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## Equator-plus camera. OLEG POMERANTZEFF.

A new fundus camera can photograph a field extending anterior to the equator (148° from the nodal point). The problems of reflection from the surfaces of the crystalline lens are minimized by fiber optic illumination from carefully chosen locations on the cornea. The camera may be used with monochromatic light of various wavelengths and is equipped with interference filters.

This camera can photograph a retinal field that extends anterior to the equator. The field covered by this camera extends 148° from the nodal point, that is, up to half the distance between the equator and the ora serrata.

However, the largest field obtained to date with no contact between the camera objective and the patient's cornea is about  $50^{\circ}$ . Thus, for any field larger than  $50^{\circ}$ , presently available optical means require contact between the objective and the patient's eye. This is, of course, an inconvenience. Since this is a costly means of increasing the field, it has to be counterbalanced by a significant gain. Any contact camera with a field greater than  $50^{\circ}$  that does not include at least the equator would probably not be worth the expense. This camera would show little more



Fig. 1. The marginal rays from the edge of the field of  $148^{\circ}$  are traced by computer through the wide-angle model of the eye. Rays that are accepted by the entrance pupil at the front surface of the crystalline lens emerge from the cornea at 4 mm. separation from the axis. The divergence of these rays in air is reduced by using a high index contact lens. The central part of the cornea (vertical cross-hatching) is used for observation, the peripheral part for illumination.



Fig. 2. The illumination of the fundus is provided by optical fibers located at the peripheral part of the cornea (cross-hatching at  $45^{\circ}$ ). The entrance pupil must be preserved from direct illumination. The field illuminated is smaller than the field that can be seen (cross-hatchings on the retina).



Fig. 3. The central haze in the picture is produced by reflection of illuminating light impinging directly on the entrance pupil.

than the posterior pole, which is easily photographed with the noncontact cameras. In addition, the increased field would be seen with less contrast and less magnification.

A camera that extends the field anterior to the equator, besides showing pathologic areas of the posterior pole, also shows those very important traumatic and degenerative lesions that are located mostly in the equatorial area. Such a camera could be used for preparing a map of the fundus, and minimizing the time and discomfort encountered in drawing with the indirect oph-thalmoscope. It could also be used for very quick screening and recording. This would bring a new dimension to photography and ophthalmoscopy and probably transform the conventional procedures of ophthalmoscopy. However, the first and *sine qua non* condition is that the camera or ophthalmoscope bring into view an equator-plus field, ideally up to the ora serrata.

Theoretical discussion. The theoretical implications of this problem were described in 1971.<sup>1</sup> Volume 14 Number 5



Fig. 4. Increased illuminated field by using two illuminating fiber bundles, one for the central field and one for the periphery, at different locations on the cornea and different inclinations of incidence.

Chief among them is the problem of reflectionless illumination of the entire field.

Fig. 1 shows the marginal rays from a field of  $148^{\circ}$ , as traced by a computer through a wideangle model of the eye.<sup>2</sup> Three points are to be noted: (1) in air, the marginal rays emerge tangentially, and they cannot be focused by conventional optics into an observable image. In highindex glass, they form a smaller angle and can be brought to focus more easily. Consequently, the contact lens must be made of high-index glass. (2) The entrance pupil of the observation system must be small to obtain a good depth of focus and minimize geometric aberrations. (3) The entrance pupil must be located at the front surface of the crystalline lens. The marginal rays from a field of 148° that cross this small entrance pupil emerge from the cornea at approximately 4 mm. from its axis. If the entrance pupil is located posterior to the front surface of the crystalline lens, the marginal rays that cross the entrance pupil emerge more peripherally on the cornea. If the entrance pupil is located anterior to the same surface, the marginal rays of the field under consideration are not accepted by this small entrance pupil. In other words, the field of view is reduced.

Fig. 1 indicates that the corneal area (cross-

hatched vertically) must be left unobstructed for the observation of the fundus. Only the peripheral area (cross-hatched at  $45^{\circ}$ ) is free for introducing the illuminating light.

