27-gauge micro incision biopsy technique for tumors of the iris and ciliary body

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Purpose. *To evaluate a 27-gauge aspiration-cutter technique for biopsy of anterior segment neoplasia.* ***Methods.*** *In this retrospective case review, all patients had clinically diagnosed iris or ciliary body tumors. Each underwent surgical iridectomy biopsy utilizing a modification of the Finger Iridectomy Technique (FIT). Specifically, a beveled clear corneal incision was created with a 27-G inked trocar. Sodium hyaluronate 1 % was infused to posteriorly displace the iris and stabilize the chamber. Then a 27-G aspiration-cutter probe was introduced into the anterior chamber, to a position over the tumor. With the aspiration portal abutting the lesion, surgical iridectomy biopsies were obtained using a cut rate of 300 cuts/min and aspiration of 600 mm Hg. After each pass, the probe was removed, its tip placed in balanced salt solution and its contents aspirated into a 3-mL syringe. Each syringe was labeled by pass number and tumor location (by clock hour). After obtaining multiple biopsies, the sodium hyaluronate 1 % was removed and the integrity of the wound checked.* ***Main Outcome Measures.*** *Cytologic, tissue histopathology, and immunohistochemical analyses were performed for diagnostic purposes. Visual acuity, intraocular pressure control and procedure-related complications were secondary outcomes measures.* ***Results.*** *Diagnostic specimens were obtained in all 8 cases (100 %). Diagnoses included unifocal melanoma (62 %), diffuse multifocal melanoma (13 %), and melanocytoma (25 %). All 27-gauge corneal wounds were self-sealing. There was no secondary glaucoma, infection, cataract, or vision loss. Two patients had small transient hyphemas which resolved within 10 days.* ***Conclusions.*** *This study reveals that 27-gauge aspiration-cutter assisted surgical iridectomy biopsy was both minimally invasive and effective. It allowed for partial and full-thickness iris biopsy. Compared to larger-gauge aspiration cutter assisted biopsy techniques, this 27-G aspiration-cutter probe allowed for even smaller corneal incisions, rapid recovery and tissue for pathology evaluation.*

Keywords: biopsy techniques, iris tumors, uveal melanoma, melanoma surgery, ophthalmic surgery.

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Biopsy-proven iris melanomas have a 10.7 % risk for metastasis [1–3]. Fortunately, suspected iris melanomas and other anterior segment tumors are accessible for biopsy through a relatively safe, clear corneal incisions, offering cytologic, histopathologic, molecular and genetic evaluation [4–7].

Methods of biopsy include: wide-incision incisional biopsy with iridectomy, excisional biopsy, fine-needle aspiration biopsy (FNAB), and aspiration-cutter techniques [8–14]. Surgical iridectomy biopsy is typically preferred by ophthalmic pathologists, in that it yields large blocks of tissue for histopathologic and immunohistopathologic analysis [15, 16]. However, surgical iridectomy requires a corneal or scleral dissection. Such wounds are relatively large, require suturing, and may cause astigmatism. This method can also result in a dysmorphic, non or poorly functional pupil as well as glare and a need for visual rehabilitation. In addition, surgical iridectomy biopsy for diffuse or multifocal iris melanomas requires even larger surgical wounds and repair.

In an effort to offer less invasive and destructive biopsy methods, FNAB and aspiration-cutter techniques have been employed. Each trades quantity of specimen for safety and speed of recovery. For example, FNAB typically involves placing a 25-G sharp needle tip (4 mm in length, 0.8 mm in circumference) through the cornea, through the aqueous and into or on the anterior segment tumor. The surgeon then scrapes, cuts and manipulates the tumor with the sharp needle-tip to liberate cells, which are manually aspirated through extension

tubing by an assistant holding a syringe. Such manual aspiration typically causes an unstable anterior chamber depth. In addition, it risks of piercing, lacerating or partially embedding the 4-millimeter-long sharp needle-tip into a mean normal thickness of iris (0.6 mm), lens (4.0 mm) or ciliary body (1.3 mm). Risks include trauma to anterior lens capsule, corneal endothelium, blood vessels and ciliary body. Understandably, hyphema is the most commonly reported complication of anterior segment FNAB [13]. While FNAB is less invasive than open surgery, it usually cannot acquire enough tissue for histopathologic or immunohistopathologic analysis.

