AN ANALYSIS OF THE SHAPE AND STRUCTURE OF GOLGI BODIES IN THE EGGS OF INVERTEBRATES WITH A NOTE ON THE PROBABLE MODES OF ORIGIN OF THE GOLGI NETWORK.

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Introduction.

Our purpose in writing this paper is not merely to record our observations on the shape and structure of the Golgi apparatus in the eggs of Invertebrates but also to suggest some probable modes of formation of a network from these various shapes. We make this attempt because we consider that the cyto-

logists working on the phenomena of oogenesis have ignored many of the suggestions of students in other branches of the study regarding the shape of the Golgi apparatus, notably those of Bowen ('20, '22 a, '26 d & '26 e).

This lack of a clear understanding of the shape and structure of the Golgi apparatus in oogenesis is reflected even in the reviews on the subject (MacBride and Hewer, '31) and hence in this paper we have made an attempt to put the available facts from our work into a connected whole, taking into consideration the already established facts in spermatogenesis and secretory phenomena.

Literature.

The historical review of the whole subject up to 1926 treating with the works of Holmgren ('03, '08 & '10), Golgi ('98), Cajal ('14), Duesberg ('12 & '14), Bensley ('10), Voinov ('25), Morelle ('24 & '25) and others has already been made by Bowen ('26 e) and hence we intend to refer to these papers only when they have some bearing on the problems with which we are concerned in this paper.

During the course of our investigations on the phenomena of yolk formation in eggs we came across Golgi bodies having different shapes. In Clibanarius (Subramaniam, '35 a) and Stomopneustes (Subramaniam and Gopala Aiyar, '36 b) we observed during the growth of the egg, the gradual transformation of a Golgi granule into a batonette. The granules which had no
differentiation into chromophilic and chromophobic regions enlarged into vesicles showing the two regions clearly. These by rupture gave rise to batonettes which reminded one of similar bodies described in invertebrate animals by Hirschler ('13, '14, '16 & '18), Gatenby ('17 a, b, c, '19 a, b, c & '22), Bowen ('20, '22 a), Beams and Goldsmith ('30) and Harvey ('31 a & b). Moreover Gatenby ('17 a) and Hirschler ('18) described rings and rods lying side by side. Even though Hirschler described ring and half-ring shaped Golgi elements in the oogenesis of Ciona intestinalis ('16) and Bowen ('26 e) from his review concluded that this cellular constituent may assume almost any shape and distribution, a reticulum being only one expression of its protean appearance, the polymorphic condition of the Golgi apparatus in eggs has been much disputed even as late as 1931. Nath in 1925 described in the earliest oocytes of Scorpion "a few clearly-defined curved rods lying on one side of the nucleus". But in the case of Pheretima (Nath, '30), Periplaneta (Nath and Mohan, '29), Otostigma (Nath and Hussain, '29), Culex (Nath, '29) and Dysdercus (Bhandari and Nath, '30) he describes vesicles and especially in Pheretima he attempts to interpret the various other shapes that he observes as caused by improper impregnation, optical sections, etc. In his latest paper (Nath, '33) he mentions that the Golgi apparatus is polymorphic but we are not able to decide whether that applies during oogenesis also. Harvey on the other hand describes "Argentophil vesicles and irregular masses" in Ciona (Harvey, '27) but in his latest paper ('31 c) he remarks while criticizing Nath's work that "there can be no connecting link between these vesicles and the netlike Golgi apparatus characteristic of vertebrate cells, which is the fundamental absolute to which all questions of the form of Golgi apparatus must ultimately be referred" (p. 451). He continues "there is one form of Golgi apparatus only in invertebrate cells, that of an ensemble of dictyosomes" (p. 452). How different is this conception from that of Bowen ('26 d & e) who considers the Golgi apparatus as a specialized cytoplasmic substance which may be moulded into any shape demanded by particular cellular conditions!! That hollow spheres have an existence may be seen from Hirschler's work ('18) whose main consideration in postulating his "Apparatinhalt" was that in the case of hollow spheres the central contents could not possibly be the same as the external cytoplasm. Students of oogenesis seem to consider oogenesis as a special phenomenon and do not refer to papers on spermatogenesis and secretory phenomena. As pointed out in another paper (Subramaniam, '35 b) we believe rather that the aim of all work on the structure or function of the Golgi apparatus should be to correlate, if possible, results obtained in one field with those recorded in others. The Golgi apparatus of secretory cells is in no way different from that found
in the various other tissues of the body, because it is the Golgi bodies in the eggs which give rise by division and distribution to the Golgi apparatus in all the cells of the body. Hence we are faced with the question, how the Golgi bodies having the same origin can be attributed different shapes and function in different cells. Either the diversity of behaviour, shape and structure should be accepted or a de novo origin of the Golgi apparatus in different types of cells should be proved. The latter possibility being negligible, the only recourse will be to accept the former one. The more we learn about the Golgi apparatus even in eggs, the more we are convinced that the variations are only the variations of a single basic procedure. Harvey's remark that the Golgi apparatus can have only the dictyosome shape disregards the work of Hirschler and Gatenby. Gatenby even in 1917 describes batonettes giving rise to vesicles. He says "At the time the mitochondrial bodies begin to run together, the acroblasts which are about three or four in number, gradually become vesicular. The process is quite easily followed out and can often be seen occurring in several acroblasts in one spermatid" (p. 439). A look at his figures reproduced below (Text-Figs. 21, 22, 23 & 24 a, b) will show that both the types, dictyosome and vesicle, occur side by side. Figures illustrating Gatenby's work on Saccocirrus ('22) especially his figures 1, 4, 19, 20, 21 & 32 will amply convince any one that there could be no hard and fast rule regarding the shape of the Golgi apparatus in oocytes, spermatocytes and spermatogonia. Similarly in secretory cells also, the diversity of shape could be gathered from the work of Ludford ('25 a, b, '27 & '31; Ludford and Cramer, '27), and Bowen ('26 a, b, c). Harvey fails to consider that even in the case of the boomerang shaped scales sections in different planes may often lead to the production of rings in some particular planes (compare the shapes in Text-Fig. 3 g.b₂, g.r₂ which is after Bowen, '20). Moreover during secretory activity it has been noticed that the apparatus during hypertrophy assumes a variety of shapes. The network at an advanced stage of secretion has been known to break up into rings, rods and granules (Ludford and Cramer, '27). Below is reproduced the pictographic summary of the secretory cycle in the Islets of Langerhans after Ludford and Cramer (Text-Fig. 1).

