Sutureless Vitrectomy for Vitreous and Retinal Biopsy

Diagnostic vitreoretinal surgery may be an alternative when noninvasive methods reveal little about disease.

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The diagnosis of posterior segment inflammatory disease and tumoral lesions is based primarily on clinical examination. At presentation, however, some patients do not have the characteristic clinical features or classic appearance of a specific disease, and the cause of their disorder remains obscure. Recent progress in basic science, particularly in the fields of molecular biology and immunology and advances in surgical instrumentation, has greatly enhanced the diagnostic armamentarium. Diagnostic vitreoretinal surgery should be considered when other noninvasive methods of diagnosis have failed to establish a pathoetiologic mechanism.

The development of the 25-gauge transconjunctival sutureless vitrectomy operating system introduces a new concept in vitreous, retinal, and choroidal biopsy. The advantage of a smaller-gauge vitrectomy instrument system is based on its ability to minimize surgically induced trauma from conjunctival peritomy and sclerotomy sites, allow self-sealing (sutureless) sclerotomies, improve operative efficiency, and hasten postoperative recovery.[1-5]

In this article, we report the feasibility and safety of using 25-gauge transconjunctival sutureless vitrectomy for vitreous and retinal biopsy.

VITREOUS BIOPSY

Advances in surgical vitrectomy and laboratory techniques have expanded the options for diagnosis of posterior segment uveitis of unknown cause and cancer. With the evolution of vitrectomy techniques, 25-gauge transconjunctival sutureless vitrectomy may become the preferred method for biopsy because it is a relatively simple and safe procedure. A one-port diagnostic vitreous biopsy can be performed either by needle aspiration or by mechanical vitrectomy. Huang et al[6] have demonstrated that the cytological detail is equivalent for specimens obtained either by needle aspiration or mechanical vitrectomy. However, obtaining an undiluted vitreous specimen by vitrectomy potentially reduces the risk of vitreoretinal traction and retinal detachment that is associated with straight needle aspiration because the vitreous cutter ensures that no vitreous still attached to the retina is aspirated. In addition, the 25-gauge system has been shown to result in a faster procedure and less postoperative discomfort than 20-gauge systems.

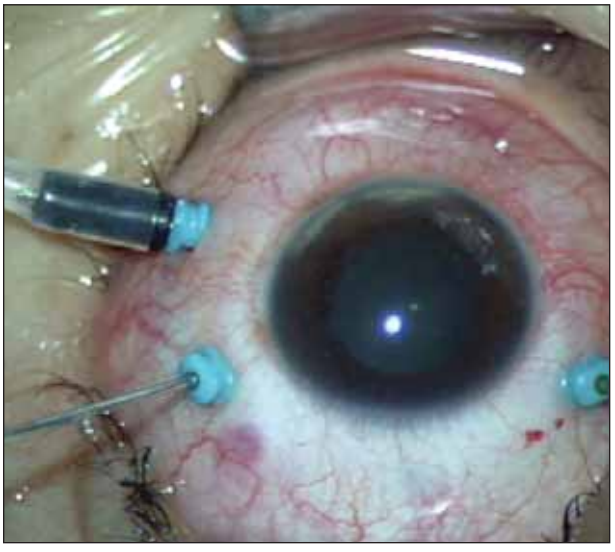

Figure 1. After insertion of the first microcannula, the infusion cannula is inserted directly into the external opening of the microcannula. The infusion cannula should be closed, (turned off) and two other microcannulas are inserted in the superotemporal and superonasal quadrants.

