Utility of tubular retractors to minimize surgical brain injury in the removal of deep intraparenchymal lesions: a quantitative analysis of FLAIR hyperintensity and apparent diffusion coefficient maps

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OBJECTIVE Brain retraction systems are frequently required to achieve surgical exposure of deep-seated brain lesions. Spatula-based systems can be associated with injury to the cortex and deep white matter, particularly adjacent to the sharp edges, which can result in uneven pressure on the parenchyma over the course of a long operation. The use of tubular retractor systems has been proposed as a method to overcome these limitations. There have been no studies assessing the degree of brain injury associated with the use of tubular retractors.

METHODS Twenty patients were retrospectively identified at Weill Cornell Medical College who underwent resection of deep-seated brain lesions between 2005 and 2014 with the aid of a METRx tubular retractor system. Using the Brainlab software, pre- and postoperative images were analyzed to assess volume, depth, extent of resection, and change in postoperative MR FLAIR hyperintensity and restricted diffusion on diffusion-weighted imaging (DWI).

RESULTS The mean preoperative tumor volume was $16.25 \pm 17.6 \text{ cm}^3$. Gross-total resection was achieved in 75%, near-total resection in 10%, and subtotal resection in 15% of patients. There was a small but not statistically significant increase in average FLAIR hyperintensity volume by $3.25 \pm 10.51 \text{ cm}^3$ ($p = 0.16$). The average postoperative volume of DWI high signal area with restricted diffusion on apparent diffusion coefficient maps was $8.35 \pm 3.05 \text{ cm}^3$. Assuming that the volume of restricted diffusion on DWI around tumor was 0 preoperatively, this represented a statistically significant increase on DWI ($p < 0.001$).

CONCLUSIONS Although tubular retractors do not appear to significantly increase FLAIR signal in the brain, DWI intensity around the retractors can be identified. These data indicate that although tubular retractors may minimize damage to surrounding tissues, they still cause cytotoxic edema and cellular damage. Objective comparison against other retraction methods, as compared by 3D volumetric analysis or similar methods, will be important in determining the true advantage of tubular retractor systems.

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KEY WORDS neurosurgery retractor; tubular; tube; FLAIR; diffusion-weighted imaging; brain injury; magnetic resonance imaging; surgical technique

BRAIN retraction systems are frequently required to achieve surgical exposure of deep-seated brain lesions. Retraction, however, is associated with complications that include brain edema, vascular compromise causing ischemia, and direct damage to surrounding cortex.²⁷ The frequency of retraction-based injury has been estimated to be approximately 10% in skull base surgery and 5% in intracranial aneurysm operations.⁴ Techniques such

as intermittent retraction,²⁴ endonasal trajectories,^{3,6,10,17} and those that use tubular retractors^{1,2,7,9,11–16,18–23,25} have been used to minimize these injuries.

Previously, we reported the successful use of a METRx spinal tubular retractor system, paired with the Brainlab frameless navigation system, for resection of deep-seated intracranial lesions.⁹ The advantages of this tubular retractor system include equal distribution of retraction pressure

ABBREVIATIONS ADC = apparent diffusion coefficient; DWI = diffusion-weighted imaging; GTR = gross-total resection; NTR = near-total resection; STR = subtotal resection.

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and gradual dilation of brain tissue to minimize damage to surrounding tissue. Since then, a number of other case reports have described the use of tubular retractor systems in adult and pediatric patients.^{2,23} However, to date, no reports have described the use of 3D volumetric analysis to determine the effectiveness of these retractor systems in terms of extent of resection and changes in preoperative versus postoperative MR FLAIR signal as well as changes in the diffusion-weighted imaging (DWI) signal. These MR sequences were chosen as correlates of postoperative trauma to surrounding brain with the FLAIR signal as a marker for extracellular fluid and vasogenic edema and restricted diffusion on DWI as a marker for brain cytotoxic edema and ischemic or cell damage.⁸

Methods

This study was approved by the institutional review board at Weill Cornell Medical College. Twenty patients who underwent resection of deep-seated brain lesions at Weill Cornell Medical College between 2005 and 2014 using a METRx tubular retractor system were retrospectively identified. A retrospective chart review of medical/surgical oncology, pathology, and radiology records was conducted. Demographic and clinical variables included age, sex, date of surgery, tumor location, retractor size, pathological diagnosis, and volumetric assessment of resection. For comparison with conventional retractors, preoperative MRI scans of all tumors removed prior to the use of tubular retractors were examined to find cases that would have been good candidates for tubular retractors. Assessments were made with the assessor blinded to postoperative FLAIR and DW images. Only a small number of suitable cases were identified ($n = 5$). The surgeon was blinded to postoperative scans and thus was not biased based on estimation of change in FLAIR or DWI. This cohort of patients underwent the same analysis as the tubular retractor patients.

