

It has already been pointed out (Hosker, 1934) that ridge formation is a passive cutting up of the intermediate cells, and these divisions curve round the feather germ in such a way that a ridge which at first lay ventrally, would ultimately fuse with the rhachis at the dorsal point. There is thus no suggestion of a movement of cells, except the early grouping of cells into barb and barbule plates, or of a movement of the two halves of the collar to fuse in the dorsal line.

After refuting Davies's conclusions regarding the method of formation of the shaft, Lillie and Juhn quote him as confirming the concrescence theory. " While

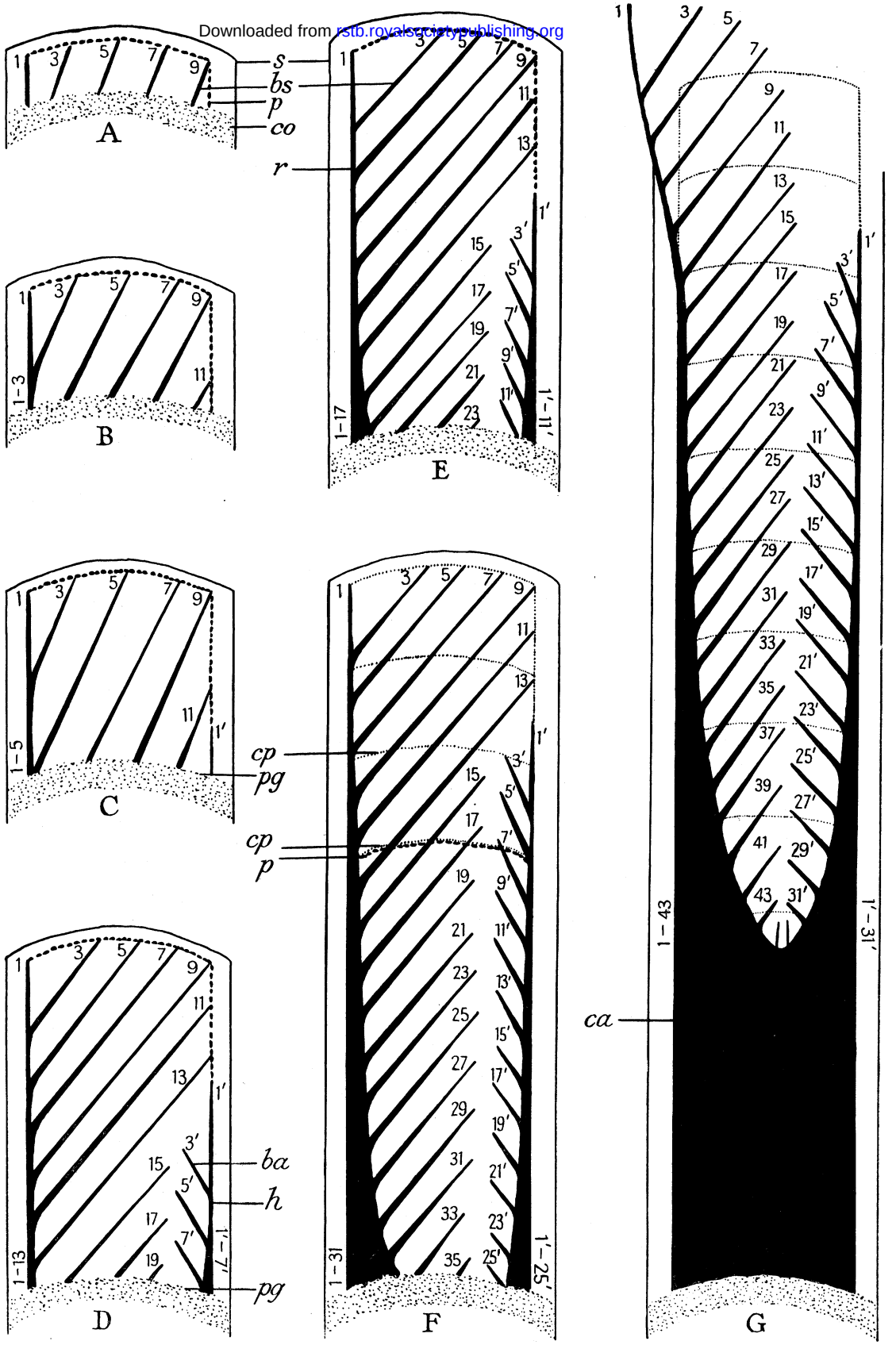

FIG. 58.—Longitudinal diagrams of hemi-sections of feather germs to show the method of formation of a feather according to the fusion of barbs theory. A.—Barbs forming at the tip of the feather germ. B.—Fusion of barbs dorsally to form the rhachis. C.—Appearance of region of "plasmatic growth" where barbs pass either dorsally or ventrally. D.—Fusion of barbs dorsally forming rhachis, and ventrally forming hyporhachis. E.—Thickening of rhachis and hyporhachis, making pulp cavity narrower. F.—Withdrawal of pulp and formation of feather caps. G.—Complete feather, showing the tip having broken from the sheath, the pulp completely withdrawn, and the calamus formed by fusion of rhachis and hyporhachis. These diagrams are of necessity foreshortened and the curvature of the barbs is not shown. The stippled region at the base of each germ represents the collar, where growth (except that due to vacuolation of cells) and pigmentation occur. *ba*—barb of aftershaft ; *bs*—barb of shaft ; *ca*—calamus ; *co*—collar ; *cp*—feather cap ; *h*—hyporhachis ; *p*—pulp ; *pg*—region of plasmatic growth ; *r*—rhachis ; *s*—sheath. 1–43—Barbs fusing to form the rhachis. 1′–31′—Barbs fusing to form the hyporhachis.

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in the primitive down the upper border of the *Spule*, to which the rays are fastened, forms a circle, in the definitive feather, in consequence of enormous prolongation of one side, it forms a long drawn-out obliquely placed ellipse, and by the great thickening of this prolongation the upper umbilicus is completely closed. In this manner there arises in the definitive feather a structure, the shaft, which in reality

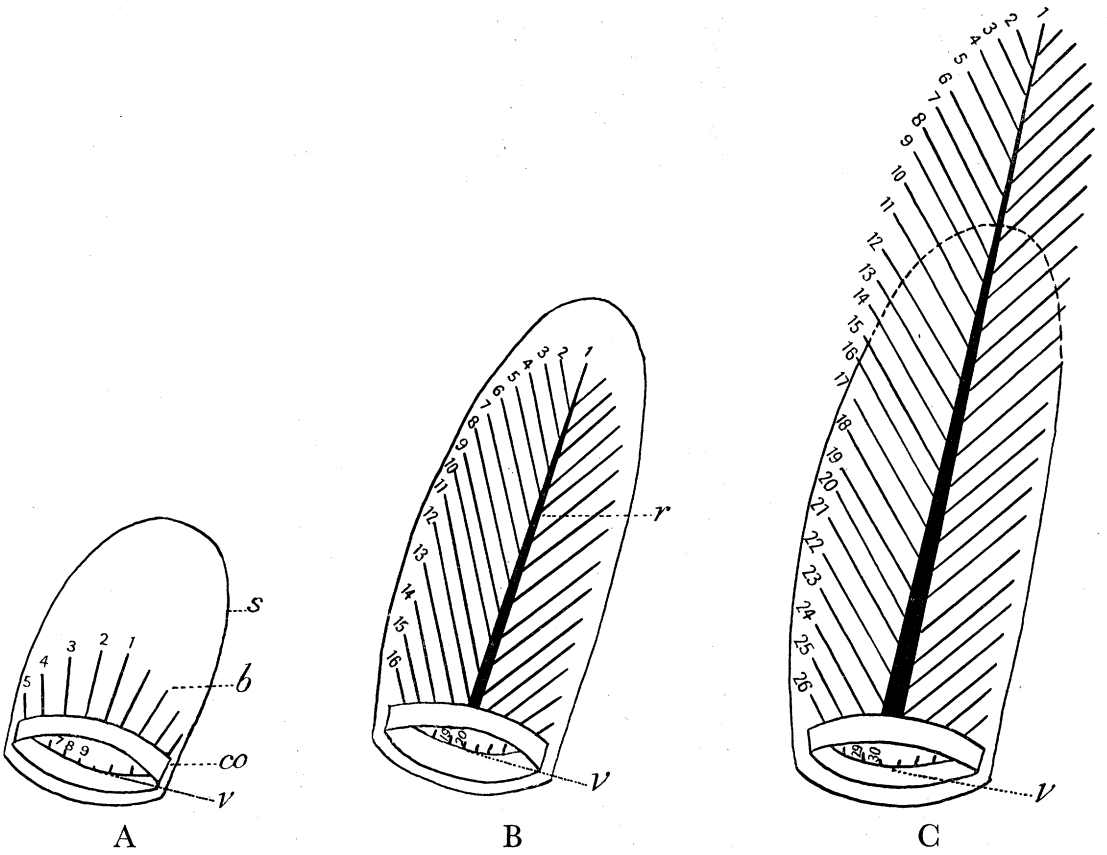

FIG. 59.—Diagrams showing the development of a feather according to the fusion of barbs theory, drawn from the same point of view as fig. 57, and omitting the method of development of the aftershaft. The whole feather arises from a ring of embryonic cells (*i.e.*, the collar) surrounding the base of the feather germ. The barbs, Nos. 1–9, fig. 59A, arise from the collar at an angle of approximately 135°. As more cells are added to the barbs from the rapidly dividing collar cells, it will follow that they will gradually approach the mid-dorsal line, and fuse with the dorsal-most barb or rhachis. This of necessity becomes broader, fig. 59B and C, and takes on the definitive shape of the rhachis. The rhachis cannot therefore be considered as a prolongation of the collar, as is implied by fig. 57, but a structure similar to a barb, and differing only from a barb in occupying the mid-dorsal position. *b*—barb ; *co*—collar ; *r*—rhachis ; *s*—sheath ; *v*—ventral.

is only part of the *Spule*, although it makes the impression of an entirely new structure."

The *Spule* LILLIE and JUHN interpret as the collar, but it really represents the calamus, which, as fig. 58 shows, is formed by the fusion of the shaft and aftershaft.

Early workers failed to realize the presence of both shaft and aftershaft in nestling down, however rudimentary they might be, but it has been shown (Part I, p. 173) that a typical feather consists of shaft, aftershaft, and calamus, although the aftershaft may be so small as to be represented by a few barbs only at the lip of the superior umbilicus, *e.g.*, remiges. Thus the *Spule* should really be drawn-out ventrally as well as dorsally to form the basis of a typical feather. Davies's statement also quoted by Lillie and Juhn that " all the parts which bear rays are only specially developed parts of the original *Spule* " may be taken as meaning that the intermediate cell layer (forming *Spule* or collar) is responsible for the entire feather, and thus has no bearing on the method of formation of the shaft.

Strong, in 1902 (*b*) published an account of an abnormal feather in a hybrid pigeon, in which an attempt at calamus formation was succeeded by the secondary division of this quill-like structure into barbs and barbules. This is difficult to interpret on the concrescence theory of development. If the rhachis arose by concrescence of the two halves of the collar which supposedly forms the primordium of the shaft, the calamus must represent the complete shaft. This admits of no explanation of the method of development of the aftershaft, unless it is assumed to be the ventral side of the shaft. The division of the calamus proximally into barbs in Strong's abnormal feather can be readily explained on the fusion of barbs theory, as the rhachis and hyporhachis can quite conceivably split apart under abnormal conditions, reversing their method of formation.

This is strikingly demonstrated in the rectrices of young ducklings, fig. 28, Plate 19, where the first generation of nestling feathers has an excessively long calamus, which is continuous with the barbs of the succeeding definitive feather. This condition has also been experimentally induced by feeding thyroid to mature fowls, fig. 55, Plate 28. A condition which occurs abnormally in some birds (*e.g.*, Strong's pigeon) and normally in others, may be taken as definite proof of Davies's statement as to the rhachis consisting of fused barbs.