In contrast, the Finger Iridectomy Technique (FIT) biopsy typically yields both cells and tissue for pathology [8]. This is accomplished using a probe that lacks sharp edges, administered under visco-elastic anterior chamber stabilization and mechanically controlled aspiration. Though we have published on larger-gauge FIT, this series reports on an even smaller "27-gauge" aspiration-cutter assisted biopsy for anterior chamber tumors. The main advantages are that these smaller-gauge cutters require even smaller "micro incisions" (Fig. 1). The question remained if we could obtain equivalent quality, diagnostic tumor samples.

MATERIALS AND METHODS

Patient Selection. This research conforms to the tenets of the Declaration of Helsinki. We obtained internal review board (IRB) approval from The New York Eye Cancer Center (http://eyecancer.com). Each patient signed release forms for the United States of America Health Insurance Portability and Accountability Act of 1996. Patients included those with iris or ciliary body tumors demonstrating growth, atypical features, or suspicious for melanoma were considered for biopsy. Less concerning tumors were included when patients sought a biopsy-proven diagnosis. Observation, biopsy, and treatment were discussed and offered as indicated for each case. When biopsy was planned, the risks, benefits, and alternatives of incisional, FNAB and FIT (25 and 27-gauge) were discussed. Written informed consent was obtained for each patient undergoing the procedure outlined below.

Surgical preparation. All patients underwent pre-admission testing and were medically ready for surgery. Topical pilocarpine hydrochloride 1 %, topical timolol maleate 0.5 %, and oral acetazolamide 500 mg were administered 1 hour prior to surgery. Upon entering the operating room the patient's clinical chart was reviewed. A surgical time-out was performed identifying the correct patient, surgical site, and procedure. The patient's eye was then treated with iodine-based topical antibiotic, draped in the sterile fashion for ophthalmic surgery and an eyelid speculum was placed.

27-gauge anterior segment microsurgery. Sitting superiorly, the operating microscope was adjusted to maximally visualize the anterior segment. A 0.3 forceps was used to for counter-traction as an inked, 27-G trocar was used to create a blue, visible, slightly expanded and shelved incision through clear juxtalimbal cornea. The clear corneal incision allowed any liberated anterior segment cells to exit the patient's body during surgery. Most incisions were made in the superior 90-degrees of the cornea, providing an easy approach for the surgeon sitting superiorly. Sodium hyaluronate 1 % was then used to fill the anterior chamber. It served to maintain both stability and the depth of the anterior chamber as well as position of the iris tumor during the biopsy.

With the saline infusion primed but not used, a primed 27-G aspiration-cutter probe was inserted into the anterior chamber. The probe was rotated along the axis of the corneal incision to iris and tumor locations. Then, the probe was rotated along its central axis as to bring its portal in contact with the iris or tumor for biopsy. During biopsy, the aspiration port was occluded by the tumor or iris and cutting obtained the surgical specimens. The initial settings were 300 mm Hg of aspiration at a cutting rate of 600 cuts per minute. These settings were adjusted as needed to maximize efficiency while keeping both suction and the cutting rates to a functional minimum.