Surgical Technique

The details of the technique have been presented elsewhere.⁹ In our patient population, a trajectory is planned to target the outer margin of the lesion, either through the crest of a gyrus or the depths of a sulcus, into the capsule of the tumor using 3D navigation planning software (Brainlab). The path is chosen to pass through the least functional white matter tracts based on anatomical information. Diffusion tensor imaging is used as needed based on the presence of adjacent critical white matter tracts, such as motor or visual tracts. A small linear corticectomy is performed, and the white matter is dissected linearly along the lengths of the white matter tracts using the tip of the navigation tool. Then, a custom-built dilator is placed into the barrel of the METRx tube and advanced to the capsule of the lesion with constant irrigation on the pia (Fig. 1). The retractor is attached to the table with a malleable arm and fixed into position. The diameter of the tube is chosen based on the size of the lesion but is generally at least 1 cm smaller than the maximal diameter of the tumor. The optimal length of the retractor is roughly 2 cm longer than the distance from the pia to the margin of

FIG. 1. The custom-built dilating tubular retractor system assembled for insertion with reflecting spheres attached.

the tumor. It is important that the retractor be longer than this distance since the METRx retractor sits 1 cm above the pial surface and often needs to be advanced deeper into the cavity as the tumor is resected. The retractor arm is frequently moved to ensure that the entire tumor is removed in a systematic fashion. The optimal instruments are similar to those used in endonasal endoscopic skull base surgery, since they are long and thin with the action at the tip and not along the shaft. Either the microscope or the endoscope or a combination can be used depending on the comfort of the surgeon, the diameter of the tube, and the visibility at the tip of the tube.

Volumetric Analysis

Two individuals conducted the volumetric analysis described below. Neither individual was involved in the surgical or postoperative care of the patients in this study, and both were blinded as to the type of resection completed by the surgical team. Radiographic MRI for each of the identified patients was imported into Brainlab software for volumetric analysis. Preoperative T1-weighted, T1-weighted with gadolinium, and T2-weighted FLAIR scans as well as postoperative T1-weighted, T1-weighted with gadolinium, T2-weighted FLAIR, and DWI scans and apparent diffusion coefficient (ADC) maps were coregistered to align the images. Using the Brainlab software SmartBrush tool, objects were drawn to measure 3D volumes. Tumor burden was defined as enhancement on pre- and postoperative gadolinium T1-weighted images. Hyperintense areas in the vicinity of lesions on T1-weighted images was defined as blood. Using the advanced manipulation tools within the program, blood was subtracted from pre- and postoperative tumor burden for a final tumor volume. Gross-total resection (GTR) was defined as 100% tumor removal. Near-total resection (NTR) was defined as $> 95\%$, and subtotal resection

(STR) as $< 95\%$. Brain edema and nonenhancing cellular infiltration around tumors was defined as hyperintensity on pre- and postoperative T2-weighted FLAIR images with the tumor burden subtracted out using the advanced manipulation tool. Change in volume of hyperintensity was determined arithmetically as postoperative edema minus preoperative edema, since shifts in brain anatomy after surgery prevent accurate 3D subtraction within the program. Using the Trajectory Planning tool in Brainlab, oblique images along the plane of the surgical path were created. The depth of tumor was then measured along this surgical plane from the entry point on the skull to the nearest edge of the tumor. DWI was used to determine areas with restricted diffusion (brain ischemia/damage) postoperatively ($n = 13$). The preoperative DWI restricted area was assumed to be 0 since DWI images were not routinely obtained on the navigation scan. Postoperative DW images were analyzed by selecting bright regions with the SmartBrush tool (Fig. 2). However, since a bright signal on T2-weighted imaging can also appear on DWI, the DW images were coregistered and fused with the patient's postoperative ADC map. The areas without low signal intensity on the ADC map were subtracted from the DWI volume so as to select only true regions with restricted diffusion. One patient in our cohort did not have preoperative MR images, and CT scans were used for the volumetric analysis in that case.