II—Growth-rate

Regarding the question of asymmetry in feathers, which according to Lillie and Juhn means " differences in growth-rate on the two sides of the feather germ at some time, or throughout development " (p. 157), there is no evidence that barbs arising simultaneously near the ventral point fuse with the rhachis more quickly on one side of the feather germ than on the opposite side. On the concrescence theory of development this might be so. When, however, ridges are passively cut out from the intermediate cells so that passing down the feather, cells are laid on to the forming barbs from below and gradually nearer to the rhachis, making a barb lying at first near the ventral point ultimately fuse with the rhachis—it is difficult to see how growth-rate can enter into the scheme at all.

'Espinasse (1934) has recently pointed out some difficulties preventing complete acceptance of the growth-rate theory of Lillie and Juhn. He considers that

growth-rates on the two sides of the collar can only differ if one of two sets of circumstances holds for that feather germ. "Either (*a*) the barbs on the two sides must be of different lengths, and the rhachis curved, since one side of it has grown faster than the other; or (*b*) the feather germ must have an asymmetry of just such a kind and degree as to compensate for the difference in growth-rate and give a straight feather. This asymmetry might be in fact a displacement of the ventral growing point from its theoretical position diametrically opposite the forming rhachis; then the more rapidly growing side would get carried out of the region of growth so much sooner than the more slowly growing side, having the less distance to travel, as to be the same size or even smaller."

Now in certain feathers of the fowl, *e.g.*, remiges, all these conditions are present. The barbs of one side are shorter than on the other; the rhachis tends to be curved away from the shorter side, fig. 54, Plate 28, and also the ventral point is deflected towards the side of the rhachis bearing the shorter barbs, fig. 53, Plate 28.

The curvature of the feather, and also of the rhachis is possibly due to the habit of the bird in holding the wings pressed against the sides of the body. As the follicles are very close together in this position, constant pressure on the developing germ would perhaps cause the feather to assume the curved shape. This, however, does not explain the curvature in other feathers, *e.g.*, the sickle feathers of the male, although in such cases the follicles are invariably crowded. The two facts of (*a*) the barbs on one side of the vane being shorter than on the other side, and (*b*) the deflexion of the ventral point are self-explanatory without considering growth-rate.

Lillie and Juhn emphasize the point that they consider narrowness of the vane to be correlated with rates of growth, but if of two barbs arising simultaneously on either side of the ventral point, one is at a shorter distance from the rhachis than its neighbour, then the vane on that side will be narrower. From the figures given, it is obvious that the right-hand side of the vane in fig. 53, Plate 28, will be much narrower than on the opposite side, while in fig. 52, Plate 28, the vane on the right-hand side will be slightly wider than on the opposite side.

It sometimes happens that the barbs of a feather are not of uniform length along one side of the vane, fig. 54, Plate 28. Occasionally barbs arise which are twice the length of their neighbours. This indicates that the so-called ventral point is not fixed, but in this region of plasmatic growth, a barb which lay nearer to the left side of the feather germ might grow towards the right side of the rhachis. On the concrescence theory, a reversal of the streaming movement of the cells of the collar, carrying with them only one barb centre would be necessary to explain this condition—a hypothesis which is difficult to visualize and which would be still more difficult to prove. Again, it would seem that such a barb would arise on one side of the rapidly growing region, which is supposedly concentrated at the ventral point, and would grow through this region, its rate of growth decreasing at a much later period than in barbs which actually arose at the ventral point. It is difficult to conceive of the growth-rate of such a barb in terms of the diagram of organization of the 12 day feather germ figured by Lillie and Juhn (1932), p. 129. It is true

that this is for a breast feather, whereas the feather figured here is a secondary flight feather, but such discrepancies have been observed in feathers from all pterylae, although not in such a marked degree.

A recent paper from the Chicago laboratories (JUHN and FRAPS, 1934) states that the " transposition of the definitive feather pattern indicates that the differences in growth-rates in the germ in respect of barb level are relatively small " (p. 1182) and later (p. 1183) with regard to transposing fault bars to the collar, " the apparent differences in barb growth-rates thus arrived at are smaller than are the differences called for by the original curve of barb growth ". Thus a more gradual curve is now indicated, but even so, irregular barbs as in fig. 54, Plate 28, do not satisfy the conditions necessary for such a graph.

III—SUMMARY

There are no apparent histological grounds for the concrescence theory of development of a feather, as the rhachis and hyporhachis form through fusion of barbs, and the lateral fusion of these structures gives rise to the calamus.

Asymmetry in feather form arises through the deflexion of the region of " plasmatic growth " nearer to one side of the rhachis, instead of its being fixed diametrically opposite the dorsal point. This results in narrowing of the vane on one side, irrespective of growth-rate in the barbs, and occasionally in lack of uniformity in length of individual barbs.

III—REGENERATION OF FEATHERS AFTER PLUCKING

I—INTRODUCTION

A common method of studying plumage reactions in relation to internal secretions consists of plucking numbers of feathers from different pterylae, before or after the administration of certain hormones. The feathers which are subsequently regenerated are presumed to have grown under the influence of this hormone, and may be expected to show its full effect. In an unplucked bird as the majority of feathers are fully grown and therefore dead structures, the hormone can have little effect. It is generally understood that the annual moult is marked by, and probably the result of a general upset in the metabolism of the bird, and hence such a time of new feather growth is unreliable for the progress of experiments. Although a considerable amount of work has been done on experimental moulting, as contrasted with deliberate plucking (Zawadowsky, 1925 ; Zawadowsky and Rochlin, 1927) it would be obviously unsuitable to make use of the resultant new growth of feathers for the assay of the effects of hormones.

The method usually adopted, therefore, consists of denuding areas on the birds some time before, or immediately after the experiment is scheduled to begin. On the whole, this appears satisfactory, but actually the results may be deceptive. For instance, it is well known that after plucking, some feathers regenerate more rapidly than adjacent feathers of the same tract. This has been assumed to be due to the follicles of the more rapidly regenerating feathers being nearer the normal period of moult than the more slowly growing feathers (Lillie and Juhn, 1932), but when one regenerating feather may be fully grown before any sign of regeneration in its neighbour occurs, this assumption breaks down. Again, plucking will cause some injury to the papilla, unless the plucked feather is on the point of moulting, and thus the method of replacement of plucked feathers cannot be identical with that of feathers moulted in the ordinary way.

The mechanism of regeneration has not yet been fully worked out. Lillie and Juhn (1932) sectioned papillae immediately after plucking, and found the epidermis completely lacking at the tip. When some time had elapsed after plucking, the feather germ was seen by these authors to consist of (*a*) an apical zone where the pulp is constantly dying off ; (*b*) a middle zone occupied by barb primordia ; and (*c*) a basal zone occupied by the collar and without barbs.

It seemed advisable, therefore, to make a detailed study of the interval between the stages noted by Lillie and Juhn, and to compare the formation of the feather regenerating after plucking with the normal replacement already described (Part I, p. 167).

II—Material and Methods

For the detailed study of early stages of regeneration, feathers were plucked from the back and thigh of Rhode Island Red fowls aged 8 weeks, at intervals of 72, 51, 27 and 4 hours before killing. Later stages were obtained by plucking a 16-weeks old fowl of the same breed in the following areas : (1) neck, 8 days ; (2) anterior and posterior breast, 7 days ; (3) thigh and abdomen, 6 days ; (4) anterior back, 4 days before killing. This bird had been addicted to cannibalism, and the posterior back showed various stages of regenerating feathers of unknown age.

In all cases, the skin of the plucked region was fixed in Bouin's solution ; feathers embedded in celloidin and wax by the long method (Part I, p. 145) ; cut from 6–8 μ ; stained in iron haematoxylin (short method) and counterstained in picro-fuchsin.

III—Early Stages

Even in the early stages of regeneration (4–72 hours after plucking) great variety exists between individual germs. All feathers are still deep within the follicle, but occasionally a small plug of cornified material projects from the mouth. This is probably due to profuse bleeding, and cannot be considered as due to the activity of the papilla. Microscopic differences are apparent showing differences in the

amount of healing and regeneration in adjacent follicles, but these are not so pronounced as in the later stages studied (4–8 days after plucking). Whereas in the early stages it is possible to judge the age of regenerating feathers with a fair degree of accuracy, in the later stages it is quite impossible in individual feathers. In the area as a whole, by taking the average lengths of numerous regenerating feathers, the period elapsed since plucking can be ascertained, but not in individual cases. The early stages will therefore be considered in detail, and the later stages without reference to time.

4 *hours*—Profuse bleeding into the follicle occurs on plucking, which partially obscures the development of the new papilla. The torn base of the old papilla may be seen projecting as an irregular mass of dermal tissue into the cavity of the follicle, fig. 39, Plate 23. There are only traces of epidermis on this papilla. The skin is often torn away from the base of the follicle, and sometimes also from the walls, if the plucked feather happens to be a very young one and therefore with an uncornified sheath still indistinguishable from the sheath of the follicle.

27 *hours*—Transverse sections through the base of a thigh feather 27 hours after plucking, show the epidermis of the feather and follicle to be fully formed and differing from the normal condition in that a wide, blood-filled space separates them. Very slight traces of ridge formation may be present in the intermediate cells.

In other feathers of the same age, the epidermis of the follicle may be complete, while the papilla is still without an epidermis except for an incomplete ring at the base.

In longitudinal sections of back feathers 27 hours after plucking, the epidermal covering at the base of the papilla is seen to be continuous with the epidermis of the follicle. At the tip of the papilla it is incomplete and broken by blood spaces. Strands of torn tissue project into this space, fig. 38, Plate 23, and may cornify with the follicle sheath if lying in close proximity to it. The epidermis of the papilla becomes constricted below this irregular tip, completely cutting it off.

A plug of cornified tissue is usually present in the mouth of the follicle, and cornified particles line the sides. Examination with the oil immersion lens shows these particles to be chiefly blood cells.

51 *hours*—The dermal papilla is now more definite, with a complete epidermal covering, and the intermediate cells show signs of grouping into ridges. These incipient ridges are continued below the blood space at the base. Presumably therefore, this region will be pushed into the follicle cavity when more dermal tissue is drawn into the papilla.

Transverse sections of back feathers at this stage show an increase in the amount of cornified tissue in the blood space, fig. 43, Plate 23. It is assumed that the whole of the torn tissue and the blood in the follicle cornify and are carried upwards by the growth of the new feather, being finally pushed out of the follicle and shed with the sheath.

The feather papilla becomes progressively wider in diameter towards the proximal end, while the blood space decreases, until the actual base presents the appearance

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of a normal feather, in which the sheaths of both feather and follicle are closely adhering.

In one feather, serial transverse sections from tip to base, show the intermediate cells and cylinder cells projecting inwards and ultimately cutting off a circular feather from the rest of the papilla. Thus the appearance of a feather within a feather is obtained, but this is more marked in the next stage.

72 *hours*.—The double nature of the feather is continued to the base, where a double crescent of epithelium is seen in transverse section, fig. 41, Plate 23.

Longitudinal sections of back feathers show the whole of the tissue within the previously blood-filled part of the follicle to be cornified, the tip of the small feather cornifying, and the double region still uncornified, fig. 42, Plate 23.

IV—Later Stages

The difference in the ages of the newly regenerated papillae is clearly seen in sections of later stages. Some feathers are already extruded from the mouth of the follicle, while others are no further advanced than the 72-hour stage. In a large number of birds plucked for other purposes, this difference is particularly noticeable. Twenty-eight days after plucking, feathers in certain areas are not visible macroscopically, while others are 4 cm or more in length.

The type of development after the third day of regeneration appears dependent on the amount of injury sustained by the papilla. If only slightly injured—whether due to more careful plucking or to the greater age of the old feather (and consequently greater withdrawal of the pulp proximally) the papilla beneath the cornified plug of torn tissue and blood may regenerate a complete new feather. In this case there is little difference between the feather regenerated after plucking, and a feather regenerated after the normal moult (Part I, p. 167). Such a feather will not have barbs extending quite to the tip, as the epidermis there is too thin to give rise to ridges, as described by Lillie and Juhn (1932). This thin part of the epidermis will be differentiated into two regions, (*a*) a thin layer of intermediate cells which cornify with or without forming incipient ridges, and (*b*) the cylinder cell layer which forms the first cap of the new feather. The whole of this region will eventually be sloughed with the cornified plug when the feather breaks from the sheath.

