Laminin and basement membrane-associated microfilaments in wild-type and mutant Drosophila ovarian follicles

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*We are greatly saddened by the early and tragic death of our collaborator Eugen Gratwohl

Summary
The localization of the extracellular matrix glycoprotein laminin was studied using polyclonal anti-laminin antibodies. The laminin patterns of the basement membranes of the muscular epithelial sheath that envelops the ovariole were conspicuously different from those of the basement membrane of the follicular epithelium. In the latter structure laminin was stained in a pattern of parallel stripes oriented perpendicular to the long axis of the follicle; microfilament bundles at the adjacent basal side of the follicle cells have the same orientation. At late vitellogenic stages the orientation of the microfilaments remained the same while the laminin stripes were no longer visible. The orientation of laminin and F-actin was abnormal in follicles of the egg-shape mutant kugel, which produces shorter and thicker eggs than wild-type flies. This phenotype might result from the disturbance of the normal circular microfilament and/or laminin pattern.

Key words: basement membrane, microfilaments, laminin, Drosophila, oogenesis, egg shape.

Introduction
Each ovariole of the Drosophila ovary contains a chain of follicles of different developmental stages. The youngest follicle is located near the gerarium at the anterior end of the ovariole and increasingly older stages are lined up in the anteroposterior direction. The wall of the ovariole is formed by a muscular and highly elastic epithelial sheath (King, 1970; Fig. 2A). The 16 germ-line cells of each follicle, consisting of 15 nurse cells and the oocyte, are enveloped by a follicular epithelium that is lined with a basement membrane.

The follicular epithelium has important functions during oogenesis. For example, this follicle cell layer synthesizes yolk polypeptides, promotes the uptake of vitellogenin from the haemolymph into the oocytes (for reviews see Tejler et al. 1982; Bownes, 1982) and, during the final stages of oogenesis, synthesizes the highly complex egg shell (Margaritis, 1985). It has recently become clear that in Drosophila the follicle cells also have essential functions in the establishment of the embryonic axes (Nüsslein-Volhard, 1990).

The follicle cells differentiate early in development; this has, for example, been demonstrated with antibodies against fasciclin III (Brower et al. 1981) or by the enhancer trap method (Grossniklaus et al. 1989). Specific groups of cells could be labelled by these methods even at previtellogenic stages when the cells form a morphologically homogeneous layer around the germ-line cells.

Cytological studies on the differentiation of the follicular epithelium during previtellogenic and early vitellogenic stages have not been carried out systematically. We have begun to analyze the cytoskeleton of the follicular epithelium and shown that F-actin at the basal side of the follicle cells (bordering the basement membrane) is organized in parallel microfilament (MF) bundles oriented perpendicular to the long axis of the follicle (Gutzeit, 1990). At late previtellogenic stages and early vitellogenic stages the MF pattern indicates a circular polarity of the follicular epithelium that had not been recognized previously (Gutzeit, 1991). During vitellogenesis the MF bundles become more numerous in each cell and at stage 10 (for stages see King, 1970) a layer of dense parallel MFs forms just underneath the basal cell membrane of the columnar follicle cells enveloping the oocyte. Indirect evidence suggests that the circular MF bundles promote increased adhesiveness to the basement membrane. The pattern in the spread-out follicle cells covering the nurse cells is different (Gutzeit, 1990).

Since the basement membrane may play a role in shaping the follicle as well as organizing the basal cytoskeleton in the follicle cells we began to analyze the distribution of laminin, a major and ubiquitous component of basement membranes in vertebrates and invertebrates. Assays of the biological activity of laminin have revealed that the molecule may affect cell growth, motility and differentiation. Also, cultured cells may adopt a certain shape, depending on the presence of laminin and other components of the extracellular matrix and on the origin of the cells (reviewed by Watt, 1986). A change in cell shape may necessitate the reorganization of cytoskeletal elements, in particular actin (see for example Couchman...
The molecular properties of Drosophila laminin are known in some detail (reviewed by Fessler and Fessler, 1989; Beck et al. 1990; Timpl, 1989) and its distribution during embryogenesis has been studied with anti-laminin antibodies (Montell and Goodman, 1989; Fessler et al. 1987; Garzino et al. 1989).

In this study we have analyzed the laminin pattern in the basement membranes of the Drosophila ovary. Our analysis has revealed striking local differences with respect to concentration and organization of laminin. Since the observed pattern suggested to us that laminin might play a role in shaping the follicle during the course of oogenesis, we also analyzed the pattern in the egg-shape mutant kugel, in which mature follicles are almost spherical. We show here that the laminin pattern as well as the MF pattern is disturbed in the mutant.

