Use of Urinary Biomarkers to Quantify Degree of Renal Parenchymal and Urothelial Damage During Ureteroscopy
PI: Mantu Gupta
NCT05350423
Document Date: 1-10-2023

Use of Urinary Biomarkers to Quantify Degree of Renal Parenchymal and Urothelial Damage During Ureteroscopy

Protocol Number*: 21-00084

Principal Investigator*: Mantu Gupta MD.

Sponsor: Mount Sinai, Department of Urology

Grant Title: Use of Urinary Biomarkers to Quantify Degree of Renal Parenchymal and Urothelial Damage During Ureteroscopy

Funded by: New York Academy of Medicine

Version Date: 23 August 2021

INVESTIGATOR'S SIGNATURE

The signature below constitutes the approval of this protocol and provides the necessary assurances that this study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable US federal regulations and ICH guidelines, as described in the *Statement of Compliance* above.

Principal Investigator or Clinical Site Investigator:

Signed: Mantu Gupta Date: 1/14/2021

Name*: Mantu Gupta, MD.

Title*: Chairman of Urology, Mount Sinai West

Chairman of Urology, Mount Sinai Morningside

Director of Endourology, Mount Sinai Health Care System

Director, Mount Sinai Kidney Stone Center

Professor of Urology, Icahn School of Medicine at Mount Sinai

Investigator Contact Information
Affiliation*: Icahn School of Medicine at Mount Sinai

Address: [redacted]

New York, NY 10019

Telephone: (212) 241-1272

Email: drmantugupta@gmail.com

Effective Date: 1/10/2023
End Date:1/9/2024

1.1 SYNOPSIS

Title:	**Selection process and evaluation of endourology fellows**
Grant Number:	
Study Description:	While ureteroscopic laser lithotripsy (URS) is a well-established operative technique, new modalities exist with regards to laser energy in URS. This study will attempt to quantify the degree of renal damage associated with different intraoperative variables by measuring urinary biomarkers specific for renal damage in order to determine a safety profile for different intraoperative variables during URS.
Objectives*:	**Primary Objectives:** 1) To determine if during ureteroscopic laser lithotripsy, laser type (Ho:Yag Moses 2.0 vs Thulium Fiber Laser) predicts differences in renal-damage associated biomarkers **Secondary Objectives:** To determine if any additional operative factors (i.e. operative time, patient positioning, use of pressurized irrigation, use of a ureteral access sheath) predict differences in levels of renal-damage associated biomarkers.
Endpoints*:	**Primary Endpoint**: The primary endpoints will be: 1) Determination of association between type of laser used during URS and degree of renal damage (as measured by urinary biomarkers). **Secondary Endpoint:** The secondary endpoint will be the determination of association between additional operative variables (see secondary objective) and renal damage during URS.
Study Population:	*Specify the sample size, gender, age, demographic group, general health status, and geographic location.* We anticipate a sample size of 100 subjects enrolled in this study. We will not discriminate by gender, demographic group, or general health status. All patients will be over the age of 18 years and from the general New York area.
Phase* or Stage:	*Indicate Phase or Stage, as appropriate. Institutes and Centers may differ in their preferences for categorizing research. Consult with your Program Official (PO)* N/A

Description of Sites/Facilities Enrolling Participants:	*Provide a brief description of planned facilities/participating sites enrolling participants. Indicate general number (quantity) of sites only and indicate if the study is intended to include sites outside of the United States* All procedures and subject consent will be performed at the following locations: [redacted] which are all Mount Sinai approved sites in which the PI practices and operates.
Description of Study Intervention/Experimental Manipulation:	*Describe the study intervention (a.k.a, experimental manipulation; hereafter referred to as "study intervention"). Include intervention dose (length and frequency) and how it will be administered. Include method of delivery (e.g., group vs. individual, web-based, etc.).* Subjects enrolled will undergo surgical treatment of their kidney stones with URS based on clinician judgement and urologic guidelines. Once enrolled, patients will be randomized into one of two groups based on the type of laser used to break apart the stones: Thulium Laser or Hol:YAG Laser. Both lasers are used routinely and interchangeable for this procedure according to the standard of care. The only involvement of patients will be collection of urine samples before and after surgery.
Study Duration*:	*Estimated time (in months) from when the study opens to enrollment until completion of data collection.* 12-18 months
Participant Duration:	*Time (e.g., in months) it will take for each individual participant to complete all study-related tasks.* 10 days

