INTRODUCTION

A great deal of work has been done in the past on the systematics of Protozoa, but very little attention has been paid to the study of their cytoplasmic inclusions and nuclei. The majority of the earlier workers have employed the old classical techniques; hence there is a good deal of confusion with regard to the identity and morphology of the various cell-structures. It will not be an exaggeration to say that even in the same protozoan, the accounts of the morphology of the various cytoplasmic inclusions vary with the different authors.

Kudo (1954) remarks: 'It seems impossible at present to indicate just exactly what the Golgi apparatus is, since the so-called Golgi techniques, ........... are not specific and the results obtained by using same method often vary a great deal'(Kudo- Protozoology).

The publications of Hirsch (1939), Roth (1944, 1957b and 1959), Baker (1944, 1949, 1957a, 1957b and 1959), Chou and Meek (1958), and others, based on morphological, histochernical, and electronmicroscopical investigations, have more or less solved the Golgi problem in Metazoa; but in Protozoa, in spite of the publication of numerous reviews by earlier workers like King (1927), Hill (1933), Maclennan (1941), and Smyth (1944) on the Golgi apparatus, the position is still far from clear.

Using the classical techniques of long osmication and silver impregnation, previous authors regard the following structures of Protozoa as the Golgi apparatus:
1. Wall of the contractile vacuole and its feeding canals.
2. Osmiophilic material associated with the wall of the contractile vacuole.
3. Parabasal body of the flagellates.
4. Stigma of the Euglenoids.
5. Osmiophil, neutral red-stainable vacuoles (vacuole).
6. Completely non-osmiophil, neutral red-stainable vacuoles (vacuole).
7. Nuclear membrane (including knob-like bodies on the surface of meganucleus).
8. Osmiophil granules, rings, crescents, and duplex spheres.

With electron microscopy also, the following three structures which are osmiophil, represent the so-called Golgi apparatus.
1. Wall of the contractile vacuole.
2. The Dalton complex.

Even in some protozoans, unrelated structures have been described under the heading of so-called Golgi apparatus.
1) In opalinids, for example, the Golgi apparatus is said to be represented by:
   (a) A single type of lipid inclusion which also represents the mitochondrion (Hirschler, 1924).
   (b) Densely osmiophil, pyriform or irregular bodies (Ring and Gatenby, 1976).
   (c) Disc-like Golgi element, each consisting of an outer lipidiferous membrane forming a loop.
(d) Irregular granules to highly twisted snake-like elements (Richardson and Homing, 1931).
(e) Rounded or twisted, snake-like bodies (Patter, 1932).
(f) Granules (Khajuria, 1950).

(2) In Paramecium the Golgi apparatus is represented by:
(a) Wall of the contractile vacuole (K Cassonov, 1934).
(b) Contractile vacuoles and certain knob-like bodies on the surface of meganucleus (Park, 1939).
(c) Osmiophil, neutral red-stainable globules (vacuoles) (all and Fiselli, 1937).
(d) Wall of collecting canals of the contractile vacuole (Catenby and others, 1955).
(e) Lipid granules (seen in low power micrograph Pl. 109, fig.1, of Cedar and Porter, 1955). They are comparable to the Golgi bodies.
(f) 'Crescentic' and spherical lipid bodies (Cedar and Rudzinska, 1956).
(g) Lipid granules (Sen Gupta and Ray, 1958).

In view of this confusion, I was asked by Professor Vishwa Nath to work out the histochemical studies of some Protozoa with a view to establish the identity of the cytoplasmic inclusions.

From the observations recorded here, we are now in a position to conclude that the 'Golgi problem' in Protozoa is practically solved. A uniform structural pattern of the cytoplasmic inclusions has been observed in all the five classes of Protozoa.

As described by Baker (1939), in Protozoa also, only two categories of permanent cytoplasmic inclusions
have been recognized with light microscopy, namely (1) lipid bodies and (?) mitochondria. In the parasitic eugregarines and parasitic ciliates, however, there is a third category of cytoplasmic inclusions, i.e. carbohydrate bodies.

Lipid bodies are either solid or duplex spheres; the latter show an externum and an internum. They are comparable to the Golgi bodies described in Metazoa by Hirsch, Nath and Baker. In Protozoa also, they are the only inclusions which are consistently osmiophil and which can be assigned to the category of the 'Golgi apparatus'. The solid lipid bodies or the externum of the duplex lipid bodies is composed of lipids and some proteins. The internum of the duplex bodies is probably watery in nature.

The duplex parabasal body of _Hematomonas muscarum_ (Trypanosomidae) is morphologically similar to the duplex lipid body. The externum of the duplex parabasal body, however, contains DNA in addition to some lipids and proteins. The so-called 'Golgi apparatus' has never been shown to contain DNA; for this reason, the parabasal body of the Trypanosomidae cannot be perhaps homologized with the Golgi apparatus despite its duplex structure.

The mitochondria represent the second category of cytoplasmic inclusions, the presence of which has been recognized in all the classes of Protozoa. With electron microscopy the ultrastructure of the mitochondria has been established in a very large number of Protozoa except in the sub-class Opalinata and family Trypanosomidae. During the present investigation their presence has been established in the various classes of Protozoa, including sub-class
Opalinata and family Trypanosomidae.

Certain carbohydrate bodies have been recognized in the parasitic eugregarines and the parasitic ciliates, while the free living ciliates do not reveal any such bodies. Some of the previous authors have described the carbohydrate bodies which could not be stained with the routine staining techniques, as 'vegetative grains' or 'nutritive spheres' or 'paraglycogen bodies'.

As the result of my investigations, limited as they are, I have provisionally concluded that in parasitic ciliates carbohydrate material appears in the form of well-defined bodies, whereas in the free forms this material is diffused. This conclusion needs confirmation by studying many more free-living as well as parasitic types, with the histochemical techniques.

The study of nucleic acids was undertaken to investigate the distribution of DNA and RNA. It was considered important because workers on eugregarines have denied the existence of Feulgen positive material in them. Present investigation, however, has established the presence of DNA in very refractile rods present in the trophozoite. The nucleus of the eugregarines has been shown to be non-chromatic being composed of RNA and some proteins.