# INTERCISTERNAL MATERIAL IN THE GOLGI BODY OF *TRICHOMONAS*

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In the flagellate *Trichomonas*, the Golgi apparatus is present in the form of an organelle which has long been called the parabasal body. This term was applied before it was recognized that the structure is a Golgi system. The parabasal body is a long, cylindrical organelle, about 1 $\mu$ in diameter and, in one species (*T. gigantea*), up to 200 $\mu$ long. It has the typical fine structure of a Golgi body, consisting of a stack of flattened, longitudinally oriented cisternae, together with associated vesicles. It differs from most other Golgi bodies in that a fiber, the parabasal filament (Figs. 4, 6), runs along the whole length of the organelle and terminates on one of the flagellar basal bodies situated at the anterior end of the cell. Such a filament is present in the Golgi bodies of some other flagellates, and in *Trichonympha* it has been shown that this filament lies on the concave, proximal face of the organelle, where new cisternae are believed to be formed (Grimstone, 1959). The opposite, distal face of the parabasal body is thought to be the site of vacuole formation.

This paper is chiefly concerned with the parabasal bodies of some large members of the genus *Trichomonas* which live as symbionts in the gut of termites. In these forms, as in other flagellates, the parabasal body is readily seen in the light microscope after staining by Bodian's silver-proteinate method. Kirby (1944), studying *T. termopsidis*, noted that in such preparations the organelle has a nodose appearance, consisting of alternating light and dark regions (Fig. 1). It is difficult to observe the parabasal body in life in *T. termopsidis*, but it can be seen quite readily in an as yet undescribed species from the termite *Porotermes adamsoni*, and here the nodose structure is again apparent (Fig. 2). The fine-structural basis of this nodose appearance has recently been determined in *T. termopsidis* and *T. gigantea* and appears to merit brief description, since this appearance results from a type of differentiation seemingly unlike that found in other Golgi bodies.

*T. termopsidis* used in this study was obtained from the termite *Zootermopsis angusticollis*, and *T. gigantea* from *Porotermes adamsoni*. No essential difference has been detected between the parabasal bodies of these two species. The flagellates were fixed in cacodylate-buffered 2.5% glutaraldehyde, postfixed in buffered 1% osmium tetroxide, dehydrated in ethanol, embedded in Araldite epoxy resin, and sectioned in the usual way. Some material was fixed in osmium tetroxide alone. Sections were stained with both uranyl acetate and lead citrate.

Fig. 3 shows an approximately longitudinal section of a small part of a parabasal body in