As this paper also deals with the structure of the Golgi apparatus we give below a review of the work dealing with the structure as conceived by different cytologists. Even at the outset it may be mentioned that except Hirschler ('18) and Bowen ('26 d & e) few seem to have considered the difference in structure between the network-like Golgi apparatus and the dictyosome. The dictyosomes according to Gatenby and Bowen have a duplex structure consisting of chromophilic and chromophobic parts. But in the network-
like Golgi apparatus of vertebrate somatic cells the chromophobic part seems to be lacking. As even in mammalian germ cells (Gatenby and Woodger, '21; Gatenby and Beams, '35) an idiosome has been demonstrated one naturally inquires how the chromophobic part comes to be absent in the network-like Golgi apparatus. Unless a clearer understanding of the structure of the Golgi apparatus is available a minute analysis of the part played by the apparatus in secretion is not possible. We have always been faced with the question whether the secretion of the acrosome is comparable to the secretion of mucus, serous and lipoid granules in secretory cells. It will be seen from papers on spermatogenesis (Bowen, '26 d) that the acrosome always arises near and in relation to the chromophobic part. No case has been recorded in which the chromophilic part is next to the acrosomal vesicle or granule and the chromophobic part farther away and having no relation with the vesicle or granule. In spite of the differences of opinion among students of oogenesis all are agreed (Nath, '29, '30 & '33 ; Harvey, '27, '31 a, b & c ; Subramaniam, '34, '35a, '37 ; Subramaniam and Gopala Aiyar, '35, '36a & '36b) that fat, fatty
yolk and yolk when they are formed as products of the activity of the Golgi apparatus always arise in relation with the chromophobic part of the dictyosomes or vesicles. Such being the case, the fact has to be admitted that the chromophobic part plays as essential a role as the chromophilic part in cells where the apparatus has a duplex structure.

If this is so, how is the secretion of the acrosome comparable to the secretion of mucus, serous and lipoid granules in secretory cells? Bowen in his papers definitely mentions that in the network he could never find the chromophobic component. Hence in order to compare secretory granule formation the first problem to be solved is the process how the chromophobic part disappears. We have come across a phenomenon (Subramaniam and Gopala Aiyar, '36 c & d) by which a vesicle by infolding gives rise to a dictyosome with double rims in which the two parts by closer apposition give rise to Golgi batonettes in which no chromophobic part is visible.

Harvey's ('31b & c) main objection as pointed out before is the method by which the vesicle gives rise to a network. His statement ('31c) that "The netlike Golgi apparatus of most vertebrate cells is foreshadowed in the vortex-like complex forms assumed in young eggs, of which Antedon provides an excellent example and in the somatic cells of many adult forms (vide Hirschler, 1914, 1918)" (p. 452), clearly demonstrates that he has not considered the problem of the structure of the Golgi apparatus at all. Hirschler ('18) though he had considered the duplex structure (his Apparathulle and Apparatinhalt) has not interpreted the loss of the chromophobic part in the networks. In addition to all these Nassonov ('24), Ludford ('25) and Bowen ('26a, b, c) record double-rimmed batonettes, hollow tubular networks and vacuolated networks. Are all these artifacts or are they only the variations of a single basic structure? If they are real they may, in a way, give a clue to the solution of the duplex structure of the Golgi bodies.

Even networks found in secretory cells are different from those found in cells of tissue cultures and tumour cells. In tissue culture cells Ludford ('27) records both a network and a collection of granules. Ludford, while discussing the structure of the Golgi apparatus, says that though he finds a chromophobic component in relation with the network, yet as the chromophil threads become drawn out, he has been unable to make out any differentiation into the two above-mentioned regions. Similar experience is also recorded by him in connection with the changes in the form of the apparatus during hypertrophy in the cells of the fibroblasts of sar tumours. His own interpretation ('27) is that "It is possible in both these cases that the idiosomic substance is either spread in a thin film over a chromophil core, or it may be
that the two are combined in such a way that the two substances are not
distinguishable optically'" (p. 418). His conclusions regarding the dis-
appearance of the chromophobie portion as presented in his paper on the
tar tumours are slightly different. He is of opinion that the chromophobie
c part may persist 'as a fine layer surrounding the reticulum of the apparatus
or else it may form the basis of the reticulum over which the material of the
apparatus is spread in a similar manner to the insulating material covering
an electric wire' (p. 589, 1925b). Whatever may be the process there should
be intermediate stages between the one with the duplex structure and that
with the chromophobie part either inside or outside. Ludford and for the
matter of that no worker offers any evidence which may suggest the possibility
of there being intermediate stages between the discrete bodies and the network.

Now we shall mention those papers in which double-rimmed batonettes-
tubular networks, and vesiculated networks are mentioned. In the spermato-
gonia of Euschistus euschistoides (Bowen, '20) the Golgi bodies presented a
characteristic appearance (Text-Figs. 2, 3 & 4). Viewed on edge the individual

![Text-Fig. 2] Schematic representation of a single Golgi body. (Text-Fig. 1 A, p. 329, Bowen, 1920.)

