



Home bladder pressure measurements correlate with urodynamic storage pressures and high-grade hydronephrosis in children with spina bifida

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Keywords

Hydronephrosis; Meningomyelocele; Spinal dysraphism; Urodynamics; Urinary bladder; Neurogenic; Pediatrics

Abbreviations

VUR, vesicoureteral reflux; UUTD, upper urinary tract deterioration; SFU, Society of Fetal Urology; EBC, expected bladder capacity; MCC, maximum cystometric capacity; AUC, area under curve; ROC, receiver operating characteristic; RBUS, renal bladder ultrasound; Pdet, detrusor pressure; UDS, urodynamic study; DLPP, detrusor leak point pressure; CIC, clean intermittent catheterization; SB, spina bifida

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Summary

Background

Patients with spina bifida are at risk for developing bladder and renal deterioration secondary to increased bladder storage pressures.

Objectives

To determine the association of home bladder volume and pressure measurements (home manometry) to: 1) detrusor storage pressures on urodynamics (UDS); and 2) the presence of Society of Fetal Urology (SFU) grades 3–4 hydronephrosis on renal bladder ultrasound in patients with spina bifida.

Methods

Data were prospectively collected on patients with spina bifida and neurogenic bladder requiring clean intermittent catheterization. Patients used a ruler and typical catheterization equipment to measure bladder pressures and volumes at home. Home measurements were compared to UDS detrusor pressures and SFU hydronephrosis grade. Detrusor pressure <20 cm H₂O at 50% maximal cystometric capacity (MCC) on UDS was used as a measure of safe storage pressures on UDS; conversely, detrusor pressure >20 cm H₂O was used as a measure to capture both unsafe storage pressures and those with potential for unsafe storage pressures. Receiver-operator characteristic curves and area under curve (AUC) were calculated to depict the association between home manometry variables with detrusor

pressures on UDS and SFU grades 3–4 hydronephrosis.

Results

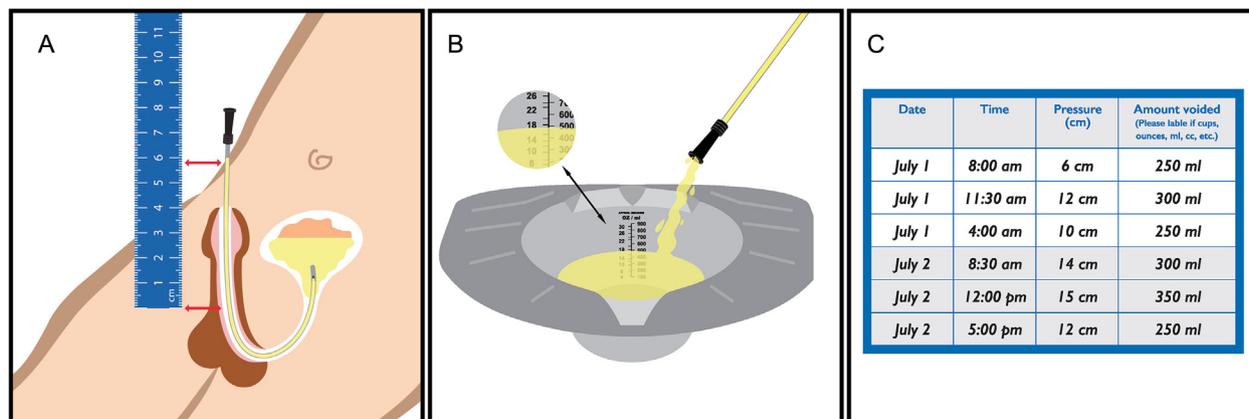
Included were 52 patients with a median age of 10.3 years (interquartile range 6.3–14.4 years). Three home manometry measurements (maximum bladder pressure, bladder pressure at maximum catheterized volume, and mean bladder pressure) > 20 cm H₂O were sensitive for Pdet >20 cm H₂O at 50% MCC. Maximal bladder pressure >20 cm H₂O was the most sensitive among home manometry measures (sensitivity 100%, specificity 70%, AUC 0.92 for Pdet >20 cm H₂O at 50% MCC on UDS; sensitivity 100%, specificity 62%, AUC 0.89 for SFU grade 3–4 hydronephrosis). None of the patients who had maximum home bladder pressure <20 cm H₂O had SFU grades 3–4 hydronephrosis; conversely, individuals with maximal home bladder pressure >20 cm had a wide range of hydronephrosis grades.

