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## BLADDER

### Effects of aging on urinary tract epithelial homeostasis and immunity

Ligon MM, **Joshi CS**, **Fashemi BE**, **Salazar AM**, **Mysorekar IU**

A global increase in older individuals creates an increasing demand to understand numerous healthcare challenges related to aging. This population is subject to changes in tissue physiology and the immune response network. Older individuals are particularly susceptible to infectious diseases, with one of the most common being urinary tract infections (UTIs). Postmenopausal and older women have the highest risk of recurrent UTIs (rUTIs); however, why rUTIs become more frequent after menopause and during old age is incompletely understood. This increased susceptibility and severity among older individuals may involve functional changes to the immune system with age. Aging also has substantial effects on the epithelium and the immune system that led to impaired protection against pathogens, yet heightened and prolonged inflammation. How the immune system and its responses to infection changes within the bladder mucosa during aging has largely remained poorly understood. In this review, we highlight our understanding of bladder innate and adaptive immunity and the impact of aging and hormones and hormone therapy on bladder epithelial homeostasis and immunity.

### Prospective, Randomized, Double-blind, Placebo-controlled, Pilot Study of Extracorporeal Shock Wave Therapy for Detrusor Underactivity/Underactive Bladder

Shen YC, Chen CH, **Chancellor MB**, Chuang YC

Detrusor underactivity/underactive bladder (DU/UAB) is a disease with great unmet needs and no current approved drug treatment. Extracorporeal shock wave therapy (ESWT) has been shown to improve regeneration of tissue and increase detrusor contractility in preclinical studies of DU/UAB. The objective of this study is to assess ESWT as a treatment of DU/UAB. Patients with DU/UAB were enrolled in this phase 2 randomized, double-blind, placebo-controlled, physician-initiated study. The primary endpoint was the average changes in postvoid residual urine (PVR) from baseline to 4 wk after treatment. Other endpoints included the average changes in 3-d voiding diary, global response assessment of patient satisfaction, Underactive Bladder Questionnaire (UAB-Q) score, and urodynamic evaluation. The conclusion showed ESWT was well tolerated with a statistically significant decrease of DU/UAB symptoms and a trend to decrease PVR versus placebo. These results indicate that ESWT may be a promising treatment for DU/UAB and multicenter studies are needed.

### Supraphysiologic Vaginal Estrogen Therapy in Aged Mice Mitigates Age-Associated Bladder Inflammatory Response to Urinary Tract Infections

**Fashemi BE**, Wang C, Chappidi RR, Morsy H, **Mysorekar IU**

Bladder diseases characterized by chronic inflammation are highly prevalent in older women, as are recurrent urinary tract infections (rUTIs). Recurrent urinary tract infections lead to chronic inflammation of the bladder mucosa and cause lower urinary tract symptoms that persist even after the infection is cleared. Vaginal estrogen therapy (VET) has long been used for the treatment of rUTIs; however, its mechanism of action remains unclear. The objective of this study was to examine the mechanism(s) by which VET affects bladder inflammation and response to rUTIs. Here, we induced surgical menopause in aged (18 months old) mice followed by VET. Mice were then infected with uropathogenic Escherichia coli, and course of infection was investigated. Inflammatory cytokine response was assessed before and during infection using enzyme-linked immunosorbent assay. RNA sequencing analysis was used to compare the inflammatory status of the young versus aged bladder and principal changes confirmed via quantitative reverse transcriptase-polymerase chain reaction to determine the effects of VET on bladder inflammation. Impact on age-associated bladder tertiary lymphoid tissue formation was evaluated histologically. Our data suggest that VET is effective by reducing age-associated hyperinflammatory conditions in bladder mucosa and in enhancing the host response to infection.

## KIDNEY

### Genetics of Kidney Disease: The Unexpected Role of Rare Disorders

Elliott MD, **Rasouly HM**, **Gharavi AG**

Hundreds of different genetic causes of chronic kidney disease are now recognized, and while individually rare, taken together they are significant contributors to both adult and pediatric diseases. Traditional genetics approaches relied heavily on the identification of large families with multiple affected members and have been fundamental to the identification of genetic kidney diseases. With the increased utilization of massively parallel sequencing and improvements to genotype imputation, we can analyze rare variants in large cohorts of unrelated individuals, leading to personalized care for patients and significant research advancements. This review evaluates the contribution of rare disorders to patient care and the study of genetic kidney diseases and highlights key advancements that utilize new techniques to improve our ability to identify new gene-disease associations.