Statistical Analysis

Scatterplots were made comparing the independent variables of preoperative tumor volume, preoperative FLAIR volume, and tumor depth versus the dependent variable of either change in FLAIR or DWI signal. Linear regression trend lines were then fit to these scatterplots. The coefficient of determination (R^2) was used to determine the fit of this regression and the proportion of variability that could be explained by that model. Paired t-tests were used to compare pre- and postoperative FLAIR hyperintensity and restricted diffusion volume on DWI. Preoperative restricted diffusion on DWI was assumed to be 0 for all cases with postoperative DWI and ADC images. Interobserver variability was determined using the Pearson correlation coefficient, and p values were reported for each variable. For comparisons between pre- and postoperative volumes, the volumes of the more senior individual were used. A p value < 0.05 was considered statistically significant. Statistical analysis was performed using Microsoft Excel.

Results

Demographics

Twenty patients (Table 1) with deep-seated brain lesions underwent resection using the METRx tubular dilating retractor system in combination with Brainlab frameless stereotactic navigation. The patients ranged in age from 23 to 81 years. The study population included 9 females and 11 males. The pathological diagnoses included metastatic non-small cell lung carcinoma ($n = 5$), metastatic breast carcinoma ($n = 1$), meningioma ($n = 2$), cavernous malformation ($n = 1$), metastatic melanoma ($n = 2$), glioblastoma

FIG. 2. Brainlab volumetric analysis tools. **Upper:** SmartBrush selection of DWI signal postoperatively. **Lower:** 3D reconstruction and volumetric measurement.

($n = 5$), ependymoma ($n = 1$), granular cell tumor ($n = 1$), toxoplasmosis ($n = 1$), and craniopharyngioma ($n = 1$). Tumors were diverse in location, including intraventricular ($n = 6$), frontal ($n = 4$), temporal ($n = 4$), and parietal ($n = 5$) regions. The diameter of the tubular retractor used ranged from 14 to 26 mm, and the length of the retractor ranged from 3 to 7 cm. The frequency of use of each is demonstrated in Fig. 3; operative notes had recorded tubular retractor diameter for 10 patients and the retractor length for 16 patients. The depth of the lesion from the pial surface averaged 45.97 ± 12.7 mm. Complications occurred postoperatively in 3 of 20 patients and included transient worsening of preoperative aphasia, transient arm pain, and 1 event of asymptomatic cardiac demand ischemia.

Interobserver Reliability

The Pearson correlation coefficients for the volumetric analysis measurements are reported in Table 2. The correlation coefficients ranged from 0.82 to 0.997, demonstrating very strong correlation for all measurements ($p < 0.0001$).

TABLE 1. Demographics

Variable	Value (%)
Sex	
Female	9 (45)
Male	11 (55)
Age in yrs	
Min	23
Median	60.5
Mean	58.35
Max	81
Tumor type	
Glioblastoma	5 (25)
Metastatic carcinoma	6 (30)
Melanoma	2 (10)
Meningioma	2 (10)
Other	5 (25)
Tumor location	
Intraventricular	6 (30)
Frontal	4 (20)
Parietal	5 (25)
Temporal	4 (20)
Other	1 (5)
Retractor diameter in mm	
Min	14
Max	26
Retractor length in cm	
Min	3
Max	7

Illustrative Cases

Case 1

This 77-year-old woman harbored a left atrial meningioma. The trajectory taken during the surgery can be seen in Fig. 4A and B. The preoperative tumor volume was 19.5 cm³ and the preoperative FLAIR volume was 29.048 cm³. Figure 4C–E shows how the FLAIR signal can initially be selected to include the tumor volume but is subtracted out using advanced manipulations. A GTR was achieved in this patient, as no residual tumor volume could be identified postoperatively. The postoperative FLAIR volume was slightly increased compared with the preoperative FLAIR volume, at a volume of 34.40 cm³, as seen in Fig. 4F.

Case 2

This 58-year-old man harbored a glioblastoma (Fig. 5). The preoperative tumor volume was 57.53 cm³, and the preoperative FLAIR volume was 27.75 cm³. The postoperative tumor volume was 2.308 cm³, giving an extent of resection of 96%. The postoperative FLAIR volume was not significantly increased at 28.42 cm³. Figure 3C illustrates that the true DWI signal from ischemic tissue is bright on DWI and dark on the ADC map. This DWI hyperintensity tracks along the path of the retractor and has a volume of 8.029 cm³.