1.2 SCHEMA

Patient eligibility will be confirmed during initial presentation of kidney stone, wherein Dr. Gupta will determine if the patient is eligible to participate based on their clinical characteristics. Dr. Gupta will determine if URS will be performed based upon available imaging studies and urologic standard of care. If so, Dr. Gupta will explain the specifics of the study to the patient and ask if he or she is willing to participate. All patients will have pre- and postoperative urine samples collected which will be stored and analyzed for protein biomarkers at a later date.

A member of the research team will approach the patient after Dr. Gupta has introduced the study and the patient agrees to learn more. Once informed consent has been achieved, patients will be randomly placed into one of two groups that will indicate which type of laser lithotripsy they will receive:

1) Hol:YAG laser
2) Thulium laser

1.3 SCHEDULE OF ACTIVITIES

After randomization, the patient will undergo URS using either the Thulium or Hol:YAG laser. Both lasers are used as part of the standard of care and are used interchangeably. At the beginning of all procedures, the surgeon will empty the bladder and collect a 2 mL sample of urine that will serve as the "Preoperative Urine Sample". It is the standard of care to collect urine at the beginning of the surgery to send to pathology. The only deviation is that a portion of the urine will be for research. One hour following the end of the procedure, a member of Dr. Gupta's research team will collect a second 2 mL sample of urine that will serve as the "Immediate Post-Op Urine Sample". This will be collected using the catheter that was placed during surgery. If no catheter was placedb the patient will provide the urine sample.

After collection, the samples will be subsequently frozen in a freezer at -80 ºC freezer operated by the pathology department at Mount Sinai West hospital.

A third and final sample of urine will be collected at the patients' 10-day follow up appointment for stent removal which will serve as the "Follow-up Urine Sample". To standardize the duration in which the last urine samples are collected, only patients who have been stented will be included in the study, as standard of care for stent removal is ten days. Upon receiving the third and final specimen 10 days postoperatively, the subject's enrollment will be considered complete. All procedures and appointments are the standard of care.

After sufficient patient enrollment and specimen collection, all the samples will be analyzed for kidney and urothelial injury biomarkers using urinary assays (ELISA, CLIA, etc.). The biomarkers that will be qualified and quantified in the urine include Cystatin C, Kidney Injury Molecule-1 (KIM-1), Neutrophil gelatinase-associated lipocalin (NGAL), Beta-2-microglobulin, Microalbumin urine, spot creatinine, total protein, and glycosaminoglycans.

2 INTRODUCTION

2.1 STUDY RATIONALE

State the problem or question (e.g., describe the population, disease, current standard of care, if one exists, and limitations of knowledge or available therapy), the reason for conducting the clinical trial and the rationale underlying the intervention. State the name and the nature of the intervention, the hypothesized target(s) of the intervention (i.e., the putative cognitive, affective, behavioral, social, community,

organizational, etc., target necessary to produce the behavior change relevant to the clinical outcome), and the clinical outcome of interest.

Standard operations used for the surgical removal of kidney stones include ureteroscopic laser lithotripsy (URS) and percutaneous nephrolithotomy (PCNL). In recent years new modalities have been introduced to both URS and PCNL including new laser technologies such as the Thulium laser and antiretropulsion properties with Holmium lasers (Moses technology). While the overall safety and efficacy of these procedures has been well established, these relatively new modalities have yet to be rigorously studied for potential deleterious effects on the kidney. To date, several studies have attempted to use these biomarkers to determine degree of tissue injury after kidney stone operations, however these studies have been limited by small cohorts and lack of data regarding the newest energy modalities and surgical techniques [1-6]. This study is intended to determine whether different intraoperative variables (type of laser, patient positioning, renal pelvis pressure) correlate with renal damage, as measured by urinary biomarkers.