### Genetic regulation of serum IgA levels and susceptibility to common immune, infectious, kidney, and cardio-metabolic traits

Liu L, Khan A, Sanchez-Rodriguez E, Zanoni F, Li Y, **Steers N**, Balderes O, Zhang J, Krithivasan P, LeDesma RA, Fischman C, Hebbring SJ, Harley JB, Moncrieffe H, Kottyan LC, Namjou-Khales B, Walunas TL, Knevel R, Raychaudhuri S, Karlson EW, Denny JC, Stanaway IB, Crosslin D, Rauen T, Floege J, Eitner F, Moldoveanu Z, Reily C, Knoppova B, Hall S, Sheff JT, Julian BA, Wyatt RJ, Suzuki H, Xie J, Chen N, Zhou X, Zhang H, Hammarström L, Viktorin A, Magnusson PKE, Shang N, Hripcsak G, Weng C, Rundek T, Elkind MSV, Oelsner EC, Barr RG, Ionita-Laza I, Novak J, **Gharavi AG**, Kiryluk K

Immunoglobulin A (IgA) mediates mucosal responses to food antigens and the intestinal microbiome and is involved in susceptibility to mucosal pathogens, celiac disease, inflammatory bowel disease, and IgA nephropathy. We performed a genome-wide association study of serum IgA levels in 41,263 individuals of diverse ancestries and identified 20 genome-wide significant loci, including 9 known and 11 novel loci. Co-localization analyses with expression QTLs prioritized candidate genes for 14 of 20 significant loci. Most loci encoded genes that produced immune defects and IgA abnormalities when genetically manipulated in mice. We also observed positive genetic correlations of serum IgA levels with IgA nephropathy, type 2 diabetes, and body mass index, and negative correlations with celiac disease, inflammatory bowel disease, and several infections. Mendelian randomization supported elevated serum IgA as a causal factor in IgA nephropathy. African ancestry was consistently associated with higher serum IgA levels and greater frequency of IgA-increasing alleles compared to other ancestries. Our findings provide novel insights into the genetic regulation of IgA levels and its potential role in human disease.

[Precision nephrology identified tumor necrosis factor activation variability in minimal change disease and focal segmental glomerulosclerosis]

Mariani LH, Eddy S, AlAkwaa FM, McCown PJ, Harder JL, Nair V, Eichinger F, Martini S, Ademola AD, Boima V, Reich HN, El Saghir J, Godfrey B, Ju W, Tanner EC, Vega-Warner V, Wys NL, Adler SG, Appel GB, Athavale A, Atkinson MA, Bagnasco SM, Barisoni L, Brown E, Cattran DC, Coppock GM, Dell KM, Derebail VK, Fervenza FC, Fornoni A, Gadegbeku CA, Gibson KL, Greenbaum LA, Hingorani SR, Hladunewich MA, Hodgin JB, Hogan M, Holzman LB, Jefferson JA, Kaskel FJ, Kopp JB, Lafayette RA, Lemley KV, **Lieske JC**, Lin JJ, Menon R, Meyers KE, Nachman PH, Nast CC, O'Shaughnessy MM, Otto EA, Reidy KJ, Sambandam KK, Sedor JR, Sethna CB, Singer P, Srivastava T, Tran CL, Tuttle KR, Vento S, Wang CS, Ojo AO, Adu D, Gipson DS, Trachtman H, Kretzler M

The diagnosis of nephrotic syndrome relies on clinical presentation and descriptive patterns of injury on kidney biopsies, but not specific to underlying pathobiology. Consequently, there are variable rates of progression and response to therapy within diagnoses. Here, an unbiased transcriptomic-driven approach was used to identify molecular pathways which are shared by subgroups of patients with either minimal change disease (MCD) or focal segmental glomerulosclerosis (FSGS). Kidney tissue transcriptomic profile-based clustering identified three patient subgroups with shared molecular signatures across independent, North American, European, and African cohorts. One subgroup had significantly greater disease progression (Hazard Ratio 5.2) which persisted after adjusting for diagnosis and clinical measures (Hazard Ratio 3.8). Inclusion in this subgroup was retained even when clustering was limited to those with less than 25% interstitial fibrosis. The molecular profile of this subgroup was largely consistent with tumor necrosis factor (TNF) pathway activation. Two TNF pathway urine markers were identified, tissue inhibitor of metalloproteinases-1 (TIMP-1) and monocyte chemoattractant protein-1 (MCP-1), that could be used to predict an individual's TNF pathway activation score. Kidney organoids and single nucleus RNA-sequencing of participant kidney biopsies, validated TNF-dependent increases in pathway activation score, transcript and protein levels of TIMP-1 and MCP-1, in resident kidney cells. Thus, molecular profiling identified a subgroup of patients with either MCD or FSGS who shared kidney TNF pathway activation and poor outcomes. A clinical trial testing targeted therapies in patients selected using urinary markers of TNF pathway activation is ongoing.

