

Transmission electron microscopy of the vitreous body tissue in chronic hemophthalmos

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ABSTRACT: Haemolytic products arising in chronic hemophthalmos cause cellular infiltration, necrosis of the vitreous structure, and fibrous membrane formation. In this process, retinal pigment epithelium plays an important role for its antioxidant properties and the capability to phagocyte the decay products.

Keywords: retinal pigment epithelium; proliferative vitreoretinopathy

The vitreous body is a bradytrophic tissue. Under physiological conditions, it primarily contains a feltwork of collagen fibres, permeated by mucopolysaccharides, sporadic hyalocytes and up to 80% of water. The vitreous body has a gel-like consistence. Clinical experience shows that prolonged presence of erythrocytes and erythrocyte haemolytic-decay products, containing predominantly trivalent ferric ions, has toxic effects on the vitreous body and on the retina (Ballinger et al., 1999). The above-mentioned effect is manifested as the loosening of the vitreous fibrillar network, cellular infiltration and, macroscopically, as liquefaction of the vitreous body. Morphologically, this reaction is manifested as inflammatory cellular infiltration, connective tissue proliferation, and as biochemical changes in the vitreous body and retina. Later on, cicatricial processes develop in which fibrous strips form. Epiretinal membranes, which negatively affect the vitreous body transparency and cause traction detachment, play an essential part in numerous complications of proliferating retinopathies, uveitides, vessel occlusions, traumas, primary detachments, and excessive application of laser coagulations. Moreover, they are a dominating factor in the aetiology of chronic hemophthalmos. Vitrectomy enabled the examination of the vitreous body ultrastructure and biochemical

examination *in vivo*. In our case, the ultrastructure of the vitreous body ultrastructure was examined in a patient with hemophthalmos lasting for several months.

MATERIAL AND METHODS

A patient aged 65 with right-eye hemophthalmos lasting 6 months was referred to our clinic. The visual acuity of the right eye equalled to light perception, with faulty light projection. On the left eye, the visual acuity was 6/6. Ultrasound examination showed beginning cicatricial changes in the vitreous body, the retina was not detached. The patient was operated on using pars plana vitrectomy, and the material obtained was used for examinations after thickening with ultracentrifugation. The tissues were fixated in 2.5% glutaraldehyde solution in phosphate buffer for 3 hours and post-fixated in 2% solution of osmium tetroxide. After dehydration in rising ethylalcohol concentrations, the specimens were embedded into Durcupan ACM Fluca. Thin and ultra-thin sections were prepared on the ultramicrotome Reichert, Jung Ultracut E. After fixation with uranyl acetate and lead citrate, the ultrathin sections were examined with the FEI Morgagni transmission electron microscope.

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RESULTS

The specimen contained, first and foremost, a large quantity of erythrocytes in various stages of haemolysis (Figure 1). In addition, we found fragments of collagen fibres, residuals of blood clots, and pigment cells containing numerous lysosomes and cytoplasmic organelles (Figure 2). The finding was indicative of high metabolic activity of pigment cells. In a part of the fibrous membrane, we also observed fibroblast-like cells, containing numerous mitochondria, cisterns of endoplasmic reticulum and ribosomes. Here and there, ingested foreign particles could be seen (Figure 3).