![Text-Fig. 3] Euschistus euschistoides. Spermatogonium late growth period. Note the
double rim in all the bodies and also ring and rod shapes lying side by side.
(Pl. 1, Fig. 7, Bowen, 1920.)

![Text-Fig. 4] Brochymena quadripustulata spermatids showing Golgi remnant. (Pl. II,
Fig. 31, Bowen, 1920.)

Golgi bodies gave the impression of a pair of rods lying side by side (Text-
Fig. 2 chr.), and separated by a clear space (chb1). This slit was observed by
Bowen from the very early stages upto the condensation and even the Golgi
remnant (Text-Fig. 4 g.r.r.) in his figures shows this identical structure. It
is possible that such a feature may be of wide occurrence for it is more frequently
figured but rarely referred to in several published accounts of the Golgi
apparatus. Bowen is certain that the division of the Golgi bodies is always at right angles to the slit and the latter therefore however well adapted it may be for purposes of division never serves that end. Even after the fusion of the Golgi bodies to form a single acroblast, the structure has a double rim as could be made out from Bowen's figure 31 (Text-Fig. 4 g.r.r.).

Nassonov ('24) and Ludford ('25a) while working on the secretory phenomena in the epididymis came across Golgi networks composed of hollow tubes. Figures representing the hollow nature as drawn by them are given below. Text-Figs. 5 and 6 are from Nassonov's paper and Text-Figs. 7, 8, 9 and 10 are from Ludford's. The liquid form of secretion (s.) is mentioned by both as originating inside these hollow tubes (t.g.a.). Bowen re-examined the phenomena in the epididymis of the cat and the rabbit and observes that he also found examples similar to Nassonov's figures in great numbers in the various parts of the male reproductive tract of both the cat and the rabbit. In addition, he found another closely related appearance in which the blackened Golgi contours were bordered externally by a light zone similar to the one which, in the other cases, appeared on the inner border of a blackened contour. Some of his figures illustrating these appearances are shown in Text-Figs. 11, 12, 13, 14, 15, 16, 17 & 18. He has observed these identical appearances even in longitudinal sections (Text-Figs. 15 & 16). As no relation existed
between secretory granules and these spaces Bowen feels that these appearances may be artifacts caused partially by the fixative due to the massive development of the Golgi apparatus in these cells. Even after suggesting that such appearances result from shrinkage, he considers it possible that such a

Text-Figs. 11 & 12.—Golgi strands having a tubular appearance. Vas deferens of the Rabbit. (Pl. 28, Figs. 87 and 86, Bowen, 1926c.)

Text-Figs. 13 & 14.—Golgi contours bordered by a light zone. Both are cross-sections. (Pl. 26, Figs. 33 and 48, Bowen, 1926c.)

Text-Figs. 15 & 16.—Golgi apparatus having a tubular appearance. Longitudinal section of a cell from the proximal portion of the epididymis of the Cat. (Pl. 26, Figs. 39 and 40, Bowen, 1926c.)

Text-Figs. 17 & 18.—Vesicular Golgi networks in some cells in the vas deferens of the Rabbit. (Pl. 28, Figs. 83 and 85, Bowen, 1926c.)
tendency to react to osmic acid will eventually throw some light on the intimate structural details of the Golgi apparatus.

It is a wonder that after having described double-rimmed batonettes in *Euschistus* he considers the double outlines of the Golgi bodies in the salivary gland cells of *Limax* as pure artifacts. In cross-sections of secretory cells of the vas deferens of the rabbit, Bowen records some interesting details. The network in such sections lacked uniformity of diameter and consisted of blackened materials connected together by delicate cords. When the impregnation was light these networks presented a vacuolated appearance. In Text-Figs. 17 and 18, we have reproduced some of the appearances seen by Bowen. Such appearances, he remarks, give one a distinct impression that the swollen masses of the Golgi network are formed by a cluster of small vacuoles. He continues "Such vesicular developments occur in other gland cells, but I know of none where these appearances are so prominent and of such frequent occurrence. Much interest attaches to the interpretation which is to be given to these vesicles. Are they artifacts caused by some swelling action of the fixatives or are they actually the beginnings of secretory granules? The latter conception would immediately explain the appearances and demonstrate in a remarkable way the relation between the Golgi material and the secretory products which, it is suggested, are differentiated under the action of the Golgi apparatus. Definite evidence of the reality of these vesiculated Golgi masses would obviously provide the final demonstration of the views which I have elsewhere suggested (Bowen, '24). Unfortunately in the present instance there are reasons for admitting the possibility that the appearance is an artifact, and while it is hard to believe that such an explanation can be complete, nevertheless in a critical case of this kind it seems best for the present to be very cautious" (pp. 414–15, '26 c). That these vesiculated masses have a real existence may be gathered from Ludford's paper. Even though he does not refer to such vesiculated masses in the text, yet figures such masses in his figs. 17 and 19 (Text-Fig. 9 *v.g.a*.). Bowen does not seem to have considered the possibility that the chromophobic part may become differentiated inside some of the strands of the network. We say chromophobic part, because it is the part which has been shown to be almost in intimate relation with the acrosomal vesicle as also with forming fat and yolk droplets in eggs. Ludford's later work on the Islets of Langerhans ('27) as also in vital staining of acinar cells of the pancreas ('31) clearly demonstrates the possibility that in some cases at least the secretory or segregation products may arise inside the strands. Figures in his pictographic summary of the behaviour of the Golgi apparatus during secretion in the Islets of Langerhans cells make our suggestion clear even though Ludford does not refer to
any such differentiation into chromophilic and chromophobic parts in this particular case. Three workers working on the epididymis and vas deferens and seeing an identical structure constitute a sure proof of the reality of its appearance especially as one of them (Ludford) figures them without making any mention of the occurrence in the text of the paper. Bowen concludes by expressing the belief that the idiosomic substance is a derivative of the Golgi material and that while it may become very clearly individualized as in male germ cells, it may in others be latent or may be so intimately related to the lipoidal constituent as to be lost in the impregnation of the latter. It will be of considerable comparative interest that the differentiation not to be made out in somatic cells is also experienced in a few cases in the Lepidoptera and Grasshoppers. Such a conception according to Bowen while accounting for observed facts also permits the possibility that in secretory cells "the idiosomic or chromophobic material may be more or less differentiated within the Golgi area, but invisible with the technique employed. I am inclined to think, however, that such a development does not exist" (p. 438, '26 d). We have not been able to decide whether Bowen's statement "differentiated within the Golgi area" means differentiated inside the strands or between the strands. The latter seems to be Bowen's opinion for, if differentiation is inside the strands of the network the invisibility will be due to optical difficulties and not due to technique. Whatever may be the interpretation Bowen does not seem to have believed in the possibility of the chromophobic part forming a core to the strands of the network. Nassonov's view that the Golgi material forms a surface of separation between the granule and the surrounding plasma throws some light on the structure of the Golgi apparatus. Even though he does not seem to have paid much attention to the structure of the apparatus his suggestion mentioned above is in accord with his observations. Nassonov's suggestion of the origin of secretory droplets is more comparable to that of acrosome formation than Bowen's own modification of Nassonov's view. Many of the instances mentioned above suggest that the chromophobic part may form a core around which the chromophilic part may lie just like an insulating material. But in the majority of the cases the core may be absent. We have to consider now how the chromophobic part comes to lie inside and also whether there are any recorded instances of such behaviour.

