

Subretinal fluid ferning test in rhegmatogenous retinal detachment

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PURPOSE. *The ferning test involves a process of crystallization achieved simply by removing water and is feasible for all ocular fluids. The ferning test of subretinal fluid (SRF) from patients with rhegmatogenous retinal detachment (RRD) reveals three different patterns: type 1 showing thin crystals, type 2 with larger crystals and type 3 with small, curvilinear structures with no tree-like appearance. The present study was designed to determine whether the SRF ferning test is correlated with the clinical features and the surgical outcome of RRD.*

METHODS. *A series of 65 consecutive patients with RRD at the first onset were considered. Particular attention was paid to duration, extension, and surgical outcome of RRD. SRF samples were collected during scleral buckling surgery. The fluid was dropped onto the slide of a light microscope, left to dry, and examined under the microscope.*

RESULTS. *There was a significant difference between SRF ferning types 1 and 2 as regards duration and extension (both $p < 0.001$). There was also a significant difference between SRF ferning types 2 and 3 as regards duration ($p < 0.001$), extension ($p < 0.001$), and surgical outcome ($p < 0.05$).*

CONCLUSIONS. *The ferning test of SRF is quick and simple and can be regarded as a useful tool for obtaining information about clinical features, such as duration or extension of first onset in uncomplicated cases of RRD especially for purposes of forensic medicine. (Eur J Ophthalmol 2001; 11: 156-9)*

KEY WORDS. *Subretinal fluid, Rhegmatogenous retinal detachment, Ferning test*

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INTRODUCTION

The chemical composition of subretinal fluid (SRF) from patients with rhegmatogenous retinal detachment (RRD) has been correlated with their clinical characteristics: in particular, the longer the duration of RRD, the higher the concentration of SRF proteins (1-5).

The ferning test is based on a process of crystallization by simple water extraction, feasible for all ocular fluids (6). The ferning test of SRF in RRD reveals three different patterns, and a preliminary study suggested a link between these crystallization patterns and clinical aspects (7).

The present study aimed to determine whether the SRF ferning test is correlated with the clinical features and the surgical outcome of RRD.

METHODS

A series of 65 consecutive patients referred to our center with RRD was considered. The inclusion criterion was a diagnosis of RRD at the first onset. Exclusion criteria were macular holes, proliferative vitreoretinopathy, giant retinal tears, vitreous hemorrhage, previous vitrectomy, cryopexy and laser photocoagulation.

Particular attention was paid to the duration and extension of RRD. The duration was expressed in days, from the first symptoms. The extension was indicated as the number of quadrants involved. Surgical success was judged on the basis of complete retinal reattachment after scleral buckling.

SRF samples were collected during scleral buckling surgery, with transscleral diathermy to avoid bleeding; the choroid was penetrated using a blunt needle. SRF was collected with a blunt-tip aspiration needle, avoiding blood contamination. The fluid was dropped onto a light microscope slide and left to dry at room temperature (20 °C, 50% humidity) then it was examined in the light microscope at 100x magnification. The SRF ferning pattern was classified as three different types: type 1 showing thin, linear crystals with small empty spaces between them (Fig. 1); type 2 with larger crystals and variable empty spaces (Fig. 2); type 3, with small, thin, curvilinear crystals without tree-like appearance (6) (Fig. 3).

The SRF ferning test patterns were interpreted in masked fashion by two of us (MBP, SS); agreement was achieved in 92% of cases, and a third author (GR) was consulted to define uncertain cases.

Patients were informed of the purpose of the study and provided informed consent. Statistical analyses were done using Student's t test and the chi-square test.

RESULTS

The Table I summarizes the results. We obtained SRF samples from 65 patients, 41 males and 24 females, mean age 62.6 ± 9.8 SD.

TABLE I - CORRELATIONS BETWEEN SRF FERNING TEST PATTERNS, DURATION, EXTENSION AND POSITIVE SURGICAL OUTCOME OF RRD

Ferning Test type	Duration	Extension	Surgical success
1	10.9	1.5	12/16 (75%)
2	20.8	2.3	28/39 (71.8%)
3	48.8	3.2	3/10 (30%)

In 16 cases in which we detected the type 1 ferning pattern the mean duration of RRD was 10.9 ± 2.4 days, with a mean of 1.5 ± 0.5 quadrants involved and a positive surgical outcome in 12 patients. The cause of surgical failure in the remaining four cases was missed retinal breaks.

In the 39 cases with type 2 ferning pattern the mean duration was 20.8 ± 15.6 days, with a mean value of quadrants 2.3 ± 0.6 involved and positive surgical outcome in 28 cases. Surgery failed because of missed retinal breaks in six cases, development of proliferative vitreoretinopathy in four and inadequate scleral buckling in one.

