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Endourology Society 2019 Summer Student Scholarship

Project Summary: Evaluation of Renal Papillary Regeneration in a Porcine Model

Statement of Appreciation:
I would like to express my gratitude to the Endourology Society for supporting my research project this summer. The Society's financial generosity made this resource intensive project possible- a project that I hope will meaningfully influence the clinical practice of endourology. Furthermore, I am thankful for the Endourology Society and my faculty mentor's endorsement of my continued pursuit of a career in the field of urology. It is my goal to be able to one day support medical students in the same way that I have been supported.

Introduction:
Percutaneous nephrolithotomy (PCNL) is the first line therapy for treating large renal stones. When performing a PCNL it is required to create a percutaneous tract into the renal collecting system such that endoscopes can be inserted into the kidney and the renal stone can be fragmented and removed. These tracts are created at the discretion of the surgeon and traverse the renal papilla, calyceal fornix, or infundibulum. Recent work in our laboratory has identified the central area of the renal papilla as the least vascular of these three structures. However, it is unknown how the location of each of these three points of entry might affect the kidney function. Similarly, while some scarring is inevitable, it remains unknown as to whether the tissue damaged by the tract has the ability to regenerate and thereby return to baseline function. In this study, we intend to evaluate whether the location of the percutaneous access affects the renal function and whether the tissue damaged by the tract can regenerate. The porcine kidney and its collecting system bear many similarities to the human. By establishing percutaneous access in porcine models, we have aimed to quantify the acute and chronic damage to three distinct areas of the kidney.

Methods:
In 3 experimental pigs, a total of 6 kidneys, percutaneous tracts were obtained via the renal papilla, or infundibulum and dilated to 24 Fr (4 renal units with infundibular access and 2 with papillary access). Two additional pigs (4 renal units) have served as control and underwent only flexible ureteroscopy. Forniceal punctures are slated later in the planned study.

Initial Procedure: Under general anesthesia, using fluoroscopy and ureteroscopic control, we established multiple percutaneous accesses (24Fr) in the renal units of 3 female pigs. This resulted in 2 entry points for each of the kidneys tested. The control group received no nephrostomy tracts, but underwent ureteroscopy similar to the experimental groups. At the beginning and the end of each procedure, serum and split urine (from each kidney individually) samples were collected for analysis of kidney function and renal injury. Urine was specifically tested for creatinine, sodium, osmolality, neutrophil gelatinase-associated lipocalin (NGAL), and kidney injury molecule-1 (KIM-1). Serum was tested for electrolytes, creatinine, and urea. Fractional excretion of sodium (FENa) was calculated using the measured sodium and creatinine values from both serum and urine collection. Of note, photographs of each site of entry were taken immediately prior to entry and at the time of entry. After removal of each sheath, suture was used for subcuticular closure of the skin.

Second Procedure (Day 7): Under general anesthesia, flexible ureterorenoscopy was performed. The intrarenal collecting system was inspected and the entry points were photographed. Split urine and serum samples were obtained for measurement of the aforementioned substances.

Terminal Procedure (Week 6): The second procedure was repeated. Both kidneys were harvested and sent for histological processing and pathological analysis. The pig was euthanized.

Current Progress and Unforeseen Difficulties:

Currently, a total of 5 pigs have begun the procedure (as outlined above). Of the initial 5, 2 experimental and 1 control pig have completed the procedure. One control and one experimental pig were euthanized prior to completion of the full 6 week procedure as noted in the following explanations. In the first experimental pig, a retrograde laser exit method for obtaining renal access was attempted and resulted in a severe hemorrhage. We concluded that the retrograde laser access technique was unsafe as it resulted in an errant trajectory of the percutaneous access. We therefore submitted a modification to the protocol to create a percutaneous tract using the standard fluoroscopic and endoscopic guided needle approach, instead of the laser exit. Since the approval of this modification, we have successfully completed the full protocol for 3 pigs with no adverse effects. During the second diagnostic procedure performed on a control pig, we were not able to gain access to one of the ureters due to complete obstruction near the ureterovesical junction. Complete obstruction was initially assumed after multiple attempts to pass wires/fluoroscopic contrast past the distal ureter were unsuccessful and was confirmed by gross examination following euthanasia. Data collection was not completed for this control pig and in order to avoid any pain or distress associated with an obstructed kidney we elected to euthanize the pig. Since this complication we have been able to access all ureters in the subsequent pigs.

