# DIFFERENTIATION OF THE CORTICAL CYTOPLASM AND INCLUSIONS IN OOCYTES OF THE FROG*

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Studies on differentiation of endodermal derivatives in amphibian embryos (Kemp, 1946, 1951) brought to my attention the well known fact that endodermal cells generally retain their stored yolk much longer than ectodermal or mesodermal cells. This observation aroused my curiosity concerning the general problems of synthesis and utilization of yolk in oocytes and embryos. The opportunity to apply electron microscopy to these problems came while I was working with Dr. T. N. Tahmisian at the Argonne National Laboratory during the summers of 1954 and 1955 and has been continued at the University of Michigan, where Dr. R. E. Hartman has kindly permitted me to use the electron microscope at the School of Public Health. This short report is based on work to be published in detail elsewhere (Kemp, 1956).

Before the young oocyte starts to synthesize lipide or protein inclusions, it has already started to grow (Kemp, 1953). Lampbrush chromosomes (Dodson, 1948; Gall, 1954) produce many nucleoli, which move to the periphery of the nucleus as they grow (Figs. 1 and 2). I have not observed the direct passage of nucleoli or nucleolar fragments through the porous nuclear membrane, but small granules and rods appear in the perinuclear cytoplasm after the nuclear membrane starts to fold (Fig. 3). Lipide droplets called lipochondria by Holtfreter (1946) soon appear throughout the cytoplasm either independently or clustered about yolk nuclei (Fig. 4). At the same time, the follicular epithelial cells adjacent to the oocyte pull away partially, thus leaving spaces into which grow microvilli erupting from the surface of the oocyte.

Yolk platelets first appear in the peripheral endoplasm. In an oocyte at stage $Y_1$, measuring about 350 $\mu$ in diameter, the cortical cytoplasm has differentiated into three distinct regions (Fig. 5). Bordering on the peripheral endoplasm is a layer containing distinctive cortical granules, first described for the frog by Motomura (1952). External to these granules is a basal cortical layer from which emerge the microvilli comprising the outermost cortical zone. Processes from follicular cells extend inward toward the microvillous layer and sometimes appear to interdigitate with microvilli (Fig. 6).

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By the time yolk has been deposited in the outer three fourths of the cytoplasm, the basal cortical cytoplasm has become folded into alternating ridges and valleys (Fig. 7). The microvilli now extend from the summits of the cortical ridges. Shortly after this stage is reached, pigment appears in the peripheral endoplasm. The appearance of yolk platelets, pigment granules, and lipochondria in the endoplasm of a stage $Y_4$ oocyte (yolk filling the entire cytosome) is shown in Fig. 8.

There is considerable presumptive evidence in support of Caspersson's (1950) theory (see Goldschmidt, 1955) that nucleoli transport RNA to the cytoplasm, which is thereby stimulated to synthesize proteins. Presumably one link in the chain of cause and effect is the stimulation of enzyme synthesis. Differentiation of the cell surface and synthesis of the visible inclusions—lipochondria, cortical granules, yolk platelets, and pigment granules—may all be manifestations of the processes set in motion by the release of RNA into the cytoplasm.

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PLATE

## EXPLANATION OF PLATE 63

FIG. 1. Photomicrograph of a portion of two oocytes at stage $Y_1$. Large nucleoli (*n*) are close to the nuclear membrane. Yolk platelets (*y*) have been deposited in a thin layer just beneath the cortical cytoplasm. Follicle cells (*f*) surround the oocyte. × 120.

FIG. 2. Low power electron micrograph showing appearance of nucleus and nucleolus (*n*), below, and cytoplasm of section of a young oocyte prior to the appearance of inclusions. Cytoplasm contains abundant mitochondria (*m*). Nucleus of follicle cell (*f*) peripheral to oocyte. × 8,135.

FIG. 3. Small granules (*g*) in the perinuclear cytoplasm of a young oocyte in which the nuclear membrane has started to fold. Mitochondria (*m*) are conspicuously larger. Nucleolus (*n*) inside nucleus. × 14,300.

FIG. 4. Peripheral cytoplasm and portion of follicle cell of young oocyte showing a yolk nucleus which appears to consist of a cluster of mitochondria (*m*) and lipochondria (*l*) of various sizes. Microvilli (*v*) are beginning to protrude into space between oocyte and follicle cell. × 8,135.

FIG. 5. Surface of an oocyte at stage $Y_1$ after yolk platelets (*y*) have appeared in the peripheral endoplasm. The cortical cytoplasm has differentiated into 3 layers: a layer containing cortical granules (*c*), a basal cortical layer (*bc*), and a layer of microvilli (*v*). Follicular cell (*f*) external to oocyte. × 7,525.