Materials and methods

Fly stocks
Well-fed Oregon R wild-type females no older than 1 week of age were used. The stock of kugel was kindly provided by Dr Ch. Nüsslein-Volhard (Tübingen).

Preparation and specificity of anti-laminin antibodies
Laminin was isolated from the media of Drosophila melanogaster Kc cells according to Fessler et al. (1987). The cell line was kindly provided by Professor W. J. Gehring (Basel). For preparation of antibodies the laminin-containing fractions of a heparin-Sepharose column were analyzed by SDS–PAGE (3% to 8% gradient) under denaturing conditions. The laminin band (Mr=850000) was excised from the gel and used for immunizing two rabbits (no. 329 and no. 330). The sera were tested by immunoprecipitation of laminin from conditioned media of Drosophila ovaries (10 pairs of ovaries/100 μl antiserum) for lh at 37°C. Both antisera have similar specificity, since radioactive proteins with Mr values estimated to be about 400000 (A-chains of laminin) using high Mr markers (Sigma). Under non-denaturing conditions the only visible band had an Mr of about 900000 (C,D); this is expected for the intact laminin molecule, which consists of 3 chains linked by disulphide bridges. The gel analysis showed that the specificities of both antisera were very similar. No radioactive material was precipitated using the preimmune sera of the same rabbits.

Frozen section
Ovaries were fixed with 4% formaldehyde dissolved in half-strength DPBS (Dulbecco and Vogt, 1954) and frozen sections were prepared as described before (Grau and Gutzeit, 1990). Non-specific binding sites were blocked by incubating the sections in 1% BSA in PBS (1 h at 37°C). The antibody (rabbit no. 329) was diluted 1:500 with PBS. The Vectorstain ABC kit (Vector laboratories, Burlingame, CA) was used as a detection system; bound peroxidase was visualized by incubating the sections for up to 15 min in a developer consisting of 10 mM Tris–HCl, pH 7.5, 9% NaCl, 0.033% diamobenzidin, 0.033% H2O2, which was added just before use.

Whole mounts
In some experiments ovarioles were carefully dissected in R-14 medium (Robb, 1969) to leave the epithelial sheath intact. When the MF pattern or the laminin pattern of the basement membrane of the follicular epithelium with purified laminin was analyzed by immunoprecipitation using electrophoretically sharpened tungsten needles. During this procedure the epithelial sheath was generally removed. The follicles were fixed for 30 min in ice in freshly dissolved paraformaldehyde in DPBS or MF-buffer (Bond and Somlyo, 1982). Anti-laminin (rabbit) was used at a 1:500 dilution in 2% BSA in PBS, and biotin-SP-affinipure goat anti-rabbit IgG (Jackson ImmunoResearch Laboratories, West Grove, PA) was diluted 1:100. Anti-Drosophila activities in the latter antiserum were largely removed by incubating the antibodies with a homogenate of Drosophila ovaries (10 pairs of ovaries/100 μl antiserum) for 1 h at 37°C. Dichtlorotriazinyl amino fluorescein (DTAF)-conjugated streptavidin or Texas Red-conjugated streptavidin (both from Jackson) was diluted 1:1000. The follicles were incubated overnight at 4°C in the primary antibody solution and for 30 min (37°C) in each of the above solutions of the detection system. The follicles were washed extensively with PBS between incubations. The preparations were viewed in a Zeiss Universal microscope equipped with epifluorescence optics. Control preparations in which the incubation with the primary antibody had been omitted showed weak diffuse staining of the follicles. Control follicles incubated with the preimmune serum did not show the characteristic staining pattern seen with the anti-laminin antiserum.

Microfilament staining
F-actin was stained with rhodaminyl–phalloidin kindly provided by Professor Th. Wieland (Heidelberg). Details of the staining procedure are published elsewhere (Gutzeit, 1990).

Concanavalin A binding sites
Follicles isolated and fixed as described above were incubated for 1 h at 37°C in a solution of purified concanavalin A conjugated to FITC (E-Y Laboratories, San Mateo, CA) diluted 1:20 with PBS.

Collagenase treatment
Follicles isolated in Robb's R-14 medium (see above) were treated for 1 h (30°C) with purified collagenase (Sigma, about 3800 Mandl units ml⁻¹; for details see Gutzeit, 1991). The follicles were washed with PBS and processed for immunostaining as described above.