Conclusion

None of the patients with maximal home bladder pressure <20 cm H₂O had grade 3–4 hydronephrosis. Home measurements of maximal bladder pressure, bladder pressure at maximum catheterized volume and mean bladder pressure of >20 cm H₂O were all sensitive for Pdet >20 cm H₂O at 50% MCC on UDS. Home manometry is an inexpensive and simple technique to identify patients at risk for and to monitor individuals at high risk of upper tract dilation, without incurring significant cost or morbidity.

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Summary Figure Representative illustration of home bladder pressure and volume measurements in a male patient with spina bifida.

Introduction

Patients with spina bifida (SB) are at risk of developing bladder and renal deterioration secondary to elevated intravesical pressures from a neurogenic bladder [1]. One of the main objectives of urological management is to monitor bladder function and prevent upper tract damage. This is typically accomplished with serial ultrasounds, voiding cystourethrograms and urodynamic studies (UDS) [2]. Monitoring and management of patients with spina bifida has a high cost, and individuals with spina bifida average greater than \$300,000 in lifetime economic impact [3]. Furthermore, voiding cystourethrograms and UDS may only be available at certain facilities, which can be a geographic limitation for patients who live far from specialty centers.

We have published previously on home manometry, which allows patients and their families to record bladder pressure and volume at home. We demonstrated that elevated home bladder pressures correlated with elevated detrusor pressures on UDS [4]. However, the cohort was small, and was intended as a feasibility study.

For this study, we had two aims. First, to confirm the association between home manometry measurements and detrusor storage pressures on UDS. We used a separate cohort of patients from our previous study. Second, to examine the association between home manometry measurements and the presence of Society of Fetal Urology (SFU) grades 3–4 hydronephrosis on renal bladder ultrasound (RBUS). We hypothesized that elevated home bladder pressures would correlate with elevated detrusor pressures on UDS, and that elevated home bladder pressures would correlate with the presence of grade 3–4 hydronephrosis on RBUS.

Materials and methods

Patient selection

Patients with neurogenic bladder secondary to SB who perform clean intermittent catheterization (CIC), who had

both RBUS and home manometry data obtained within a three-month period, were identified in a prospectively maintained database between May 2010 and May 2019. Demographic and clinical data were collected and managed using REdCap (Research Electronic Data Capture, Vanderbilt University, Nashville, TN) [5]. This cohort represents a separate patient population from our prior study [4]. Patients with primary or secondary grade IV and V vesicoureteral reflux, or neurogenic bladder not secondary to SB were excluded. This study was approved by the Institutional Review Board (IRB# 180209).

Home manometry

Patient caregivers were instructed by experienced nurses on how to perform home manometry, with verification of understanding demonstrated through verbal teach-back and caregiver-patient demonstration of technique (Fig. 1). With the patient in a relaxed supine position, the caregiver inserts the catheter into the bladder and holds the catheter upwards, perpendicular to the body. A ruler is aligned where zero is level with the penoscrotal junction (in males), the urethral meatus (in females), or the anterior abdominal wall (in patients who catheterize through a continent catheterizable channel). The height of the urine column in the catheter is measured and recorded (Fig. 1). The bladder is then emptied via the catheter, and the volume of urine is measured and recorded.

An instructional handout and a link to an explanatory video tailored to patient gender, channel of catheterization (continent catheterizable channel or native urethra), and language (English or Spanish) were given to patients' caregivers (<https://www.choc.org/programs-services/urology/home-manometry>). Caregivers were provided a measurement container for volume measurement, a ruler, urethral catheters with a clear tubing (corresponding to the diameter of catheters the patient uses at home), and additional clear tubing to serve as a connector when the pressure column of the urine exceeds the length of the catheter.