2.2 BACKGROUND

Nephrolithiasis is one of the most common urologic conditions with a lifetime incidence in the United States approaching 10% and rising [7]. While many kidney stones will pass spontaneously, a significant portion ultimately require surgical intervention in the form of ureteroscopic laser lithotripsy (URS), percutaneous nephrolithotomy (PCNL), or shock wave lithotripsy (SWL) to eradicate the stone burden. The ideal surgical procedure for eradication of kidney stones would have maximal efficiency in stone removal while minimizing collateral damage to the renal parenchyma and the epithelial lining of the collecting system (urothelium). While much is known about the physical properties of stone fragmentation techniques, relatively little is known about the unwanted effect these techniques may have on renal tubular and urothelial cells.

In daily practice, clinicians almost exclusively use a patient's estimated glomerular filtration rate (eGFR), obtained by measuring serum creatinine as a biomarker of renal parenchymal injury. The drawbacks of using serum creatinine as a surrogate for renal function are well known. First, serum creatinine can change with many confounding variables that are unrelated to kidney function such as diet, hydration status, and muscle mass to name a few. More importantly, serum creatinine (eGFR) is considered more a measure of global renal function and has a poor sensitivity for detecting early or small insults to renal tubular cells. In other words, eGFR does not change in any detectable way until a relatively large insult to the kidney has taken place. Lastly, eGFR is a better measure of renal blood flow and is not a direct marker of cellular injury. In recent years, several urinary protein biomarkers have been found that are now known to be markers of renal cellular injury. When compared to eGFR, these markers can detect renal insults earlier and have significantly higher sensitivity. Indeed, they can be used as early predictors of impending acute kidney injury (AKI) [8].

Kidney stone removal operations such as URS involve a number of modifiable factors which may contribute to the degree of renal or urothelial injury. The goal of this study is to use sensitive urinary biomarkers to determine which aspects of URS are associated with the greatest degree of injury to renal tubular cells and urothelium. In this study, we will quantify urinary biomarkers specific for kidney and urothelial injury before and after URS and correlate these markers to different intraoperative variables. We postulate that our results will shed a new light on urological endoscopic procedures.

2.3 RISK/BENEFIT ASSESSMENT

2.3.1 KNOWN POTENTIAL RISKS

There will be no risks to physical harm of the subjects due to study interventions. Both lasers are used interchangeably during URS for kidney stones. The only other intervention performed solely for the purpose of the study is collection of postoperative urine samples. Pre-operative urine samples and follow up urine samples are routinely collected from Dr. Gupta's surgical patients. Surgical intervention and follow up for kidney stones will be performed regardless of a patient's willingness to participate in this study according to the standard of care. Since we will be storing patient samples and data, there is a small risk that personal data will be compromised. This risk will be mitigated by encryption of all data storage devices and use of unique numerical patient identifiers, which will be separately linked to the patient's protected health information.

2.3.2 KNOWN POTENTIAL BENEFITS

There are no potential benefits to participants.

2.3.3 ASSESSMENT OF POTENTIAL RISKS AND BENEFITS

Other than the extremely small risk of compromising protected health information, we do not foresee any risks or benefits to participants in this study. The extent of participation for each subject involves collection of three perioperative urine samples (collected via cystoscope, Foley catheter, or clean void). No invasive procedure will be performed to collect the samples (for example, a Foley catheter will not be placed solely for the purpose of collecting a urine sample for this study).