Electron microscopy
Follicles were dissected in R-14 medium and transferred in a small volume of medium to diluted fixative solution (0.2% glutaraldehyde and 0.1% osmium tetroxide in 0.1 M cacodylate buffer, pH 7.4). After about 10 min the fixation was continued in a solution of 2% glutaraldehyde and 1% osmium tetroxide in the same buffer (1.5 h on ice). For details of the embedding procedure, sectioning and staining of ultrathin sections see Wenzel et al. (1990).
Results

Basement membranes of the somatic ovarian envelopes: ultrastructure and presence of laminin

Each ovariole is enveloped by a muscular epithelial sheath (King, 1970; Cummings, 1974). The follicular epithelium and the epithelial sheath are lined by basement membranes (Fig. 2A, B). The muscle cells of the epithelial sheath form long strands, which are, as a rule, oriented in a circumferential direction (perpendicular to the long axis of the follicle) and are sandwiched between two basement membranes. A distinctive lamina lucida was not visible in our preparations. The lamina densa was prominent in the basement membrane of the follicular epithelium but was considerably thinner in the basement membranes of the epithelial sheath (Fig. 2B). There was always a conspicuous pars fibroreticularis with electron-dense globular structures that could still be recognized in cases where the basement membranes of the epithelial sheath and of the follicular epithelium came into close contact (Fig. 2B, inset). Apparently, the opposing basement membranes do not fuse as is typically observed in other systems where double basement membranes form (Inoue, 1989). This might be so because during the course of oogenesis the growing follicles slide down the ovariole within the epithelial sheath, the external surfaces (pars fibroreticularis) of the respective basement membranes are required to slide against each other smoothly.

We have localized laminin in frozen sections through Drosophila ovaries using a specific polyclonal antibody (see Fig. 1 and Materials and methods). A superficial thin layer around each follicle was heavily stained. In some areas where two adjacent follicles were sectioned the epithelial sheath extending from one follicle to the next had lost its intimate contact with the follicles’ surface and in such cases it was apparent that the antigen was most concentrated in the basement membrane(s) of the epithelial sheath (Fig. 2C, arrowhead). The intercellular space between follicle cells and between follicle cells and oocyte (perivitelline space) was weakly stained (Fig. 2C, arrows).

Laminin and F-actin patterns in the basement membranes and muscle strands of the epithelial sheath

After staining with rhodamine-phalloidin, F-actin in the muscle strands showed up as brightly fluorescing bands (Fig. 2D). In a double-labeling experiment laminin was also stained using biotinylated goat anti-rabbit IgG antibodies followed by DTAF-labelled streptavidin. Little laminin was present where the muscle strands were located (Fig. 2E). However, laminin was present at high concentrations at the rims of the muscle strands. In addition, laminin strands bridged the space between adjacent muscle strands, thus forming a ladder-like structure whose rungs were spaced at a distance ranging from about 2.5 to 7 μm (Fig. 2E). Since the fixation was optimized for the immunological localization of laminin (no permeabilization with Triton X-100), the normal staining pattern of F-actin in the muscle strands (Gutzeit, 1986a; Gutzeit and Haas-Assenbaum, 1991) was usually not seen.

Regional differences of the laminin pattern in the basement membrane of the follicular epithelium

The staining of the basement membrane of the follicular epithelium was generally much weaker than the staining intensity of the basement membranes of the epithelial sheath, indicating a lower laminin concentration. In preparations in which the epithelial sheath, had been mechanically removed prior to fixation the laminin pattern of the basement membrane of the follicular epithelium could be analyzed in whole-mount preparations.

In the basement membrane of late previtellogenic and early vitellogenic follicles laminin was organized in a regular pattern: circular stripes of staining material with very precise circumferential orientation were observed (Fig. 3A). At midvitellogenic stages the circular pattern was maintained only at the anterior end and became progressively more diffuse towards the posterior end of the follicle (compare Fig. 3A and B).