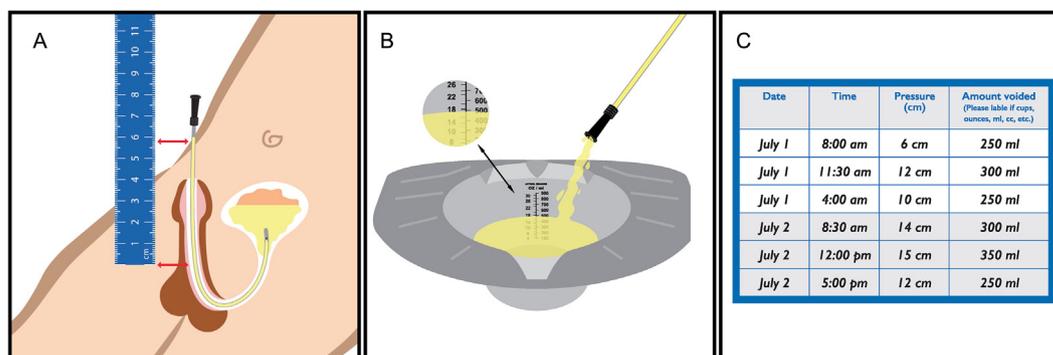


Fig. 1 Home bladder pressure and volume measurements. A male patient is represented here, but similar principles may be applied to female patients and children with continent catheterizable channels. (A) With male patient lying flat with his head down, a caregiver other than the patient inserts the catheter. Once urine is seen in the catheter, the catheter is held straight up. Once urine levels out in the catheter, the “0” of the ruler is held at the penoscrotal junction, an approximation of the middle of the bladder. The height of the urine is recorded in cm into the measurement log. (B) The caregiver fully empties the urine from the bladder into a container and the amount of urine is recoded into the log under “Amount voided.” Units of measurement are per patient preference. (C) The caregiver records the date, time, bladder pressure, and amount emptied or voided on the log. A minimum of three recordings are requested, and the log is reviewed at the next appointment.

Home manometry variables

Patients were instructed to record home manometry measurements (volume [mL] and pressure [cm H₂O]) from CIC for a minimum of three days of their choosing, which did not have to be consecutive. For patients who had UDS, home manometry measurements were typically obtained one month prior to their UDS visit. For each patient, we extracted the following variables from their home manometry logs: largest recorded volume; highest recorded bladder pressure; bladder pressure recorded at maximal catheterized volume; and “mean bladder pressure,” which represented the mean of all home bladder pressure across multiple entries. Expected bladder capacity (EBC) in mL, for patients who were 13 years of age or younger, was calculated with the linear equation of $(\text{age in years} + 2) \times 30$ [6]. Mean largest observed volume as ratio of EBC was calculated by dividing largest recorded volume in home manometry by EBC. We used 450 mL (mean of 400 mL and 500 mL [7]) as the estimated bladder capacity for individuals over 13 years old.

Urodynamic evaluation

Urodynamic assessment was conducted according to International Children’s Continence Society’s guidelines [8]. Urine cultures were completed prior to UDS in all children. Prophylactic antibiotics were given to all patients with vesicoureteral reflux or recurrent urinary tract infections (UTIs). A computerized urodynamic system (AQUARIUS[®] XT, Laborie Medical Technologies, Portsmouth, NH) was used in combination with fluoroscopy for evaluation. Standard cystometry was performed with a 7 Fr dual-lumen urethral catheter and 10 Fr rectal catheter. Filling was performed at a rate of less than 10% of expected bladder capacity, or bladder capacity estimated on prior UDS or imaging. UDS was performed first in supine position then in Fowler’s position for each patient.

Urodynamics variables

Maximal cystometric capacity (MCC) on UDS was defined as the infused fluid volume up to either the detrusor leak point pressure or the total infused volume. Detrusor leak point pressures were measured as the lowest detrusor pressure at which urine leakage occurs in the absence of increased abdominal pressure [9]. Detrusor pressure (Pdet) at 50% and 85% of MCC was calculated by a pediatric urologist based on UDS tracings.

Outcome measures

Pdet <20 cm H₂O at 50% MCC was used as a measure of safe storage pressures on UDS in our cohort based on the study by Houle et al. [10]. Based on review of catheterized volumes on home manometry entries, we deemed 50% MCC on UDS a reasonable reflection of the home volumes at which patients catheterize. RBUS grading of hydronephrosis in accordance to the SFU criteria and the presence of hydro-ureter was assessed by a pediatric urologist [11]. A thickened bladder on RBUS was recorded if it was included in the radiology report. Bladder trabeculation on fluoroscopy or VCUG was graded by a pediatric urologist according to previously published criteria [12].