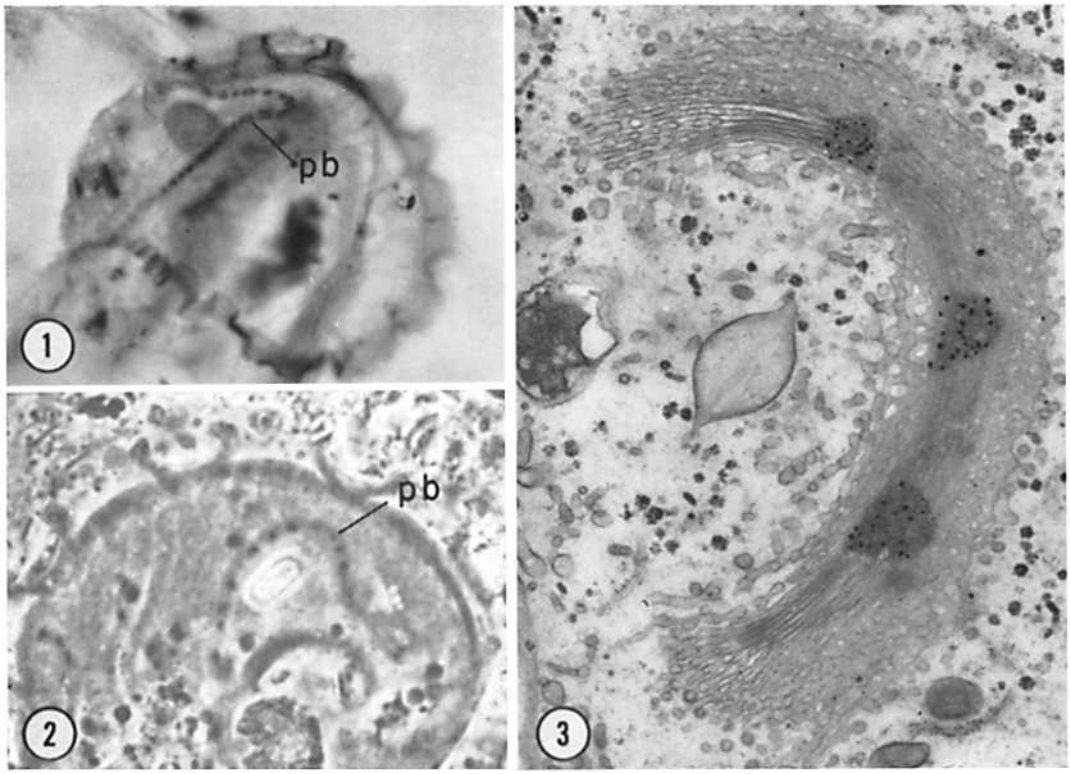


FIGURE 1 Photomicrograph of *T. termopsidis* stained by the Bodian technique (*pb*, parabasal body.) × 1,500.

FIGURE 2 Photomicrograph of living *Trichomonas* of an undescribed species from *Porotermes adamsoni*, showing alternating light and dark regions in the parabasal body (*pb*). Phase contrast. × 1,500.

FIGURE 3 Section of *T. termopsidis* showing a small part of the parabasal body in oblique longitudinal section. Note the three dense nodes, with small dense granules characteristic of $OsO_4$-fixed material. × 25,000.

which there are three prominent areas of increased density. These areas will be called nodes. From their size and spacing (about 1 $\mu$ apart, center to center) there can be little doubt that they correspond to the dense nodes seen in living material or in material stained by Bodian's method. The nature of the nodes is more readily seen in Figs. 4–6, in which it is apparent that they arise from the presence of electron-opaque material between the central regions of the more distal cisternae. The nodes display a superficial resemblance to the grana of chloroplasts.

The distribution of the intercisternal material across the parabasal body may be significant from a functional point of view, as will be indicated later, and will, therefore, be described in some detail. In a large number of micrographs which have been examined, the dense material has never been seen in the five most proximal intercisternal spaces, and, typically, the six proximal spaces are free of it. In a very few cases, it is apparently restricted to a more distal location, and in Fig. 5, which is the most extreme example encountered, there is no dense material between the 13 proximal intercisternal spaces. The situation in this micrograph could, however, be accounted for by the fact that the section passes through the margin of a node. At the opposite face of the parabasal body the dense material is usually present between all the uninflated cisternae except the two most distal. No dense material can usually be found associated with