The basement membrane of the follicular epithelium is continuous over the surface of the follicles and the stalk cells. Fig. 3C shows the anterior part of a stage 9 follicle with the attachment site of a brightly fluorescing stalk cell located a short distance away from the follicle’s anterior pole (encircled area in Fig. 2A). This preparation had been stained with FITC-labelled concanavalin A. The observed staining pattern closely resembles the pattern observed with anti-laminin antibodies, thus indicating that laminin is the major glycoprotein in the basement membrane of the follicular epithelium. A distinctive ring at the base of the stalk was visible in both laminin-stained (Fig. 3A) and concanavalin A-labelled preparations (Fig. 3C). This also holds true for the brightly stained fibres that radiate out in the plane of the basement membrane (Fig. 3C, compare with laminin-stained preparation shown in the inset). The basement membrane around the stalk cells was also differentiated; long laminin fibres could be recognized in preparations with low fluorescence intensity (Fig. 3C, inset). At the attachment site of the stalk cells to the follicular epithelium an area of weak staining was often observed around the intensely stained ring in concanavalin A-labelled (Fig. 3C) and laminin-stained preparations (not shown).

In follicles treated with purified collagenase laminin was still present in the basement membrane. An intricate network of immunopositive material became visible. The predominantly circular orientation of the remaining fibrous material could still be recognized in anterior regions of mid-vitellogenic follicles (Fig. 3D). This observation indicates that collagenase digestion left the laminin network in the basement membrane essentially intact.

Laminin and microfilament patterns in kugel follicles

The circular orientation of basal MF (Gutzeit, 1990) and of laminin (Fig. 3A) suggested to us that these structures might act as a circumferential corsettle, so that the follicle grows mainly in the direction of the anteroposterior axis. For this reason we also analyzed the MF and the laminin patterns of the egg-shape mutant kugel. Follicles of this mutant are shorter and thicker than wild-type follicles (Fig. 4F). In extreme cases the mature egg was almost spherical.

The follicle cells of wild-type follicles are polarized (Fig. 4A), which is best seen in phalloidin-stained stage 7 and 8 follicles. Cell protrusions form basally only at one side of each cell and extend in a circumferential direction over the neighboring follicle cell (Gutzeit, 1991). The circular polarity is abolished in homozygous kugel follicles. The polar cell protrusions were usually not present and the few remaining were oriented in different directions (Fig. 4B). Interestingly, the laminin pattern in kugel follicles was also abnormal. The circular orientation was
Fig. 2. (A) Schematic drawing of an idealized section through an ovariole containing two early vitellogenic follicles. For clarity the cell dimensions are not drawn to scale (cf. King, 1970). The germ-line cells, i.e. the 15 nurse cells (nc) and the oocyte (ooc) are shaded. These germ-line cells are enveloped by an epithelium of somatic follicle cells (fc). Neighbouring follicles are connected by stalk cells (sc). The follicle cells including the stalk cells are lined by a basement membrane (bm, thick line). The epithelial sheath (es) forms an elastic tube within which the follicles develop. The ultrastructure of the boxed area is shown in B; the circle indicates the region shown in Fig. 3C. The length of the larger follicle is about 250 µm. (B) Electron micrograph of the basal region of the columnar follicle cells covering the oocyte (stage 9 follicle). The oocyte is enveloped by an epithelial sheath (es) consisting of circular muscle strands seen in cross-section and two thin basement membranes (arrowheads) between which the muscle cells are sandwiched. The basement membrane of the follicular epithelium (bm, arrow) is considerably thicker. Adjacent to the basal plasmalemma is a layer of microfilaments oriented circumferentially like the muscle strands (Gutzeit, 1990); ics, intercellular space between two follicle cells. ×30,000. Inset: in some areas the basement membranes of the epithelial sheath and of the follicular epithelium were in close contact. In these cases the opposing basement membranes did not fuse and the globular electron-dense structures (pars fibroreticularis) on the surface of the respective basement membranes were still visible. (C) Frozen section (about 10 µm thick) through a wild-type ovary. The localization of laminin was studied with a polyclonal antibody and a detection system based on the activity of peroxidase using diaminobenzidine as a substrate; the insoluble reaction product appears dark. The epithelial sheath was strongly labelled (arrowhead) while the follicular epithelium was labelled only weakly at the basal side (basement membrane) and the intercellular spaces between the follicle cells, and between follicle cells and oocyte. The strong labelling of the epithelial sheath is clearly seen in areas where the epithelial sheath does not contact the basement membrane of the follicular epithelium, e.g. between adjacent follicles (see also Fig. 2A). Control sections that had not been incubated with the primary antibody were unstained. Bar, 25 µm. (D,E) In a double-labelling experiment F-actin in the muscle strands of the epithelial sheath was visualized by rhodaminyl-phalloidin (D) and laminin was demonstrated immunologically using streptavidin-DTAF as a detection system (E). The rhodamine and DTAF fluorescence could be viewed separately using the appropriate filters (D and E, respectively). However, the brightly stained F-actin bands were also visible in the DTAF channel (arrowheads; both fluorescent images were not exactly aligned). Note that little laminin is present in the area of the muscle strands. Their rims, however, are strongly labelled and extending from the rims there is periodically stronger staining between the muscle strands, thus giving rise to a pattern resembling rungs of a ladder. Bar, 10 µm.
Fig. 3. (A) Laminin staining in the basement membrane of the follicular epithelium after removal of the epithelial sheath. Near the anterior pole of the follicle (anterior pole to the left in A–D) immunopositive material is organized in circumferential stripes (i.e. perpendicular to the long axis of the follicle). This pattern was characteristic of stage 6–9 follicles and was also observed at the anterior end of stage 10 follicles (A), while at this stage of development at the posterior end the pattern was more diffuse (B). Staining with FITC-labelled concanavalin A (C) gave essentially the same pattern as anti-laminin (compare with A). The stalk cells always stained intensely. At reduced fluorescence intensity a pattern of stripes (parallel to the long axis of the chain of stalk cells) could be seen (inset: laminin-stained preparation). Note filaments radiating out from the base of the stalk cells and the ring around the area of the cell attachment. Bar: (A–C), 10 μm. (D) Stage 9 follicle treated with purified collagenase in R-14 medium for 30 min. The undigested material in the basement membrane forms a network that seems to consist mainly of laminin (stained as in A). The preferential orientation of laminin in circumferential direction is still discernible. Bar, 10 μm.