In the numerous papers dealing with the behaviour of the Golgi bodies and mitochondria scanty attention has been paid to structure. Even though pointed references are absent there are in studies on the behaviour of the mitochondria instances where threads originate from vesicles. As mitochondria are also polymorphic occurring in the form of grains, vesicles and threads
M. K. Subramaniam and R. Gopala Aiyar

we believe that references to recorded instances of change of shape from vesicle to thread form may not be out of place. Gatenby's and Bowen's papers on Lepidopteran and Insect spermatogenesis give a clue as to how thread formation may take place. We have to point out here that as in the case of Golgi granules, mitochondrial granules also when they enlarge have been noticed to differentiate into chromophobic and chromophilic regions. Gatenby's figure (Text-Fig. 19, m.g., m.v.) of the process given below will make it clear that even as early as 1917 the process has been described. The above-mentioned paper is especially interesting in that Gatenby figures and describes some peculiar behaviour of the mitochondria. According to him near the end of growth stage the mitochondria have a tendency to run together to form vesicles as shown in our Text-Fig. 20 at V. He seems to believe

![Text-Fig. 19.—Spermatocyte of *Smerinthus populi* showing mitochondrial granules and vesicles. (Pl. 23, Fig. 7, Gatenby, 1917a.)](image1)

![Text-Fig. 20.—Equatorial view of the first maturation division of *Smerinthus populi* showing precocious running together of mitochondria. (Pl. 23, Fig. 11, Gatenby, 1917a.)](image2)

that in such cases the running together may be caused by the close contact and subsequent fusion of the outer rim of the mitochondrial bodies and the flowing together of the chromophobic fluid core of the mitochondria taking part in the formation of such large vesicles. While the macromitosome is formed the following behaviour is described. After the second maturation division while the nucleus is becoming reorganized the mitochondrial bodies show a tendency to collect in a horn-shaped mass near the nucleus. This preliminary massing being over there is a tendency for the chromophilic rims to flow together and give rise to a spireme. The most significant fact to be observed here is that while the outer layer forms the spireme, the inner chromophobic areas which have coalesced together form the substance in which the spireme lies. The whole process could be made out in Text-Figs. 21, 22 and 23. Having traced the behaviour of the mitochondria so far
we shall now go back to the behaviour of the same cell organ during spermatogonial division. During division the mitochondria are affected by what is taking place in the cell. Some of them become elongated in the longitudinal axis of the spindle and others run together with their neighbours and also become elongated in the same direction. Text-Figs. 24 and 25 which are taken from Gatenby's paper illustrate this behaviour very clearly especially in the anaphase and telophase. In Text-Fig. 24 is shown the mitochondria forming funnel-like masses with their narrow ends applied to each other representing the region where the cells constrict. Gatenby suggests that such elongation might be due to mere mechanical reasons, such as the pressing of individual mitochondria one against the other.
Gatenby records some other peculiar processes also. In addition to the formation of large vesicles by fusion of a number of smaller mitochondrial vesicles filament formation also occurs. In places where the mitochondria are densest they run together to form filaments and cords as shown in Text-Figs. 25 and 26 *m.t.* His explanation seems to be "that such peculiarities are due to the effect of the fixative on the matter around which the true mitochondrial substance is applied; thus the filamentous condition, of which Pl. 25, fig. 31 is an example, is probably due to the rupture of the outer layer by the 'brutality' of the fixative and the consequent running together of the rims of the mitochondria. This last process is one which the mitochondrial bodies always have a tendency to undergo. It may, then, be stated that the running together, though a natural process in later stages, is artificially hastened by the action of the fixative" (p. 424, '17 a). We shall now consider whether the elongation and running together is a precocious exhibition of a similar power possessed by them in later stages of spermatogenesis or whether they are artifacts. Gatenby records coalescence and formation of large vacuoles as a natural process. He further records that these mitochondrial bodies finally fuse together to give rise to the macromitosomal spireme. If these two are natural processes, Gatenby's evidence for the running together of the mitochondria during division stages as artifacts is not sufficient. As Gatenby's later work has demonstrated variation even among the spermagonia of the same generation we feel that there are reasons to consider the thread formation and elongation as caused by a precocious exhibition of a latent power. That mutual pressure and other mechanical causes do not explain such lengthening of the vesicles may be gathered from his fig. 48 (Text-Fig. 25) which is in the prophase of the first maturation division. Brutality of fixation does not also explain why the mitochondria in the centre should have run together to form rods while the mitochondria in the other regions instead of being distorted in various ways retain their vesicular shape. Figs. 49 and 31 in his paper are not comparable since in Fig. 49 all the mitochondria have been distorted. What happens to the chromophobic parts of the vesicles when they become filamentous is not clear from his explanation. Do these filaments have the chromophobic part outside or does the chromophobic part come to lie inside the filaments and thus become obliterated during the process of elongation?