OBJECTIVES AND ENDPOINTS

OBJECTIVES	ENDPOINTS	JUSTIFICATION FOR ENDPOINTS	PUTATIVE MECHANISMS OF ACTION
Primary			

OBJECTIVES	ENDPOINTS	JUSTIFICATION FOR ENDPOINTS	PUTATIVE MECHANISMS OF ACTION
1) To determine if during ureteroscopic laser lithotripsy, laser type (Ho:Yag Moses 2.0 vs Thulium Fiber Laser) predicts differences in renal-damage associated biomarkers	1) Determination of association between type of laser used during URS and degree of renal damage (as measured by urinary biomarkers).	Quantification of the concentrations of specific urinary biomarkers will allow us to compare the relative tissue damage caused by each laser type.	*N/A*
Secondary			
To determine if any additional operative factors (i.e. operative time, patient positioning, use of pressurized irrigation, use of a ureteral access sheath) predict differences in levels of renal-damage associated biomarkers.	The secondary endpoint will be determination of association between additional operative variables (see secondary objective) and renal damage during URS.	Quantification of the concentrations of specific urinary biomarkers will allow us to compare the relative tissue damage caused by different variables during URS.	*N/A*

3 STUDY DESIGN

3.1 OVERALL DESIGN

The overall design is to evaluate the degree of renal damage caused by different laser energy modalities and access sheath calibers used during URS. Pre- and postoperative urine samples will be collected from subjects to quantify urinary biomarkers which are specific for renal damage.

Subjects enrolled will be randomized into two arms, one in which subjects will undergo laser lithotripsy with the Thulium fiber laser (TFL), and the other in which the procedure will be performed with the Holmium:YAG laser.

3.2 SCIENTIFIC RATIONALE FOR STUDY DESIGN

Randomization by laser type is being performed. We chose to do this as this would eliminate any potential bias on the part of the surgeon for preference of one laser type over another. Both lasers are used interchangeable during URS for kidney stones, and surgeons do not typically choose a specific laser to use prior to surgery. We believe that from the subjects' standpoint, there is little to no perceived difference in use of TFL or Ho:YAG lasers, and both are used interchangeably as standard of care.

3.3 JUSTIFICATION FOR INTERVENTION

Surgical interventions for kidney stones will be performed regardless of participation in this study.

3.4 END-OF-STUDY DEFINITION

This investigation will finish once all urine samples have been collected and analyzed for urinary biomarkers for both surgical cohorts.

4 STUDY POPULATION

4.1 INCLUSION CRITERIA

Inclusion Criteria:

- Age ≥ 18 years
- Solitary kidney stone within the ureter or kidney < 2cm
- Scheduled to undergo URS with Dr. Gupta

4.2 EXCLUSION CRITERIA

-Exclusion Criteria:

- Multiple or bilateral renal/ureteral stones
- Staghorn calculi
- History of chronic kidney disease or ESRD
- Presence of indwelling ureteral stent or nephrostomy tube at time of recruitment
- Urinary tract infection (cystitis or pyelonephritis) within 2 weeks of recruitment
- Development of postoperative septic shock (defined as persistent need for pressors >1 hour after end of procedure)
- Documented ureteral/renal pelvis perforation

4.3 STRATEGIES FOR RECRUITMENT AND RETENTION

Patients will be recruited from the practice of Dr. Mantu Gupta. Dr. Gupta has one of the busiest kidney stone practices in the area providing ample patients to recruit into the study. We do not anticipate any issues with retention as patient involvement will occur at two visits. The first 2 of 3 total urine samples will be collected on the day of the surgical intervention. The 3rd and final urine sample will be collected at an obligatory outpatient follow up visit. The follow up visit is considered obligatory as patients will have their ureteral stents removed at this visit, at which time urine samples may be collected.

5 STUDY INTERVENTION(S) OR EXPERIMENTAL MANIPULATION(S)

5.1 STUDY INTERVENTION(S) OR EXPERIMENTAL MANIPULATION(S) ADMINISTRATION

5.1.1 STUDY INTERVENTION OR EXPERIMENTAL MANIPULATION DESCRIPTION

For subjects receiving URS, randomization will occur preoperatively. These subjects will be randomized to undergo lithotripsy with either Ho:YAG or TFL lasers (100 subjects in each arm).

