the appendage was stationary or whether nucleoplasm of the oocyte nucleus was pushed through the constriction into the nuclear bud in a pulsating movement. In none of the 13 cases in which the occurrence of nuclear appendages was filmed, could the origin of the appendages be ascertained. The site of the appendages suggested that they were formed by budding from the oocyte nuclei; in any case, involvement of pulsation of oocyte nuclei in the formation of the nuclear appendages could not be confirmed.

Some of these appendages were found in oocytes of ovaries which showed rapid development, i.e. which in all probability produced only female-determined eggs[11]. We were not able to follow these appendages in the films during oocyte growth, for accumulation of yolk granules obstructed the view. However, small nuclear-like vesicles were found in fixed material of immature and mature female-determined eggs produced in ovary cultures in vitro of a different series (figure 3). These nuclear-like vesicles were in most cases not (no longer?) attached to the oocyte nuclei. Chromosomal material was not found in them.

The observation that nuclear appendages were found in oocytes which most probably developed to female-determined eggs makes it unlikely that the nuclear appendages were transformed into small nuclei. The latter do not appear in female-determined eggs. Besides, it would be difficult to explain the occurrence of 10 chromosomes in the small nuclei. It seems more plausible to assume that the nuclear appendages were identical with the nuclear-like vesicles found in later stages of oocyte development.

The nuclear appendages and nuclear-like vesicles can be compared to the so-called 'accessory nuclei' which may occur in great numbers in oocytes of Hymenoptera and other insects[12,13]. These accessory nuclei probably arise by budding from the oocyte nucleus. Their function is still controversial. As yet we have found no indication of a possible function of the nuclear appendages in development of *Heteropeza pygmaea* oocytes.

1 This study was supported in part by the Swiss National Science Foundation.
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Secretory cell in the medulla of the bursa of Fabricius

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Summary. Light microscopy has revealed a possible secretory cell in the medulla of the bursa of Fabricius. Cyclophosphamide increased the presence of the secretory cell.

Research with the bursa of Fabricius has contributed significantly to our understanding of immunoglobulin synthesis in vertebrates[2]. We describe here the presence of a possible secretory cell in the medulla of the bursa and its cytological changes subsequent to treatment with cyclophosphamide.

Materials and methods. In our morphological studies, newly hatched 4-, 14-, 35-, 50-, and 120-day-old chickens were used. The bursal tissue samples were fixed in 4% buffered (0.2 M phosphate buffer) glutaraldehyde (2 h), rinsed 30 min in 0.2 M phosphate buffer (3 times), post-fixed in 2% osmium tetroxide (1–1.5 h), rinsed in buffer, dehydrated in ethanol, carried through propylene oxide, and embedded in araldite (Durcupan).

Results and discussion. Secretory cells were observed in all ages studied. They were localized in the medullary area and were usually parallel to the cortico-medullary border. We have found the largest number of cells in those follicles which were sectioned close to and parallel to the cortico-medullary border. In general, the secretory cell is elongated in shape, and the nucleus possesses a similar chromatin pattern to the small lymphocyte. The cytoplasm is dark and has 1 or 2 thick, long processes which contain dark granules of less than 1 μm in diameter (figure 1).

The secretory function of these cells became evident after cyclophosphamide (Cy) treatment. 7-week-old chickens (900–1100 g b.wt) were injected i.p. with Cy on 5 consecutive days. On the 1st day they were injected with 50 mg Cy/kg b.wt and on the following 4 days with 25 mg Cy/kg b.wt. The animals were killed 4 days after the last injection. The bursal tissues were prepared for histological investigation.

The lymphoid cells were eliminated from the follicles of Cy-treated birds. Histological structure of the follicles was the same as described by others[3–8]. In 1-μm sections stained with toluidine blue the bursa medulla of Cy-treated birds revealed a large amount of an intercellular dark substance (figure 2). The follicles of Cy-treated birds contained only epithelial cells, secretory cells, small lymphocyte-like cells, and plasma cells. Secretory cells were second in abundance to epithelial cells in the follicles of Cy-treated birds. The changes in secretory cell structure subsequent to Cy-treatment included modifications of the nucleus and the cytoplasm. The nucleus became round and revealed a leptochromatic pattern instead of the rough heterochromatic one of the normal animal. The cytoplasm was abundant and pale with fewer granules than in normal animals. Instead of long thick processes the cytoplasm was bulky, and exhibited spike-like processes embedded into the intercellular substance (figure 3). This cell type may be the same as mentioned by Glick[3] as large pale reticular cells or by Hoffman-Fezer et al.[8] as solitary thioninophilic cells. The intercellular substance appears initially around the cells containing granules (figure 4). When the light micrographs showed large amounts of an intercellular substance the