FIG. 6. Highly magnified view of surface of oocyte and follicle cell showing a follicular process (*f*) extending down into the layer of microvilli (*v*). × 28,820.

FIG. 7. Surface of oocyte at stage $Y_3$ after yolk has been deposited in outer three fourths of cytoplasm. Basal cortical layer (*bc*) has become folded and contains numerous droplets. Microvilli (*v*) extend from summits of cortical ridges. × 14,300.

FIG. 8. View of inclusions in subsurface endoplasm of oocyte at stage $Y_4$ when yolk fills the cytoplasm. Yolk platelets (*y*), lipochondria (*l*), and pigment granules (*p*) are abundant. × 8,135.

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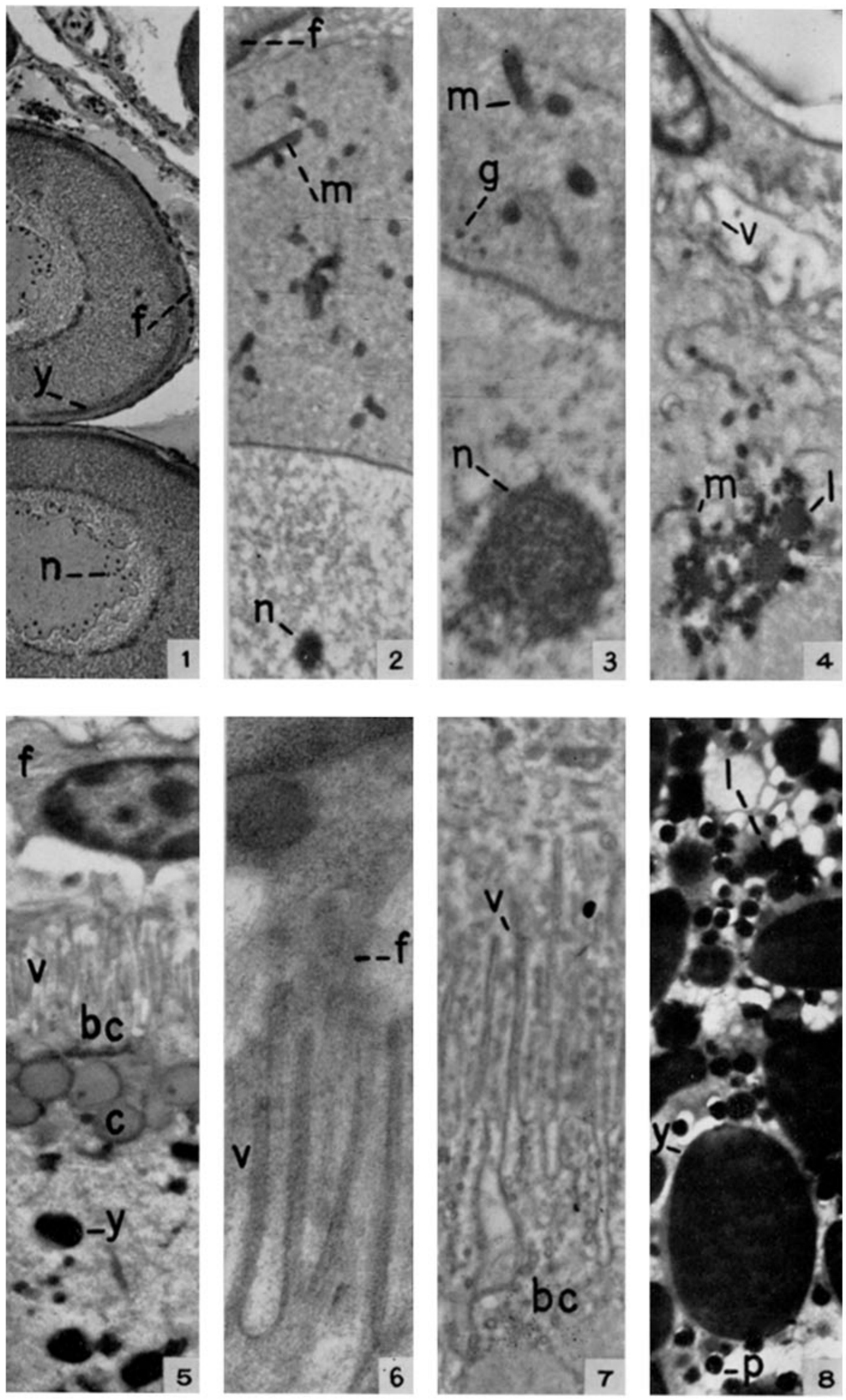


(Kemp: Cortical cytoplasm and inclusions in frog oocytes)