By far the commoner type of regeneration is that where a transitory papilla is formed after plucking, as indicated above for the 72-hour stage, but intermediate grades are seen. The tip of the new papilla may be split, fig. 44, Plate 24, and the cornified plug extend between the two parts. Regeneration of the epidermis over this split papilla will take place, but no definite barbs are formed in this region. Eventually, the pulp withdraws from the split tip (as in normal withdrawal of pulp) and the intermediate cells cornify, becoming continuous with the sheath. These are ultimately shed, but definite barbs are formed lower down.

If the tip of the papilla is badly torn, the whole of this region is constricted off from a new papilla which forms beneath. Strictly speaking, the "new papilla" is

merely the base of the old one, but since the papilla grows from the base, the torn tip is carried distally, and when the constriction occurs between the tip and the basal region cutting off the former, the latter gives the impression of an entirely new structure, figs. 40 and 42, Plate 23. It sometimes happens that the first papilla is so badly torn that the intermediate cells and pulp are intermingled in an indefinite mass. The intermediate cells, however, appear to possess an inherent capacity for forming ridges, no matter how badly injured or how much they are displaced from their normal position. Fig. 45, Plate 25, shows the dense, partly cornified first papilla with definite signs of ridge formation, the details of which are given in fig. 45*a*. More proximally, a new papilla is being formed which will actually give rise to the new feather. In extremely bad cases of injury, the tip of the second papilla forms incipient barbs without definite ridge formation. These cornify, but are sloughed with the sheath, and more proximally the true barbs are formed.

The follicle generally assumes a curved shape during regeneration, which is probably due to the distortion caused by plucking, and also to the profuse bleeding into the follicle. This blood is prevented from escaping by the narrow follicle mouth which becomes sealed with coagulated blood. Thus the follicle after plucking seldom retains its original shape, and the new feather follows a curved course in regaining the obliquity to the surface which is characteristic of feathers, fig. 44, Plate 24.

This curvature is seen by the " crease " in the follicle wall which marks off the enlarged cavity of the torn region of the follicle, and forms a definite constriction between the part of the papilla destined to give rise to permanent barbs, and the first papilla or the cornified plug, according to the degree of injury and consequent type of regeneration. This is illustrated by the diagrams of cross-sections in figs. 45–51, Plates 25–27. Fig. 45, Plate 25, is through the tip of the follicle, and shows an indefinite mass of cornifying material within a thick sheath. The sheath of the follicle is also unusually thick, and the distinction between the two sheaths is very obscure. In this cornifying mass attempts at ridge formation may be seen, fig. 45*a*, Plate 25. Fig. 46, Plate 26, is slightly lower down, and shows the tip of the second papilla ; fig. 47, Plate 26, shows the beginning of the crease which marks the region where a constriction will take place between the distal and proximal parts of the second papilla. Above this crease, traces of barbs are to be seen among the intermediate cells of the second papilla. These never form true barbs, although they cornify and become separated from the rest of the epidermis, figs. 48–50, Plates 26–27. The curvature of the whole follicle is well illustrated by the fact that in these serial transverse sections, both first and second, and finally all three papillae are present in the same section, for a certain distance down the follicle. Near the base, the second and third papillae are completely separated by dermal tissue. This is foreshadowed in fig. 51, Plate 27, where the proximal and distal parts of the follicle are separated by intermediate cells.

Regeneration of feathers after plucking, therefore, differs from normal regeneration, owing to the base of the follicle in the former condition having to regenerate

a papilla before the feather can be formed. The completeness of the papilla in normal regeneration, is due to the outer layers of the intermediate cells at the base of the feather germ forming the proximal end of the calamus of the old feather, while the inner ones are differentiated into a protective sheath persisting as a covering to the papilla when the old feather is shed. Regeneration after plucking is further complicated by the extent of the injury sustained by the papilla, and also by the amount of bleeding into the follicle. These two factors are also responsible for the decided curvature in the follicle, through which the new feather has to steer a diagonal course to the surface. As a result of this, the papilla may make several abortive attempts to form a new feather. When the distortion in the follicle and the injury to both follicle and papilla are completely overcome, the regenerated feather is an exact replica of its predecessor.

V—Summary

Regeneration after plucking takes place in the following stages, according to the degree of injury sustained.

A.—Slight Injury.

(1) Cornification of blood and torn tissue in follicle.
(2) Proliferation of epidermis over torn papilla.
(3) Formation of new feather from healed papilla.

B.—Moderate Injury.

(1) and (2) As in A.
(3) Widening of basal part of papilla.
(4) Formation of second papilla by constriction between narrow, distal part (*i.e.*, first papilla) and wide base (*i.e.*, second papilla).
(5) Cornification of first papilla.
(6) Formation of new feather from second papilla.

C.—Great Injury.

(1) and (2) As in A and B.
(3) Possible grouping of intermediate cells in first papilla to form incipient barbs.
(4) Formation of second papilla (as in 3B) and formation of incipient barbs from intermediate cells.
(5) Cornification of first papilla.
(6) Third papilla constricted off from second papilla.
(7) Formation of new feather from third papilla.

In all cases, the excess of tissue formed above the actual feather regenerated, is shed with the sheath.

Regeneration of feathers after plucking differs from normal regeneration in that a functional papilla must first be formed, instead of the old papilla giving rise directly to a new feather within the base of the calamus of its predecessor.

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Description of Plates

Ant., anterior ; *b*, barb ; *ba*, barb of aftershaft ; *bl*, barbule ; *bs*, barb of shaft ; *bv*, blood vessel ; *c*, cortex ; *ca*, calamus ; *cc*, cylinder cell layer ; *co*, collar ; *cp*, cap ; *d*, dorsal ; *de*, dermis ; *e*, epitrichial layer ; *ep*, epidermis ; *ep. f*, epidermis of follicle ; *h*, hyporhachis ; *i*, intermediate cells ; *m*, medulla ; *ml*, Malpighian layer ; *p*, pulp ; *pa*, papilla ; *pc*, pigment cell ; *post.*, posterior ; *r*, rhachis ; *ri*, ridge ; *s*, sheath ; *sc*, stratum corneum ; *v*, ventral ; *pg*, region of " plasmatic growth".

Plate 16

Fig. 1—Transverse section of skin from the spinal tract of a 10 days embryo Aylesbury duckling, showing the elongation of the Malpighian cells and the concentration of the dermis.

Fig. 2—Transverse section through a feather filament of a 16 days embryo Aylesbury duckling, showing the intermediate cells of each ridge grouped together into median and lateral plates.

Fig. 3—Transverse section through the skin of the spinal tract of an 11 days embryo Aylesbury duckling, showing the elevation of the feather germ with the steeper slope on the anterior side, the whole greatly increased in size from the previous day.

Fig. 4—Transverse section through the base of a feather filament of a 17 days embryo Aylesbury duckling, showing the aggregation of intermediate cells into ridges, while the cylinder cell layer has not yet passed between them.

Fig. 5—Detailed drawing of the part of 6 lying between the lines a and b, showing the preliminary stages of pigmentation.

Fig. 6—Diagram of a transverse section through a prepenna and two preplumulae of a 15 days embryo Khaki Campbell duckling, showing the relative size of the two and the arrangement of the ridges.

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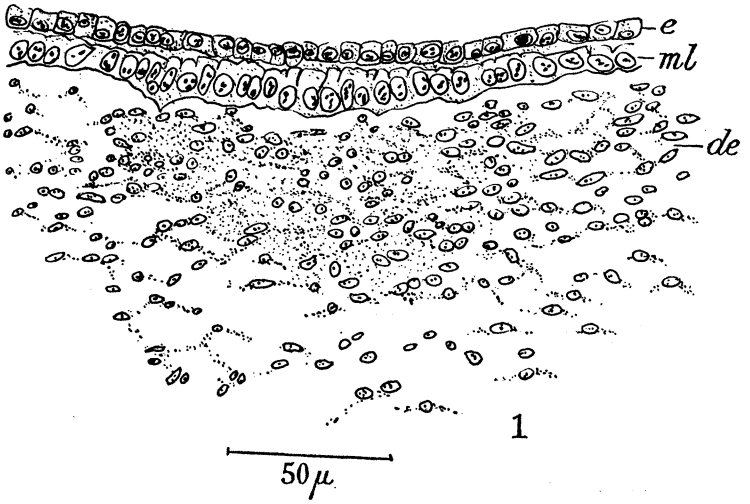

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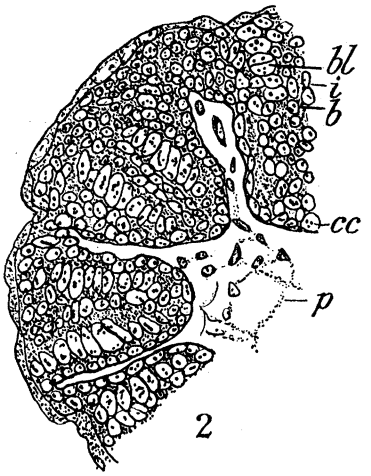

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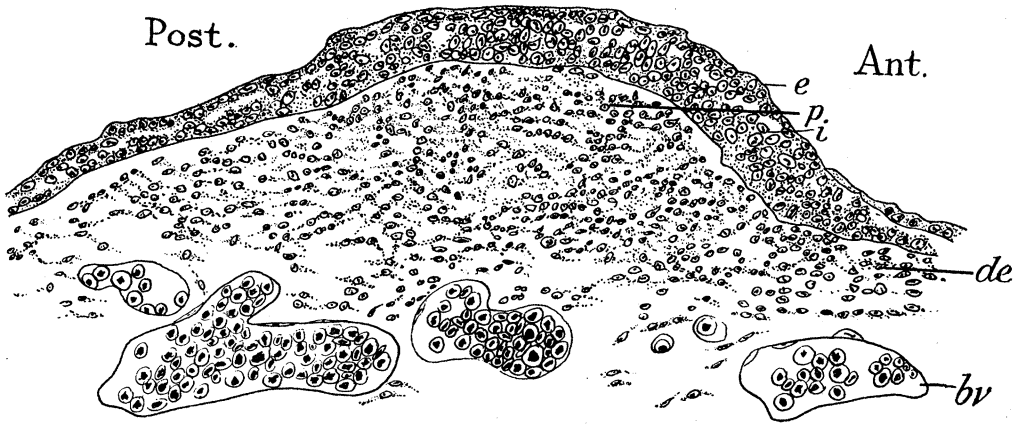

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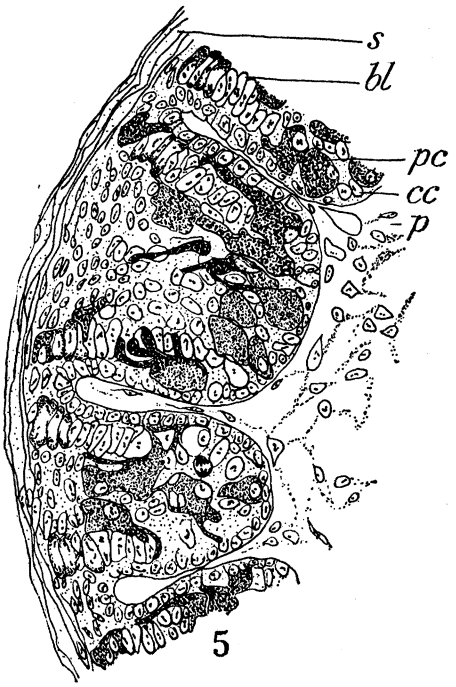

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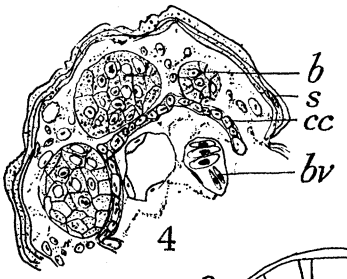

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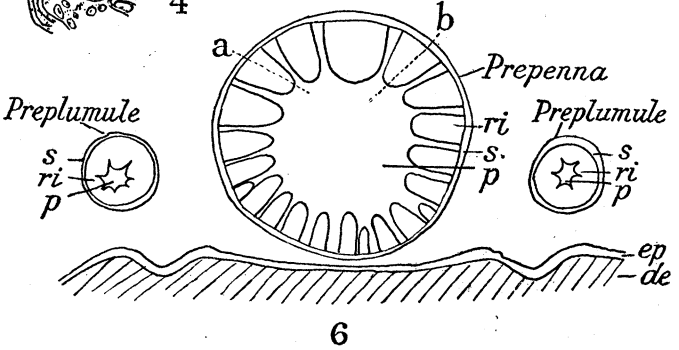

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Plate 17

Fig. 7—Diagram of a longitudinal section of a 15 days embryo Aylesbury duckling, showing the feather follicle and the continuity between corresponding layers.