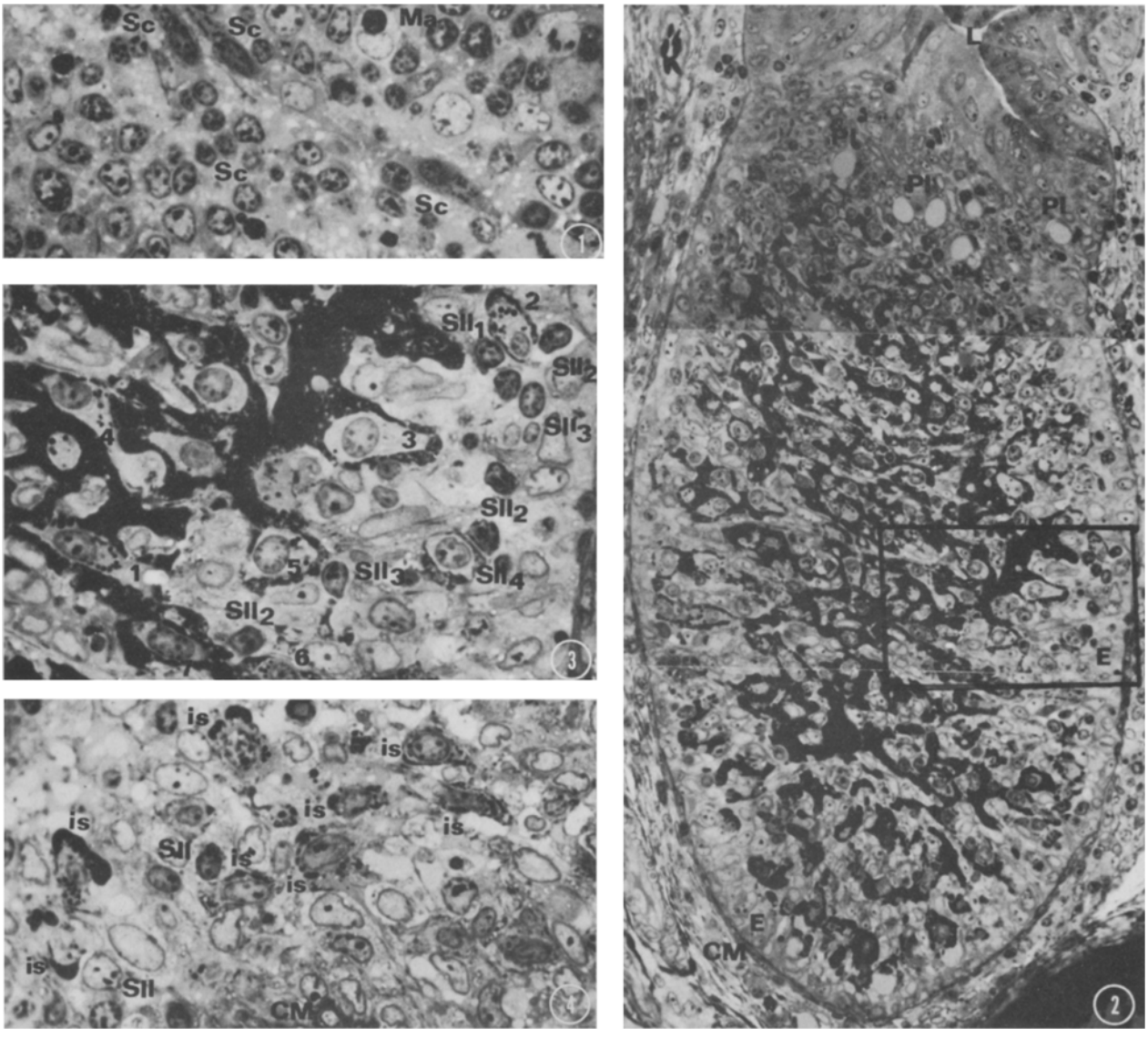

Fig. 1. 35-day-old normal animal. The secretory cells (Sc) show an elongated darkly stained nucleus with 1 or 2 nucleoli. The cytoplasm usually has 1 long thick process which contains several dark, spherical granules. Ma, macrophage; 1 μM toluidine blue. × 1200. Fig. 2. 50-day-old Cy-treated animal. The montage shows an excessive amount of extracellular substance filling the medulla. L, lumen of the bursa of Fabricius; CM, corticomedullary border; Pl, plasma cells with highly dilated ergastoplasmic cisternae; E, epithelial cell layer along the corticomedullary border. The outlined area is shown in figure 3. 1 μM toluidine blue. × 480. Fig. 3. The activated secretory cells (1–7) have a large nucleus with a leptochromatic pattern. The cytoplasm is pale, bulky, and contains few, if any, granules. Small spike-like cytoplasmic processes are embedded in the dark intercellular substance. The several small lymphocyte-like cells can be distinguished by the presence of heterochromatin, reduced cytoplasm (SII_1), pale cytoplasm (SII_2), and increased size with an intermediary chromatin pattern ($SII_{3,4}$). × 1200. Fig. 4. The dark intercellular substance (is) appears around the secretory cells which contain a large rounded nucleus and several granules. Small lymphocyte-like cells (SII) are seen close to the corticomedullary border (CM). 1 μM toluidine blue. × 1200.

electron micrographs revealed in the majority of the granules an intracellular granulolysis, which suggested that the granules had discharged their contents. 2 morphologically distinct types of small lymphocyte-like cells appeared close to the epithelial cells forming a continuous layer along the basement membrane (figures 3 and 4). The larger small lymphocyte-like cell had an intermediary chromatin pattern between that of the smaller small lymphocyte-like cell and secretory cell. The phenomenon suggested that the small lymphocyte-like cells may be the precursors of the secretory cells. The secretory cell appeared to be not only resistant to Cy, but also activated by Cy. The chemical nature and the functional significance of the secretory substance are under investigation.

1 We thank Karen Anderson for typing the manuscript and Dr Zygmunt (Meade-Johnson) for the gift of Cyclophosphamide. This is Journal Article No. 3732 from the Mississippi Agricultural and Forestry Experiment Station. Supported, in part, by grant No. R01 CA 20169, National Cancer Institute, DHEW.

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