Biopsies included partial and full-thickness iridectomies as well as partial and full-thickness tumor biopsies. After each pass, the cutter was removed from the eye and an empty 3-cc syringe was attached to the effluent connector. Then the aspiration-cutter portal was placed in sterile saline solution, while the 3 cc syringe plunger was withdrawn as to flush the biopsy specimen from the tubing into the syringe. Approximately 0.5 mL of saline was typically required to retrieve the specimens. The syringe

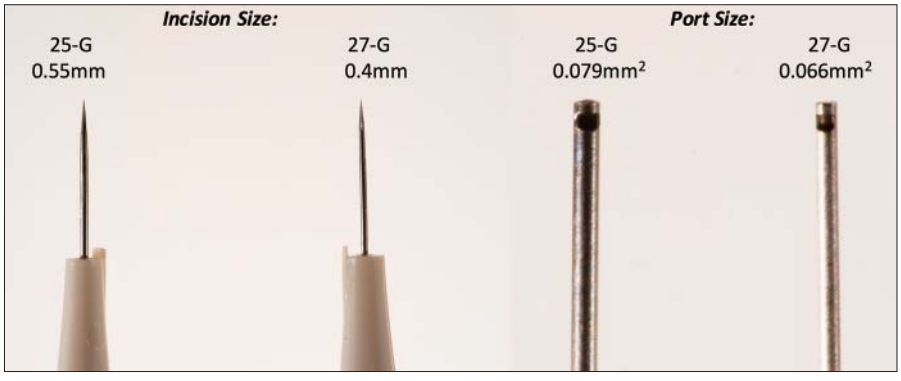

Fig. 1. Comparison between the 25-G and 27-G Trocars and Aspiration-Cutters. Photographic images demonstrate the smaller size trocar (0.4 mm) and aspiration-cutter port (0.07 mm^2) of the 27-G setup as compared to the 25-G instruments. Performing the procedure with smaller instruments allowed for smaller, self-sealing wounds and a more rapid visual recovery.

contents were visualized under the microscope to assess the adequacy of the sample. Biopsy typically yielded pieces easily visualized within the syringe when viewed under the microscope. The syringe was labeled according to pass and clock hour from which the specimen was obtained. This process was usually repeated at least 3 times.

The syringes were immediately sent to pathology for analysis. The residual sodium hyaluronate 1 % was removed from the eye and replaced with balanced salt solution. The corneal incision was checked for leakage utilizing Seidel's test. A subconjunctival antibiotic-steroid solution was injected (superiorly). Antibiotic-steroid ointment was placed on the eye and the eyelid speculum was removed. Due to sedation, the operative eye was temporarily patched and shielded. Our cytopathology, histopathology, and immunopathology methods for specimen analysis have been previously described [8, 11, 17]. In short, the specimens were concentrated by cytospin, placed on glass slides and stained with hematoxylin and eosin. Immunohistochemical analysis and genetic profiling were employed when possible [18, 19].

RESULTS

Eight iris biopsies were performed utilizing the 27-G aspiration-cutter technique [14]. The patients' mean age was 51.38 ± 26.46 years and 63 % were female. Demographic information and clinical findings for all cases are included in Table 1. This table reviews the demographic information and clinical characteristics of the iris or iridociliary tumors that underwent biopsy. Postoperative course and pathology results are summarized in Table 2. This table summarizes the number of specimens obtained in each case, the resulting diagnosis, and the postoperative course of each patient. Vision and intraocular pressure were stable postoperatively. Note that no sutures were required for wound closure.

Table 1. Patient and tumor characteristics

Case	Eye	Age (Yrs)	Gender	Tumor Color	Iris			Pigment	UBM	
					Correctopia	Ectropion Uvea	Vasculopathy	Dispersion	IPE Involved	CB Involved
1	OS	33	F	Dark Brown	Yes	Yes	No	Yes	Yes	Yes
2	OS	20	F	Dark Brown	No	No	No	No	Yes	No
3	OD	71	F	Dark Brown	Yes	No	No	No	Yes	No
4	OS	26	M	Dark Brown	Yes	No	No	No	No	Yes
5	OS	73	M	Light Brown	No	No	No	No	Yes	Yes
6	OS	90	M	Dark Brown	No	No	Yes	No	No	Yes
7	OD	66	F	Amelanotic	No	No	Yes	No	Yes	Yes
8	OD	32	F	Dark Brown	No	Yes	No	No	Yes	No

Note. M = male; F = female; UBM = ultrasound biomicroscopy; IPE = iris pigment epithelium; CB = ciliary body.