Fig. 8—Detailed drawing of the part of 7 between the lines a and b, showing the differentiation of the intermediate cells of each ridge into barbs and barbules, and the withdrawal of the cylinder cell layer.

Fig. 9—Transverse section of a filament from an 18 days Khaki Campbell duckling embryo, showing the detailed structure of a barb and the withdrawal of the cylinder cell layer.

Fig. 10—Diagram of a transverse section near the base of a teleoptile from the wing of a chick (20 days) ; the smaller ridges on the left hand side represent the aftershaft.

Fig. 11—Transverse section near the tip of a filament of a 20 days chick embryo, showing the crowded arrangement of barbs and barbules, and their appearance after cornification.

Fig. 12—Detailed drawing of the part between the lines a and b in 10, showing the fusion of barbs to form the rhachis.

Fig. 13—Detailed drawing of the rhachis and adjacent barbs of the teleoptile of a chick embryo (20 days), showing the typical shape of the barbs and the elongation of the barbule cells.

Figs. 14–16—Diagrams of sections taken at the levels marked 14, 15, 16, in fig. 27. 14 showing the decrease in size of the barbs from the rhachis towards the ventral point. Fig. 15 shows the barbs of the shaft, the barbs of the aftershaft not being present. Fig. 16—Section through the junction of a protopile and a teleoptile. The darkened barbs belong to the teleoptile and the light ones to the protoptile.

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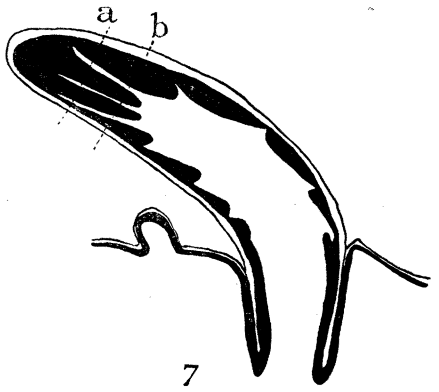

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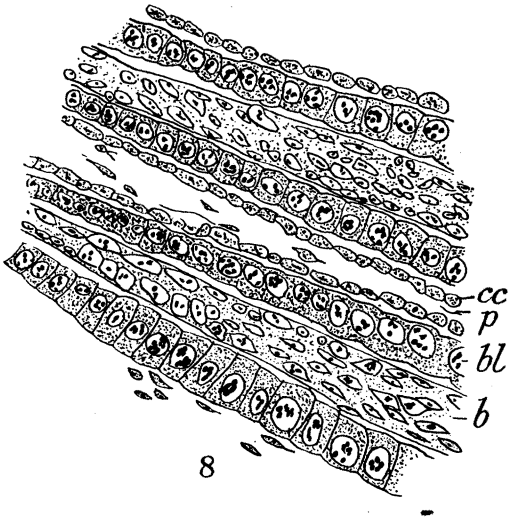

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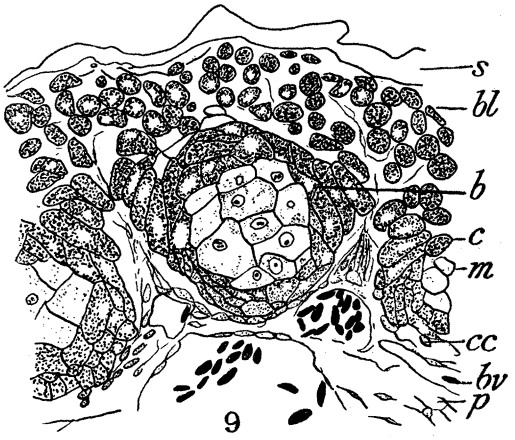

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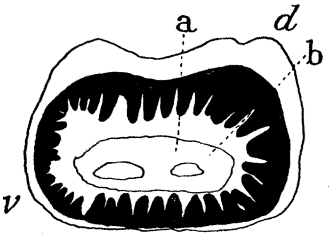

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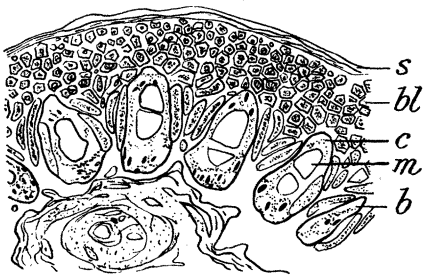
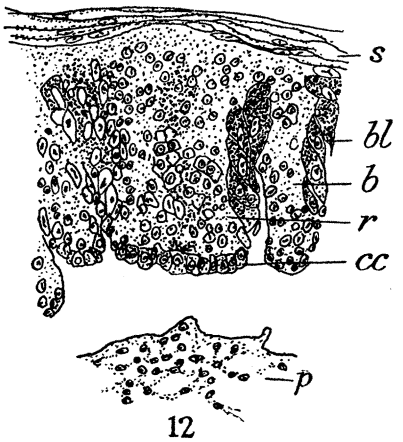


11

50 μ



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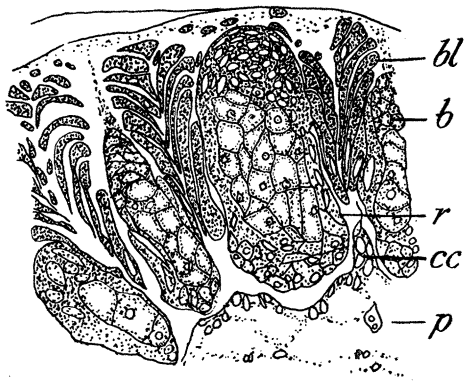

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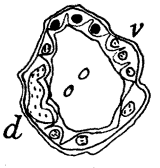

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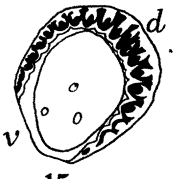

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Plate 18

Figs. 17–21—Transverse sections taken at different levels of a protoptile of a 24 days Khaki Campbell duckling embryo.

Figs. 22–26—Transverse sections taken at corresponding levels of a preplumule of a 24 days Khaki Campbell duckling embryo.

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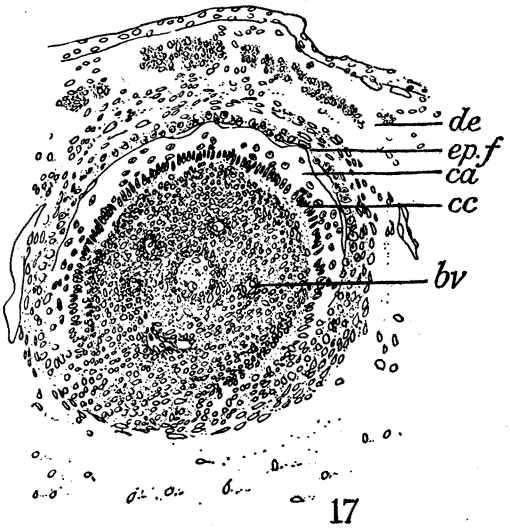
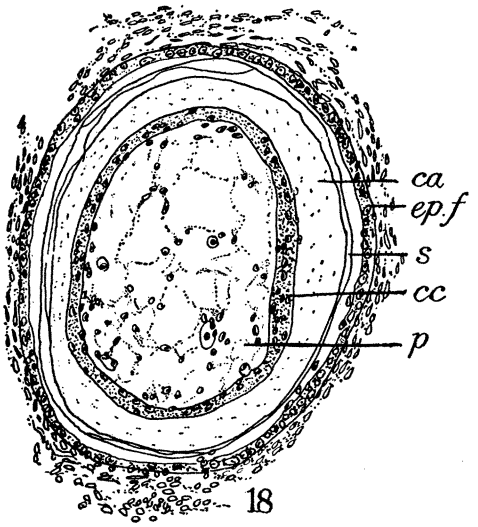
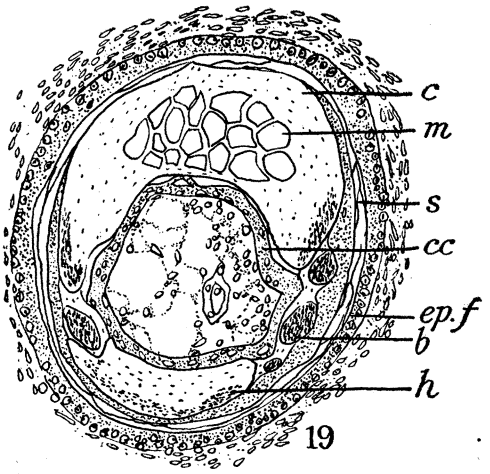
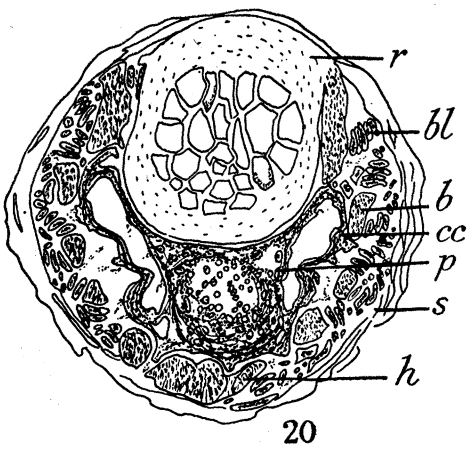
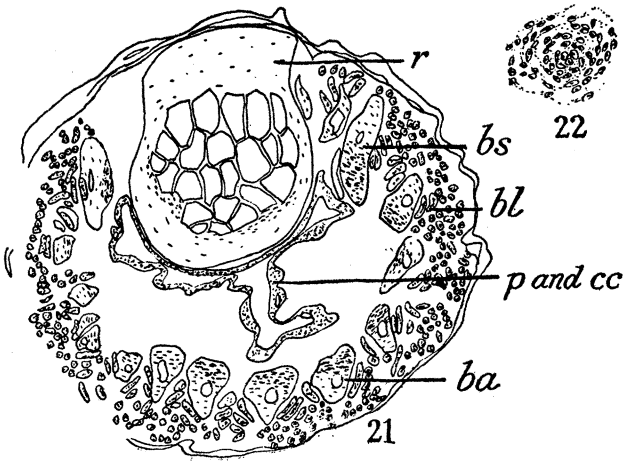