FIG. 3. Bar graph showing the tubular retractor lengths (upper) and diameters (lower) used in the cases. The lengths and diameters were not available for all cases

3D Volumetric Analysis

The average preoperative tumor volume was 16.25 ± 17.6 cm³ (Table 3). Volumes ranged from 0.22 to 61.52 cm³. The average extent of resection was 96.94%. We obtained a GTR in 75%, NTR in 10%, and STR in 15% of patients.

Volumetric Analysis of Vasogenic Edema (FLAIR Hyperintensity) and Cytotoxic Edema (Restricted Diffusion)

The average preoperative FLAIR hyperintensity was

TABLE 2. Interobserver correlations

Variable	r Value	p Value
Preop tumor vol	0.992	<0.00001
Preop FLAIR vol	0.997	<0.00001
Postop tumor vol	0.959	<0.00001
Postop FLAIR vol	0.996	<0.00001
Postop DWI vol	0.876	0.00009
Δ FLAIR vol	0.821	<0.00001

* Values in boldface are statistically significant.

FIG. 4. Case 1. **A:** Trajectory taken by the surgeon. **B:** In-plane/oblique image of the trajectory used for measuring depth of tumor from the pia. **C–E:** Subtraction of tumor volume from FLAIR. The orange outline is the preoperative tumor volume (C); and the orange outline is the FLAIR volume plus tumor volume (D). **E:** The preoperative FLAIR volume is calculated by subtracting out the preoperative tumor volume from the FLAIR plus tumor volume by advanced manipulations. **F:** The postoperative FLAIR volume surrounding the operative cavity.

$41.71 \pm 45.55 \text{ cm}^3$. This value ranged substantially from 0 to 157.54 cm^3 due to the wide variety of lesions. Postoperatively, the average FLAIR hyperintense signal volume was $43.75 \pm 41.61 \text{ cm}^3$. Although there was a small increase in average FLAIR intensity volume by $3.25 \pm 10.51 \text{ cm}^3$, this was not a statistically significant change in FLAIR volume ($p = 0.16$). The average postoperative volume of restricted diffusion on DWI was $8.35 \pm 3.05 \text{ cm}^3$. Assuming that volume of DWI-restricted diffusion was 0 preoperatively, this represented a statistically significant increase in DWI ($p < 0.001$).

Linear regression analysis was performed to assess whether the change in FLAIR hyperintensity and volume with restricted diffusion on DWI after surgery correlated with preoperative tumor volume, preoperative FLAIR volume, or depth of tumor. Based on a coefficient of determination (R^2), none of these factors explained a significant amount of variation in the changes in FLAIR or DWI signal after surgery (Table 3).

FIG. 5. Case 2. **A:** The preoperative tumor volume is outlined in orange. **B:** The postoperative residual tumor is outlined in blue. **C and D:** DWI hyperintensity postoperatively (C) lines up with the dark area on the ADC map (D).

Comparison With Traditional Retraction

Five patients were retrospectively identified as harboring deep-seated tumors that would have been suitable for resection using tubular retractors, but they underwent surgery prior to the use of that retraction system (Table 4). Two patients underwent GTR, 2 had NTR, and 1 had an STR. The average preoperative tumor volume in this cohort was $34.46 \pm 14.9 \text{ cm}^3$ (Table 5). The average change in FLAIR volume was $2.39 \pm 13.93 \text{ cm}^3$, which was not significantly different from the change in FLAIR volume seen in the tubular retractor cohort ($p = 0.9$). The average postoperative DWI volume was $16.51 \pm 8.9 \text{ cm}^3$. This increase in DWI volume showed a trend toward higher DWI volume in the traditional retractor compared with the tubular retractor cohort ($p = 0.16$); however, it did not reach statistical significance due to the small sample size.