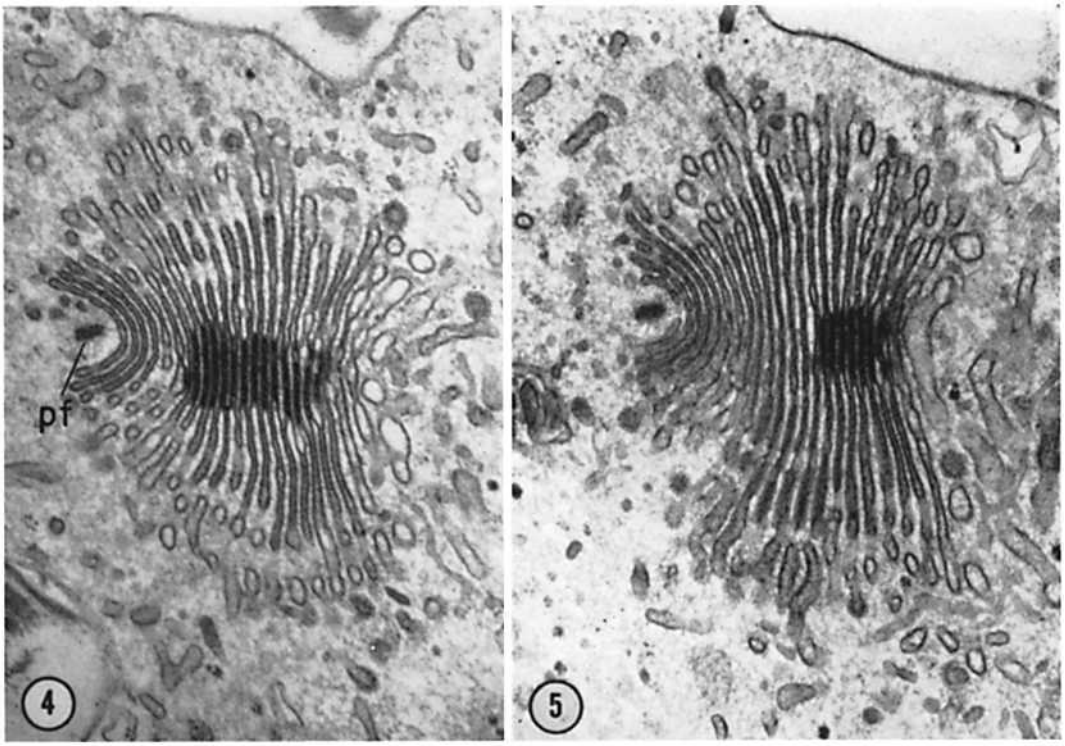


FIGURE 4 Parabasal body in transverse section. Note that where the dense intercisternal material is present, the membranes are more closely and regularly spaced than elsewhere. The parabasal filament (*pf*) lies on the proximal face of the parabasal body. Glutaraldehyde fixation. × 38,000.

FIGURE 5 Transverse section of a parabasal body showing intercisternal material restricted to the seven distal intercisternal spaces, the 13 proximal ones being free of it. Glutaraldehyde fixation. × 38,000.

distal cisternae which are partly inflated (Figs. 5, 6). In any node the intercisternal material is present between all adjacent cisternae; no case has been seen of an intercisternal space lacking it interpolated between others where it is present. Examination of longitudinal sections shows that different nodes in the same parabasal body usually have substantially the same distribution of intercisternal material in the proximo-distal direction: the distribution of the material in two adjacent nodes has not been found to differ by more than one intercisternal space. The location of the intercisternal material across the parabasal body may be summarized by saying that a typical parabasal body consists of 21 uninflated cisternae, giving 20 intercisternal spaces, and that if these are numbered from the proximal face, then the dense material of the nodes will typically be found in spaces 7–19.

The lateral extent of the dense material along the intercisternal spaces, as seen in transverse sections of the parabasal body, varies considerably, both from one node to another and from one intercisternal space to the next (compare Figs. 5 and 6). It never extends across the whole intercisternal space, but is restricted to the central third or less. A precise description of the extent of the material would require serial sectioning, which has not been attempted, but it is clear that the areas of dense material are usually less extensive in the more proximal spaces than distally (Figs. 5, 6).

Where dense material is present, the membranes of the cisternae run markedly straighter, closer together, and more nearly parallel to each other than where it is absent. This is apparent from a comparison of proximal cisternae, where no dense material is present, with distal ones (Fig. 4), or of