Table 2. Multifocal iris biopsy results and postoperative course

Case	Biopsy Sites	IOP		Vision (20/*)		Suture Needed	Hyphema After 1 Day	Complications After 1 Month	Histopathology	Additional Testing
		After 1 Month	Change	After 1 Month	Change					
1	4	22	-2	20	0	No	Yes	None	Mixed cell melanoma	MART 1+, HMB45+ Genetic analysis
2	3	14	0	20	0	No	No	None	Melanocytoma	
3	3	13	-1	20	0	No	No	None	Malignant melanoma	MART 1+, HMB45+
4	3	15	0	HM	0	No	Yes	None	Melanocytoma	Rare GFAP and S-100 reactive cells, MART1+
5	3	8	-1	20	0	No	No	None	Malignant melanoma	
6	3	21	0	100	0	No	No	None	Malignant melanoma	MART 1+, HMB45+, Ki-67+ Enucleation showed RPE adenoCA
7	3	12	-2	20	0	No	No	None	Malignant Melanoma	MART 1+, HMB45+
8	4	8	-4	20	0	No	No	None	Malignant Melanoma	

Note. IOP measured in mm Hg; Vision measured in snellen visual acuity 20/*; + = positive.

This minimally invasive iridectomy technique was capable of yielding specimens adequate for cytologic, histopathologic, and immunopathologic analysis in all cases (100 %). Case 1 also underwent genetic analysis. Malignant melanoma was diagnosed in six cases, including one diffuse iridociliary melanoma. There were two cases of melanocytoma. The mean number of iris biopsies was 3.3 (range 3–4). No 10–0 nylon sutures were required for wound closure. No patient developed secondary glaucoma. Specifically, the mean preoperative intraocular pressure (IOP) was 14.1 ± 5.2 mm Hg, and mean postoperative IOP was 12.9 ± 5.9 mm Hg. As noted in our prior series, a total of 63% of patients had a decrease in IOP at the 1-month postoperative visit. This suggested an improvement in aqueous humor dynamics. Two patients with the largest tumor burden experienced a small hyphema postoperatively (less than 1 mm) that resolved in 7 to 10 days.

Within 1 month after biopsy, all visual acuity measurements were within one line of vision compared to preoperative visual acuity using the Early Treatment Diabetic Retinopathy Study protocol (Table 2). This includes a patient who underwent brachytherapy with an anterior segment plaque 2 weeks after biopsy. No patients experience transient corneal edema, glare, diplopia, or visual distortion after biopsy. This also includes cases where full-thickness biopsies were obtained in the inferior 90 degrees of the iris. There was no trauma to the lens capsule, iris, or ciliary body.

Case Example. Case 1. A 33-year-old female was referred after reporting an increased pigmentation of her left iris over the last 6 months. Her Snellen visual acuity was 20/20 and IOP was 24 mm Hg. Slit-lamp examination revealed blue irises with multiple diffuse dark brown tumors, corectopia and ectropion uvea of the left eye (Fig. 2, 1a). The anterior chamber was deep and quiet, with the exception of a shallow chamber inferiorly, resulting from a ciliary body mass bowing the iris forward. Pigmentary deposits were present on the anterior lens capsule. Gonioscopy revealed the presence of tumor in the inferior and nasal angle. 35 MHz high-frequency ultrasound imaging revealed diffuse uveal (iris and ciliary body) tumors with blunting of the angle inferiorly in the location of the region of the ciliary body mass. The dilated fundus exam was unremarkable. The options of biopsy prior to initiating treatment was discussed with the patient, and she preferred to proceed with a 27-G aspiration-cutter assisted biopsy to establish a tissue diagnosis prior to treatment (Fig. 2, 1b).