mostly lost. In some areas parallel laminin fibres could still be recognized, but the direction of the fibres was variable (Fig. 4C).

At stage 10 the MF density increases and a nearly homogeneous layer of F-actin forms below the basal cell membrane (Gutzeit, 1990). The MF bundles also form in kugel follicles and in some cases their density and structure appeared normal. However, the circular orientation typically observed in wild-type follicles was disturbed in the mutant. The difference can be seen best in stage 13 follicles, since in wild-type follicles of this developmental stage the circular MF pattern is very regular (Fig. 4D) and local disturbances concerning the direction of the MFs are rare and mostly restricted to the areas close to the follicle’s poles. In contrast, the spatial orientation of the MFs in kugel follicles was much less regular (Fig. 4E). The degree of abnormality was variable, ranging from large areas comprising many follicle cells in which the normal circular pattern was maintained (but there were always abnormal areas too) to a chaotic organization with regard to the orientation of the MF bundles. Yet within one cell the MF remained mostly parallel.

Neighbouring cells appeared to influence each other to
some degree with regard to the MF orientation, so that groups of cells typically had the same orientation (Fig. 4E). Similarly, the direction of laminin stripes (wherever they could be discerned in the disturbed pattern of the mutant) often remained nearly constant in areas several cell diameters wide (Fig. 4C).

**Discussion**

**Known molecular and biological properties of laminin**

Laminin is a ubiquitous major component of the basement membrane, which consists of three covalently linked polypeptide chains. *Drosophila* and mouse laminin share considerable sequence homology. Electron micrographs show that the large laminin molecule (Mr=850,000) is typically cross-shaped with a long axis of about 105 nm (reviewed by Fessler and Fessler, 1989; Timpl, 1989; Beck et al. 1990). The molecule may self-associate and form large polymers under suitable conditions in vitro (Yurchenco and Schittny, 1990). It is conceivable that the striped laminin pattern results from an oriented deposition of the molecule in the basement membrane.

A number of cellular reactions have been attributed to interactions of laminin with the cell surface. These biological effects of laminin may be instrumental in developmental processes. Laminin has, for example, been implicated in vertebrate morphogenesis (Ekblom et al. 1986; Bard, 1990) and has been shown to affect the organization of the epithelium (see for example, Sugrue and Hay, 1981; Grover et al. 1983). Laminin is deposited prior to collagen IV and glutactin during early embryogenesis in *Drosophila* (Fessler and Fessler, 1989) and is known to self-associate and also to bind to several other constituents of the basement membrane (Yurchenco and Schittny, 1990). Hence the formation and organization of the basement membrane may be initiated by laminin.