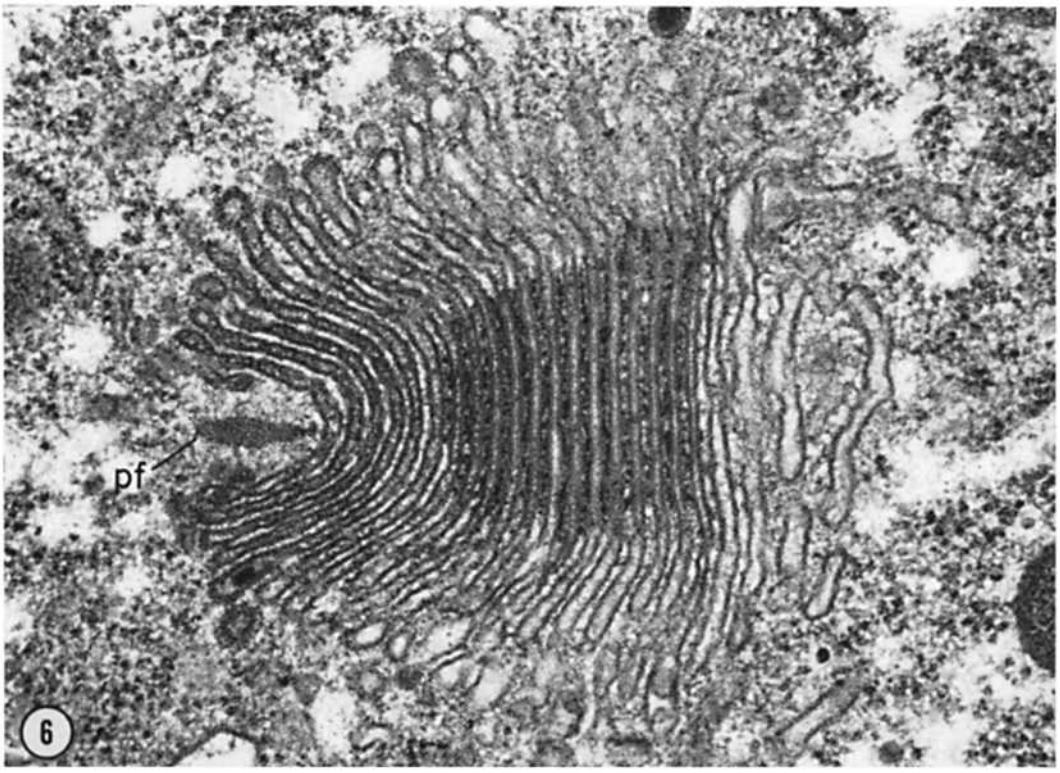


FIGURE 6 Transverse section of parabasal body, showing dense material between 14 of the more distal cisternae. Note the parabasal filament (*pf*), and the absence of intercisternal material between the six proximal cisternae adjacent to it. Glutaraldehyde fixation. × 69,000.

the lateral regions of distal cisternae with their central areas (Figs. 4, 6). In accurately longitudinal sections it can also be seen that the cisternal membranes are in close and regular parallel array in the nodes, and are more irregularly arranged in the regions between. Where intercisternal material is present, the separation of the membranes, measured from their outer surfaces, is variable, ranging from about 50 to 200 A.

The membranes themselves, (which are trilaminar unit membranes as described in other flagellates (Cleveland and Grimstone, 1964), have a total thickness of 60 A, and the width of the intracisternal space in the uninflated condition is about 100 A.

After glutaraldehyde fixation the intercisternal material may appear uniformly dense (Fig. 4), but in most sections it has a discontinuous, irregularly granular structure (Fig. 7). The granules, or aggregates of them, seem to vary in size, sometimes filling the entire intercisternal space, sometimes appearing smaller. In osmium tetroxide–fixed material (Figs. 3, 8) the bulk of the intercisternal material appears less dense than after glutaraldehyde fixation, but there are present a number of small, extremely dense spherical granules, about 200 A in diameter. These granules are larger than the thickness of the intercisternal space and, therefore, cause the membranes to lose their parallel configuration. The dense granules are not present in glutaraldehyde-fixed material: Fig. 6 shows one apparent granule of large size, but it is superposed on the section.

Nothing is known about the chemical composition of the intercisternal material, about its functional significance, or ultimate fate. The most nearly comparable observations seem to be those of Mollenhauer (1965), who described intercisternal material in the Golgi bodies of maize root-tip cells. In those cells, the material appears to be located rather precisely in the middle of the intercisternal space, sometimes in the form of a