Case 7. A 66-year-old female was referred for an iris tumor in the right eye. Her Snellen visual acuity was 20/20 OU and was IOP 14 mm Hg in the right eye. Slit-lamp examination revealed blue iridies, an iris tumor in the right eye extending into the pupil of the right eye (Fig. 2, 7a). Dilated fundus exam was unremarkable. High-frequency ultrasound imaging (35 MHz) showed a variable reflective club- or mushroom-shaped iridociliary tumor with invasion into the supraciliary space with a thickness of 1.9 mm at the iris and 1.5 mm at the ciliary body. Again, the options of biopsy prior to initiating treatment was discussed with the patient and she elected to undergo an aspiration-cutter assisted biopsy. Multiple biopsies were obtained adjacent to mushroomed-shape lesion to reduce the risk of biopsy-related bleeding. (Fig. 2, 7b).

Pathology. Pathologic specimens obtained from several cases are shown in Figure 3. Each case underwent cytologic, histopathologic and immunohistopathologic analysis (Table 2). The detailed characteristics of each specimen, clinical course and outcomes of each case are beyond the scope of this case series.

Complications. Our aspiration-cutter assisted technique had a low complication rate. While this is a small series, postoperative evaluations revealed no wound leaks, endophthalmitis, cataract, intraocular pressure spikes or tumor dissemination. None of the patients experienced

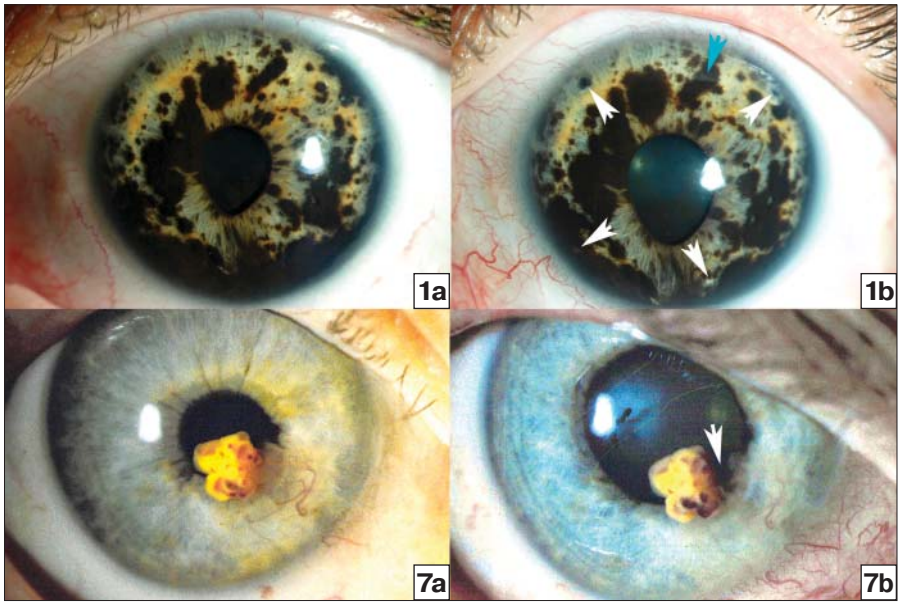

Fig. 2. Slit-lamp Photography Pre- and Post 27-G MFIT. Slit-lamp photographic images taken prior to (left) and after (right 27-gauge FIT biopsy). Case #1 shows a diffuse iris melanoma with multifocal areas of biopsy (white arrows). The 27-gauge corneal entry point is also visible (blue arrow). Lower left slit lamp photographs of case #7 reveals a pedunculated, hypervascular, relatively amelanotic iridociliary melanoma (left image). On the bottom right, the base of the tumor was biopsied (white arrow).