**Locally different laminin patterns**

Our results show that the basement membranes of *Drosophila* ovarioles are differentiated with respect to the concentration and distribution of laminin. The staining pattern of the basement membranes of the epithelial sheath differs strongly from that of the basement membrane of the follicle cells. Furthermore, a very different pattern was observed around the attachment site of stalk cells that form a link between neighbouring follicles. It is remarkable that those structures that are
presumably under considerable mechanical stress contain high concentrations of laminin. The epithelial sheet and its basement membranes become strongly deformed during muscular contraction, which can be seen to proceed rhythmically in anteroposterior direction over the ovariole in vitro. Also, the stalk cells may become stretched during these contraction cycles when follicles become temporarily displaced in the ovariole as was observed by video microscopy of cultured ovarioles (A. Haas-Assenbaum and Gutzeit, unpublished). It is, therefore, conceivable that the laminin pattern is determined by the requirements for mechanical stability. On the basis of circumstantial evidence, Bard (1990) states that the ‘extracellular matrix can be organized over long distances by the tractional forces exerted by cells.’

If the intrafollicular transport of macromolecules and organelles from the nurse cells to the oocyte is based on a pressure difference between these cells, as was suggested earlier (Gutzeit, 1986c), the basement membrane might limit the expansion of the nurse cells (up to stage 10A) that are thought to be under higher hydrostatic pressure than the oocyte. The observed circular laminin pattern might, therefore, be interpreted as a structural adaptation to resist expansion of the nurse cells. At later developmental stages (stages 10B-12) this function is no longer required as the nurse cells regress, presumably due to the contraction of microfilament bundles in the nurse cells (Gutzeit, 1986).

The observed local differences in the laminin content and organization lend indirect support for the polymorphic polymerization model (Furthmayr, 1988). Differences in the basement membrane composition have been noticed in other systems (Linsenmayer et al., 1984).

Laminin–microfilament interactions

Our observations on wild-type and kugel follicles suggest that the spatial organization of laminin and F-actin is coordinated. The molecular mechanisms of the inferred interaction between laminin and cortical actin are not fully understood. Both molecules are located in close proximity to the cell membrane. The MF bundles are closely attached to the cell membrane on the basal side of the follicle cells (Gutzeit, 1990). High-resolution immunoelectron microscopy (Schittny et al. 1989) revealed that in the basement membrane of the mouse cornea the center of the laminin molecule is located near the lamina lucida/ lamina densa border. Some domains may extend into the lamina densa while others transverse the lamina lucida and presumably make contact with the epithelial cell surface. A periodicity in the laminin distribution was not observed. Likely candidates for providing the inferred molecular link between laminin and the cytoskeleton of the follicle cells are the integrins, a family of membrane glycoproteins (reviewed by Ruoslahti and Giancotti, 1989).

The basal MF pattern and the distribution of laminin in kugel follicles suggests that neighbouring follicle cells influence each other with regard to the orientation of the respective molecules. Within one group of cells the MF orientation may be (nearly) the same, while between different groups of cells the orientation may vary. Local interactions are thought to synchronize the cellular orientation in, for example, fibroblasts and the insect epidermis (Albrecht-Buehler, 1979; Nübler-Jung, 1987). The biological and biochemical properties of laminin suggest that the molecule may be capable of organizing the basal cytoskeleton of epithelial cells in concert with other matrix proteins. For example, human skin fibroblasts readily attach to laminin-coated Petri dishes and form focal adhesions with associated MF bundles (Couchman et al. 1983). With the techniques employed we could not decide if MFs or laminin were first oriented circumferentially. Presumably, the basal cytoskeleton and different components interact dynamically during oogenesis. Laminin is unlikely to specify the organization of the basal MFs alone, since the laminin pattern in the basement membrane covering the columnar follicle cells (stage 10–14) became diffuse at a time when the number of circular MF bundles increased, thus forming a rather homogeneous layer below the basal cell membrane.

Is the follicular shape determined by the organization of laminin?

There is ample evidence that components of the extra- cellular matrix (including laminin) modulate cell shape (Watt, 1986). For example, anti-laminin antibodies injected into sea urchin embryos led to changes in cell shape and deformation of the embryonic epithelium (McCarthy and Burger, 1987). Recently, we have obtained evidence that the normal shape of the Drosophila follicle critically depends on the integrity of the basement membrane (Gutzeit and Haas-Assenbaum, 1991). The present observations on the abnormal patterns of F-actin and laminin in follicles of the egg-shape mutant kugel suggest a functional role for these molecules in shaping the follicle.

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References


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