100 μ



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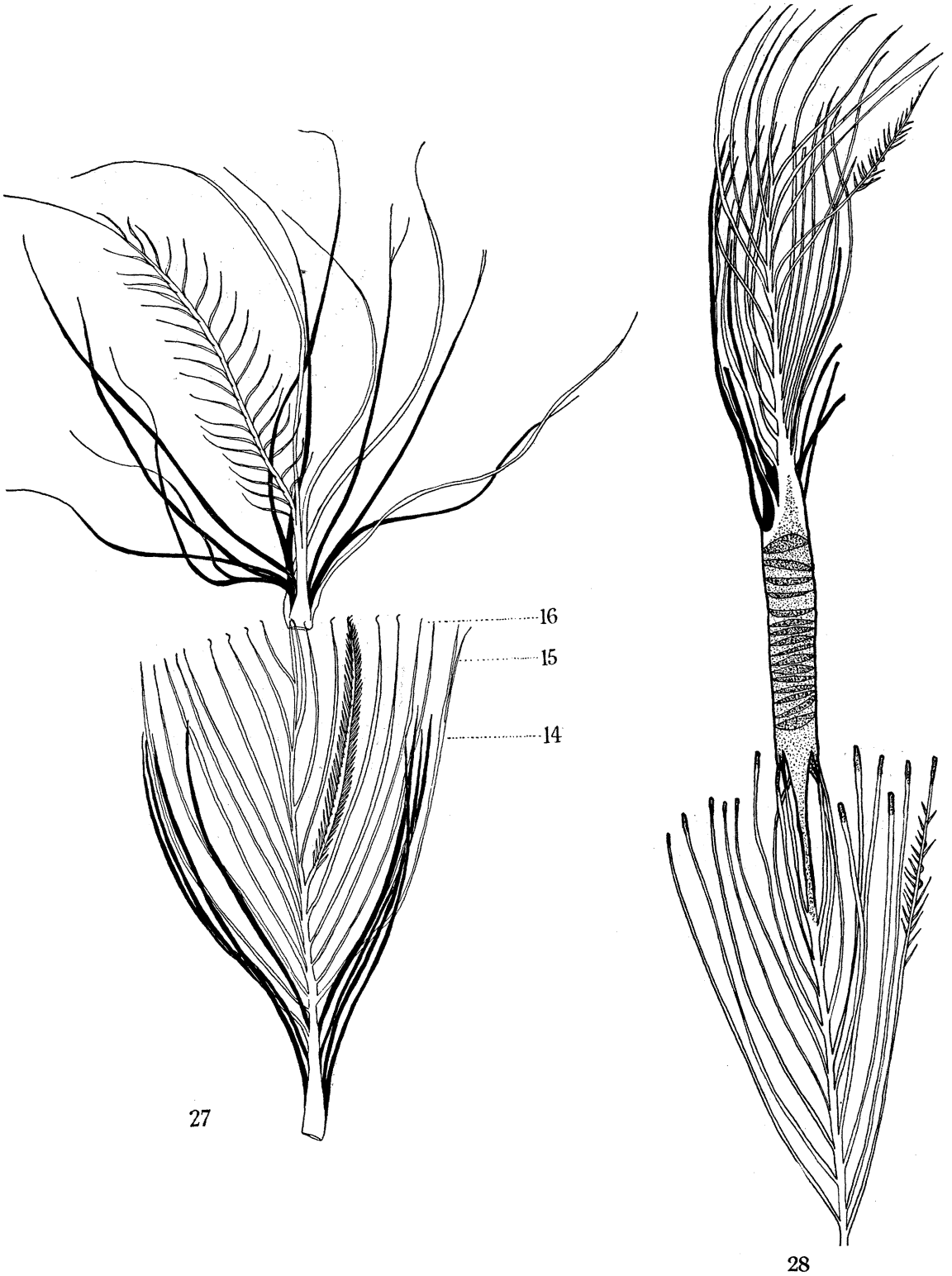

PLATE 19

FIG. 27—Protoptile and teleoptile from the wing of a day old chick.
FIG. 28—Protoptile and teleoptile from the tail of a 6 weeks old Khaki Campbell duckling.

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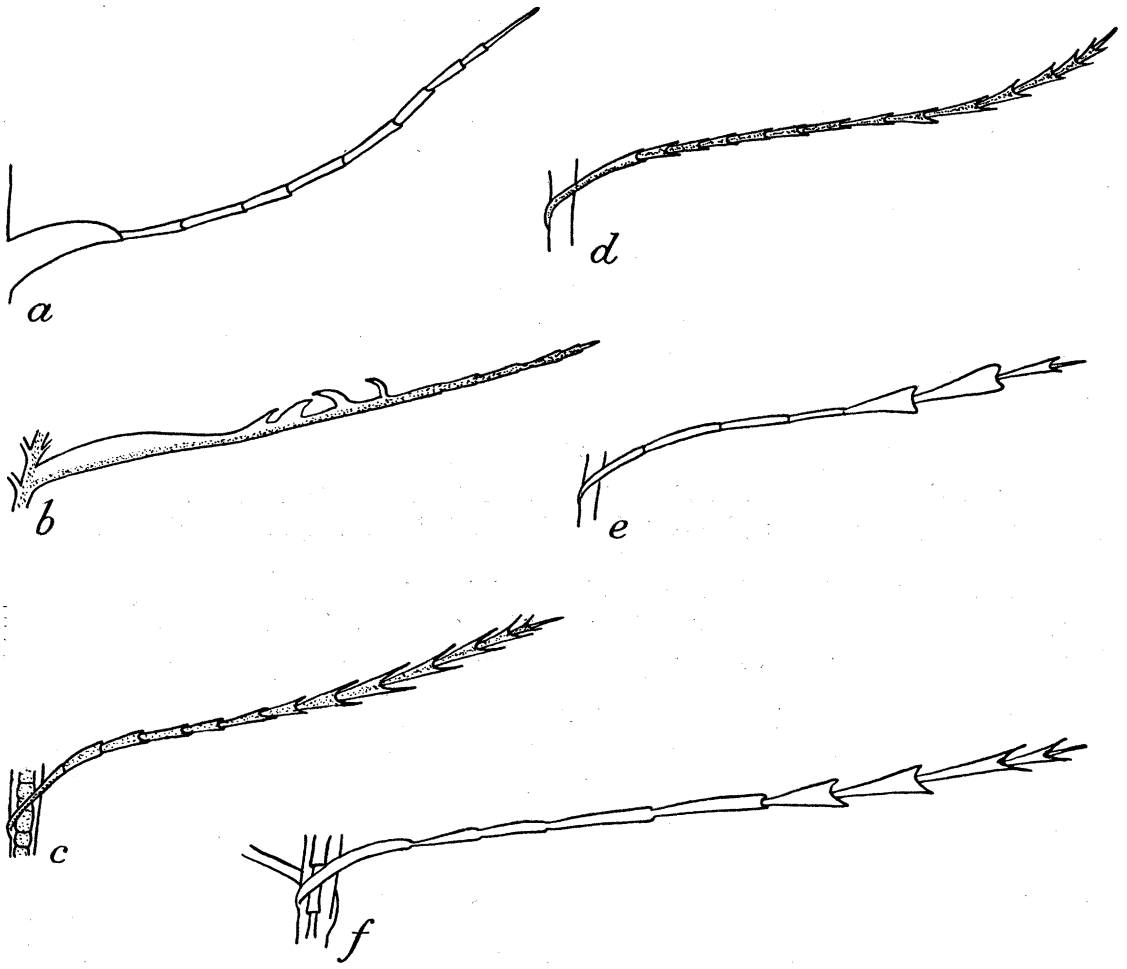

PLATE 20

FIG. 29—Barbules from (*a*) prepenna of chick ; (*b*) tip of remex of fowl ; (*c*) prepenna of Chinese gosling ; (*d*) prepenna of domestic duckling ; (*e*) plumule of wild duck ; (*f*) plumule of Chinese goose.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Plate 21

Fig. 30—Diagram of a transverse section of a back feather of a 2 months old Rhode Island Red fowl.

Fig. 31—Detailed structure of the region of " plasmatic growth " in fig. 30 where barbs pass either dorsally or ventrally.

Fig. 32—Detailed structure of part of fig. 30, showing the fusion of a barb with the rhachis.

Fig. 33—(*a*) Detailed structure of the barb and barbules from one ridge of a tail feather of a 2 months old Rhode Island Red fowl ; (*b*) Two barbs of fig. 33*a* under high magnification.

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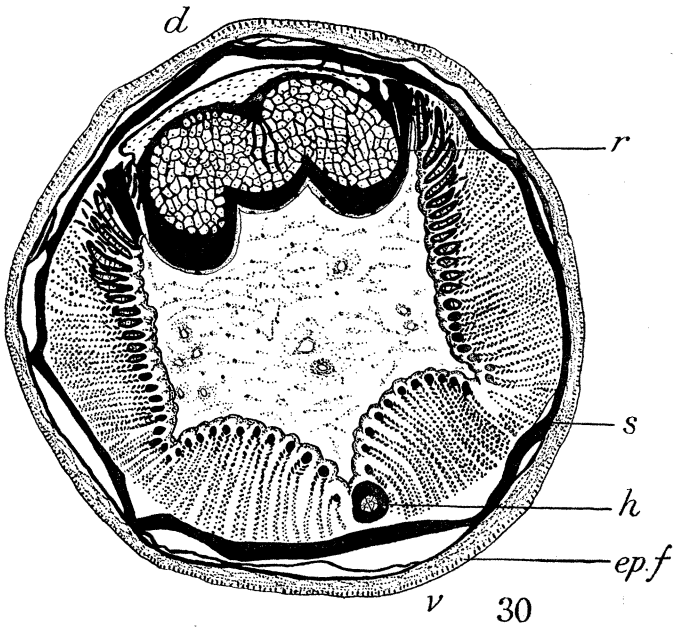

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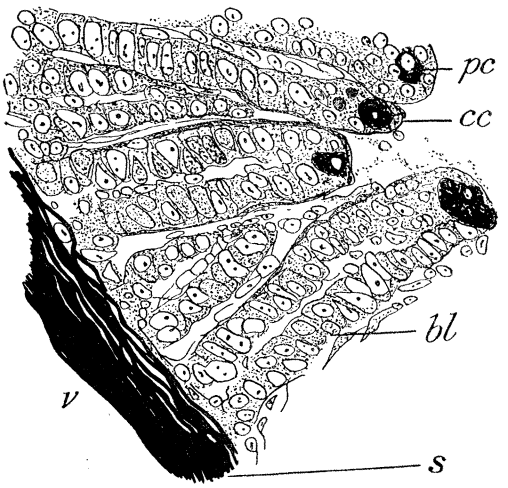

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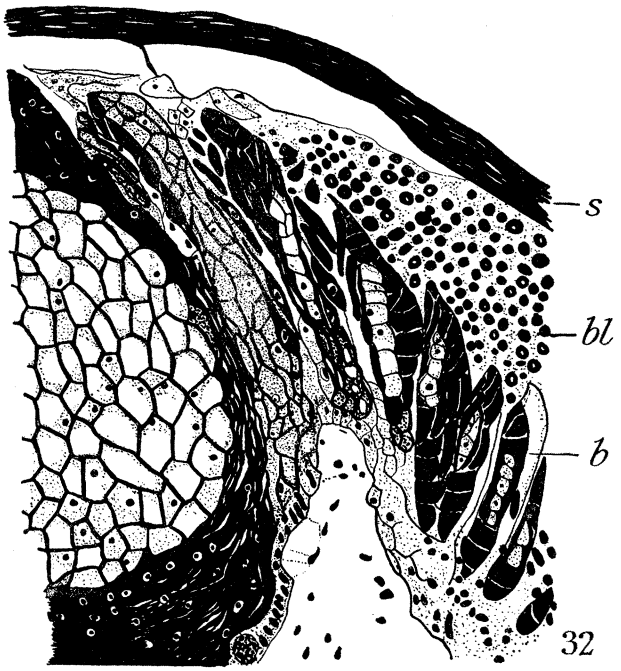

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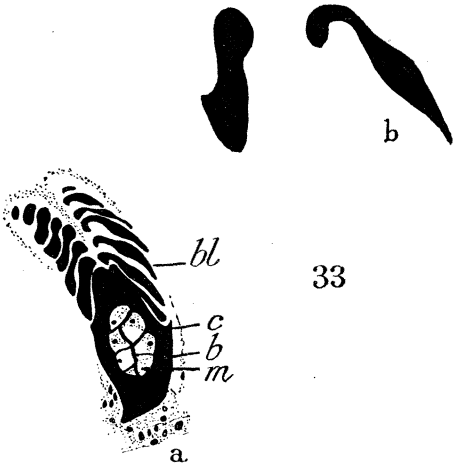

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Plate 22

Fig. 34—Diagram of a longitudinal section through the tip of a regenerating feather, showing the feather caps (10 weeks old Rhode Island Red, tectrice majores).