All subjects will undergo surgical interventions that abide by broadly accepted guidelines and standards of care, allowing for slight variations in technique as seen necessary by the attending surgeon Dr. Gupta.

The type of laser that is chosen in SOC is based on hospital availability. That is, most hospitals only have one laser type to choose from (and that laser type is generally contingent on the biomedical contracts the hospital holds with the various laser manufacturers). We are uniquely situated at Mount Sinai West in that we have multiple laser systems to choose from. Multiple surgeons share the lasers, and typically Dr. Gupta will use whichever laser is already in the room or readily available. Approximately 50% of ureteroscopies were performed with each laser, +/- 10%.

5.2 FIDELITY

5.2.1 INTERVENTIONIST TRAINING AND TRACKING

Dr. Mantu Gupta, professor of urology at Mount Sinai West Hospital will be performing the procedures. Dr. Gupta is a world-renowned expert in these operative techniques.

5.3 MEASURES TO MINIMIZE BIAS: RANDOMIZATION AND BLINDING

Patients will be randomized to receive either Thulium or Holmium laser lithotripsy. Patients will be blinded as they will be anesthetized during the operation. Blinding of the operators would not be possible owing to the differences in laser modalities.

We will utilize random.org which is a validated tool that utilizes atmospheric noise to generate random numbers. Each enrolled patient will be given a subject ID (1-108, consecutively assigned) and these study IDs will be entered as string of integers to random.org and randomized prior to the initiation of the study.

This process will determine group assignments. This will ensure both true randomization and equal allocation… The laser systems are bulky and require transfer into and out of the operating room preoperatively as the OR staff prepares for the upcoming case. Thus, though it only takes a few minutes to actually switch the laser systems, we tend to use the laser that is already in the operating room. The purpose of randomization is to remove this 'last case' bias and ensure equal distribution of laser the modality used.

6 STUDY INTERVENTION/EXPERIMENTAL MANIPULATION DISCONTINUATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

6.1 DISCONTINUATION OF STUDY INTERVENTION/EXPERIMENTAL MANIPULATION

No discontinuation will take place as the surgical procedures being performed are part of subjects' standard clinical care.

6.2 PARTICIPANT DISCONTINUATION/WITHDRAWAL FROM THE STUDY

Patients will be considered enrolled in the study once consent has been signed. Participants may discontinue or withdraw from the study for any reason and at any point in time through the conclusion of the study. If a participant desires withdrawal after some or all of their urine samples have been collected or analyzed, their urine samples will be discarded in a biohazard-safe and anonymous fashion.

6.3 LOST TO FOLLOW-UP

A single follow up visit is required to complete enrollment criteria for this study. Participants who fail to show up to their first follow up appointment for stent removal will be automatically withdrawn from the study and previously collected urine samples will be discarded as above.

7 STUDY ASSESSMENTS AND PROCEDURES

7.1 ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

7.1.1 DEFINITION OF ADVERSE EVENTS

Any untoward medical occurrence, unintended sign, symptom, illness or disease temporally associated with the study protocol regardless of the suspected cause.

7.1.2 DEFINITION OF SERIOUS ADVERSE EVENTS

An Adverse Event that is considered "serious" if it meets at minimum one of the three Seriousness reporting criterions below:

1. Led to death,
2. Led to a serious injury which:
    a. Resulted in a life-threatening illness or injury, or
    b. Resulted in a permanent impairment of a body structure or a body function, or
    c. Resulted in medical or surgical intervention to prevent life-threatening illness or injury or permanent impairment to a body structure or a body function

7.1.2.1 RELATIONSHIP TO STUDY INTERVENTION/EXPERIMENTAL MANIPULATION

All AEs will be assessed for relationship to the study protocol based on the following definitions:

Not Related: There is no clear evidence that the AE has a relationship to the study protocol and can be attributed to an underlying or concurrent illness/clinical condition or an effect of another device, drug or treatment.