A second method of thread formation has been observed by Meves in the honeybee ('07) and Gatenby in *Tenebrio* ('19). In the latter Gatenby believes that "the mitochondria either exist as spheres containing a chromophobe zone inside or else as elongated tubes containing internally a chromophobe substance. These tubes are formed by fusion of granules and when
viewed under monocular vision look like apposed solid rods which are really hollow tubes (I use hollow in the sense that their centre is chromophobe, though it contains a fluid)” (p. 251, '19 a).

**Text-Fig. 25.**—Prophase of first maturation division in *S. populi* showing elongated mitochondria. (Pl. 25, Fig. 48, Gatenby, 1917a.)

**Text-Fig. 26.**—Spermatocyte in which the mitochondrial vesicles have given rise to thread-like mitochondria. (Pl. 25, Fig. 31, Gatenby, 1917a.)

**Text-Fig. 27.**—An early growing oocyte of *Meretrix* showing Golgi masses, Golgi granules and Golgi vesicles. Mann Kopsch × 2,000 (Original).
Observations.

During the later stages of the study of oogenesis in various animals, we paid particular attention to the structure of the Golgi bodies. In addition we have examined many of our old preparations. In Clibanarius (Subramaniam, '35 a) and in Meretrix (Subramaniam, '37) the earliest oocytes contained a Golgi mass in which no differentiation into chromophilic and chromophobic regions could be observed (Text-Fig. 27 g.m.). In Salmacis (Subramaniam, '34), Meretrix and Clibanarius the initial mass breaks up into a number of granules. This granular condition in which the individual bodies have no duplex structure is retained in Salmacis throughout oogenesis. In Meretrix, Stomopneustes (Subramaniam and Gopala Aiyar, '36 b) and Clibanarius the granular components of the Golgi apparatus give rise to a number of vesicles. The processes leading up to the formation of vesicles is interesting in Meretrix. Innumerable variations in behaviour remind one of the variations observed in the cells of Islets of Langerhans (Ludford and Cramer, '27). The initial Golgi mass divides into two one of which generally migrates to the other side of the nucleus. One of the two masses so formed may divide again. These masses may immediately become resolved first into a number of granules and later into a number of vesicles or one of them before the others may give rise to vesicles with a chromophobic interior (Text-Fig. 27 g.g., g.v.). The initial mass may break up into a number of vesicles before division. Such a collection of vesicles may divide into two and both may begin to scatter their individual elements at the same time or one group may remain inactive for a longer time and may even divide into two. In a few cases, the vesicles never divided but began to scatter from the place of their occurrence.

In Stomopneustes (Subramaniam and Gopala Aiyar, '36 b), Clibanarius and Dasychone (Subramaniam and Gopala Aiyar, '36 a) rod-like dictyosomes occur. But the three differ in that in Dasychone the scale-like dictyosomes alone are present throughout oogenesis whereas in both Stomopneustes and Clibanarius dictyosomes arise by rupture of the vesicles. Plate VII, Fig. 1, is from a Nasonov preparation of Clibanarius ovary. At g.g. are the Golgi grains without any differentiation into chromophilic and chromophobic regions. At g.v.a. and g.v.b. are vesicles formed by the enlargement of the grains in which the two regions could be seen. At g.v.c. is shown the rupture of the vesicle and the resultant batonette could be seen at g.b. A look at the Plate will show how the chromophobic area enclosed by the chromophilic region in the vesicle comes into relation with the cytoplasm.

In Stomopneustes during the secretion of albuminous yolk the Golgi bodies present a variety of behaviour. Their behaviour may roughly be classified
into two different types based on the structure of the bodies when found attached to the mitochondrial clumps. (1) After slight enlargement of the Golgi vesicles rupture on one side takes place resulting in the formation of semi-lunar shaped elements as in the case of Clibanarius. (2) In some cases the Golgi vesicle attached to a mitochondrial clump becomes irregularly plastered on the mitochondrial clump without rupture. Whereas in the first case the chromophobic part comes into relation with the mitochondrial clump, in the second instance the chromophobic part is not visible at all. The different shapes assumed by the Golgi after the different modes of attachment is diagrammatically represented in Text-Fig. 28.

![Text-Fig. 28](image)

Text-Fig. 28.—1. Various shapes assumed by the Golgi when found attached to mitochondrial clumps. 2. A diagrammatic representation of the formation of a Golgi batonette by rupture of a vesicle and its attachment to a mitochondrial clump. The chromophobc part becomes reduced to invisibility during the process. (Figs. 2 and 3, p. 579, Subramaniam and Gopala Aiyar, 1936b.)