Fig. 35—Drawing through the base of the feather shown in fig. 34, showing the continuity between intermediate cells round the base of the calamus of the feather about to be shed.

Fig. 36—Diagram of the feather papilla shown in detail in fig. 35.

Fig. 37—Drawing of a longitudinal section through a developing feather cap in the tail feather of a Starling.

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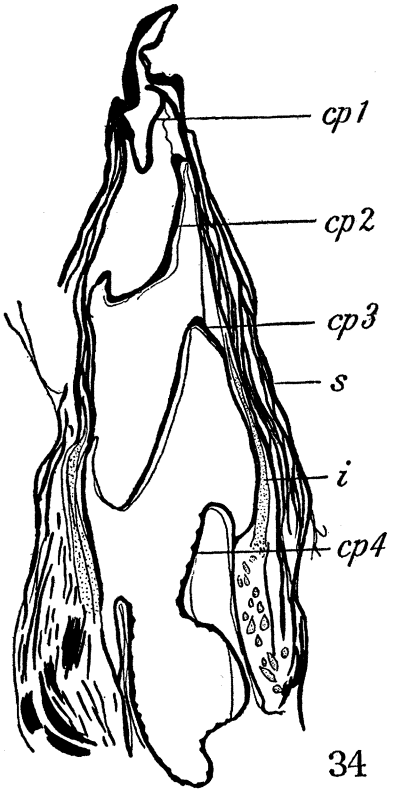

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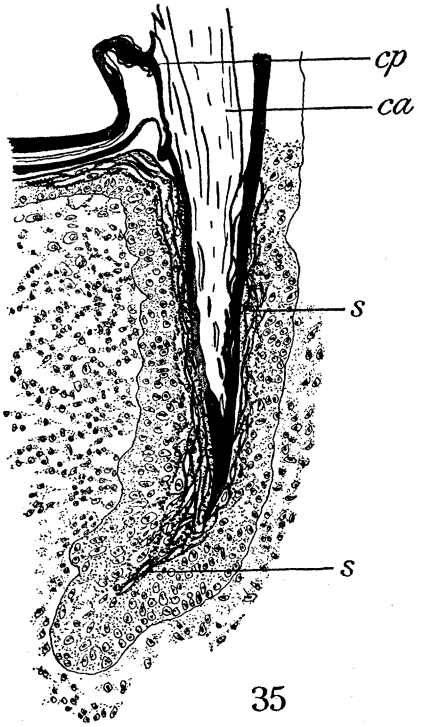

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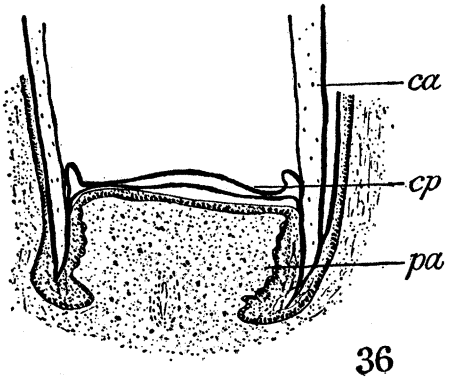

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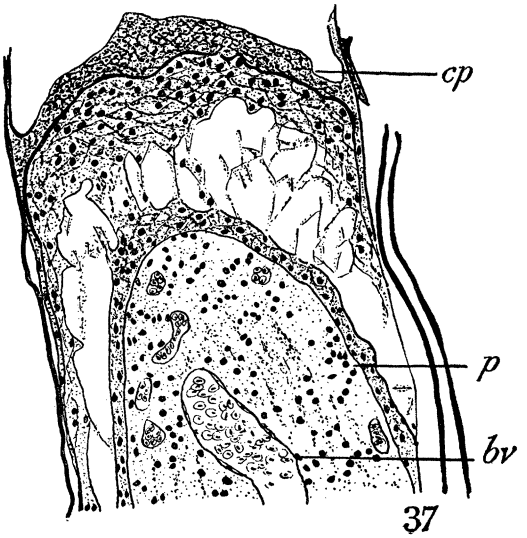

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Plate 23

Fig. 38—Longitudinal section of 27 hours regenerating back feather.
Fig. 39—Transverse section of 4 hours regenerating back feather.
Fig. 40—Longitudinal section of 51 hours regenerating thigh feather.
Fig. 41—Transverse section of 72 hours regenerating back feather.
Fig. 42—Longitudinal section of 72 hours regenerating back feather.
Fig. 43—Transverse section of 51 hours regenerating back feather.

bf, blood in follicle; *ct*, cornified tissue; *sf*, sheath of follicle.

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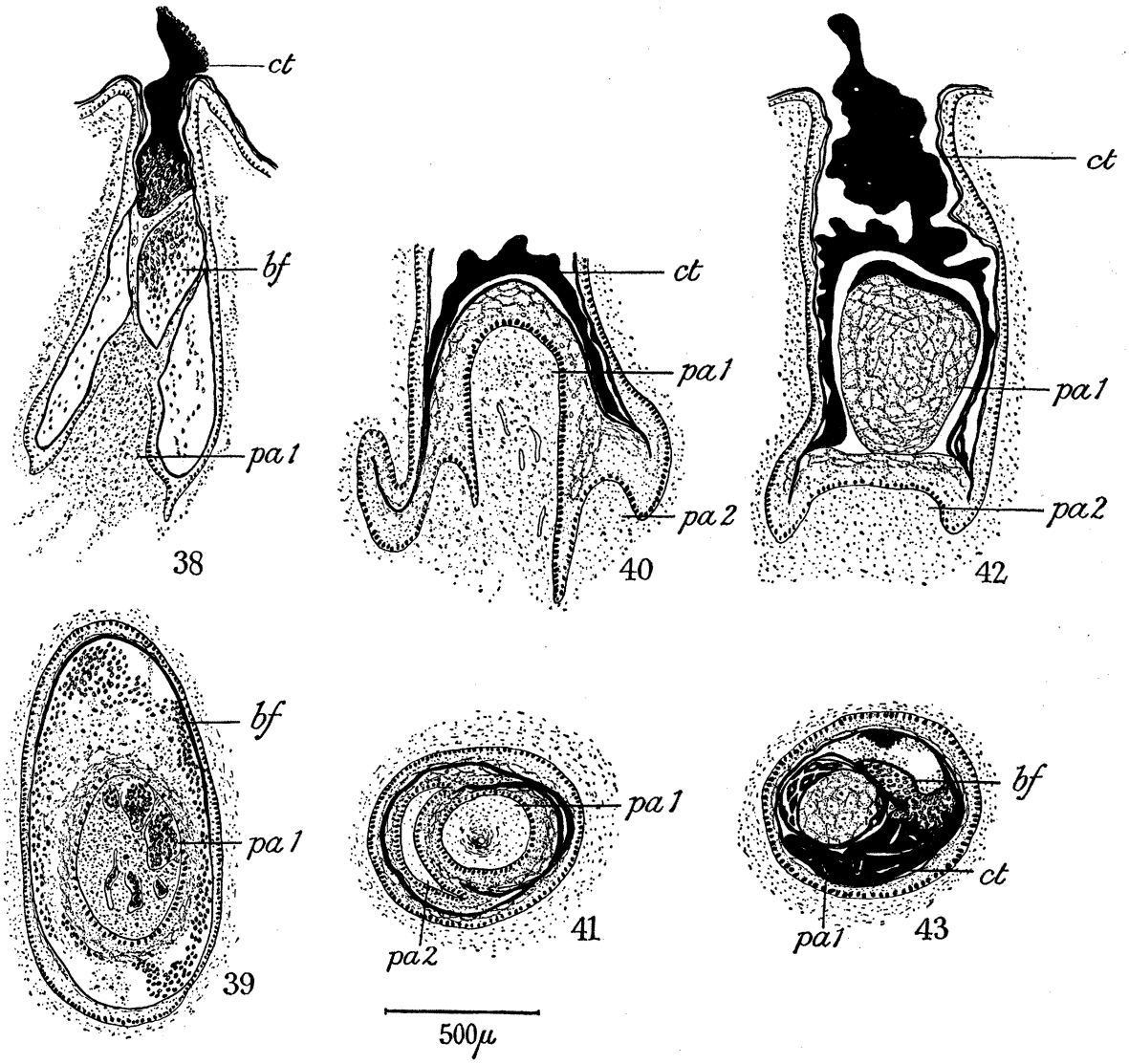

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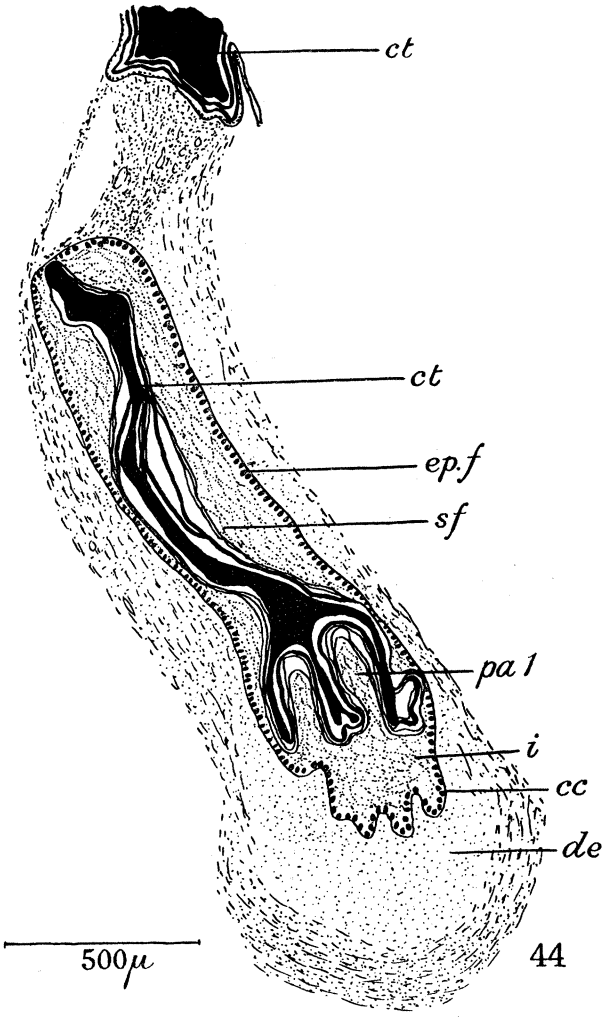

PLATE 24

FIG. 44—Longitudinal section of 6 days regenerating thigh feather.