Related: There is a clear causal relationship of the AE to the marketed device or procedure beyond reasonable doubt.

7.1.3 TIME PERIOD AND FREQUENCY FOR EVENT ASSESSMENT AND FOLLOW-UP

All enrolled subjects will be monitored for adverse events by review of the medical record on a monthly basis by Dr. Gupta. Subjects will have routine follow up scheduled 10 days postoperatively at which time in-person visit will be performed and assessment of adverse events may be performed.

7.1.4 ADVERSE EVENT REPORTING

Adverse events must be reported to the IRB as soon as possible and no later than **2 working days** after the PI first becomes aware of the event. The PI or designee must record all AE information that can be gathered within the reporting timeframe and enter it onto the AE eCRF.
Relevant follow-up information should be submitted to the IRB as soon as it becomes available and/or upon request.

7.1.5 SERIOUS ADVERSE EVENT REPORTING

See section 7.1.4

7.1.6 REPORTING EVENTS TO PARTICIPANTS

All SAE's or AE's will be reported to affected participants by the principal investigator directly.

8 STATISTICAL CONSIDERATIONS

8.1 STATISTICAL HYPOTHESES

After sufficient patient enrollment and specimen collection, all the samples will be analyzed for kidney and urothelial injury biomarkers using urinary assays (ELISA, CLIA, etc.). The biomarkers that will be qualified and quantified in the urine include Cystatin C, Kidney Injury Molecule-1 (KIM-1), Neutrophil gelatinase-associated lipocalin (NGAL), Beta-2-microglobulin, Microalbumin urine, spot creatinine, total protein, and glycosaminoglycans.

Abundance of each kidney injury biomarker for each laser will be analyzed using the Kruskal-Wallis statistical test. Multiple multivariate regression will also be used to control for stone size, operative time, and other potential confounders.

8.2 SAMPLE SIZE DETERMINATION

A sample size of 108 patients will be accrued locally. We are enrolling 108 patients to allow for any potential exclusions due to failure to follow up or for patients who were not stented.

A study by Fahmy et al. prospectively evaluated urinary Kidney Injury Marker-1 (uKIM-1) levels in patients undergoing kidney stone surgery to compare the effect of Extracorporeal Shockwave Lithotripsy (ESWL) and Ureteroscopy/Laser Lithotripsy (URS) on biomarkers of renal damage1. In this study, the 50 patients in the ESWL arm had a mean preoperative uKIM-1 of 5.78ng/mL (SE=0.8) and the 10 patients in the URS arm similarly had a mean preoperative uKIM-1 of 5.78ng/mL (SE=2.0). Two hours post operatively, the ESWL group had a mean uKIM1 of 10.14ng/mL (SE=1.4) and the URS group had a uKIM-1 of 5.49ng/mL (SE 0.7, SD=2.21). Another study by Timmeren et al evaluated uKIM1 in 11 health controls and 53 patients with varying degrees of kidney disease and found a -0.37 correlation (p=0.016) between uKIM-1 levels and eGFR.2 That is, a 1ng/mL rise in uKIM1 correlated to a 0.37mL/min decline in eGFR. The purpose of our statistical analysis is to evaluate for non-inferiority of the two laser modalities in regard to occult kidney damage. Given that kidney stone disease is a recurrent illness often requiring multiple surgical interventions over the course of a patient's lifetime, we defined non-inferiority as producing a change in eGFR 1mg/mL). With an allocation ratio of 1:1, 99% power, non-inferiority limit of 40%, and alpha of 5%, using the noninferiority power calculation, our sample size is 49 patients in each study arm for 98 patients in total (~100). Based on these calculations and factoring in a 10% dropout rate, we seek to enroll 108 total patients.

Power Calculation References

1. Fahmy N, Sener A, Sabbisetti V, et al. Urinary expression of novel tissue markers of kidney injury after ureteroscopy, shockwave lithotripsy, and in normal healthy controls. Journal of endourology. 2013;27(12):1455-1462.