Fig. 2 shows the illumination system. Two points are important: (1) from the preceding discussion, it follows that the optical fibers that provide illumination cannot be located closer than



Fig. 5. The equator is located on the picture by the location of the vortex ampulae (white arrows).

4 mm, to the axis of the eye. The patient's pupil must be at least 7.5 mm. in diameter to allow the rightmost marginal illuminating ray, which touches the right edge of the pupil, to reach the retina on the right side of the axis of the eye. If the patient's pupil is smaller, this ray will reach the retina on the left side of the axis and the posterior pole will not be illuminated. (2) In order to avoid reflections from the crystalline lens, the entrance pupil, marked by vertical crosshatching, should be left free of direct light. Therefore, the left marginal ray of this bundle will, at most, touch the right edge at the entrance pupil. The total retinal area on the left side of the field that is illuminated by this bundle is marked by the cross-hatching inclined to the right. Symmetrically, the bundle from the opposite side illuminates the right part of the field marked by the cross-hatching inclined 45° to the left.

However, only a relatively small field (approximately 70° to 90°) can be illuminated in this way. Increasing the inclination of the fiber bundles on the cornea or increasing their numerical aperture would increase the illuminated field in the periphery. However, it would result in producing reflections from the crystalline lens that cover the central part of the field with a haze, as shown in Fig. 3.

This problem, which at first appeared extremely difficult, has been overcome by placing another ring of illuminating fibers more peripherally using



Fig. 6. Equator plus camera with safety device limiting the pressure of the contact lens on the patient's eye.



Fig. 7. Picture of the fundus in yellow light showing retinal circulation in high contrast.



Fig. 8. Picture of the fundus in red light showing the choroidal circulation.

another angle of incidence and another numerical aperture as shown in Fig. 4.

This arrangement avoids the central "veiling" of the field and permits reflectionless pictures with a field of view extending anterior to the equator as shown in Fig. 5. The equator can be seen in the photograph by the location of the vortex ampulae (marked by arrows).

The equator-plus camera is shown in Fig. 6. Its mechanical parts were designed by Mr. Robert Thompson, a mechanical designer at the Eyc Research Institute, and machined in our Instrument Shop by Mr. George Bearse and Mr. Frank Hughes. One technical detail worth mentioning is that the contact lens at the tip of the camera



Fig. 9. Picture of the fundus in combined yellow and red light showing at the same time retinal and choroidal circulations.

has a very smooth axial sliding motion. This mechanism is spring-loaded with a constant force spring calculated to exert a maximum pressure on the eye of 20 mm. Hg. A buzzer indicates if this pressure is exceeded. When the media are clear, the buzzer is not needed. However, if the media present opacities, the operator may be tempted to push the camera forward and possibly exceed the permitted pressure.

The camera is equipped with interference filters, which are used to better visualize different structures in the fundus. As an example, Fig. 7 shows a fundus in yellow light and Fig. 8 shows the same fundus in red light. It is remarkable that in red light, the retinal circulation, which is revealed in high contrast by yellow light, disappears almost completely, while the choroidal circulation is highly visible. A combination of these two wavelengths renders both circulatory systems relatively visible (Fig. 9).

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## Subretinal fluids: lipid analyses. K. W. LAM, W. A. J. VAN HEUVEN, G. STEWART RAY, AND STEPHEN FEMAN.

Lipid was found to accumulate in the subretinal space during rhegmatogenous retinal detachment. The concentration of lipid in the subretinal fluid varied between 0.1 to 2.4 mg. per milliliter. Based upon a comparison of lipid profiles and lipoprotein profiles, the amount of lipid observed in the subretinal fluids was not directly related to the permeability of the ocular vessels. Thinlayer chromatography analyses of lipid in subretinal fluids showed differences between the lipid composition of the subretinal fluid and blood. The major characteristic of lipid composition of subretinal fluid is the low concentration of lecithin, which is the major lipid compound in the serum. The present data indicate that the lipids of ocular tissues are released into the subretinal space during rhegmatogenous retinal detachment, and are adsorbed on albumin.