In the 10 cases with type 3 ferning pattern, the mean duration was 48.8 ± 8.5 days with mean extension 3.2 ± 0.5 and positive surgical outcome in three cases (30%). Unsuccessful surgery depended on missed retinal breaks in four cases, development of proliferative vitreoretinopathy in two, and inadequate scleral buckling in one.

Statistical analysis showed significant differences ($p < 0.001$) between SRF ferning test types 1 and 2 as regards duration and extension. A significant difference was also seen between SRF ferning test types 2 and 3 as regards duration and extension ($p < 0.001$), and surgical outcome ($p < 0.05$). No correlation was found with the development of proliferative vitreoretinopathy.

DISCUSSION

The chemical composition of SRF in RRD is extraordinarily complex and variable. Vitreous, retina, and choroid contribute to its composition, each of these structures being impaired to varying degrees during the course of RRD (4). Weve and Fischer, in 1940, were the first to suggest that SRF might provide useful information about pathogenesis and prognosis in RRD (8).

The ferning test can be regarded as a process of crystallization obtained simply by removing the water and reveals three different patterns in SRF of RRD (6). Assuming that the ferning test reflects the different composition of SRF in RRD, the present study set out to correlate the test findings with the clinical characteristics and surgical prognosis of RRD.

Several factors may affect the outcome of scleral



Fig. 1 - Ferning test type 1 (100x).



Fig. 2 - Ferning test type 2 (100x).

buckling surgery, including the development of proliferative vitreoretinopathy, extension of RRD, size, number and type of retinal tears, previous vitrectomy, cryopexy or laser treatment, degree of vitreous traction (9). An important feature is the retinal adhesion capacity, which is influenced by other elements such as pressure on the retina from the vitreous gel, interdigitations between outer segments and retinal pigment epithelium microvilli, metabolic activity of retinal pigment epithelium, and the progressive breakdown of the interphotoreceptor matrix, whose constituents contribute to the composition of SRF (10-14).

We studied a selected group of patients, excluding complicated types of RRD, in order to analyze better the relationship between SRF and the clinical characteristics and surgical outcome. Our results indicate a link between the SRF ferning test patterns and the duration and extension of RRD. Ferning test type 3, detected in cases with longer duration and greater extension was often associated with a bad surgical outcome. No clear correlation was found between the development of proliferative vitreoretinopathy and ferning test.

Research regarding the structural composition of tear ferns indicates that the fern crystals are mainly sodium chloride and potassium chloride, and protein may be deposited on the salt crystals, controlling the crystallization indirectly by coating the crystal faces and blocking extension of the fern (15). A previous study reproducing an experimental model of SRF combining three main chemical compounds, hyaluronic

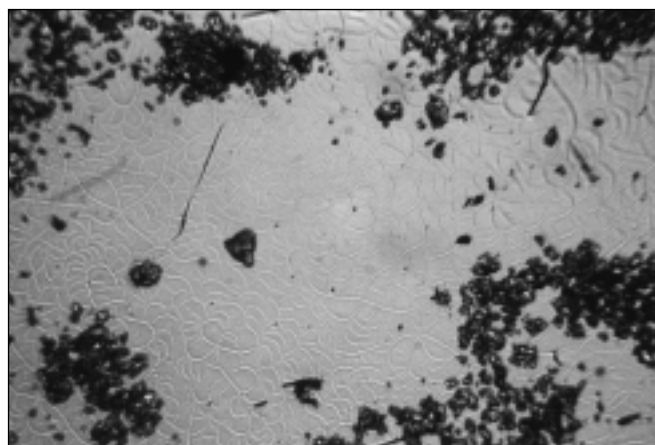


Fig. 3 - Ferning test type 3 (100x).

acid, albumin, and sodium chloride, at different concentrations, indicated that it is the increasing protein concentration that governs the passage from type 1 to type 2, up to type 3 ferning pattern, type 3 having the highest protein concentration (16).

We suggest that the thin curvilinear crystals, typical of ferning test type 3, with their lack of tree-like pattern reflects the higher protein concentration of the SRF samples collected during the scleral buckling procedure. The relationship between ferning test type 3 and the clinical characteristics and surgical outcome may be explained in the light of previous reports of a correlation between long-standing RRD, higher SRF protein concentration, and an unsuccessful surgical outcome (3-5).

In our experience the SRF ferning test is a quick

and simple examination, and can give useful information, especially for forensic medicine, about clinical features such as duration of disease, extension of first onset uncomplicated cases of RRD. As regards the surgical outcome, even though the ferning test type 3 is associated with a statistically significant proportion of failed surgery, we do not believe it is a reliable means of obtaining information about surgery. Scleral buckling can fail for several reasons, including the development of proliferative vitreoretinopa-

thy, missing or incomplete closure of retinal breaks, or inadequate surgical technique. The full clinical significance of SRF ferning test findings in different types of retinal detachments awaits further study.

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