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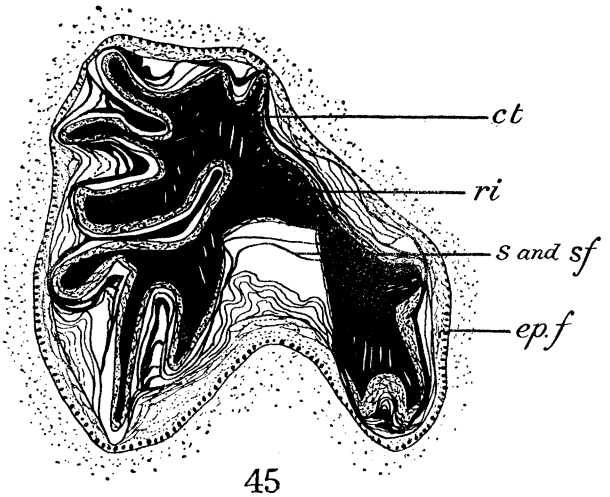
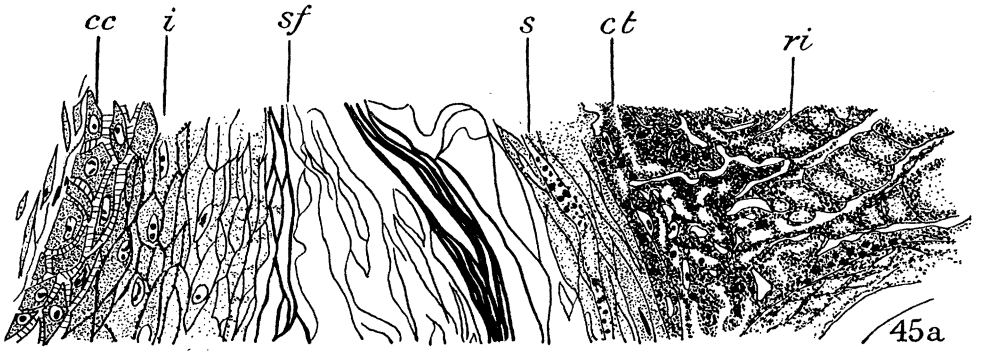


45

500 μ



50 μ

PLATE 25

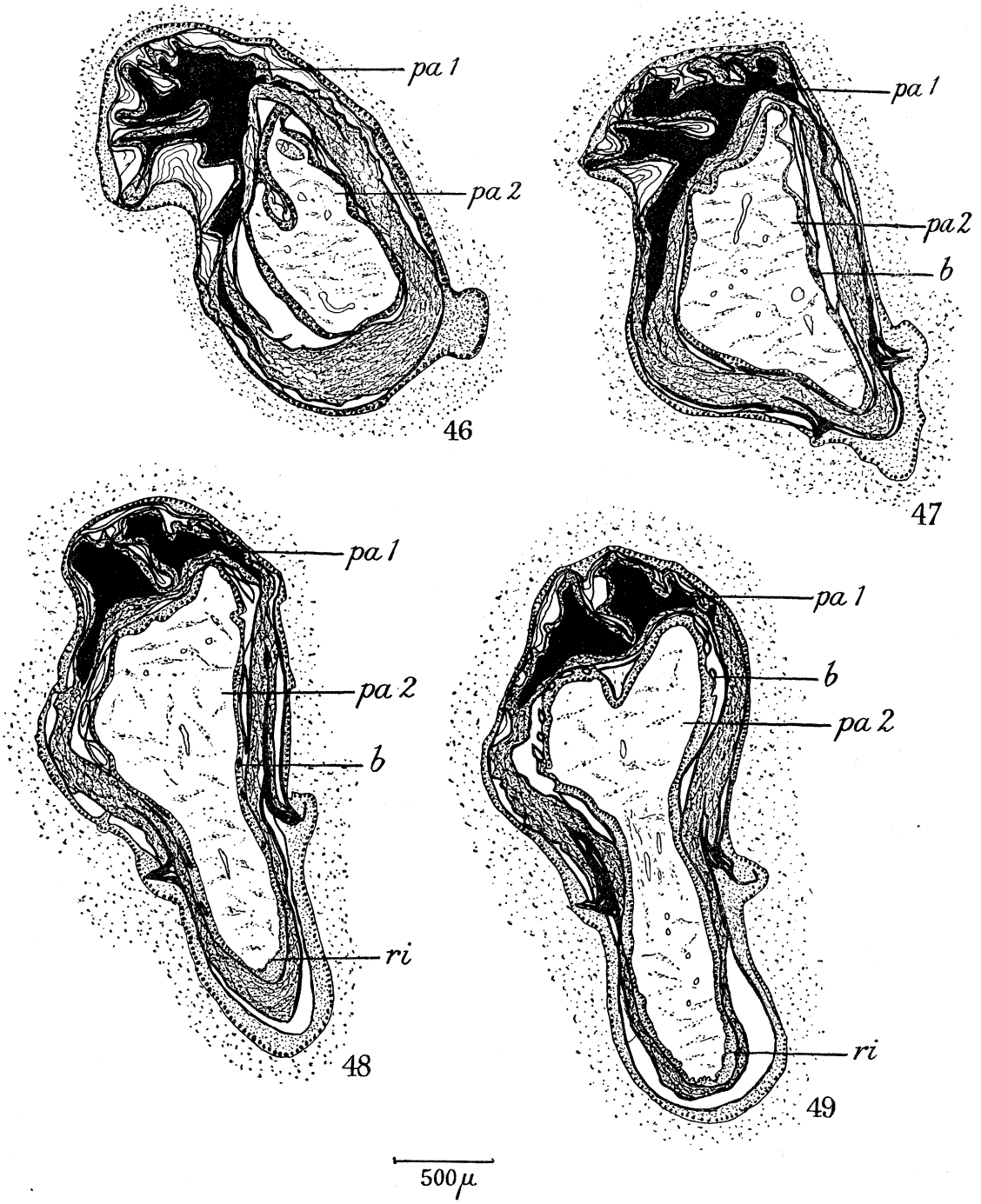
FIG. 45—Transverse section of tip of 7 days regenerating posterior breast feather.
FIG. 45*a*—Drawing of part of fig. 45 under oil immersion lens.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Plate 26

Figs. 46–49—Transverse sections of same feather as in fig. 45, from tip to base.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

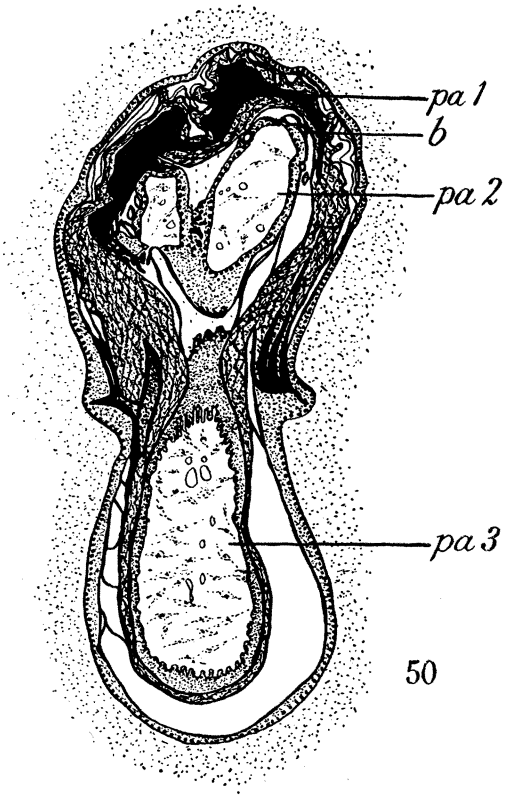


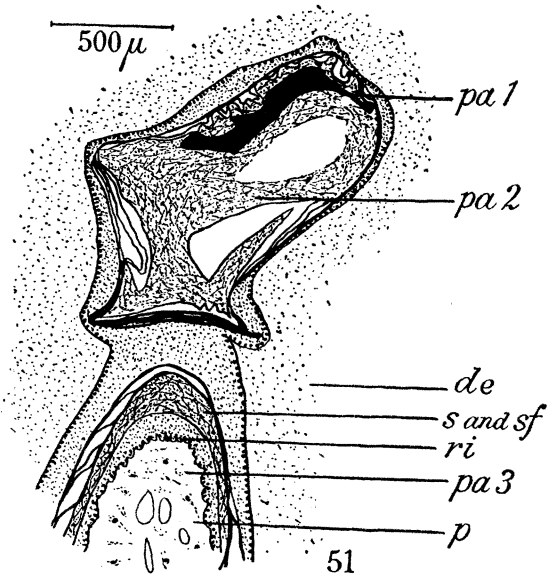
PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Plate 27

Figs. 50 and 51—Transverse sections of same feather as in fig. 45, from tip to base.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES





PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Plate 28

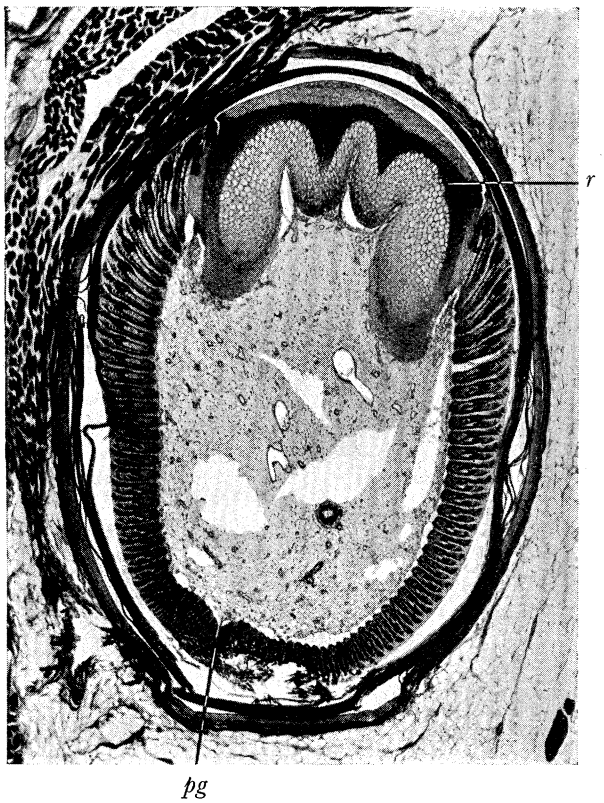
Fig. 52—Microphotograph of transverse section near the base of a thigh feather of an 8 weeks old Rhode Island Red fowl, showing the deflexion of the region of plasmatic growth nearer to the left side of the rhachis.

Fig. 53—Microphotograph of transverse section near the tip of a primary of an 8 weeks old Rhode Island Red fowl, showing the deflexion of the region of plasmatic growth nearer the right side of the rhachis.

Fig. 54—Photograph of a secondary from the wing of a 6 weeks old Rhode Island Red fowl, showing (*a*) asymmetry in the width of the vane ; (*b*) curvature of the rhachis towards the wider side of the vane ; and (*c*) unequal length of adjacent barbs.

Fig. 55—Microphotograph of the junction between two generations of feathers from the back of a Rhode Island Red fowl fed 30 grams thyroid, showing continuity between barbs of the new and the calamus of the old feather.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES



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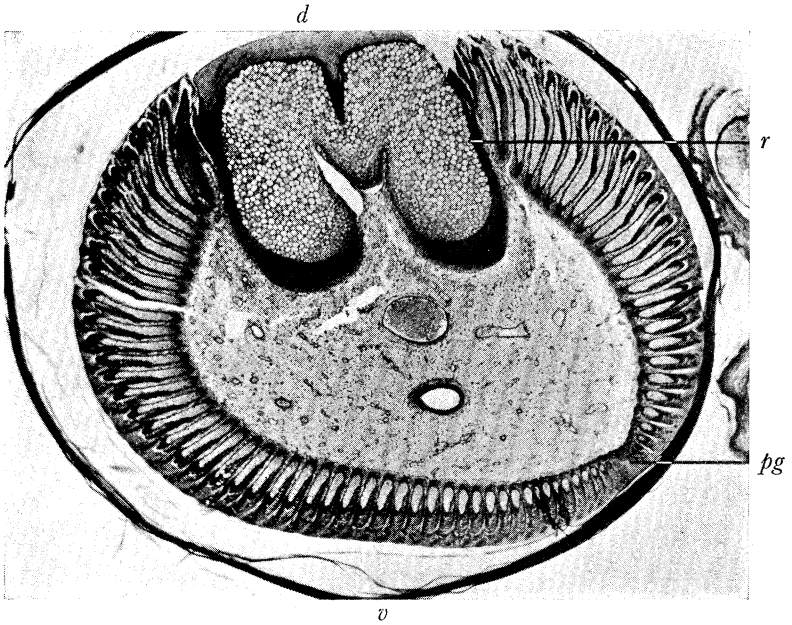