One of us very recently (Aiyar, R. G., '35) showed that Lycestis indica, a common polychaete worm, occurring in the brackish waters in and around Madras, is a hermaphrodite. Its eggs are highly plastic and we thought that a study of the phenomena of oogenesis in Lycestis would be of interest (Subramaniam and Gopala Aiyar, '36 d). We are recording below some peculiar behaviour and structure of Golgi bodies which have not been recorded previously in eggs. Incidentally, we may mention here that the Golgi bodies could be seen even in Champy fixed material. The Golgi bodies in the growing oocytes could be seen as vesicles. These vesicles during the growth of the egg become converted into double-rimmed batonettes. In Plate VII, Fig. 2,
the stages leading up from the vesicle to the double-rimmed batonette could be seen. At g.v. is shown a typical vesicle showing chromophilic and chromophobic regions. This vesicle by a process of tucking in gives rise to a gastrula-like structure which later gives rise to a double-rimmed batonette (Plate VII, Fig. 2, Plate VIII, Figs. 3 & 4). The whole series could be seen in Plate VII, Fig. 2. At g.v.1, is shown the beginning of the process of folding. This becomes deeper and deeper (g.v.2 & g.v.3) and the resulting double-rimmed batonette could be seen at g.b. Thus the batonette comes to have the chromophic part between the two chromophic rims. In Plate IX, Fig. 5 at g.r. and in Text-Fig. 29 A at 4 could be made out sections of the same bodies at right angles to the plane in which they lie in Plate VIII, Figs. 3 and 4. In Plate IX, Fig. 5, the mitochondria are visible as scattered granules in the cytoplasm. Finally in the fully grown oocytes, the majority of the batonettes do not seem to have any chromophic component at all. Such absence of the chromophic part has been observed by Nath in Scorpions (’25) and Bowen and others in a few Lepidoptera and Grasshoppers. Plate VIII, Fig. 4, shows how the chromophic part is completely absorbed. At g.b.2x, is shown a double-rimmed batonette in which the two rims have united at one of the ends. This process goes on till no chromophic part is discernible. A pictographic summary of the process is given below (Text-Fig. 29 A).

**Text-Fig. 29.** A diagrammatic representation of the formation of a double-rimmed batonette as well as a batonette in which the chromophic part is absent from a Golgi vesicle. At 4 is shown a cross-section of stage 3. (Slightly modified after Fig. 13, Subramaniam and Gopala Aiyar, 1936d.)

B. The different methods of multiple folding. At 3 is shown a cross-section of a slightly later stage of infolding than those shown in 1 and 4. At 7 is shown one of the shapes that result after complex multiple folding. (Original.)

Multiple folding of the walls of the vesicles has also been observed (Plate VII, Fig. 2 and Plate VIII, Fig. 3). In Plate VII, Fig. 2, is shown (m.f.1) an instance of double folding. In Text-Fig. 29 is shown some of the
methods of multiple folding observed by us. Similar bodies could be seen in the Plates. Perusal of Plate VII, Fig. 2, Plate VIII, Fig. 3 and Plate X, Fig. 6 and Text-Fig. 29 B, 7, reminds one of the vacuolated networks figured by Bowen, Nassonov and Ludford. It will be seen that in the figure showing the extremely vacuolated condition (Plate VIII, Fig. 3, \( m_f_2, m_f_3 \)) the chromophobic part is reduced to invisibility by the fusion of the walls of the folds lying near.

**Discussion.**

Even though the Golgi apparatus was discovered some 40 years ago, little is known about its structure. During recent years Parat's papers ('26, '28; Parat and Painleve, '24 a, b, c & '25) diverted the attention of cytologists to a study of the vacuome. Enough work has been done (Gatenby, '29, '32 & '33) to show that the Golgi apparatus is a permanent cell inclusion quite different from the vacuome. Further, we have reasons to believe (Subramaniam, '37) that neutral red vacuoles in eggs which we have examined represent enzymes secreted by the Golgi apparatus.

Passing now to a consideration of the shape of the Golgi bodies we feel that the photographs will convince anyone that the apparatus in eggs may have any shape ranging from that of a granule to a dictyosome. In this we are only bringing the diversity of shape of the Golgi bodies observed by us in eggs into line with facts recorded by a host of workers in cells from other tissues. In many eggs as in *Salmacis* and *Dasychone* a particular shape persists throughout the major part of the growth period of the oocyte. *Dasychone* may only be one of the many animals in which the shape of the Golgi bodies is fixed. But that fact alone does not entitle one to define the shape of the Golgi apparatus in all types of eggs. Gatenby's statement in the case of the oocytes of *Helix* will, we believe, prove that even in 1917 there has been observed variations even in cells lying in a single section. "I have already mentioned that great variation was found in the appearance of the male cells. This, I think, applies even more strongly to the case of female cells......." (p. 588, '17 c). That this variation is not restricted to the mitochondria alone may be gathered from his observation "I can, at present, think of at least several sorts of spermatogonia; by this I mean that it is quite possible to find a large number of cells which are in the spermatogonial generation of male cells and which differ markedly either in their nucleus, their nebenkern or their mitochondria" (p. 571, '17 c).

The process of formation of a typical batonette from a granule is analogous to the behaviour of the mitochondria during the formation of a macromitosomal spireme. The only difference between the rupture of the mitochondria and the rupture of the Golgi vesicle seems to be that in the case of the mitochondria
the process is only an intermediate stage, where soon after rupture all the chromophilic portions fuse together end to end. In the case of the Golgi bodies of *Clibanarius* the process seems to stop with rupture. Later, each batonette comes into relation with mitochondrial or Golgi clumps and secretes albuminous yolk.