INDICATIONS FOR DIAGNOSTIC VITRECTOMY

In general, the indications for diagnostic vitrectomy can be grouped into atypical diseases unresponsive to standard therapy and conditions in which infection or cancer are suspected. Cytopathologic diagnosis has proved valuable in reported series or case reports of patients with endogenous and exogenous bacterial and fungal infections, nematode endophthalmitis, inflammatory pseudotumor of the iris and ciliary body, pars planitis, phacoanaphylaxis, intraocular lymphoma, leukemia, metastatic melanoma, metastatic carcinoma, medulloepithelioma of the ciliary body, epithelial ingrowth, proliferative vitreoretinopathy, and miscellaneous conditions with vitreous opacities (chronic inflammation in patients with suspected lymphoma, acute retinal necrosis, birdshot chorioretinopathy, toxoplasmosis, Whipple's disease, amyloidosis).[6-11]

TECHNIQUE FOR DIAGNOSTIC VITRECTOMY

Vitreous biopsy is limited by its dependence on the spillover of cells from the diseased tissue in question into the vitreous cavity to allow diagnosis. A good clinical evaluation of the case is important before choosing the technique to use. If the patient does not need a therapeutic vitrectomy, the approach to be used may be a one-port mechanical vitrectomy; otherwise, a three-port pars plana vitrectomy (PPV) approach may be preferred. We always prefer a three-port PPV because intraocular illumination during the procedure facilitates the attainment of a good and large enough sample and prevents accidental damage to the retina or crystalline lens.

To perform a diagnostic vitrectomy, an undiluted vitreous sample is taken after preparation for a 25-gauge transconjunctival sutureless vitrectomy as described by Fujii et al[1] with some technical modifications. After appropriate anesthesia, the operative field is prepared for surgery. We have had excellent experience with peribulbar or sub-Tenon's lidocaine anesthesia for 25-gauge cases. Some reports have indicated that topical anesthesia might be adequate in simple cases. The microcannulas are inserted through the conjunctiva into the eye, 3.5 mm to 4 mm posterior to the limbus, by means of a trocar.

In order to reduce the chance of communication between the ocular contents and the external environment, the conjunctiva is first grasped with the point of the trocar or conjunctival forceps and then moved into a position 3.5 mm to 4 mm from the limbus where beveled biplanar sclerotomies (to prevent leaks and hypotony) are to be made. In this way, the conjunctival and scleral openings are offset, and when the instrument is removed the openings return to their original position and there is a better seal of the sclerotomy site. Immediately after insertion through the eye wall, the microcannula can be held by the collar to provide stability when the trocar is withdrawn.

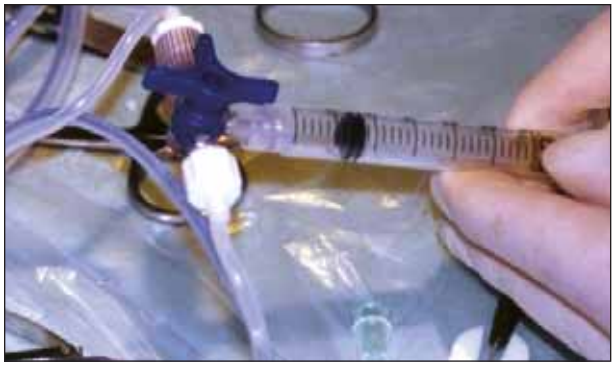

Figure 2. A 10 mL Syringe is spliced via a three-way stopcock into the aspiration line. The vitrectomy handpiece is placed in mid-vitreous cavity with the infusion turned off. Automated cutting and manual aspiration of the vitreous without concurrent infusion is then performed. The vitrector is withdrawn from the eye, and the vitreous specimen is aspirated into the syringe.

After insertion of the first microcannula, the infusion cannula is inserted directly into the external opening of the microcannula. The infusion cannula should be closed (turned off) if an undiluted specimen is to be taken. Alternatively, air may be used in the infusion to pressurize the eye, and two other microcannulas are inserted in the superotemporal and superonasal quadrants once the eye is pressurized (Figure 1). A 10 mL syringe is spliced via a three-way stopcock into the aspiration line. The vitrectomy handpiece is placed in mid-vitreous cavity with the infusion turned off. Automated cutting and manual aspiration of the vitreous without concurrent infusion is then performed (Figure 2). The infusion is turned on before the eye begins to collapse.