Discussion

The use of tubular retractors for the resection of intracranial tumors, such as thalamic astrocytomas, was first demonstrated by Kelly and colleagues and expanded upon by Moshel et al.^{15,16,18} Since then, a number of case series/reports using a variety of retractors and stylets, such as transparent plastic sheaths with metal stylets¹⁹ or inflatable vinyl retractor tubes,¹² have been reported in the literature. Previously, we reported a series of 10 cases in which an METRx retractor system with frameless stereotactic navigation was safely used for resection of intracranial lesions. Fahim et al. further demonstrated the use of a similar system in a resection of a pediatric arteriovenous malformation.⁷ Almenawer et al. subsequently used a METRx retractor system in a 30-patient cohort.² These additional reports support the feasibility and reproducibility of the technique.

TABLE 3. Volumetric analysis and R² regression analysis

Variable	Value	R ² vs Δ in FLAIR	R ² vs Δ in DWI
Preop tumor vol in cm ³		0.003	0.016
Min	0.22		
Median	10.69		
Mean $\pm$ SD	16.25 $\pm$ 17.61		
Max	61.52		
Depth of tumor in mm		0.13	0.003
Min	20.80		
Median	48.80		
Mean $\pm$ SD	45.97 $\pm$ 12.67		
Max	70.10		
Extent of resection			
Min	74.93%		
Median	100.00%		
Mean $\pm$ SD	96.94 $\pm$ 7.16%		
Max	100.00%		
GTR	15 (75%)		
NTR	2 (10%)		
STR	3 (15%)		
Preop FLAIR vol in cm ³		0.17	0.14
Min	0.00		
Median	25.90		
Mean $\pm$ SD	41.71 $\pm$ 45.55		
Max	157.54		
Postop FLAIR vol in cm ³			
Min	2.90		
Median	31.41		
Mean $\pm$ SD	43.75 $\pm$ 41.61		
Max	142.54		
Δ FLAIR vol in cm ³			
Min	-17.41		
Median	4.39		
Mean $\pm$ SD	3.25 $\pm$ 10.51		
Max	30.26		
Postop DWI vol in cm ³			
Min	2.97		
Median	7.92		
Mean $\pm$ SD	7.53 $\pm$ 3.13		
Max	14.29		

The concept underlying the use of tubular retractor systems is that damage to the brain can be reduced by applying an equal distribution of pressure radially around a tube compared with the more concentrated pressure applied on smaller surface areas with older retraction methods. Studies in animals have shown that pressures as low as 25 mm Hg can cause electroencephalographic changes⁵ and those as low as 30 mm Hg can reduce cerebral blood flow.²⁴ Furthermore, with the use of progressive dilation using blunt instruments, there is less transection of white matter tracts.

TABLE 4. Traditional retractor control group

Variable	Value*
Sex	
Female	3 (60)
Male	2 (40)
Age in yrs	
Min	27
Median	48
Mean	50.6
Max	75
Extent of resection	
Min	85.94
Median	99.45
Mean $\pm$ SD	95.55 $\pm$ 6.28
Max	100.00
GTR vs NTR vs STR	
GTR	2 (40)
NTR	2 (40)
STR	1 (5)

* Values are presented as the number of patients (%) unless stated otherwise.

Although these theories have been suggested, most studies in humans have been limited to only assessing the feasibility and safety of these retractor systems. Outcomes have by and large been subjective, surgeon-dependent assessments of GTR versus STR, patient length of stay, and occurrence of postoperative complications. Objective methods such as those used in the aforementioned animal studies to assess retraction damage have not been feasible as measures in human populations. This study not only expands on our original case series but represents the first report of the use of 3D volumetric analysis to objectively measure extent of resection as well as quantify cortical damage associated with the use of tubular retractors.

Using Brainlab software, 3D volumetric analysis confirmed that tubular retractors could be successful in achieving GTR of tumors of a variety of pathologies at a variety of depths, locations, and preoperative volumes. Changes in hyperintensity on FLAIR and areas with restricted diffusion on DW images were used to assess postoperative vasogenic edema and cytotoxic edema (ischemia/cellular damage), respectively. Vasogenic edema from trauma results from damage to the blood-brain barrier and causes swelling in the brain with an increase in T2 FLAIR signal that does not show restricted diffusion.²⁶ In our data, no statistically significant increase in FLAIR signal occurred after surgery. This suggests that the blood-brain barrier was not significantly damaged and there was not a significant increase in vasogenic edema postoperatively, which supports the concept that tubular retractors are potentially less traumatic than other retraction techniques. However, there are no data available in the literature for other spatula-based techniques for comparison. This study provides a benchmark for comparison.