## PROSTATE

[Impact of the bladder detrusor muscular ring on lower urinary tract symptoms due to benign prostatic hyperplasia: A quantitative MRI analysis]

Nandalur KR, Walker D, Ye H, Al-Katib S, Seifman B, Gangwish D, Dhaliwal A, Connor E, Dobies K, Sesoko C, Dejoie W, **Zwaans B**, Nandalur S, Nguyen J, Hafron J

The etiology of lower urinary tract symptoms secondary to benign prostatic hyperplasia (LUTS/BPH) remains uncertain. The purpose of our study was to quantitatively analyze anatomic characteristics on magnetic resonance imaging (MRI) to assess novel independent factors for symptoms. This retrospective single-institution study evaluated treatment-naïve men who underwent prostate MRI within 3 months of international prostate symptom score (IPSS) scoring from June 2021 to February 2022. Factors measured on MRI included: size of the detrusor muscular ring (DMR) surrounding the bladder outlet, central gland (CG) mean apparent diffusion coefficient (ADC), levator hiatus (LH) volume, intrapelvic volume, intravesicular prostate protrusion (IPP) volume, CG volume, peripheral zone (PZ) volume, prostate urethra angle (PUA), and PZ background ordinal score. Multivariable logistic regression and receiver operating characteristic analysis were used to analyze factors for moderate/severe (IPSS ≥ 8) and severe LUTS/BPH (IPSS ≥ 20). Expansion of the DMR surrounding the bladder outlet is a novel anatomic factor independently associated with moderate and severe LUTS/BPH, taking into account prostate volumes, including quantified IPP volume, which were unrelated. Detrusor ring diameter, easily and reliably measured on routine prostate MRI, may relate to detrusor dysfunction from chronic stretching of this histologically distinct smooth muscle around the bladder neck.

## STONES

[GeoBioMed perspectives on kidney stone recurrence from the reactive surface area of SWL-derived particles]

Todorov LG, Sivaguru M, Krambeck AE, Lee MS, **Lieske JC**, Fouke BW

Shock wave lithotripsy (SWL) is an effective and commonly applied clinical treatment for human kidney stones. Yet the success of SWL is counterbalanced by the risk of retained fragments causing recurrent stone formation, which may require retreatment. This study has applied GeoBioMed experimental and analytical approaches to determine the size frequency distribution, fracture

patterns, and reactive surface area of SWL-derived particles within the context of their original crystal growth structure (crystalline architecture) as revealed by confocal autofluorescence (CAF) and super-resolution autofluorescence (SRAF) microscopy. Multiple calcium oxalate (CaOx) stones were removed from a Mayo Clinic patient using standard percutaneous nephrolithotomy (PCNL) and shock pulse lithotripsy (SPL). This produced approximately 4-12 mm-diameter PCNL-derived fragments that were experimentally treated ex vivo with SWL to form hundreds of smaller particles. Fractures propagated through the crystalline architecture of PCNL-derived fragments in a variety of geometric orientations to form rectangular, pointed, concentrically spalled, and irregular SWL-derived particles. Size frequency distributions ranged from fine silt (4-8 µm) to very fine pebbles (2-4 mm), according to the Wentworth grain size scale, with a mean size of fine sand (125-250 µm). Importantly, these SWL-derived particles are smaller than the 3-4 mm-diameter detection limit of clinical computed tomography (CT) techniques and can be retained on internal kidney membrane surfaces. This creates clinically undetectable crystallization seed points with extremely high reactive surface areas, which dramatically enhance the multiple events of crystallization and dissolution (diagenetic phase transitions) that may lead to the high rates of CaOx kidney stone recurrence after SWL treatment.

