Homologous Pairing and Synaptinemal Complexes in the Nurse-Cell Nuclei of Carabid Ovaries (Ins. Coleoptera)

The trophocytes of meroistic insect ovaries develop by differential mitosis from oogonia[1–3]. It has been shown by light- and electron-microscopic studies that the cellular derivates of an oogonium are connected by cytoplasmic bridges in accordance with the scheme of their mitotic cleavages. Thus a specially defined arrangement of cellular connections is established between all germ line elements of a follicle[4–6].

In view of the germ line nature of trophocytes, it is interesting that polycomplexes were found in the nurse-cell nuclei of the Nematocere *Aedes*[7]. These structures may represent stapels of synaptinemal complexes liberated by dysjunction of the paired chromosomes. In young *Drosophila* egg follicles synaptinemal complexes were found in the nucleus of the oocyte as well as in one of the 15 nurse-cells[6]. Synaptinemal complexes are nucleus organelles with a characteristic ultrastructure[8]. They are formed at the chromosomes during the leptotene stage of the meiotic prophase and presumably they guarantee the exact pairing of the homologous chromosomes[9].

Methods. See captions.

Results. During our investigations of oogenesis in some Carabid species (*Carabus*, *Pterostichus*, *Abax*) we found that the nuclei of young egg nurse-cell clusters in squash preparations all show a typical bouquet arrangement (Figure 1). If at all, the oocyte can only be distinguished from the protrophocytes by a slightly enlarged nucleus volume. Feulgen squashes clearly demonstrate the homologous pairing of the chromosomes in both cell types. In this respect the oocyte and protrophocytes show the same lightoptical aspects as described for Lepidoptera[10], which also have meroistic ovaries.

In electron-microscopic studies very young Carabid egg nurse-cell units exhibit no distinct morphological differences between oocyte and protrophocytes. But in accordance with the cytological properties examined by light-microscopy, these follicles showed various nuclei with clearly demonstrable synaptinemal complexes. Such cases suggest that young nurse-cell nuclei of Carabid ovaries build up the auxiliary structures characteristic of homologous chromosomes pairing during meiosis: from the genesis of an egg nurse-cell cluster it must be con-

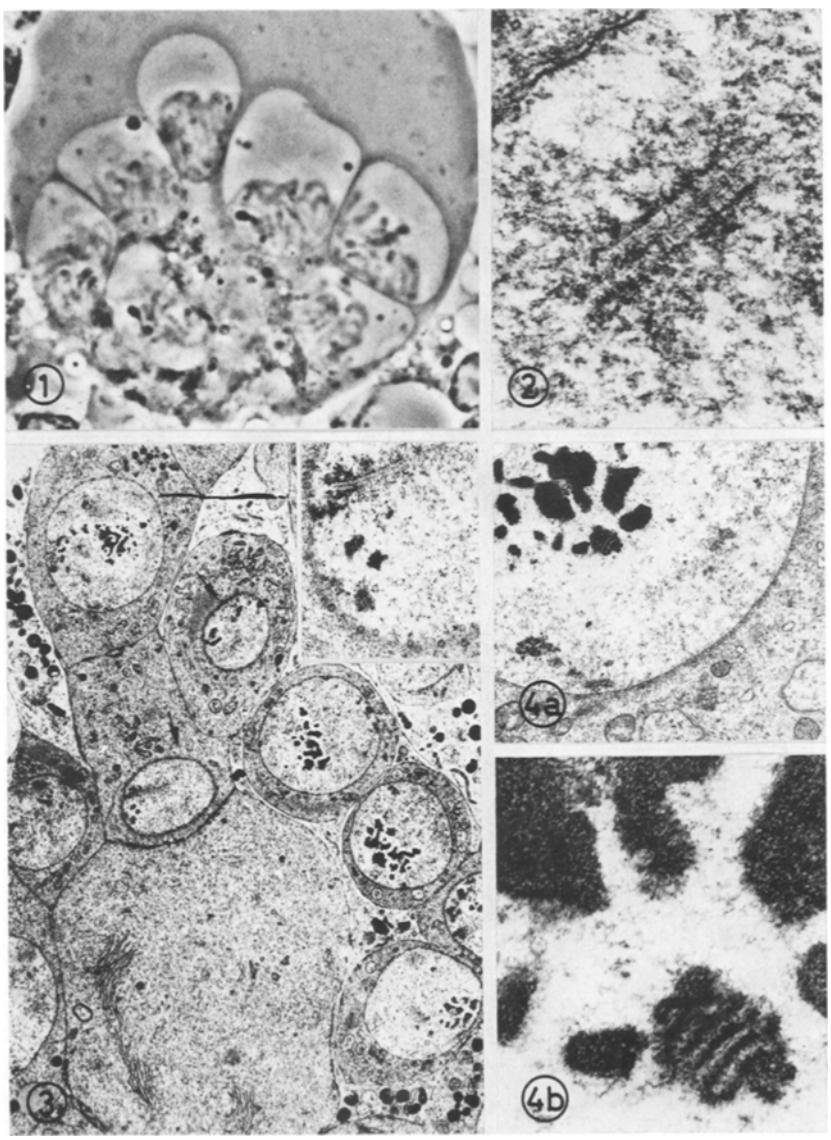

Fig. 1. Young egg nurse-cell cluster of *Abax ater* after unfixed squashing. All nuclei exhibit zygo-pachytene (bouquet arrangement). Ph, × 1680.