Golgi bodies with duplex structure and Golgi networks with a conspicuous absence of a duplex structure are comparable to vesicular mitochondria with chromophilic and chromophobic regions and filamentous mitochondria in which the chromophobic part is absent. Yet, a clearer understanding of the origin of the filamentous mitochondria as also of the Golgi network is not yet available. That even in the case of the filamentous mitochondria which have no duplex structure a necessity for some conception that they should have originated from structures with duplex structure may be gathered from Bowen's work ('22 b) where the filamentous mitochondria when they form the nebennkern have the power of differentiating a chromophobic portion. In his example Bowen ('22b) finds thread-like mitochondria in which he has not been able to find a differentiation into the two components. Yet, after the mitochondrial threads fuse together the chromophobic part puts in its appearance as a number of vacuoles in the periphery of the mitochondrial mass. In *Murgantia histrionica* Bowen finds in the early spermatocyte period mitochondrial granules possessing a duplex structure. He deduces from the above observation that as the vesicles produce the long thread-like mitochondria typical of *Euschistus*, both the parts should be present and that the threads should have an inner core of chromophobic material as in the honey-bee (Meves, '07) and *Tenebri* (Gatenby, '19). Thus even in observed cases of mitochondrial behaviour there are two types of thread formation. No previous worker seems to have given any clear account of the process which leads to the formation of a thread from a granule. In another part of the paper while we were discussing the behaviour of the mitochondria in Lepidoptera, we pointed out that some of the shapes of the mitochondria in Lepidoptera may not after all be due to the action of the fixatives but may be a precocious exhibition of a latent power. We suggest that the elongated tubes shown in Text-Figs. 25 and 26 may after all be stages leading up to the formation of the thread-like mitochondria where the chromophoric inner core is not visible.

Similar to observations in mitochondria there are also recorded instances of double-rimmed batonettes and hollow tubular networks. But Bowen is not prepared to extend his suggestion of the supposed presence of an inner core to the mitochondrial threads, to the strands composing the Golgi network. Even observations regarding the presence and position of the chromophoric
areas are highly contradictory. Bowen in his figures of the double-rimmed batonettes in *Euschistus euschistoides* shows two chromophobic areas: (1) between the two rims (Text-Fig. 2, *chb*₁) and (2) in the cup-like area between the two arms of the U of the batonette (*chb*₂). The lamellar view of the structure of the Golgi apparatus suggested by Hirschler (‘18) and applied by Bowen to the elucidation of the structure of the nebenkern and the Golgi network may explain the figures of Bowen. Such a conception is not considered at all by Bowen in *Euschistus* but that the Golgi bodies have three dimensions may be made out from the different shapes of the batonettes in his figures. A careful examination of Bowen’s figures especially his figs. 8, 9 and 10 also demonstrate that the real chromophoric part may only exist between the two chromophil rims and that the apparent chromophobic part between the arms of the U may be the lamellar structure in surface view.

Ludford who studied the net-like Golgi apparatus records the presence of a chromophoric component in relation with the network in tissue culture cells and tar tumours. Ludford’s views have been presented elsewhere but the processes in tissue cultures and tar tumours seem to be entirely different. In the fibroblasts of tar tumours the Golgi apparatus which exists in the form of rods having a duplex structure when they anastomose and give rise to the complicated network lose their chromophobic component (p. 569, ‘25). In the tissue culture cells, on the contrary, the network which has in relation with it an idiosome loses the above part when it draws out into threads. The processes are directly contradictory and both have been observed in tissues which show wide departures from the normal. Bowen in his wide search for the chromophoric part could not come across any network in which he could find an unmistakable idiosomic component. We have discussed in the resume of literature some of Bowen’s observations in relation to similar work on similar tissues. There seems to be a distinct possibility that some networks at least differentiate an area during some part of secretory activity which is comparable to the chromophoric component of the discrete dictyosome.

The above considerations lead to the question of the nature and presence of the chromophoric part. Bowen (‘26e) suggested two possibilities: (1) That the idiosome may arise as a differentiation product of the Golgi apparatus and (2) that the Golgi apparatus is constantly differentiated into two components which are only morphologically distinguishable with the present technique in the germ cells. Though Bowen found it difficult to decide between these two possibilities our results make it difficult to accept both the suggestions. The behaviour of the Golgi grains in the formation of a batonette in *Clibanarius* and the behaviour of the vesicle in *Lycastis* during the formation of a batonette having no duplex structure suggest that the
idiosome is masked in the grain and the network and hence could not be resolved under the powers of the microscope. But when a granule enlarges into a vesicle both the parts become apparent. There also seems to be a relation between the chromophilic and chromophobic regions in the various shapes of the apparatus. Thus the chromophobic part which appears when the grain enlarges into a vesicle may later be reduced to such an extent as to be absolutely invisible as in the case of the dictyosome in Lycastis. It will be seen that the above concept is directly opposed to that of Morelle ("22, '24, '25 & '26) who suggested that the space enclosed by the Golgi apparatus as a whole is the Golgi substance, the peripheral layer alone of which is blackened by osmic acid to give the characteristic appearances. And we also find no reason to concur with Beams and Goldsmith ('30) that the chromophobic part is a result of the immediate expression of the metabolic activity of the Golgi body, nor with Voinov's view ('25) that the whole complex is to be viewed as a single thing—the idiosome being without any significance. The above suggestion also differs from Bowen's conception of the structure of the Golgi apparatus. The conclusion arrived at by Bowen is that "the Golgi apparatus is a substance which may be scattered through the cell in the form of discrete bodies, often plate-like in form, or the substance may be concentrated leading to a network with strands sometimes in all probability like mere threads, but at other times certainly spreading out to form extensive lamellar surfaces. In other words, the important thing is that the Golgi apparatus is a substance, the exact modelling of which in the cell is purely a matter of secondary interest". Though agreeing with the lamellar structure of the batonettes and networks, we would like to add to Bowen's description that the idiosome masked in the granule and present in the vesicle is secondarily reduced in the network. In other words, networks, like granules, are occasionally able to differentiate a chromophobic region or tubular networks with a chromophobic core to the strands may be present in some cells.

