HHS Public Access
Author manuscript
Continence (Amst). Author manuscript; available in PMC 2022 July 03.

Published in final edited form as:
Continence (Amst). 2022 June ; 2: . doi:10.1016/j.cont.2022.100032.

Effects of vasopressin receptor agonists on detrusor smooth muscle tone in young and aged bladders: Implications for nocturia treatment

Youko Ikeda[a,b,*], **Irina Zabbarova**[a], **Mathijs de Rijk**[c], **Anthony Kanai**[a,b], **Amanda Wolf-Johnston**[a], **Jeffrey P. Weiss**[d], **Edwin Jackson**[b], **Lori Birder**[a,b]

[a]University of Pittsburgh, School of Medicine, Renal-Electrolyte division, United States of America

[b]University of Pittsburgh, School of Medicine, Department of Pharmacology and Chemical Biology, United States of America

[c]Maastricht University, Faculty of Health, Medicine, and Life Sciences, School for Mental Health and Neurosciences, Department of Urology, the Netherlands

[d]SUNY Downstate Health Sciences University, Department of Urology, United States of America

Abstract

Purpose: The main goal of this study was to determine the effects of arginine vasopressin (AVP) and desmopressin on bladder contractility and to examine whether the effects of these vasopressin receptor (VR) agonists differ in young versus aged animals. These aims were addressed using urinary bladders from young (3 months) and aged (24 month) female Fischer 344 rats that were isolated and dissected into strips for isometric tension recordings. Bladder strips were exposed to AVP and desmopressin through the perfusate, and tension changes recorded.

Results: In young rat bladders, AVP, an agonist at both vasopressin-1 receptors (V_1Rs) and vasopressin-2 receptor (V_2Rs), concentration-dependently caused contraction of bladder strips with a sensitivity that was greater in young versus aged bladder strips. Removal of the mucosa did not alter the sensitivity of young bladder strips to AVP yet enhanced the AVP sensitivity of aged bladder strips. The differential sensitivity to AVP between young denuded and aged denuded bladder strips was similar. In contrast to AVP, desmopressin (V_2R selective agonist) relaxed bladder strips. This response was reduced by removal of the mucosa in young, but not aged, bladder strips.

Conclusion: These findings support a direct role for VRs in regulating detrusor tone with V_1Rs causing contraction and V_2Rs relaxation. In aged bladders, the contractile response to V_1R activation is attenuated due to release of a mucosal factor that attenuates V_1R-induced contractions. Also in aged bladders, the relaxation response to V_2R activation is attenuated by lack of release of a mucosal factor that contributes to V_2R-induced relaxation. Thus age-associated

[*]Correspondence to: A1219 Scaife Hall, 3550 Terrace St, Pittsburgh PA 15261, United States of America. Yoi4@pitt.edu (Y. Ikeda).

Declaration of competing interest
The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

changes in the bladder mucosa impair the effects of VRs on bladder tone. Because the V_2R signaling system is impaired in the older bladder, administering an exogenous V_2R agonist (e.g., desmopressin) could counteract this defect. Thus, desmopressin could potentially increase nighttime bladder capacity through detrusor relaxation in concert with decreased urine production, reducing nocturnal voiding frequency.

Keywords

Arginine vasopressin; Vasopressin-2 receptor; Urinary bladder; Spontaneous contractions

1. Introduction

Advanced age is associated with increased incidence of nocturia [1] which is defined by the International Continence Society as waking two or more times a night to void during the main sleep period with each urination followed by sleep or intention to sleep [2]. The pathophysiology of nocturia is multi-factorial involving both urologic (*e.g.*, nocturnal polyuria, lower urinary tract symptoms) and non-urologic (*e.g.*, heart failure, sleep apnea, diabetes insipidus) conditions that cause dysregulation of urine production [3]. Nocturia can be highly bothersome particularly when associated with poor sleep quality [4]. The condition can adversely impact daily activities and is associated with increased mortality risk due to effects on sleep and other comorbid syndromes [5]. Nocturia can be ameliorated by behavioral modifications (*e.g.*, decreased fluid intake before sleep) or pharmacological interventions to reduce nighttime urine production and/or ameliorate lower urinary tract symptoms [6-8].