2. van Timmeren MM, van den Heuvel MC, Bailly V, Bakker SJ, van Goor H, Stegeman CA. Tubular kidney injury molecule-1 (KIM-1) in human renal disease. The Journal of pathology. 2007;212(2):209-217.

8.3 POPULATIONS FOR ANALYSES

All enrolled subjects will be included for analyses.

8.4 STATISTICAL ANALYSES

8.4.1 GENERAL APPROACH

The biomarkers that will be qualified and quantified in the urine include Cystatin C, Kidney Injury Molecule-1 (KIM-1), Neutrophil gelatinase-associated lipocalin (NGAL), Beta-2-microglobulin, Microalbumin urine, spot creatinine, total protein, and glycosaminoglycans.

8.4.2 ANALYSIS OF THE PRIMARY ENDPOINT(S)

Abundance of each kidney injury biomarker for each laser will be analyzed using the Kruskal-Wallis statistical test. Multiple multivariate regression will also be used to control for stone size, operative time, and other potential confounders.

8.4.3 ANALYSIS OF THE SECONDARY ENDPOINT(S)

Operative time, patient positioning, use of pressurized irrigation, and use of a ureteral access sheath) will be analyzed with Kruskal-Wallis statistical tests and multivariate regression to predict differences in levels of renal-damage associated biomarkers.

9 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

9.1 REGULATORY, ETHICAL, AND STUDY OVERSIGHT CONSIDERATIONS

9.1.1 INFORMED CONSENT PROCESS

The standard informed consent process for research outlined in SOP HRP-090 will be followed.

9.1.1.1 CONSENT PROCEDURES AND DOCUMENTATION

Subjects will be recruited from Dr. Mantu Gupta's practice. Patients will be recruited during initial presentation to the office. Consent will be obtained at the initial office visit when decision to proceed to surgery has been made.

Potential subjects will be informed of the study at their preoperative visit (see above point) and may sign the consent at that time if they feel comfortable. If subjects require time for further contemplation, the consent may be signed on the day of surgery in the preoperative area (prior to administration of any anesthesia). This gives potential subjects at least 24 hours to consider the study and review the consent form.

Given the simplicity of the nature of subject involvement, we anticipate that no more than 5 minutes will generally be required to explain the consent form. However, longer and more extensive discussions will be available to those who request or require. All potential subjects will be verbally informed that their participation is completely optional and that their decision of whether or not to participate does not impact their care in any manner. Subjects will be asked to verbally repeat and summarize their involvement in the study. A copy of the consent form will be provided to the subject directly after signing the form.

9.1.2 CONFIDENTIALITY AND PRIVACY

The data will be housed in Mount Sinai's REDCap database. The data is anonymous, and no identifiers are collected. The data will be stored with protected passwords. The data will be reported to the sites in aggregate and anonymized. The publication will also not mention any center by name. The results in the publication will be in aggregate form and anonymous.

Urine and blood samples will be kept in a secure freezer in the Mount Sinai West pathology lab, and then will be transported to our main lab at the Icahn School of medicine for analysis. Only study staff will have access to these samples.

9.1.3 FUTURE USE OF STORED SPECIMENS AND DATA

The data will be deleted 6 years after publication, per regulations. Data will not be shared with any outside organizations. Urine specimens will be stored with identifiable information removed. After this removal, samples may be the information and/or samples may be used for future research studies or shared

with other research teams for future research studies. Subjects will not be informed of the details of specific research that is done with the specimens.

9.1.3.1 DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

Data collection will be the responsibility of the research staff at the site under the supervision of the site investigator. The investigator will be responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

All source documents will be completed in a neat, legible manner to ensure accurate interpretation of data.

Hardcopies of the study visit worksheets will be provided for use as source document worksheets for recording data for each participant consented/enrolled in the study. Data recorded in the electronic case report form (eCRF) derived from source documents will be consistent with the data recorded on the source documents.