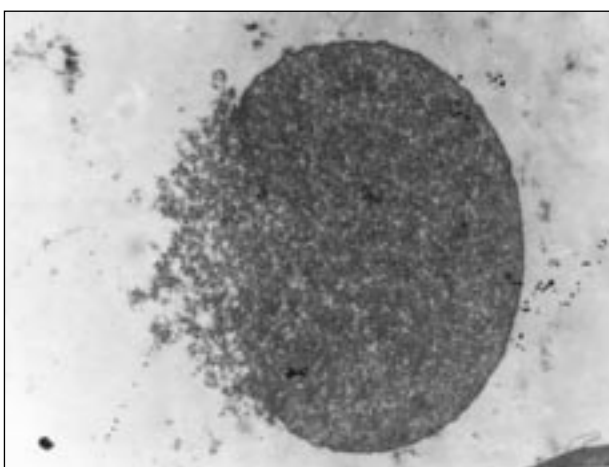


Figure 1. Erythrocyte with haemolytic cellular membrane. Magnified about 15 000×

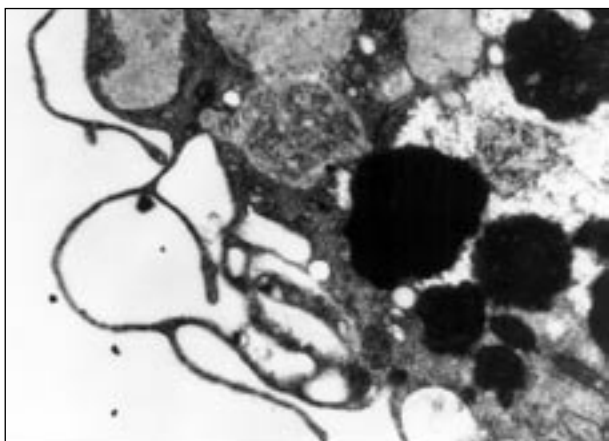


Figure 2. Detail of a pigment cell from the superficial part of the membrane. The cytoplasm contains numerous cisterns of granular endoplasmic reticulum, ribosomes and mitochondria. Conspicuous parts of the cytoplasm are large phagosomes containing haemosiderin. Magnified about 20 000×

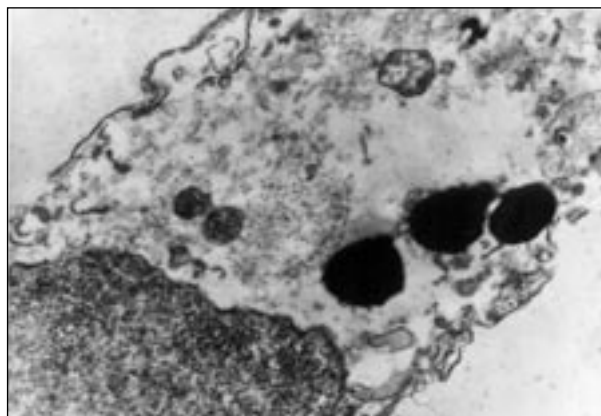


Figure 3. Detail of fibroblast-like cell, containing numerous organelles and ingested foreign particles are seen. Magnified about 23 500×

DISCUSSION

Proliferative vitreoretinopathy (PVR) stands for the formation of fibrous membranes pre- and retro-retinally and in the vitreous body. It begins as cellular infiltration; retinal pigment epithelium (RPE) is present, which very often gets into the vitreous body after cryocoagulation or as reaction to erythrocyte decay products (Lincoff et al., 2003). Oxidative stress is at the beginning of the cellular infiltration (Bailey et al., 2004). This, after the exhaustion of the reserve antioxidant capacity of the vitreous tissue (Berra et al., 2002) manifests itself as damage to the RPE organelles (Ballinger et al., 1999). After prolonged action of stress factors, retinal pigment epithelia necrose (Cai et al., 1999; Jin et al., 2001), and the production of granulation tissue is stimulated (Kono et al., 1995) under the development of fibrous membranes on the retina surface, subretinally and in the vitreous body. The production of fibrous membranes can be prevented by corticoids or antimetabolites (Kon et al., 1998). Its transformation into fibroblast-like cells has been described; they are present in contractile membranes. Another component is glia cells developing from Müller's radial cells, astrocytes, microglia and perivascular glia. They form a skeleton for complex membranes. Strictly glial membranes are not contractile. Strips and taut membranes form subretinal membranes. Diffuse membranes are made of glia; they are not contractile, while the strips are mixed, formed of RPE, fibroblast-like cells, macrophages and glia. Fibroblasts develop through RPE

transformation or they originate from vascular epithelia, glia or hyalocytes. The cells change in both morphology and histochemistry. Fibroblast-like cells (FLC) contain myofibrils and they are capable of active contraction. Inflammatory cells are represented by macrophages and lymphocytes. Transformation of RPE to macrophages was also described. The extracellular matrix contains type I and III collagen, or type II collagen from the vitreous body. RPE and glia can produce collagen, namely type IV, which is part of basal membranes, then heparan sulphate and laminin. Fibronectin is another important component; it is produced by all above-stated cell types. Eyes with PVR are more inclined to fibrin formation after vitreoectomy, which provokes RPE to clustering and to the formation of FLC layers.

Cytokines play an important part. This is a complex process, which has different activities in different stages of the illness. Tissue infiltration with macrophages and lymphocytes, together with increased growth factor level, develops as the normal healing reaction, as proliferative vitreoretinopathy is, after all, an exaggerated regeneration process. The presence of the above-mentioned factors explains the favourable effect of the intravitreally-administered triamcinolone on the course of the disease.

CONCLUSION

Haemolytic products arising in chronic hemophthalmos cause cellular infiltration, necrosis of the vitreous structure, and fibrous membrane formation. In this process, retinal pigment epithelium plays an important role for its antioxidant properties and the capability to phagocyte the decay products.

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