Statistical analysis

Descriptive data were summarized using medians and percentages. Comparisons of home manometry and UDS measurements between patients with grade 3–4 hydronephrosis on ultrasound versus patients without grade 3–4 hydronephrosis was conducted using two-sample t-tests for continuous data and Fisher’s exact tests for categorical data.

We dichotomized the home manometry pressure variables to <20 cm H₂O or to >20 cm H₂O to illustrate sensitivity and specificity of home manometry measures when compared to the specific outcome measures that we were

investigating. Receiver operating characteristic (ROC) curves and area under the curve (AUC) were calculated to determine the relationship between home manometry variables (mean pressure, maximum pressure, and bladder pressure at maximum volume) and 1) Pdet >20 cm H₂O at 50% MCC on UDS and 2) grade 3–4 hydronephrosis on RBUS. Statistical significance was set at $p < 0.05$ and analyses were performed using SAS, version 9.4 (SAS Institute, Cary, NC, USA).

Results

Fifty-two individuals with spina bifida who completed home manometry and had RBUS within a 3-month period were included in the study. Nine of 52 patients had grade I–III VUR (17%), with five of the nine patients having grade I VUR. Forty-four individuals (84.6%) had UDS data for analysis. Patients recorded home bladder pressures for a median number of 5.0 days (interquartile range [IQR] 3.0–6.5 days), with a median of 10 entries (IQR 8.0–13.5 entries).

The demographics and clinical characteristics of the cohort are described in Table 1. Four patients (7.6%) had grades 3–4 hydronephrosis; 14 patients (26.9%) had grades 1–2 hydronephrosis, and 34 (65%) had no hydronephrosis. Four of 52 patients (7.6%) had hydroureter. Of these four patients with hydroureter, one was associated with grade 4 hydronephrosis, with the remaining three associated with grades 1–2 hydronephrosis. More than 50% of the patients

were female, Hispanic, and had public insurance. Most were on oral anticholinergic medication (75%) and performed CIC 3–5 times daily (67%). Twenty-nine percent used an overnight indwelling foley catheter.

Home manometry measurements and urodynamics were compared between patients with no hydronephrosis or grades 1–2 hydronephrosis and patients with grades 3–4 hydronephrosis on RBUS (Table 2). For home manometry measures, mean bladder pressure, mean highest recorded bladder pressure, and the mean bladder pressure at maximal catheterized volume were all significantly higher in patients with grades 3–4 hydronephrosis as compared to patients with no hydronephrosis or grades 1–2 hydronephrosis on univariate analysis (29 vs. 15 cm H₂O, $p = 0.003$; 40 vs. 21 cm H₂O, $p = 0.002$; 32 vs. 19 cm H₂O, $p = 0.03$; respectively). Individuals with grades 3–4 hydronephrosis all had thickened bladder walls on RBUS (4/4, 100%) and grade 2 bladder trabeculation on fluoroscopy during video UDS (4/4, 100%). They were significantly more likely to have a thickened bladder on RBUS (100% vs. 29%, $p = 0.004$) and grade 2 bladder trabeculation on video UDS (100% vs. 17%, $p = 0.002$) as compared with individuals with no hydronephrosis or grades 1–2 hydronephrosis.

On UDS measures, patients with grades 3–4 hydronephrosis had a higher mean detrusor pressure at both 85% of MCC (39 vs. 23 cm H₂O, $p = 0.14$) and 50% of MCC (18 vs. 11 cm H₂O, $p = 0.36$) as compared to patients without hydronephrosis or grades 1–2 hydronephrosis, although differences were not statistically significant. The ratio of mean largest observed volume on home manometry to expected bladder capacity was 0.65 in individuals with grades 3–4 hydronephrosis, as compared to 0.78 in individuals with no hydronephrosis or grades 1–2 hydronephrosis ($p = 0.98$).

None of the patients who had maximum home bladder pressure <20 cm H₂O had grades 3–4 hydronephrosis. Individuals with maximal home bladder pressure >20 cm had a wide range of hydronephrosis grades ($n = 28$). This includes those with no hydronephrosis (12/28, 42%), grade 1 hydronephrosis (3/28, 11%), grade 2 hydronephrosis (3/28, 11%), grade 3 hydronephrosis (2/28, 7%), and grade 4 hydronephrosis (2/28, 7%). We also observed that the proportion of patients with maximal home bladder pressure >20 cm H₂O increased as we moved to increasing grades of hydronephrosis (No hydronephrosis, 35%; grade 1, 38%; grade 2, 60%; grade 3, 100%, grade 4, 100%).