Pigs 6 and 7 have just completed the initial procedure and the experimental portion of the project is projected to be completed by early 2020.

While the project was initially estimated to be completed by Fall 2019, it became evident in early procedures that this goal was unachievable. Several changes were required to the project's IUCAC protocol early in the summer because of difficulty with the initially proposed laser exit method of tract creation. There was also unforeseen loss of operating time, as the building that contains the animal procedure rooms was closed for 4 weeks for construction upgrades.

Preliminary Results:

With only 6 kidneys having completed the full protocol (2 in the papillary access experimental group, 2 in the infundibular access experimental group, and 2 in the control group) there are limited data for analysis. For the purpose of normalization, kidney specific injury proteins evaluated with ELSIA (NGAL and KIM-1) and pathological analysis will not be completed until all experimental procedures are finished. However, serum and urine chemistry results are available for preliminary analysis - data for the initial collection period of each procedure can be found in Table 1.

Urine osmolality decreased by an average of 26.1% in the papillary access group and 44.1% in the infundibular access group when comparing urine concentration from the beginning of initial procedure to the 6^{th} week post access establishment. The control group experienced almost no change in urine osmolality over the same time period. Average FENa decreased by 52.3% in the papillary access group and 51.3% in the infundibular access group when comparing values from before the first procedure to the 6^{th} week post access establishment. Over the same period the control group similarly decreased by 61.4%. Similar trends are more difficult to appreciate at the one-week mark for both urine osmolality and FENa. Lastly, serum creatinine increased across all groups over the duration of the experiment: 26.7%, 28.6%, and 41.7% in the papillary, infundibular, and control groups respectively. Formal statistical analysis will be conducted when all data for the project have been collected.

Table 1: Preliminary Data

	Osm. Initial (mOsm/kg)	*Osm. Second (mOsm/kg)*	*Osm. Terminal (mOsm/kg)*	*FENa Initial*	*FENa Second*	*FENa Terminal*	*Creatinine Initial (mg/dL)*	*Creatinine Second (mg/dL)*	*Creatinine Terminal (mg/dL)*
Control 1	462	143	454	2.83	-	1.09	0.7	1.0	1.2
Control 2	470	189	478	1.83	-	0.71	0.7	1.0	1.2
Papilla Access 1	459	172	311	1.44	0.70	0.55	1.1	1.1	1.5
Papilla Access 2	365	265	292	2.24	0.71	1.28	1.1	1.1	1.5
Infundibulum Access 1	253	420	131	6.32	1.09	1.47	1.5	1.6	2.1
Infundibulum Access 2	210	390	126	5.54	1.19	4.09	1.5	1.6	2.1

Laboratory values from sample collection at the beginning of each procedure (initial, second (week 1) and terminal (week 6)) for osmolality, calculated FENa, and serum creatinine are listed. Each row references an individual renal unit. Second procedure urine samples for Control 1 and 2 were lost when sent to an outside laboratory- repeat results are pending.

Preliminary Discussion:

This project has the potential to provide physicians with a better understanding of the renal damage associated with currently used percutaneous approaches to the kidney and thereby guide their selection of the nephrostomy tract's point of entry into the collecting system. Many patients receiving minimally invasive urological procedures may present with some form of kidney dysfunction and care should be taken to avoid exacerbating a current, or introducing a new, renal pathology while simultaneously minimizing the risk of hemorrhage during the procedure. Given the limited data that are currently available, there are no conclusions that can be drawn. Indeed, the forniceal portion of the project is still in process, as are the expansion of the infundibular and renal papillary portions of the study. However, these early observations provide some interesting possibilities. In the six kidneys tested to date, infundibular access resulted in a greater decrease in urine concentrating ability, yet FENa and serum creatinine experienced similar trends in all groups, including the control. Regarding the changes in the controls, this may in part be due to transient ureteral obstruction, as ureteral stents were not deployed at the end of ureteroscopy in control or experimental kidneys.