Arginine vasopressin (AVP, also known as anti-diuretic hormone) is a key regulator of water homeostasis. AVP is released from the hypothalamic nuclei in response to changes in plasma osmolarity [3] and acts upon vasopressin receptors (VRs), predominately the vasopressin-2 receptor (V_2R) subtype on principal cells located in the collecting ducts and distal tubule. Activation of V_2Rs increases trafficking of aquaporin-2 channels to the apical membrane, which facilitates water reabsorption, concentrates the urine, decreases urine volume and regulates plasma osmolarity [9]. The release of AVP also follows a diurnal pattern attributed to its close connection with the suprachiasmatic nucleus in the hypothalamus that is the principal center for regulation of circadian rhythm [3]. AVP released into the circulation could exert an effect on the bladder wall by direct access from the circulation or from its clearance into urine [10]. Natural aging is believed to augment the circadian pattern resulting in altered behavioral, metabolic, and physiological activities. Accordingly, dysregulation of the AVP/V_2R system is believed to contribute to the increased nighttime urine production that is particularly prevalent in the older adult.

Desmopressin, an analog of AVP that acts as a potent and selective V_2R agonist is currently the only pharmacologic agent approved for treatment of nocturnal polyuria. The principal mechanism for desmopressin is thought to occur via enhancing water reabsorption by the kidneys thereby decreasing urine production and increasing urine concentration. Interestingly, desmopressin is effective in increasing nighttime bladder capacity in patients with mixed nocturia [11] and in decreasing daytime urinary frequency in trials with multiple sclerosis patients [12] indicating additional actions that promote urine storage. Previous studies have shown rat and human urinary bladders express VRs with the highest expression within the mucosa [13], however, the functional significance of these receptors within the urinary bladder has yet to be defined. Thus, we examined the role of bladder VRs on baseline tone and spontaneous contractile activity in isolated bladder strips from young and aged rat bladders to extrapolate its effects during the storage phase.

2. Methods

2.1. Animals

Young female Fischer 344 rats (3–6 months) were obtained from Envigo (Indianapolis, IN). Aged Fischer 344 rats (24–26 months) were obtained from the National Institute of Aging rodent colony. Rats were housed as pairs in microisolator cages with food and water provided *ad libitum* on a 12-hour light/dark cycle. For collection of tissue, rats were deeply anesthetized with 5% isoflurane and the urinary bladder dissected from the abdomen followed immediately by a thoracotomy and exsanguination. All animal procedures received ethical approval from the University of Pittsburgh Institutional Animal Care and Use committee.

2.2. Tension recordings

Isolated bladders were dissected into four strips in the dome to base orientation and strips were mounted in a recording chamber with the dome end attached to a tension transducer (World Precision Instruments, Sarasota, FL) for isometric recordings. A set of strips were denuded of the mucosa by blunt dissection under a stereomicroscope. The tissues were superfused with Tyrode's solution (in mM: NaCl, 118; KCl, 4.0; $NaHCO_3$, 24; NaH_2PO_4, 0.4; $MgCl_2$, 1.0; $CaCl_2$, 1.8; D-glucose, 6.1; Na pyruvate, 5; pH 7.4) that was bubbled with 95% O_2/5% CO_2 and maintained at 36 °C in a water bath. The tissues were superfused with a peristatic pump at a flow rate of 1 ml per minute. Tissues were incrementally stretched and electrically field stimulated with platinum electrodes (20 Hz, 3 s train, 0.5 msec pulse width, 15 V output). Tension was recorded through a PowerLab data acquisition system and LabChart7 Pro software (AD Instruments, Colorado Springs, CO). Tissues were stretched (*via* micromanipulator holding the tension transducer) and stimulated until evoked contractions ceased to increase in amplitude. This was determined to be the length at which there was optimal alignment of contractile fibers [14], and once this length determined, field stimulation was stopped and tissue was allowed to equilibrate for 30 min. Stocks of AVP (1 mM in water) and desmopressin (1 mM in DMSO) were added to the superfusate solution to final working concentrations.