In the setting of a vitreous biopsy, a potential disadvantage of higher cutting rates is the greater mechanical disturbance of cellular elements within the specimen. Ratanapojnard et al[12] demonstrated that the clinical utility of all specimen types collected with high-speed cutting was preserved (using a 20-gauge vitrector); however, cutting rates of 1,500 cuts per minute (cpm) using a guillotine mechanism can mildly reduce fungal yield and markedly diminish leukocyte viability. Therefore, we suggest selecting cutting rates on a case-by-case basis by comparing merits of higher rates for the surgical goals with possible effects on sample collection. This study can probably be applied to 25-gauge transconjunctival sutureless vitrectomy because this system uses a guillotine mechanism too. We tend to use between 300 cpm and 500 cpm to minimize damage to cellular material.

PROCESSING VITREOUS SAMPLES

Both the vitreous washings and undiluted vitreous aspirate should be immediately delivered for microbiologic, virologic, cytologic, and immunologic analysis. The number of investigations that can be carried out on an individual specimen depends on the amount of harvested material. It is essential to coordinate the diagnostic vitrectomy with the microbiology laboratory so that samples may be placed in appropriate culture media and may to detect the microorganisms (Table 1). Vitreous culture is a method that directly identifies replicating infectious agents; bacterial, fungal, and viral cultures may be obtained. To avoid false-negative results it is essential to instruct the laboratory to hold cultures for at least 10 to 14 days. Viral cultures may be falsely negative because of the small quantity of organisms shed into the vitreous cavity. For suspected viral infection we suggest alternative approaches, such as polymerase chain reaction (PCR).

TABLE 1. DIAGNOSTIC VITRECTOMY: SAMPLE ANALYSIS

Microbiologic	Stains	Gram Periodic acid-Schiff Giemsa Methenamine silver (fungi)
	Cultures	Aerobic bacteria Anaerobic bacteria Fungal Viral (Occasionally)
Cytophatology	Morphology Immunocytochemistry	
Antibody Studies (simultaneous vitreous and serum)		
Polymerase chain reaction (PCR)		

TRANSVITREAL RETINAL BIOPSY

In certain conditions, the inflammatory process is localized primarily in the sensory retina or the retinal pigment epithelium (RPE). The vitreous may harbor few or none of the responsible microorganisms, and idiopathic syndromes that have a poorly understood disease mechanism can primarily involve the retina. In such cases, only histologic examination of these areas can yield a diagnosis, and a diagnostic vitrectomy would yield very little information. Retina biopsy can be performed to better understand the disease process and to help establish a diagnosis.

SURGICAL TECHNIQUE FOR RETINAL BIOPSY

We always remove the cortical vitreous to gain access to the retina, to clear the media, and to allow gas tamponade. Before a retinal biopsy is performed, the location of the tissue to be sampled must be carefully considered. An appropriate site is chosen to minimize both intraoperative and postoperative complications. Location is preferably in the superior and nasal retina, at the junction of infected and uninfected retina, as peripheral as possible, and in a relatively avascular area. The specimen should include the advancing edge of the affected area (such as in retinitis if that is the case) because this is where actively replicating, viable organisms are most likely to be found. Central areas of the lesion may contain only necrotic tissue. Cautery at the area of the biopsy site with a 25-gauge diathermy probe is occasionally needed if large vessels are present. In cases in which the retina is attached, a cannula is used to inject saline under the sensory retina to create a small bleb. An incision is then made in the retina using a 25-gauge flexible subretinal needle. After, the tissue is excised with 25-gauge microvitreoretinal scissors, leaving a small area of anchoring attachment. The biopsied retina is grasped securely with 25-gauge vertical forceps so that as little as possible of the specimen is crushed and is removed from the eye. Laser is not necessary at the biopsy edges involved by inflammation, but it is placed at the edges of normal retina (Figure 4). An air-fluid exchange is performed, and occasionally a long-acting gas is injected.