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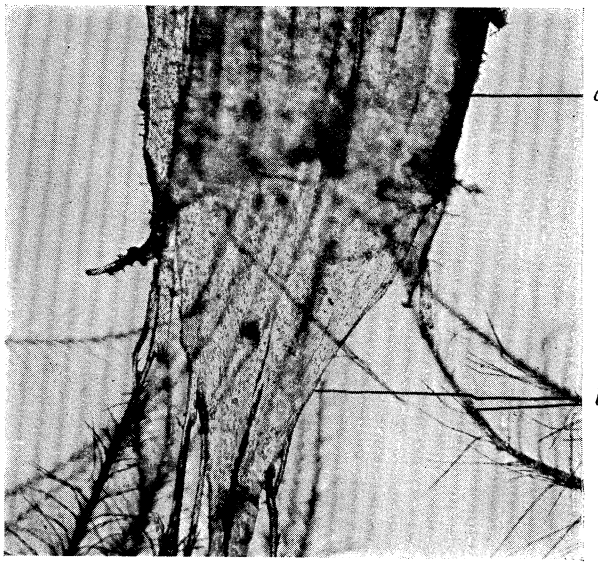

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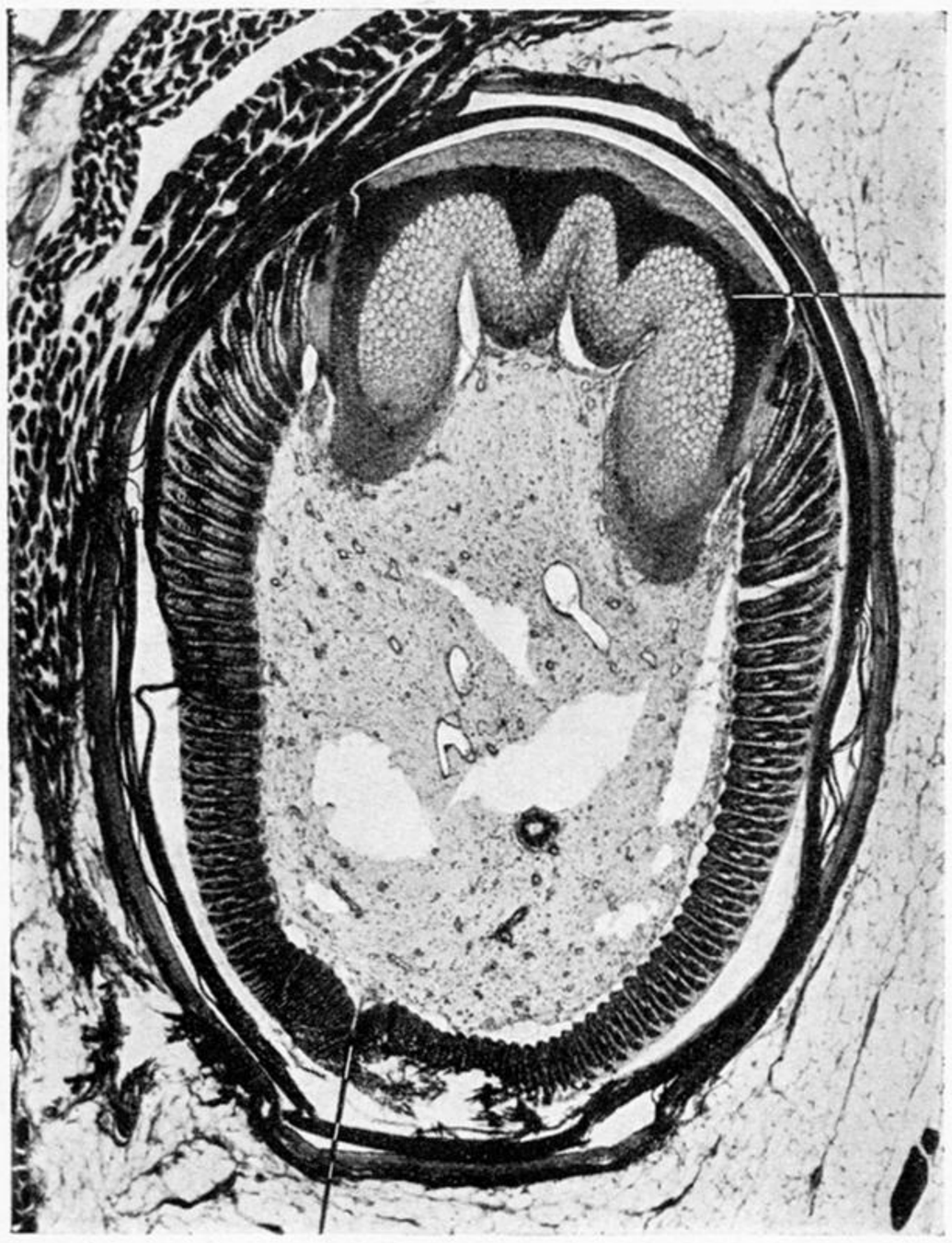

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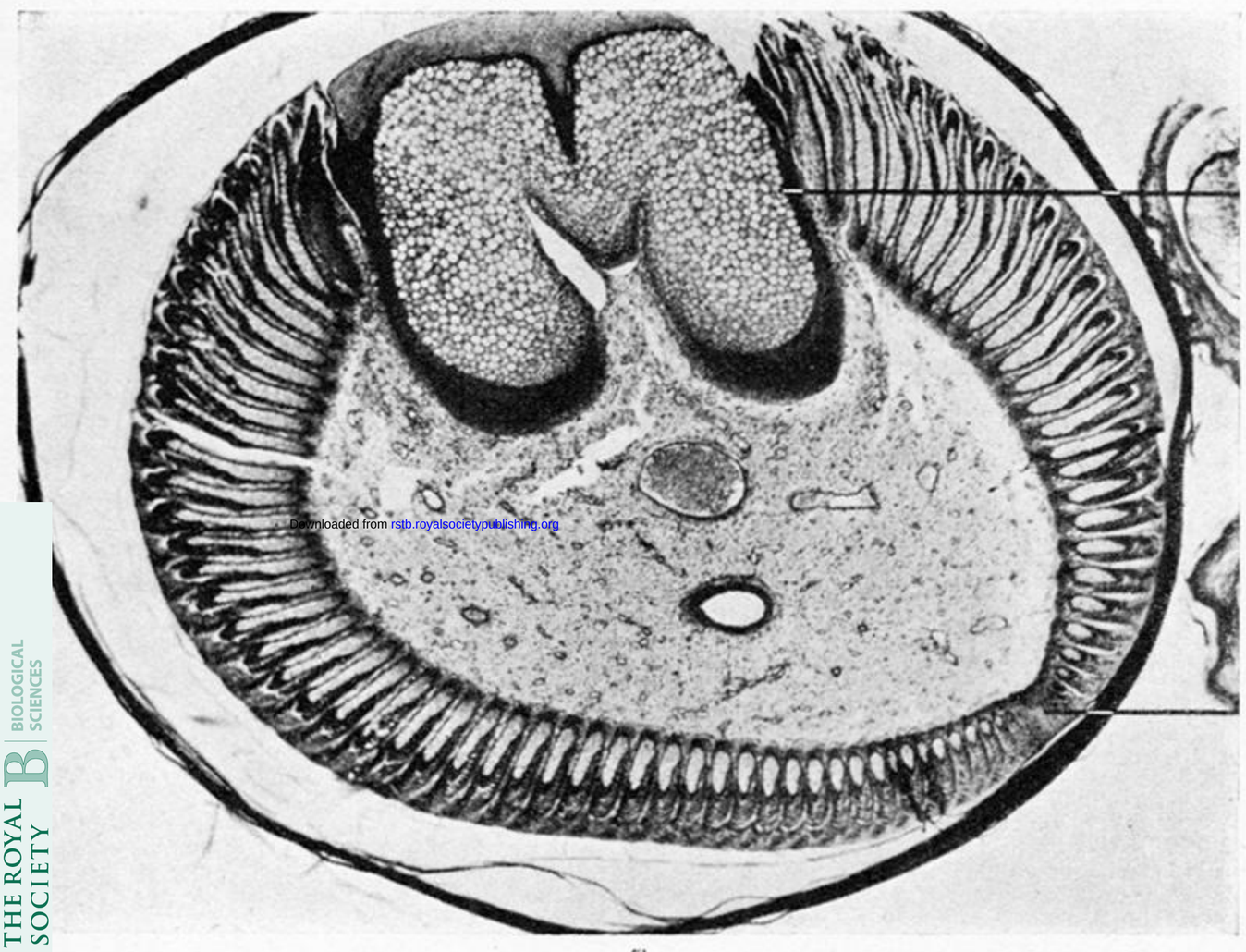

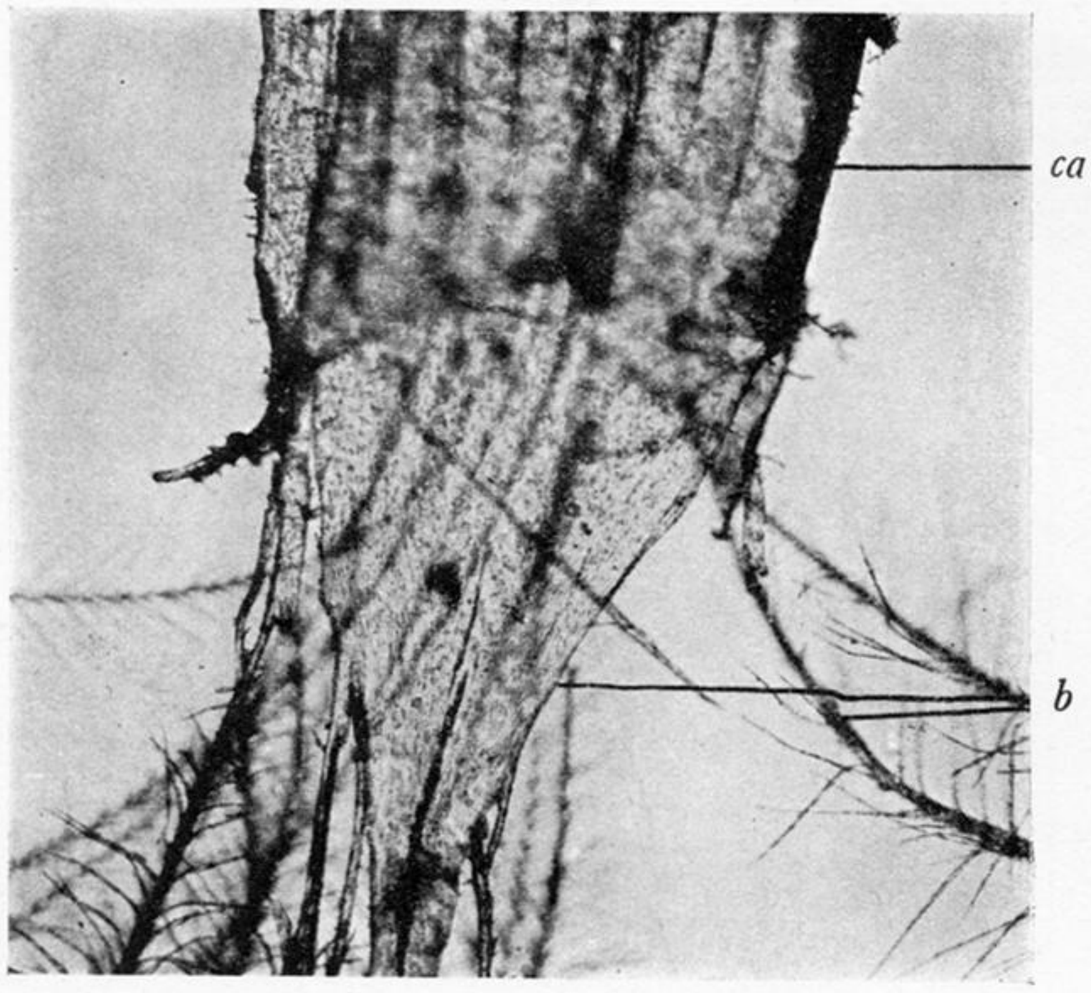

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55

Plate 28

Fig. 52—Microphotograph of transverse section near the base of a thigh feather of an 8 weeks old Rhode Island Red fowl, showing the deflexion of the region of plasmatic growth nearer to the left side of the rhachis.

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