Fig. 2. Chromatin associated synaptinemal complex with lateral and medial components of a young nurse-cell nucleus (*Carabus granulatus*). OsO_4 fixed; contrasted with uranyl acetate and lead citrate. × 57,200.

Fig. 3. A somewhat older egg nurse-cell cluster of *Carabus granulatus* than in Figure 1. The tangentially sectioned oocyte differs from the nurse-cells by a greater volume and accumulated lamellar bodies. Synaptinemal complexes in nurse-cell nuclei are indicated by arrows. The insertion demonstrates the left complex with greater enlargement. × 3900 (insertion: × 14,000).

Fig. 4. a) Nurse-cell nucleus of *Carabus granulatus* with polycomplex-like structure enlarged in b). The polycomplex is associated with the nucleolar apparatus of the nurse-cell nucleus. a) × 98,000. b) × 50,000.

cluded that at least one of any 2 cells connected by a cytoplasmic bridge must be a nurse-cell.

But we have also succeeded in finding synaptinemal complexes in several nurse-cell nuclei of somewhat more developed follicles, in which the oocyte can be distinguished from the nurse-cells by its state of differentiation and its position (Figures 2 and 3).

Older stages sometimes exhibit polycomplex-like structures in oocyte and trophocytes, which might have arisen from synaptinemal complexes after disjunction of the chromosomes[7] (Figure 4).

Discussion. Our light- and electron-microscopic findings strongly suggest that in *all* protrophocytes the homologous chromosomes pair and build up synaptinemal complexes. To this extent the situation described differs from that in *Drosophila*[6]. The differentiation between oocyte and nurse-cells after the construction of synaptinemal complexes demonstrates the germ line nature of Carabid trophocytes. The synaptinemal complexes in protrophocytes seem to have no functional significance. We believe that they may represent rudimentary organelles. This is understandable in view of the phylogenetic origin of the trophocytes from oocytes.

Not only the morphology of the young follicles suggests that the Carabids possess relatively primitive meroistic ovaries. The function of their young follicles is equally primitive. After the breakdown of chromosome pairing the oocyte enters into a lampbrush stage with extremely uncoiled chromosomes[11]. The nucleolus of the oocyte shows a considerable increase in size[12]. The rate of the RNA-synthesis is about twice as great as in nuclei of other tissues with comparable ploidy-step ($4n$)[12]. These and other observations suggest that the initial growth of the oocyte in Carabids is due to RNA which is synthesized in the oocyte nucleus. In a second previtellogenetic phase, the oocyte nucleolus disappears and – somewhat later – the chromosomes condense to form the caryosphere[11] typical of meroistic ovaries. At this point the oocyte growth is completely directed by RNA transfered from the polyploidized nurse-cells to the oocyte cytoplasm[13].

Zusammenfassung. Bei Carabiden erfolgt die Differenzierung zwischen Oocyte und Trophocyten nach der Homologenpaarung und dem Aufbau synaptischer Komplexe in beiden Zelltypen. Zusätzlich zu diesem morphologischen Aspekt deutet auch die Funktion des Oocytenkernes auf einen relativ primitiven Zustand des meroistischen Carabidenovars. In der Initialphase der Oogenese zeigt der Oocytenkern bemerkenswerte Ähnlichkeiten mit den morphologischen und funktionellen Verhältnissen des Oocytenkernes panoistischer Ovarien.

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Digestion Process of *Microsporum cookei* Spore Wall by Snail Enzyme: Scanning Electron-Microscopic Examination

Studies of the surface features of fungal wall with the scanning electron-microscope (SEM) have provided much useful information[1-5]. We have been investigating the ultrastructural and immunochemical characteristics of the cell wall of dermatophytes which cause trichophytosis in man[6,7]. Recently, observations by SEM conducted in our laboratory of the surface structure of the hyphal and spore walls of several strains of dermatophytes (*Microsporum cookei* HUT-2061; *Epidermophyton floccosum* TEF-30; *Trichophyton rubrum* T-1005; *T. megnini* 4034; *T. tonsurans* A410, *T. fluviomuniense* 3635)[8] and other non-pathogenic fungi (*Aspergillus niger* 460, *A. oryzae* 216; *Penicillium chrysogenum* 473; *Candida utilis* 553)[9], yielded some interesting results. However, there are no reports in which the surface structures of the cell walls were observed by stereoscan following treatment with mycolytic enzymes.

Materials and methods. Fungus: Cultures of *M. cookei* HUT-2061 were grown on Petri dishes containing Sabouraud's dextrose agar (4% glucose, 1% peptone, 0.5% yeast extract and 1.7% agar), for 4 weeks at 28 °C.

Preparation of spore: The compactly grown cultures, consisting almost entirely of macroconidia, were scraped off the agar medium with a spatula. The spores were freed from the accompanying mycelia by thorough washing with saline followed by centrifugation.

Digestion with snail enzyme: Snail-gut juice of *Helix pomatia*, supplied by L'Industrie Biologique Française, Seine (France), which contains the lytic enzyme complex helicase, was stored as a freeze-dried powder at −15 °C until used. The washed macroconidia were suspended in 0.05 *M* citrate-phosphate buffer (pH 5.8) containing 0.6 *M* mannitol and 0.01% cystein hydrochloride[10], to which 10 mg/ml of snail-gut juice powder was added. Digestion was then carried out by incubation at 28 °C with gentle shaking for varying periods.

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