2.3. Data and statistical analysis

Recorded tension changes were normalized and expressed as force per cross-sectional area of tissue (mN/mm^2). Spontaneous contractile activity was analyzed using the peak analysis module in LabChart7 where contractions were discriminated based on height (threshold of 0.1 g, 485 s sampling block) which also generated estimated baseline tension for each contraction detected. Data were evaluated using Prism 9 software (GraphPad, San Diego, CA) for dose–response curve fitting and statistical analyses. Continuous measures were expressed as mean ± standard deviation (SD) or mean ± standard error of mean (SEM). Multiple group comparisons were performed by one-way ANOVA with Tukey's post hoc analysis. AVP dose–response curves were fitted using nonlinear regression and 50% effective concentration (EC_{50}) of each group compared by extra sum-of-square F test. The null hypothesis was rejected at $p < 0.05$.

3. Results

3.1. Spontaneous contractions (SC) in aged rat bladders are enhanced by the mucosa

Young and aged rat bladder strips had comparable baseline tensions, regardless of whether the mucosa was intact or denuded (Fig. 1A). In preparations with an intact mucosa, SC amplitudes were similar in young and aged rat bladder strips (Fig. 1B). However, in aged, but not young, bladder strips, removal of the mucosa significantly reduced SCs (Fig. 1B). There was no significant difference in the contraction frequency between all preparations (young intact, 3.8 ± 1.5 contractions/min; young denuded, 3.3 ± 1.2 contractions/min; aged intact, 3.3 ± 0.8 contractions/min, aged denuded, 2.8 ± 0.5 contractions/min). These data suggest that a mucosal factor contributes to SCs in aged, but not, young, bladders.

3.2. AVP increases baseline tone in rat bladder strips

AVP caused concentration-dependent increase of baseline tension and SC amplitude of rat bladder strips as exemplified in Figs. 2A and 2B. The 50% effective concentration (EC_{50}) values for baseline tension were compared between young/intact (Fig. 2C), young/denuded (Fig. 2D), aged/intact (Fig. 2E) and aged/denuded (Fig. 2F) strips using the built-in method employed by Prism 9 GraphPad to compare the EC_{50}s calculated from concentration–response curves for replicate preparations. This analysis indicated a significantly increased EC_{50} to AVP in aged versus young rat intact bladder strips (EC_{50}: young 2.4 nM *versus* aged 7.5 nM, $p = 0.03$). Removal of the mucosa reduced the EC_{50} to AVP in aged, but not young, rat strips (EC_{50}: aged intact 7.5 nM *versus* aged denuded 1.5 nM). Together these results show that AVP induces contraction of rat bladder strips and that this effect is altered in aged bladder strips due to release of a mucosal factor that negatively modulates responses of the underlying detrusor smooth muscle cells to AVP.

3.3. Desmopressin relaxes detrusor smooth muscle through mucosal vasopressin receptors

A desmopressin dose–response curve could not be obtained due to a time-dependent desensitization likely due to internalization of V_2R [15] which occurred even at low concentrations (not shown). Therefore, we opted for an evaluation using a single concentration of desmopressin. Addition of 100 nM of desmopressin to the superfusate decreased the baseline tension in both young and aged intact bladder strips with a more prominent effect in young (versus aged) bladder strips (Fig. 3). Removal of the mucosa attenuated the relaxation effect of desmopressin in young but not aged bladder preparations (Fig. 3). The amplitude of SCs was also decreased by desmopressin with no significant difference between young intact, young denuded, aged intact and aged denuded bladder preparations. Together these results show that desmopressin induces relaxation of rat bladder strips and that this effect is augmented in young bladder strips due to release of a mucosal factor that modulates responses of the underlying detrusor smooth muscle cells. This factor appears to be lacking in aged bladder strips since removal of the mucosa in aged bladder strips does not change the response to desmopressin.