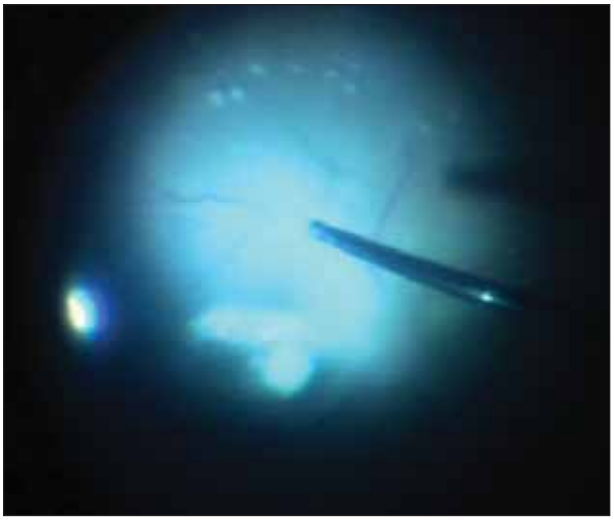

Figure 3. After the undiluted sample is taken, the therapeutic three-port vitrectomy can be completed if necessary.

INDICATIONS FOR RETINAL BIOPSY

A retinal biopsy may help explain and categorize inflammatory diseases; it also may influence therapeutic decisions. Ciulla et al[13] reported a case of ocular lymphoma in which a

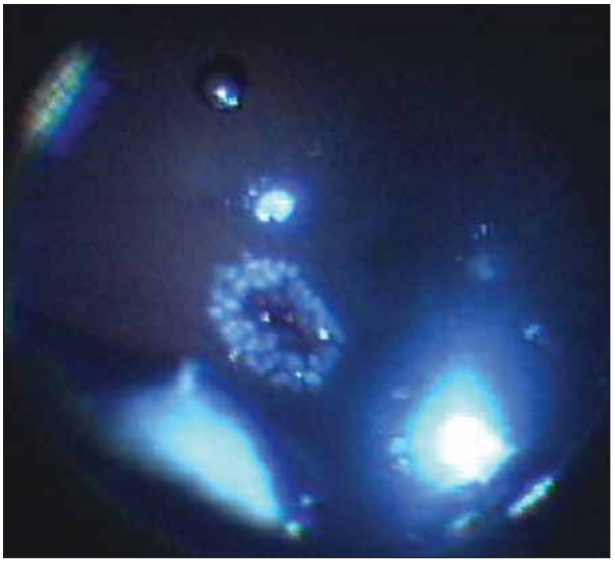

Figure 4. Laser is not necessary at the biopsy edges involved by inflammation, but it is placed at the edges of normal retina.

diagnosis of large cell lymphoma was made by transvitreal subretinal aspiration biopsy, whereas two vitreous biopsies and a concurrent retinal biopsy were nondiagnostic. Pavan et al[14] also reported a case in which subretinal aspiration of infiltrates revealed large atypical cells with positive staining for leukocyte-common antigen, confirming a diagnosis of high-grade lymphoma. Coskuncan et al[15] reported on a series of 397 patients who were followed prospectively after bone marrow transplantation, 2% of whom developed infectious retinitis requiring intraocular biopsy. In these cases, retinal biopsy was crucial to diagnostic and therapeutic decision-making.

PROCESSING RETINAL SAMPLES

Removal of the sample from the eye can be difficult using a 25-gauge port because it may be difficult to remove the retinal biopsy specimen through the relatively long length of trocar tubing protruding into the eye. For these reasons, it may be best to open the port corresponding to the forceps and use a 20-gauge instrument to remove the specimen. After the tissue sample is removed from the eye, it is immediately placed in appropriate fixative. The retina specimen can be sectioned into three pieces under the operating microscope. If orientation is important, the sample may be placed on a piece of filter paper or other material and the correct localization marked on the paper. This is an important point of discussion for the surgeon and the pathologist before the surgical procedure. One sample should be frozen in cryostat compound for immunopathology, the second piece fixed with 4% glutaraldehyde for light and electron microscopy study, and the third piece sent for culture and PCR. It should be recalled that glutaraldehyde would cross link proteins and usually make immunocytochemistry difficult.

INTRAOCULAR BIOPSY COMPLICATIONS

The risks of intraocular biopsy are those of vitreoretinal surgery in general. These include increased intraocular pressure, cataract progression, peripheral retinal tears, retinal detachment, choroidal hemorrhage, vitreous hemorrhage, endophthalmitis, exacerbation of the underlying inflammatory disease, and proliferative vitreoretinopathy. ■

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