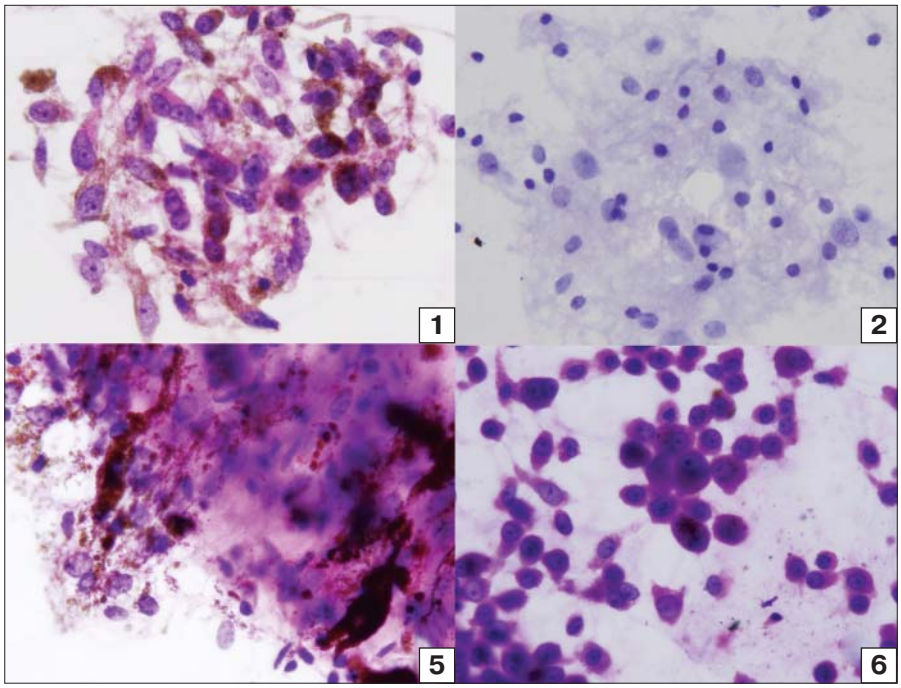

Fig. 3. Representative Pathology. A series of specimens from Cases 1, 2, 5, and 6 demonstrate the adequacy of specimens retrieved by 27-gauge FIT. Cases 1, 5, and 6 photomicrographs of Hematoxylin and eosin stained specimens under oil immersion (100 x). All cases demonstrate mixed cell malignant melanoma.

polycoria or corectopia. All patients maintained their baseline visual acuity with no complaints of photophobia, glare, or double vision. Two patients had a small hyphema postoperatively (<1 mm) that resolved in within the first 10 days. All patients were evaluated within 24 hours of the procedure and at 1 week, 1 month, and 2 months after surgery. Several patients have been followed for over 6 months, including patients with indeterminate lesions and those treated with eye and vision-sparing palladium-103 radioactive plaques.

DISCUSSION

Uveal melanoma carries a risk of morbidity and mortality. Prompt diagnosis and definitive treatment reduces this risk. This study demonstrates that iris biopsies can be obtained with a 27-G aspiration-cutter through a small, clear corneal incision. In every case, a pathologic diagnosis was achieved. While adequate tissue was typically available for genetic analysis, this testing is not routinely obtained in our center due to out of pocket cost and lack of impact on clinical management. Not one patient experienced a change in visual acuity or secondary glaucoma after biopsy. In fact, 63 % of patients had decreased IOP at 1 month follow up (off medications). We theorize that this finding is likely related to a secondary change in aqueous flow dynamics related to the newly acquired full-thickness iridectomies. Two patients experienced transient, minor post-operative hyphemas without consequence.

We have reported the success of this technique with a 25-G aspiration-cutter probe [14]. In comparison, this smaller-gauge technique was just as capable of obtaining specimens of similar size for cytologic, histopathologic, histochemical and genetic analysis (despite working through an even smaller incision). However, in our previous report, 7 % of patients required a 10–0 nylon suture for wound closure. In contrast, using this new 27-gauge technique, all wounds were self-sealing and no sutures were required. Similar to our 25-G technique, the transcorneal approach eliminated the risk of subconjunctival tumor seeding possible with open biopsy.

