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ADJUSTABLE PRELOAD AND DYNAMIC COMPLIANCE IN THE HUMAN DETRUSOR

Hypothesis / aims of study

Acute changes in detrusor tension may be important in both overactive bladder syndrome (OAB) and detrusor underactivity (DU). Total detrusor tension can be analyzed according to its component parts: the active tension (T_a) generated by detrusor smooth muscle (DSM) contraction and the preload (T_p). T_p can be further sub-divided into passive tension (T_{pas}) derived from non-regulatable structural components (i.e. collagen and elastin) and an adjustable preload (T_{ap}) that has been identified in rabbit DSM (rDSM) [1] and other smooth muscle types. Alterations of T_{ap} are observed through the process of reversible strain softening [1]. Our published data using rDSM supports the hypothesis that T_{ap} is adjusted by breakage and reformation of slowly cycling actomyosin crossbridges, permitting reduced muscle stiffness (increases compliance) at increased muscle lengths [2]. Thus, we propose that T_{ap} is a mechanism by which bladder compliance is adjusted acutely during filling (dynamic compliance). In biologic tissues, strain softening is defined as progressive loss of stiffness with repeat stretches due to cross-link breakage. Unlike the irreversible strain softening that occurs when a latex balloon is stretched and released prior to its initial inflation, T_{ap} can be actively restored after contraction at short muscle lengths [1]. Recent pre−clinical studies have established that repeated passive fills of the bladder can acutely increase compliance. Moreover, acute compliance changes can be reversed by active contraction, a process we term "dynamic compliance."

The present study was designed to examine whether human DSM (hDSM) demonstrates T_{ap} *in vitro* via manipulation of hDSM tissue samples and the macroscopic correlate of T_{ap}, dynamic compliance, during *in vivo* human urodynamics. Improved understanding of the biomechanical mechanisms that can acutely regulate detrusor tension may therefore lead to novel treatments for OAB and DU and are the focus of the current investigation.

Study design, materials and methods

Tissue strip study: After IRB approval, hDSM tissue was obtained from cystectomy specimens. A validated stair-step length-tension protocol [3] was applied to each hDSM strip to determine the reference active tension and length (L_{ref}). To quantify T_{ap} in hDSM, tissues were released to 60% L_{ref} (slack tension) and actively contracted twice to re-form cycling actomyosin crossbridges and restore T_{ap} to baseline. Tissues were then subjected to three sequential load-unload (strain softening) cycles to 130% L_{ref} designed to break established crossbridges and quantify the reduction in T_{ap}. After cycle 2, tissues were actively contracted twice at 60% L_{ref} to reform crossbridges. The difference in T_p at L_{ref} between cycle 1 ($T_{p,1}$; before strain softening) and cycle 2 ($T_{p,2}$; after strain softening) estimates T_{ap}. T_p in cycle 3 ($T_{p,3}$; after active contraction) was expected to approximate $T_{p,1}$ due to re-establishment of cycling actomyosin crossbridges lost during strain softening. All T_p values obtained in the T_{ap} protocol were normalized to $T_{p,1}$ measured at L_{ref} at the beginning of cycle 1 and reported as means ± SEM. A repeated-measures one-way ANOVA was performed with Holm-Sidak *post hoc* analysis to test for statistical significance.

Dynamic compliance study: Individuals with OAB defined as ICIq−OAB question 5a ≥3 were enrolled in an IRB−approved extended urodynamics protocol (**Figure 1**). An initial urodynamic study was performed per best practice guidelines for clinical purposes and to determine maximum cystometric capacity (C_{cap}). Four repeat fills were then initiated at a rate 10%C_{cap}/min as follows: 1) fill to 30% C_{cap} and passively empty via syringe aspiration, 2) fill to 60%C_{cap} and passively empty, 3) fill to C_{cap} and void (voluntary or involuntary) and 4) fill to C_{cap} and void. Repeat Fills 1-3 were performed to strain soften the bladder with incrementally increasing preload. Dynamic compliance was evidenced by decreases in intravesical pressure (p_{ves}) with passive filling. The void after Fill 3 was performed to demonstrate the reversibility of dynamic compliance with active bladder contraction. Fill 4 was expected to demonstrate p_{ves} similar to that seen in Fill 1. Extracted p_{ves} values at 75mL infused for Fills 1-4 were normalized to p_{ves} at 75ml in Fill 1 and significance was determined via repeated-measures one-way ANOVA with Holm-Sidak *post hoc* analysis.