In spite of the minimal and nonsignificant increase in FLAIR signal, our data showed significant increases in ar-

TABLE 5. Traditional retractor volumetric analysis

Variable	Value
Preop tumor vol in cm ³	
Min	11.89
Median	36.42
Mean $\pm$ SD	34.46 $\pm$ 14.95
Max	50.31
Depth of tumor in mm	
Min	23.40
Median	36.80
Mean $\pm$ SD	35.10 $\pm$ 7.93
Max	42.30
Preop FLAIR vol	
Min	21.69
Median	56.57
Mean $\pm$ SD	55.33 $\pm$ 28.63
Max	98.10
Postop FLAIR vol in cm ³	
Min	28.44
Median	50.93
Mean $\pm$ SD	57.71 $\pm$ 25.47
Max	93.31
Δ FLAIR vol in cm ³	
Min	-18.21
Median	6.75
Mean $\pm$ SD	2.39 $\pm$ 13.93
Max	15.95
Postop DWI vol in cm ³	
Min	5.71
Median	16.64
Mean $\pm$ SD	16.51 $\pm$ 8.89
Max	27.07

eas with restricted diffusion on ADC maps after surgery. This signal correlates with cytotoxic edema resulting from ischemic tissue and cell damage.²⁶ These results also indicate that FLAIR signal alone may not be an adequate measure of cortical injury after surgery. Although some of the increase in DWI signal occurred around the site of the resected mass, indicating trauma due to surgery and not the retractor, the majority of the signal surrounded the corridor created by the tubular retractor, as seen in 3D reconstructions of the images (Fig. 2). Since it would be difficult and somewhat subjective to differentiate the 2 causes of injury, we measured them together, which may artificially increase the reported amount of cytotoxic injury caused by the retractor. Nevertheless, this suggests that the tubular retractors, despite the equal distribution of pressure on brain tissue, may cause enough trauma to result in some degree of cytotoxic injury to surrounding white matter.

Comparison with traditional retractors is the next step in understanding the clinical relevance of these findings. Retrospectively, we were able to identify only a small co-

hort of patients in our operative caseload who underwent surgery for deep-seated lesions with traditional retractors, but would have been suitable for tubular retractors. Comparing the cohort of tubular retractors to traditional retractors, we found no difference in the change in FLAIR signal but found a nonsignificant trend toward higher DWI volumes in the traditional retractor group. This small control group was not statistically powered to test the hypothesis that tubular retractors cause less damage than traditional retractors; however, it does demonstrate a need for further comparison of surgical retractors by randomized, controlled prospective trials using the quantitative volumetric methods described. Moreover, the increase in cytotoxic edema found around the retractors should serve as an impetus for future biomedical engineering designs to improve this technology. One possible solution might be to develop softer and more flexible tubes that may cause less injury. Another possibility is the dilation method, which could be done in a less traumatic fashion with the aid of an internal expander. Our data suggest that objective tools such as 3D volumetric analysis of DWI may be a useful technique to assess postoperative damage of tubular retractors and can be used in future studies as an outcome measure to compare with other retraction methods.

Since our group has exclusively used tubular retractors since 2005, one limitation of our study was the lack of an adequately powered traditional retractor control group. Another limitation includes the lack of preoperative DW images for comparison. We cannot conclude whether tubular retractors minimize damage compared with other retraction methods; however, we demonstrate evidence that this analysis method can be used to test this hypothesis in future prospective studies.

Conclusions

We provide objective evidence that the use of tubular retractors to remove deep-seated intraparenchymal tumors does not significantly increase FLAIR signal in the brain, indicating minimal trauma to surrounding white matter. However, increases in DWI signal were identified, demonstrating that cytotoxic injury still occurs around rigid tubular retractor systems. Further refinements and improvements in these systems are needed.

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Disclosure

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Author Contributions

Conception and design: Schwartz, Bander, Kovanlikaya. Acquisition of data: all authors. Analysis and interpretation of data: all authors. Drafting the article: Schwartz, Bander, Kovanlikaya. Critically revising the article: Schwartz, Bander, Kovanlikaya. Reviewed submitted version of manuscript: Schwartz, Bander, Kovanlikaya. Approved the final version of the manuscript on behalf of all authors: Schwartz. Statistical analysis: Schwartz, Bander. Study supervision: Schwartz.

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