In the past, most biochemical analyses of subretinal fluids dealt with proteins and carbohydrate compounds, derived from the blood and vitreous.1 Recently, there has been a developing interest in the lipid content of subretinal fluid; this is because lipid is a major constituent of the outer segments of the light receptors which are adjacent to the subretinal space. Furthermore, histologic studies have shown that there is a degradation of these cells during rhegmatogenous retinal detachment.<sup>2, 3</sup> It is reasonable to speculate that a significant amount of retinal lipid is released into the subretinal space as a result of these morphological changes of the outer segments. This study is concerned with subretinal fluid lipid analyses; and the results are discussed with respect to the biochemical pathology of retinal detachment.

## Methods.

Collection of samples. Subretinal fluid was collected in a syringe attached to a short flexible catheter held over the choroidal perforation site during retinal detachment repair surgery. Particular care was taken to avoid contamination with blood or with irrigating solutions. At the same time 5 ml. of venous blood was collected.

When a sample was collected, it was divided for the following analyses: (1) ten microliters were used for counting red blood cells in a hemocytometer in order to estimate the amount of blood contamination. If the red blood cell count exceeded  $2 \times 10^3$  cells per cubic millimeter the sample was discarded. (2) Ten microliters were taken for protein analyses by the method of Lowry and co-workers.<sup>4</sup> (3) Another 0.5 µl were used for electrophoretic analyses of protein or for lipoprotein profiles. (4) The remaining sample was used for lipid analyses.

Agarose electrophoresis. Universal agarose films supplied by Fisher Scientific Company were used in this study. The agarose films consisted of 1 per cent agarose in a buffer containing 0.035 per cent disodium ethylenediaminetetraacetate, and 0.05 M barbital, pH 8.6. Samples of 0.5 µl were applied to each agarose film. The samples were subjected to electrophoresis for 30 minutes (3 milliamperes per sample). At the end of electrophoresis, the films were submerged in 5 per cent trichloroacetic acid for 15 minutes. The samples were stained with Amido black (0.5 per cent) to visualize the protein bands, or stained for lipoproteins with Oil Red O, as described by Chin and Blankenhorn<sup>5</sup> and as specified in the legend to the figure.

Lipid extraction. Lipid was extracted by the method of Bragdon.<sup>6</sup> Samples of 0.25 ml. were mixed with 5.5 ml. of chloroform-methanol (2:1, v/v), diluted to 6.25 ml. with the same solvent, mixed with 1.25 ml. of 0.02 N sulfuric acid by inversion, and centrifuged at 1,000 g for 10 minutes. In each case, the lower lipid-containing layer was used either for total lipid analysis, or for thin-layer chromatography.

Total lipid analyses. An aliquot of 0.5 ml. of each lipid extract was evaporated to dryness under nitrogen. Each of the resulting residues was dissolved in 1 ml. of potassium dichromate (68 mM dichromate dissolved in concentrated sulfuric acid) and heated at 100° C. for 30 minutes. Then, each one was diluted with water to 2.5 ml. at room temperature. The absorbance at 580 nm. was determined within one hour. Tripalmitin was used to produce the standard curve.<sup>6</sup>

Thin-layer chromatography. Each of the lipid residues obtained by the extraction of 0.25 ml. of subretinal fluid or serum was dissolved in 50  $\mu$ l of ethyl ether. One-fifth of each sample was transferred to a silica gel plate (Kodak Chromagram sheet). Solvent A was a mixture of hexaneether-acetic acid (80:20:1); and the lipid spots were stained with iodine vapor. Solvent B was a mixture of chloroform-methanol-water (65:25:4); and the phospholipid spots were also stained with iodine vapor.<sup>7</sup>

Results. The tabulation of protein and lipid concentrations for 20 samples is shown in Table I.