Clinical data (including adverse events (AEs), concomitant medications, and expected adverse reactions data) and clinical laboratory data will be entered into the electronic data capture (EDC) system, a 21 CFR Part 11-compliant data capture system. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

The data safety will be monitored by the PI

9.1.3.2 STUDY RECORDS RETENTION

.

Study documents will be retained until at least 6 years have elapsed since the formal discontinuation of clinical development of the study intervention. These documents should be retained for a longer period, however, if required by local regulations. No records will be destroyed without the written consent of the principal investigator.

The data will be kept until its publication at most six years.

9.1.4 PROTOCOL DEVIATIONS

This protocol defines a protocol deviation as any noncompliance with the clinical trial protocol, International Council on Harmonisation Good Clinical Practice (ICH GCP), or Manual of Procedures (MOP) requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions will be developed by the site and implemented promptly.

It will be the responsibility of the site investigator to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity. All deviations will be addressed in study source documents. Protocol deviations will be sent to the reviewing Institutional Review Board (IRB) per their policies. The site investigator will be responsible for knowing and adhering to the reviewing IRB requirements.

The PI will be responsible for any vigilance and monitoring of the data.

10 FUNDING

Funding for this study is provided in part by the New York Academy of Medicine through the Ferdinand C. Valentine Fellowship award for Research in Urology. This award is to support the research training of Dr. Alan Yaghoubian during his fellowship with Dr. Mantu Gupta.

Dr. Yaghoubian was awarded the grant in 2021 to pursue this research study. The aims and objectives of the study remain the same, and the funding will support the research activities outlined in the above protocol.

Dr. Gupta remains the PI for this study, and Dr. Yaghoubian is the funding PI because the grant is specifically awarded to those pursuing a fellowship in Urology.

11 REFERENCES

Include a list of relevant literature and citations for all publications referenced in the text of the protocol. Use a consistent, standard, modern format, which might be dependent upon the required format for the anticipated journal for publication (e.g., N Engl J Med, JAMA, etc.). The preferred format is International Committee of Medical Journal Editors (ICMJE).

1. Villányi KK, Székely JG, Farkas LM, Jávor E, Pusztai C. Short-term changes in renal function after extracorporeal shock wave lithotripsy in children. J Urol. 2001 Jul;166(1):222-4.
2. Nasseh H, Abdi S, Roshani A, Kazemnezhad E. Urinary Beta-2Microglobulin: An Indicator of Renal Tubular Damage after Extracorporeal Shock Wave Lithotripsy. Urol J. 2016 Dec 8;13(6):2911-2915.

3. Jobs K, Straż-Żebrowska E, Placzyńska M, et al. Interleukin-18 and NGAL in assessment of ESWL treatment safety in children with urolithiasis. Cent Eur J Immunol. 2014;39(3):384-391.
4. Daggülli M, Utangaç MM, Dede O, Bodakci MN, Hatipoglu NK, Penbegül N, Sancaktutar AA, Bozkurt Y, Söylemez H. Potential biomarkers for the early detection of acute kidney injury after percutaneous nephrolithotripsy. Ren Fail. 2016;38(1):151-6.
5. Sharifiaghdas F, Kashi AH, Eshratkhah R. Evaluating percutaneous nephrolithotomy-induced kidney damage by measuring urinary concentrations of β2-microglobulin. Urol J. 2011 Fall;8(4):277-82.
6. Balasar M, Pişkin MM, Topcu C, Demir LS, Gürbilek M, Kandemir A, Öztürk A. Urinary kidney injury molecule-1 levels in renal stone patients. World J Urol. 2016 Sep;34(9):1311-6.
7. Griffin BR, Faubel S, Edelstein CL. Biomarkers of Drug-Induced Kidney Toxicity. Ther Drug Monit. 2019 Apr;41(2):213-226.
8. Beker BM, Corleto MG, Fieiras C, Musso CG. Novel acute kidney injury biomarkers: their characteristics, utility and concerns. Int Urol Nephrol. 2018 Apr;50(4):705-713.