Three measures on home manometry (highest recorded bladder pressure >20 cm H₂O, bladder pressure at maximal catheterized volume >20 cm H₂O, mean bladder pressure >20 cm H₂O) were highly sensitive for Pdet >20 cm H₂O at 50% MCC on UDS and are illustrated graphically as ROC curves (Fig. 2a). Among the three measures, highest recorded bladder pressure at home >20 cm H₂O was the most sensitive for Pdet >20 cm H₂O at 50% MCC on UDS, with a sensitivity of 100% and specificity of 70% (Supplementary Table 1; AUC 0.92, 95% confidence interval [CI]: 0.84–1.0). Bladder pressure at maximum catheterized volume and mean bladder pressure on home manometry had, respectively, a sensitivity of 86% and 71%, and specificity of 81% and 89% (AUC 0.90, 95% CI: 0.80–1.0; AUC 0.89, 95% CI: 0.79–0.99).

Table 1 Demographics and clinical characteristics of patients with spina bifida.

	All SB patients (n = 52)
Gender, n (%)	
Male	22 (42)
Female	30 (58)
Median age at UDS, years (IQR)	10.3 (6.3–14.4)
Median follow-up time, years (IQR)	10.6 (5.2–14.8)
Race/Ethnicity, n (%)	
Non-Hispanic White	15 (29)
Hispanic	27 (52)
African American	1 (2.0)
Asian	1 (2.0)
Unknown race/ethnicity	8 (15)
Insurance, n (%)	
Private (PPO/HMO)	17 (33)
Public (Medicare/Medicaid)	35 (67)
On oral anticholinergic medication, n (%)	39 (75)
Frequency of CIC, n (%)	
Twice daily	3 (6.0)
3–5 times daily	35 (67)
6 times or more daily	14 (27)
Overnight indwelling foley catheter use at time of UDS, n (%)	15 (29)

Abbreviations: UDS, urodynamics study; SFU, Society of Fetal Urology; CIC, clean intermittent catheterization; PPO, preferred provider organization; HMO, health maintenance organization; RBUS, renal bladder ultrasound.

Table 2 Comparison of home bladder pressure and volume measurements and urodynamics characteristics between patients with no hydronephrosis or grade 1–2 hydronephrosis and patients with grade 3–4 hydronephrosis.

	SB patients with no hydronephrosis or grade 1–2 hydronephrosis	SB patients with SFU grade 3–4 hydronephrosis	P value*
Home manometry data (n = 52)	n = 48	n = 4	
Mean largest observed volume as ratio of EBC \pm SD	0.76 \pm 0.47	0.76 \pm 0.50	0.98
Mean of individual mean bladder pressures, cm H ₂ O \pm SD	15.1 \pm 7.9	28.5 \pm 10.8	0.003
Mean of individual maximal bladder pressure, cm H ₂ O \pm SD	20.7 \pm 10.7	39.5 \pm 16.1	0.002
Mean of individual bladder pressure at maximum volume, cm of H ₂ O \pm SD	18.7 \pm 11.4	32.2 \pm 10.7	0.03
RBUS data (n = 52)	n = 48	n = 4	
Thickened bladder wall, n (%)	14 (29)	4 (100)	0.004
UDS data and VCUG (n = 52)	n = 48	n = 4	
Bladder trabeculation, n (%)			0.002
None	26 (54)	0	
Grade 1	9 (17)	0	
Grade 2	8 (17)	4 (100%)	
Unknown	5 (10)	0	
UDS data only (n = 44)	n = 42	n = 2	
Mean detrusor pressure at 85% MCC, cm H ₂ O \pm SD	22.6 \pm 15.4	39.0 \pm 7.1	0.14
Mean detrusor pressure at 50% MCC, cm H ₂ O \pm SD	10.8 \pm 10.0	17.5 \pm 13.4	0.36

Abbreviations: SB, spina bifida; EBC, expected bladder capacity; SD, standard deviation; UDS, urodynamics study; MCC, maximum cystometric capacity; VCUG, voiding cystourethrogram.

*P value from two sample *t*-test or Fisher's exact test.

