of the bubble meniscus. Fig. 2 illustrates a 1.0 cm³ bubble viewed at the plane of the meniscus. In this position the meniscus appears as a thin dark line with minimal curve just visible in the pupillary aperture. If the angle of observation is lowered, less than 5 degrees, one sees progressively more of the base of the bubble (Fig. 2). Viewed through the pupil, the meniscus now has an elliptical shape, making the bubble appear to fill the superior third of the pupil, simulating a 2.0 cm³ bubble.

**Discussion.** With the use of a transparent model to demonstrate the configuration of gas bubbles in the vitreous cavity and a mathematical model to calculate bubble volumes, it is clear that small volumes of gas produce large arcs of bubble-retina contact. When this is applied to retinal detachment surgery, it becomes apparent that a great number of the problems for which an intraocular tamponade would be useful could be managed with relatively small quantities of gas. For example, in the average eye a 0.28 cm³ bubble will cover a 90-degree area of retina and be sufficient to press out undesirable folds in the retina, manage a fish-mouthing retinal hole, or simply tamponade a retinal break that is not responding to scleral buckling. Larger tears require disproportionately large increments in volume. A 90-degree tear would be just covered by a 0.28 cm³ bubble. To cover a 180-degree tear would require a 8.6-fold increase in bubble volume to 2.4 cc. To obtain a minimal overlap of 15 degrees to either side of the tear would necessitate a further increase in volume to 3.3 cm³, over two thirds of the entire vitreous cavity volume.

Estimating residual bubble size in the patient is clinically useful. Observations in the model eye have shown that small shifts in the angle of observation can produce significant changes in the level of the bubble meniscus. A 1.0 cm³ bubble can be made to simulate a 2.0 cm³ bubble simply by changing one’s angle of observation (Fig. 2). A correct evaluation of intraocular bubble size requires that the plane of observation be adjusted so that it coincides with the plane of the meniscus. Practically, this is obtained by adjusting one’s level of observation to the point where the meniscus is a thin dark line with minimal curve.


**Key words:** intraocular gas, retinal detachments, intraocular gas tamponade, estimating intraocular bubble size, retinal tamponade, intraocular volume-area relationship

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**The fine structure of nuclear changes in superior limbic keratoconjunctivitis.** H. BARRY COLLIN, PETER C. DONSHIK, S. ARTHUR BORUCHOFF, C. STEPHEN FOSTER, AND H. DWIGHT CAVANAGH.

**Superior limbic keratoconjunctivitis (SLK) is a condition of unknown etiology. Histological signs include acanthosis, dyskeratosis, keratinization, and balloon degeneration of the bulbar conjunctival epithelium. Ultrastructural examination of biopsy material from five eyes of patients with SLK shows numerous nuclear changes in the conjunctival epithelial cells. These include abnormal distribution and aggregation of nuclear chromatin, the presence of filaments within the nucleus, and dense accumulations of cytoplasmic filaments which surround the nucleus, resulting in “strangulation” and the formation of multilobed nuclei or multinucleated cells. These changes do not appear to have been described previously in any cell type.**

The clinical and histological characteristics of superior limbic keratoconjunctivitis (SLK) in man have been reported. However, there is only one report of the ultrastructural features which accompany this condition and in that publication the changes in the nuclei of the bulbar conjunctival epithelial cells were not described. Apart from the degenerative changes there was brief reference only to abnormal chromatin distribution and tortuosity of the nuclear envelope.

In this study, unusual ultrastructural appear-
Fig. 1. A, Part of an epithelial cell from a patient with SLK. The dark-staining nuclear chromatin is aligned along the long axis (between arrows) of the nucleus (N). The black cytoplasmic material is composed of aggregations of glycogen granules. (Patient 1; OPF; ×6,700.)

B, Part of a bulbar conjunctival epithelial cell from a patient with SLK. The nucleus (N) has an area of tortuous nuclear membrane and contains a large nucleolus (Nu) and several dense accumulations of dark-staining nuclear chromatin (arrows). (Patient 2; OPF; ×15,600.)
Fig. 2. A, Nucleus of a conjunctival epithelial cell from a patient with SLK. There are two large nuclei (N) and several dark-staining, smaller, membrane-bound nuclear fragments. A large dense bundle and several small bundles of filaments (large arrows) separate the various nuclei, giving the appearance that the nucleus had been strangulated into smaller pieces. There are two small, round keratohyalin granules (small arrows) in the cytoplasm. (Patient 3; normal osmium; x12,600.) B, Strangulated nucleus (N) from the bulbar conjunctival epithelium of a patient with SLK. The central compressed region is apparently surrounded by tight bundles of cytoplasmic filaments. (Patient 2; OPF; x21,600.)
Fig. 3. Larger area of the same region of bulbar conjunctiva shown in Fig. 2. B. In addition to the same abnormal cell nucleus (3), there is another strangulated nucleus (2) similar in appearance and at about the same stage of abnormal development. Another pair of nuclei (1 and 4) are also abnormal and at a similar stage of strangulation. Filament bundles are present adjacent to all four nuclei (small arrows). One large keratohyalin granule (large arrow) is also present. Glycogen granules (black) are abundant in the cytoplasm. (Patient 2; OPF; ×8,400.)
Junctiva of all patients, with SLK showed marked abnormalities. These nuclear abnormalities have not been reported previously.