Having cleared the ground thus far we shall proceed to a consideration of the probable nature of origin of the network. We believe that the papers of Bowen ('26a, o, e, d & e), Ludford ('25a, '25b, '27 & '31; Ludford and Cramer, '27), Nassonov ('23 & '24), Brambell ('25) and others sufficiently demonstrate that during the final stages of secretion, the typical network becomes broken up into discrete bodies and that a new apparatus is re-organized from these elements (see also Text-Fig. 1). Hence the possibility of a similar origin by union of the discrete dictyosomes becomes at once apparent.

Naturally considering the structure of the apparatus, the typical network of somatic cells of vertebrates can only arise from batonettes of the type seen in Lycastis. Hirschler's postulate fails to account for the disappearance of
the chromophobic part and if we conceive of the formation of a network from batonettes in which the idiosomic substance is masked such a difficulty does not arise at all. Finally variations in the structure of the network as observed by various cytologists may also be correlated with the variations in the structure of the Golgi bodies as shown in the Plates. As shown diagrammatically below three interesting methods of formation may be probable. In the first type the double-rimmed batonettes by end to end union may give rise to hollow tubular networks as observed by Nassonov and Ludford. In the second, the batonette with no visible idiosomic component may, by a similar process of multiple fusion, give rise to typical networks as figured by Bowen ('26 a, b, & c). Finally there is also the distinct possibility that the vesiculated Golgi bodies found in Lycastis where the chromophobic part has been completely reduced as to be absent by multiple folding may singly or by union with one or more similar bodies enlarge and give rise to networks by a mere process of expansion of the connecting chromophil threads. Our conclusions are given below in Text-Fig. 30 in a pictographic manner.

Text-Fig. 30.—A pictographic summary showing the three possible modes of evolution of the Golgi network. (Fig. 14, p. 69, Subramaniam and Gopala Aiyar, 1936d.)

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KEY TO LETTERING OF TEXT-FIGURES AND PLATES.

A.—Acrosome; a.b.—Acroblast; chb.a.—Chromophobic area between the two chromophilic rims; chb.g.—Chromophobic region in the cup-like area between the two arms of the U.; chr.—Chromophilic rims; g.a.—Golgi apparatus; g.b.—Golgi patonette with the idiosome in contact with the cytoplasm; g.b.g.—Golgi patonette with the idiosome between the two chromophilic rims; g.b.g.—Golgi patonette showing fusion of the two chromophilic rims leading to the formation of a patonette without any visible idiosomic component; g.b.g.—Golgi patonette without any visible idiosomic component; g.g.—Golgi granule without any differentiation into chromophilic and chromophobic regions; g.m.—Golgi mass; g.r.s.—Golgi bodies having the shape of two chromophilic rings one inside the other; g.r.r.—Golgi remnant; g.v.—Golgi vesicles; g.v.a, g.v.b, g.v.c.—Stages showing the formation of a Golgi patonette by rupture of a Golgi vesicle; m.—Mitochondria; m.f.t, m.f.t, m.f.t.—Multiple folding of the wall of the vesicle and also the vesiculated masses that are produced by such a process; m.g.—Mitochondrial granules; g.v., g.v., g.v.s.—Stages in the process of folding of the wall of the Golgi vesicle leading to the formation of double-rimmed patonettes; without a duplex structure; m.m.—Macromitosome; m.t.—Thread-like mitochondria; m.v.—Mitochondrial vesicles; n.—Nucleus; r.g.a.—Remains of the Golgi apparatus; s.—Secretion; t.g.a.—Tubular strands of the Golgi apparatus; v.g.a.—Golgi apparatus presenting a vesiculated appearance; V.—Large vacuoles formed by fusion of a large number of mitochondrial vesicles.

EXPLANATION OF PLATES.

PLATE VII.

Fig. 1.—A portion of Clibavarius egg showing Golgi granules, vesicles and patonettes. 15 x 100. Bellows extension 8°. Nassonov. (Original.)

Fig. 2.—A portion of an egg of Lycastis with the Golgi vesicles showing various stages of folding of their wall and also the resulting double-rimmed patonettes. 15 x 100. Bellows extension 6°. Modified Champy. (Original.)

PLATE VIII.

Fig. 3.—A portion of an egg of Lycastis showing double-rimmed patonettes, patonettes without any chromophilic component and vesiculated masses formed as a result of multiple folding. 15 x 100. Bellows extension 6°. Modified Champy. (Original.)

Fig. 4.—A portion of an egg of Lycastis showing double-rimmed patonettes, patonettes without any chromophilic component and vesiculated masses formed as a result of multiple folding. 15 x 100. Bellows extension 6°. (Original.)

PLATE IX.

Fig. 5.—A portion of an egg of Lycastis showing the fusion of the two rims of a double-rimmed patonette. 15 x 100. Bellows extension 6°. Modified Champy. (Original.)

PLATE X.

Fig. 6.—Showing mitochondrial granules and Golgi bodies having the shape of two chromophilic rims one inside the other. 15 x 100. Bellows extension 6°. (Fig. 11, p. 68, Subramaniam and Gopala Aiyar, 1936d.)

Fig. 7.—A portion of an egg of Lycastis showing various modes of multiple folding of the walls of the vesicle. 15 x 100. Bellows extension 8°. (Original.)

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FIG. 1.

FIG. 2.
FIG. 3.

FIG. 4.
FIG. 5.