The same three measures on home manometry (highest recorded bladder pressure >20 cm H₂O, bladder pressure at maximal catheterized volume >20 cm H₂O, mean bladder pressure >20 cm H₂O) were also highly sensitive for the presence of grades 3–4 hydronephrosis on RBUS (Fig. 2b). The maximal recorded bladder pressure on home manometry, if >20 cm H₂O, was the most sensitive measure for the presence of SFU grade 3–4 hydronephrosis on RBUS, with sensitivity 100% and specificity 60% (Supplementary Table 2; AUC 0.89, 95% CI: 0.77–1.0). Bladder pressure at maximum catheterized volume and mean bladder pressure on home manometry had, respectively, a sensitivity of 75% and 75%, and specificity of 75% and 85% (AUC 0.88, 95% CI: 0.75–1.0; AUC 0.82, 95% CI: 0.61–1.0).

Discussion

In this study, we demonstrate that maximum bladder pressure, bladder pressure at maximal catheterized volume and mean pressure measured at home >20 cm H₂O were all correlated with Pdet >20 cm H₂O at 50% MCC. We validate our previous results in a distinct cohort of patients [4]. The majority of our cohort (92%) did not have grade 3–4 hydronephrosis. None of the patients with maximal bladder pressure <20 cm H₂O on home manometry had grade 3–4 hydronephrosis, suggesting that maximal bladder pressure <20 cm H₂O on home manometry performs well as a surrogate measure for safe storage pressures.

Conversely, not all patients with maximal home bladder pressures >20 cm H₂O had grades 3–4 hydronephrosis, which is reflected in the specificity (range 60%–83%, Fig. 2b). Forty-two percent of individuals (12/28) with maximal home bladder pressure >20 cm H₂O had no hydronephrosis. There may be three contributing factors. First, upper urinary tract deterioration (UUTD) occurs over time, and we may be capturing patients prior to development of increasing UUTD on RBUS. Second, RBUS represents a snapshot in time, and anterior posterior diameters of the renal pelvis are known to vary pre- and post-catheterization. Third, the frequency of CIC introduces variation at what volumes the patient catheterizes at, which in turn can affect volume-specific bladder pressures. The value of our study lies in identifying patients with maximal home bladder pressures <20 cm H₂O as individuals who likely have safe storage pressures, while those who have maximal home bladder pressures >20 cm H₂O deserve closer monitoring and possibly further evaluation.

While a DLPP of 40 cm H₂O is the gold standard of identifying children with SB at risk of upper tract deterioration [1], contemporary studies have shown that upper tract damage can occur at pressures <40 cm H₂O [13–16]. Tarcan et al. found that 43% of spina bifida patients with DLPP between 20 and 40 cm H₂O had evidence of upper tract deterioration on RBUS or dimercaptosuccinic acid scan [16]. Pdet <20 cm H₂O at 50% MCC was used as a measure of safe storage pressures on UDS in our cohort based on the study by Houle et al. [10]. In an examination