In comparison to FNAB, where cells are aspirated through a sharp-needle-tip and manual suction; our technique utilizes a blunt-tipped aspiration-cutter with machine controlled aspiration and cutting. Thus, we have found reduced risks of secondary hyphema and potential damage to other intraocular structures. In addition, when FIT biopsy causes tumor-hemorrhage, the aspiration-cutter was also used to evacuate the blood. Lastly, when compared to more invasive methods such as surgical iridectomy, the specimens obtained from the FNAB are smaller and thus offer less insight into the extent of local invasion.

Other uses of aspiration-cutters in the anterior segment include: iridectomy for narrow angle glaucoma and during phacoemulsification in patients with iridoschisis [6, 20, 21]. However, all of these techniques have used larger gauge aspiration-cutters and several reports employ irrigation from the phacoemulsification probe or an anterior chamber maintainer [6, 21].

This retrospective, interventional case series demonstrates that a smaller, 27-gauge, minimally invasive approach to surgical iridectomy biopsy was effective in obtaining full and partial thickness iris biopsy for the diagnosis of iris tumors. All patients benefited from a diagnosis by pathology, a rapid recovery and no significant complications.

Conflict of Interest: There is no conflict of interests.

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Микроинцизионная биопсия при опухолях радужки и цилиарного тела с помощью канюли 27-го калибра

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Цель – *оценить методику биопсии при новообразованиях переднего отдела глаза с помощью аспирационной иглы 27-го калибра.* ***Методы****. В настоящем ретроспективном обзоре рассматриваются истории болезни пациентов, у которых были клинически диагностированы опухоли радужки или цилиарного тела. Каждому пациенту была проведена биопсия путем хирургической иридэктомии по модифицированному методу P.Finger. Конусообразный роговичный разрез производился с помощью меточного троакара 27-го калибра. Затем в переднюю камеру вводили 1% раствор гиалуроната натрия для смещения радужки и стабилизации камеры. После этого в переднюю камеру над опухолью вводился аспирационный зонд 27-го калибра. Аспирационный порт соприкасался с пораженным местом, и ткань радужки извлекалась для биопсии при скорости 300 срезов в минуту и аспирационном давлении 600 мм рт. ст. После каждой биопсии зонд вынимался, его конец помещался в сбалансированный солевой раствор, а содержимое перемещалось в шприц объемом 3 мл. Каждый шприц маркировался номером биопсии и локализацией опухоли (с ориентацией по часам). После получения серии биоптатов 1% раствор гиалуроната натрия удалялся, а роговичный разрез проверялся на герметичность.* ***Оценка результатов****. В диагностических целях проводился цитологический, гистологический и иммуногистохимический анализ полученной ткани. Кроме того, определяли остроту зрения, внутриглазное давление и возможные осложнения, связанные с выполнением процедуры.* ***Результаты****. Диагностические образцы были получены для всех 8 пациентов (100%). Поставлены следующие диагнозы: солитарная меланома (62%), диффузная мультифокальная меланома (13%) и меланоцитома (25%). Все роговичные разрезы, выполненные троакаром 27-го калибра были самогерметизирующимися. Среди осложнений процедуры не отмечена вторичная глаукома, инфекция, катаракта или потеря зрения. У 2 пациентов наблюдались небольшие временные гифемы, которые рассосались в течение 10 дней.* ***Заключение****. Исследование показало, что биопсия радужки, проведенная с помощью аспирационной канюли 27-го калибра, является малоинвазивной и эффективной. Она позволяет осуществить как поверхностную биопсию радужки, так и биопсию всей толщины ткани радужки. Если сравнить нашу методику с методиками биопсии с помощью режущих инструментов большего калибра, то можно заключить, что аспирационный зонд 27-го калибра при минимальном разрезе роговицы обеспечивает быстрое восстановление и достаточное количество ткани для гистологической диагностики.*

Ключевые слова: методы биопсии, опухоли радужки, увеальная меланома, хирургия меланомы, глазная хирургия.

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