Methods. A block of bulbar conjunctival tissue extending from 10:30 to 1:30 at the limbus and posteriorly 3 to 4 mm was surgically resected from one eye of each of three patients and from both eyes of a fourth patient with SLK. Approximately 2.5 mm of upper tarsal conjunctiva was also removed from one of the same patients. Since the case histories and light and preliminary electron microscopic findings have already been reported, the clinical and histological features of SLK will not be repeated here.

Controls consisted of normal superior limbal bulbar conjunctival tissue from two patients who underwent cataract surgery and of upper tarsal conjunctiva from one patient whose conjunctiva appeared normal.

The methods of preparation of tissue for electron microscopy by means of normal osmium and potassium ferrocyanide (OPF) procedures have been described previously.

Results. The epithelium from the limbal conjunctiva of all patients with SLK showed marked degenerative changes and signs of keratinization. Extensive abnormal accumulations of glycogen were prominent (Fig. 1, A and B). In some degenerating cells the nuclei appeared empty and were undergoing balloon degeneration. In other areas of apparently healthy epithelial cells, all the nuclei showed a central spindle-shaped alignment of the dark-staining nuclear chromatin (Fig. 1, A). This alignment of nuclear chromatin was present in all five eyes from patients with SLK, as also was the presence of keratohyalin granules in both nuclei and cytoplasm of epithelial cells.

Epithelial nuclei from three of the four patients—those who had not received recent treatment with silver nitrate—showed dark-staining chromatin which was aggregated into round or oval masses or “swirls” (Fig. 1, B). Some of these masses seemed to be composed of tightly packed fine filaments 10 to 15 nm in diameter.

Accumulations of cytoplasmic filaments were common in cells in the process of keratinization. These were sometimes in the form of large, dense masses close to the nuclei. In some cells the nuclei were multiple, with up to six membrane-bound portions, and dense bundles of filaments were usually present separating the various nuclei (Fig. 2, A).

In cells where the separation of the nuclei into multiple, smaller, membrane-bound nuclear fragments seemed incomplete, dense masses or tightly packed bundles of filaments appeared to be encircling the nuclei, “strangulating” them into “dumb-bell” shapes and into smaller nuclear units (Fig. 2, A and B). These strangulated cell nuclei were found in two of the four patients, neither of whom had been treated with silver nitrate. The cytoplasm of these cells had accumulations of glycogen granules and showed early signs of keratinization, including filament aggregations and keratohyalin granules (Figs. 2, A, and 3).

The strangulated cells always occurred in groups of three or four in one section, and the abnormal nuclei in such a group of cells were usually at a similar stage of abnormal development. Fig. 3 shows two pairs of aberrant cells. Within each pair the nuclear changes are almost identical, although the two pairs differ greatly. The two cells from each pair appear to have been derived from the same parent cell.

In some instances similar filaments were present within the abnormal nuclei and appeared to be continuous with the bundles of cytoplasmic filaments.

Nuclear changes of the type described were not observed in the control tissue from the limbal and upper tarsal conjunctiva of patients without signs of SLK or from the upper tarsus of one patient with SLK.

Discussion. Histological changes occurring in the conjunctival epithelial cells in SLK have been described with the use of both light and electron microscopy. There is evidence of acanthosis, dyskeratosis, keratinization, and balloon degeneration. However, the nuclear changes reported here have not been documented previously in SLK or in either normal or pathological conjunctival epithelium. In fact, they do not appear to have been described in any cell.

Nuclear abnormalities were observed in tissue from all five eyes from patients with SLK. However, the strangulated nuclei were found in only two eyes from two patients. Although this may appear to be a low incidence, it should be pointed out that two of the eyes which did not show nuclei with a strangulated appearance had been treated relatively recently, that is, 13 and 64 days previously with 0.5% silver nitrate. Advanced signs of keratinization were present in all five eyes but were considerably less marked in those same two silver nitrate-treated eyes.

An increase in cytoplasmic filaments is known to occur during keratinization of epithelial cells in man, and on occasions the filaments are formed into heavy bundles. However, the large, dense mass of filaments described in this report should be considered as a distinctive feature of SLK.
masses of cytoplasmic filaments associated with the epithelial cell nuclei and described here in SLK do not appear to have been reported elsewhere.

Binucleated or multinucleated cells or multilobulated nuclei do not appear to have been reported in normal conjunctival epithelium. Similarly, they are rare in pathological conjunctival tissue. Takakusaki described only one binucleate cell in the palpebral conjunctival epithelium from a patient with vernal conjunctivitis. In the corneal epithelium, which is continuous with that of the conjunctiva, the nuclei from a patient with preinvasive cancer were frequently enlarged and sometimes appeared to be multiple, although serial sections showed that these were more often multilobulated single nuclei. However, there is little evidence that SLK is neoplastic.

Strangulation of the nuclei into dumb-bell shapes appears to be due to the presence and/or contraction of the bundles of filaments surrounding the nuclei. The possibility exists that these nuclear changes represent an abnormality of mitosis.

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