Results

Tissue strip study: Urinary bladder tissue specimens from a total of six patients were used for the T_{ap} protocol (n=6). Following the T_{ap} protocol, T_p decreased after strain softening (**Figure 2**). Normalized $T_{p,2} = 0.46 \pm 0.11$ which was a significant loss of T_p due to strain softening when compared to $T_{p,1}$ ($p = 0.01$). Normalized $T_{p,3} = 1.2 \pm 0.30$ was similar to $T_{p,1}$ ($p = 0.50$) demonstrating the presence of T_{ap} in hDSM (T_p lost to strain softening was restored by active contraction). Two patients in the T_{ap} cohort had neurogenic bladder, however, there were no differences in normalized T_p values between neurogenic and non-neurogenic tissues (t-test, p=NS).

Dynamic compliance study: Four patients completed the study and had results available for analysis (n=4). In comparison to repeat Fill 1, dynamic compliance was evident in subsequent fills (Fills 2 and 3) as seen by decreased p_{ves} values. This was likely due to strain softening during filling not reversed by passive emptying. Voiding occurred at the end of Fill 3 and p_{ves} during Fill 4 approached p_{ves} during Fill 1, demonstrating the reversibility of dynamic compliance caused by stain softening (**representative data, Figure 3**). Average normalized p_{ves} at 75 ml infused volume demonstrated dynamic compliance after passive emptying (Fill 3: normalized $p_{ves} = 0.69 \pm 0.10$ vs. Fill 1: 1.0, $p<0.05$) that was reversed after active voiding (Fill 4: normalized $p_{ves} = 0.92 \pm 0.04$ vs. Fill 1: 1.0, p=NS) (**Figure 4**).

Interpretation of results

This study identified and quantified the *in vitro* biomechanical property of T_{ap} in hDSM tissue strips and its *in vivo* correlate, dynamic compliance, seen during an extended repeat filling cystometry protocol. T_{ap} can be decreased acutely by strain softening and restored by active contraction of hDSM. Dynamic compliance is the macroscopic manifestation of T_{ap}: repeat fills strain soften

the bladder, similar to repeat stretches in hDSM tissue strips, and the reversibility of strain softening with voiding, similar to contraction of hDSM tissue strips. It is tempting to speculate that a derangement in the active process regulating T_{ap} could contribute to the pathophysiology of an OAB-subtype – an increase in T_{ap} causing an increase in urinary bladder stiffness during filling would result in increased afferent nerve activity and thus increased urgency at lower bladder volumes. Conversely, a decrease in T_{ap} may cause impaired contractility as seen in DU due to a decreased bladder preload.

Concluding message

In this investigation, we revealed that the biomechanical process of T_{ap}, previously shown in rDSM exists in hDSM tissue, and that T_{ap} can be seen during *in vivo* human urodynamics as dynamic compliance. Future research designed to explore the regulatory mechanism of this biomechanical process may lead to improved understanding and treatment of OAB and DU.

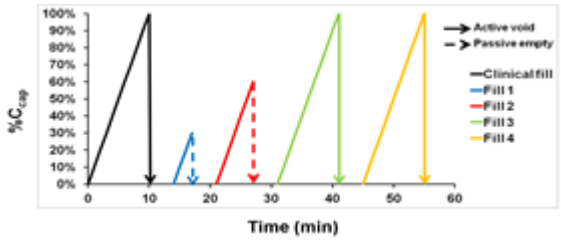

Figure 1

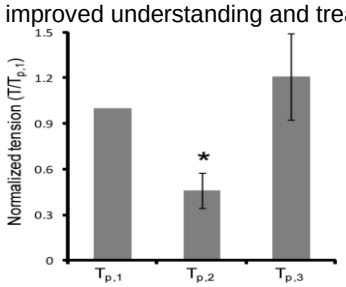

Figure 2

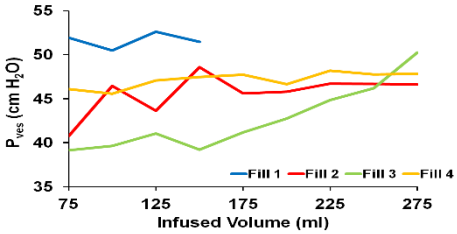

Figure 3

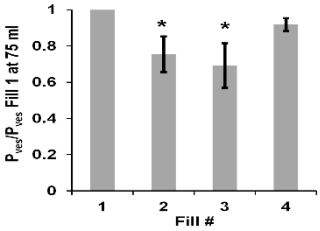

Figure 4

References

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Disclosures

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