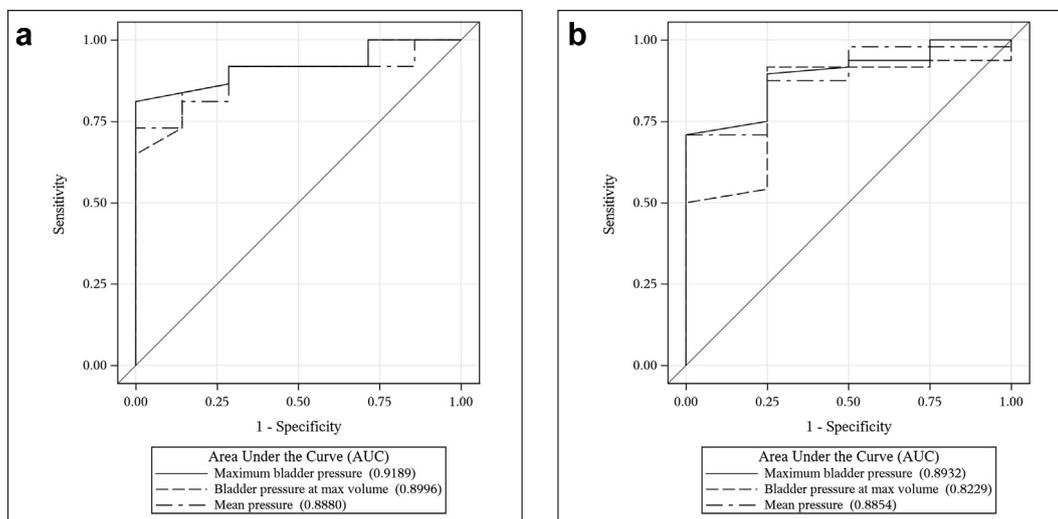


Fig. 2 (a) Receiver-operator characteristic curve and area under the curve for correlation of home manometry measurements >20 cm H₂O with urodynamics measurement of detrusor pressure >20 cm H₂O at 50% maximum cystometric capacity. (b). Receiver-operator characteristic curve and area under the curve for correlation of home manometry bladder pressure >20 cm H₂O and Society of Fetal Urology grade 3–4 hydronephrosis.

of 923 UDS tracings in children, with 69 tracings included as “normal UDS,” Houle et al. found that 95% of children spent less than 5% of the examination at a Pdet >20 cm H₂O, and 99% of the examination at Pdet <30 cm H₂O [10]. On review of home manometry entries, we found that the volumes at which patients catheterize are frequently at 50%–85% of the patient’s expected bladder capacity. We therefore deemed 50% MCC on UDS a better reflection of the home volumes at which patients catheterize.

Home manometry is a useful tool but is not intended to replace UDS. UDS provides important information, such as detrusor overactivity and bladder compliance, that home manometry is unable to measure. However, home monitoring of bladder pressures is capable of capturing bladder pressures over multiple timepoints in the patients’ home environment and with natural filling, providing longitudinal individual data that UDS, as a snapshot in time of bladder function, is unable to yield at a low cost or time-efficient manner. Therefore, home manometry can serve three distinct purposes: first, as a complementary longitudinal data source that augments the information provided at a point in time by UDS; second, as a possible screening tool to alert the clinician to the possibility of changes in bladder function; and third, as a monitoring tool when the pediatric urologist adjusts management of the bladder, such as changing the frequency of catheterizations or anticholinergic dosage. Abdominal pressures are not measured during home manometry. Despite the fact that increased abdominal pressure would translate to an increased bladder pressure measure on home manometry, we believe that the increased bladder pressure reading makes home manometry an even more sensitive tool to screen for unsafe storage pressures during UDS. Since bladder pressures can vary depending on the volume that the patient catheterizes at as well as with technique, single high bladder pressure values in home manometry entries should prompt a review of catheterization techniques and review of other entries.

Home bladder pressure monitoring has been published in other settings [17–19]. Cooper et al. developed a handheld device that records opening bladder pressure and volume evacuated by a pump. However, our home manometry protocol has the benefit of using inexpensive tools (clear catheter and ruler), and may be more easily applied in low resource settings with less opportunity for device failure. In our experience, most caregivers who have been instructed to perform home manometry have been willing to participate, as it empowers them to confirm what bladder pressures are at home.

Our study has several limitations. First, our sample size is relatively small at 52 patients, and the subset of patients with grades 3–4 hydronephrosis was small with four patients. External validation would improve our results. Second, we used Pdet <20 cm H₂O at 50% MCC on UDS as a measure of safe storage pressures. However, multiple objective measures are associated with a higher risk of UUTD: poor bladder compliance, detrusor overactivity, high DLPP, and detrusor sphincter dyssynergia [16,20], which our study does not account for. Third, despite verbal teach-back and caregiver-patient demonstration of home manometry after instructions, there can still be variability in how home manometry is performed.

While we demonstrate a reliable, inexpensive method of monitoring bladder storage pressures at home, future directions include a prospective investigation of a cohort of patients with maximal home bladder pressures >20 cm H₂O and longitudinal follow up on hydronephrosis development, with goals of elucidating predictive parameters for development of hydronephrosis.

Conclusions

None of the patients with maximal home bladder pressure <20 cm H₂O had grade 3–4 hydronephrosis. Home

measurements of maximal bladder pressure, bladder pressure at maximum catheterized volume and mean bladder pressure of >20 cm H₂O were all sensitive for Pdet >20 cm H₂O at 50% MCC on UDS. Home manometry is an inexpensive and simple technique to identify patients at risk for UUTD and to monitor individuals at high risk of UUTD, without incurring significant cost or morbidity.

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Ethical approval

This study was approved by our Institutional Review Board (IRB#180209).

Conflicts of interest

The authors have no conflicts of interest to declare.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.jpuro.2022.06.011>.