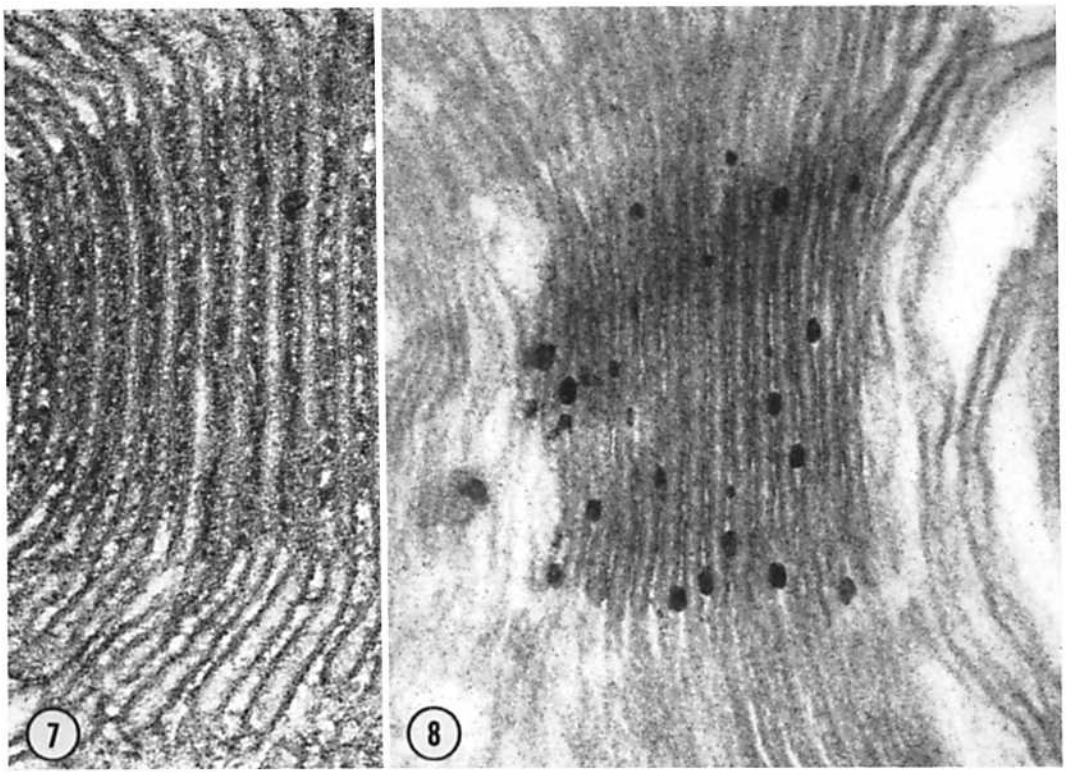


FIGURE 7 Enlargement from Fig. 5 to show the membranes of the cisternae and the material between them. × 120,000.

FIGURE 8 Parabasal body in transverse section, showing the small dense granules which are found after $OsO_4$ fixation. × 120,000.

narrow, continuous sheet. Turner and Whaley (1965) have also described intercisternal material in Golgi bodies of *Nitella*, but it is seemingly present in filamentous form. (It is not clear what relationship, if any, this material has to the seemingly fibrous, supposedly intercisternal material described in negatively stained, isolated Golgi bodies [Cunningham, Morré, and Mollenhauer, 1966].) The fact that the cisternae run parallel to each other and are uniformly spaced where the intercisternal material is present suggests that this material may link the membranes together (or, at least, that it does so after glutaraldehyde fixation), but it is not clear why this should be the case in Golgi bodies of *Trichomonas* and not in those of most other cells, including those of other flagellates, such as *Trichonympha*.

In a previous paper, evidence was reported which suggested that in *Trichonympha* the proximal face of the parabasal body is the site of formation of new cisternae, which replace those used up in vacuole formation at the distal face (Grimstone, 1959). This hypothesis, which implies that the parabasal body is a steady-state system, is probably applicable to the Golgi body in *Trichomonas* and, indeed, to most Golgi bodies. If this is the case, the intercisternal material cannot be formed at the same time as the cisternae, since it is never present between the proximal ones. Since it is rather difficult to envisage that the material could be inserted from outside the parabasal body into the central regions of the intercisternal spaces, perhaps the most plausible explanation of its distribution is that it represents a product of the cisternae which is synthesized some time after their formation. The fact that the material is not produced earlier (i.e. between more proximal cisternae) might then be an indication of the fact that these cisternae either are in some way not "mature," or simply have not been functional for

long enough. This reasoning, of course, in no way accounts for the periodic distribution of the nodes along the length of the parabasal body, and no explanation of this distribution can be offered at present. Irrespective of the mechanism of formation, however, the asymmetric distribution of the intercisternal material across the parabasal body appears to be a further manifestation of the fundamental polarity of the Golgi apparatus.


## SUMMARY

In the Golgi body (parabasal body) of *Trichomonas termopsidis* and *T. gigantea*, electron microscopy shows the presence of dense, finely granular material between some of the cisternae. This intercisternal material is not uniformly distributed throughout the Golgi body but is restricted to discrete regions, or nodes, which occur periodically along the length of this organelle. It occurs between the distal cisternae but not between the newly formed, proximal ones. The significance of these observations in terms of the functioning of the Golgi apparatus is discussed.



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