## International Alliance of Urolithiasis (IAU) guidelines on the metabolic evaluation and medical management of urolithiasis

Zeng G, Zhu W, Robertson WG, **Penniston KL**, Smith D, Pozdzik A, Tefik T, Prezioso D, Pearle MS, Chew BH, Veser J, Fiori C, Deng Y, Straub M, Türk C, Semins MJ, Wang K, Marangella M, Jia Z, Zhang L, Ye Z, Tiselius HG, Sarica K.

The aim of this study was to construct the fourth in a series of guidelines on the treatment of urolithiasis by the International Alliance of Urolithiasis (IAU) that by providing a clinical framework for the metabolic evaluation, prevention, and follow-up of patients with urolithiasis based on the best available published literature. All recommendations were summarized following a systematic review and assessment of the literature in the PubMed database from January 1976 to June 2022. Each generated recommendation was graded using a modified GRADE methodology. Guideline recommendations were developed that addressed the following topics: initial evaluation, metabolic testing, dietary measures, medical management, and follow-up of recurrent stone formers. It was emphasized by the Panel that prevention of new stone formation is as important as the surgical removal of the stones. Although general preventive measures may be effective in reducing stone recurrence rates in some patients, specific medical and dietary management should be well considered and eventually applied in an individualized manner based on the outcomes of metabolic work-up, stone analysis and some certain patient related factors. A detailed follow-up of each case is essential depending on the metabolic activity of each individual patient.

## In-vivo prediction of kidney stone fragility using radiomics-based regression models

Sudhir Pillai P, Hsieh S, Vercnocke A, Potretzke AM, **Koo K**, McCollough C, Ferrero A

The surgical technique for urinary stone removal is partly influenced by its fragility, as prognosticated by the clinician. This feasibility study aims to develop a linear regression model from computed tomography (CT)-based radiomic markers to predict kidney stone comminution time in-vivo with two ultrasonic lithotrites. Patients identified by urologists at our institution as eligible candidates for percutaneous nephrolithotomy (PCNL) were prospectively enrolled. The active engagement time of the lithotrite in breaking the stone during surgery denoted the comminution time of each stone. The comminution rate was computed as the stone volume disintegrated per minute. Stones were grouped into three fragility classes (fragile, moderate, hard), based on inverse of the comminution rates with respect to the mean. Multivariable linear regression models were trained with radiomic features extracted from clinical CT images to predict comminution times in-vivo. The model with the least root mean squared error (RMSE) on comminution times and the fewest misclassification of fragility was finally selected. The conclusion showed CT metrics-based fragility models may provide information to surgeons regarding kidney stone fragility and facilitate the selection of stone removal procedures.

## Nuclear Magnetic Resonance Metabolomic Profiling and Urine Chemistries in Incident Kidney Stone Formers Compared with Controls

Thongprayoon C, Vuckovic I, Vaughan LE, Macura S, Larson NB, D'Costa MR, **Lieske JC**, Rule AD, Denic A

The urine metabolites and chemistries that contribute to kidney stone formation are not fully understood. This study examined differences between the urine metabolic and chemistries profiles of first-time stone formers and controls. High-resolution <sup>1</sup>H-nuclear magnetic resonance (NMR) spectroscopy-based metabolomic analysis was performed in 24-hour urine samples from a prospective cohort of 418 first-time symptomatic kidney stone formers and 440 controls. In total, 48 NMR-quantified metabolites in addition to 12 standard urine chemistries were assayed. Among the standard urine chemistries, stone formers had lower urine oxalate and potassium and higher urine calcium, phosphate, and creatinine. Among NMR urine metabolites, stone formers had lower hippuric acid, trigonelline, 2-furoylglycine, imidazole, and citrate and higher creatine and alanine. A cross-validated model using urine chemistries, age, and sex yielded a mean AUC of 0.76 (95% CI, 0.73 to 0.79). A cross-validated model using urine chemistries, NMR-quantified metabolites, age, and sex did not meaningfully improve the discrimination (mean AUC, 0.78; 95% CI, 0.75 to 0.81). In this combined model, among the top ten discriminating features, four were urine chemistries and six NMR-quantified metabolites. The conclusion showed Although NMR-

quantified metabolites did not improve discrimination, several urine metabolic profiles were identified that may improve understanding of kidney stone pathogenesis.

*- Jennifer Allmaras, MPH and MuenWang*
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*Email cairibu@urology.wisc.edu to feature your newly published research in next month's communique*