4. Discussion

Nocturia is a multi-faceted condition which is primarily diagnosed based on bladder diaries along with identifiable comorbidities that affect water/salt homeostasis and lower urinary tract function [8]. Nocturia is categorized as (1) low nocturnal/global bladder capacity, (2) global polyuria, (3) nocturnal polyuria or (4) mixed etiology [6]. Desmopressin promotes antidiuresis through its actions on the V_2R and has become an established pharmacotherapy for nocturnal polyuria and enuresis. Efficacy of desmopressin has also been described for patients with mixed etiology where treatment decreased nocturnal urine production and improved bladder capacity [11]. The effect of desmopressin on nocturnal bladder capacity is intriguing, as it is anticipated that decreased urine production rather than modification of bladder capacity would be responsible for increasing intervals between voids. This suggest that the efficacy of desmopressin in nocturia may not be limited to the kidneys. This concept is further supported by studies in rats showing that desmopressin modulate bladder associated brainstem neurons to suppress isovolumetric bladder contractions [16].

The presented study confirms a functional role of mucosal VRs in the urinary bladder, specifically, the ability of mucosal VRs to modulate the detrusor tone and SCs. The effect of VR on evoked contractions were not examined in this study as our primary hypothesis is that AVP release is highest during sleep periods (*i.e.*, storage phase) and this is when VR activity would be most evident. The expression of bladder VR has been described through mRNA and western blot analyses [13,17]; however, further studies are needed to determine the receptor localization or functional roles. Further, AVP activates all VRs with equal affinity [9]; however, which subtype is dominant is determined not only by receptor affinity, but also by the relative receptor number, coupling efficiency and the presence of spare receptors [18]. The diurnal release of AVP from the hypothalamus is responsible for water reabsorption in the kidney. There are also indications that an endogenous circadian rhythm exists in the kidneys and regulates VR expression and water reabsorption [19]. Similarly, the bladder can locally synthesize AVP, and this likely enables a paracrine AVP signaling mechanism [20,21]. Whether AVP release in the bladder follows a circadian rhythm and its physiological role have yet to be established. Endogenous circadian clock regulation of various cellular processes has been described in the mouse bladder [22], raising the possibility that AVP release and/or VR expression could be regulated in this manner.

In both the young and aged bladder, AVP results in a net contraction; however, the EC_{50} for AVP in the aged bladder is significantly greater compared to that of a young bladder. In the aged, but not young, removal of the mucosa decreases the EC_{50} for AVP-induced contractions, a finding suggesting that in the aged bladder there is a release of a factor from the mucosa that is responsible for the impaired AVP-induced contractions. Indeed, in denuded bladder strips the EC_{50} for AVP-induced contraction is similar in young versus aged bladders. The role of the mucosa in an intact bladder is presumably mediated by release of diffusible modulators between the two layers. Release of diffusible factors (e.g., prostanoids) upon bladder distension are known to play a role in controlling urinary bladder motility [23] which may be altered with advanced age.

As mentioned, our findings indicate that stimulation of V_2Rs with desmopressin produces a decrease in muscle tone (e.g., increased smooth muscle relaxation), an effect that would increase bladder compliance. An effect on bladder intravesical pressure by VR activation has also been previously demonstrated in anesthetized rats [17], however, the receptor subtype responsible was not identified. In the current study, bladder relaxation induced by desmopressin is greater in young bladders versus aged bladders. Notably, removal of the mucosa reduces V_2R-induced relaxation in young but not aged bladders. Moreover, in denuded bladder strips, the effects of desmopressin are very similar in young versus aged bladders. These results suggest that V_2Rs induce bladder relaxation in both young and aged bladders; however, V_2R-signaling in aged bladders is impaired due to lack of release of a mucosal relaxing factor that has yet to be identified. In support are findings in the renal artery, whereby desmopressin causes an endothelial-dependent relaxation via V_2R stimulation [24]

In general, our results support the concept that both V_1R and V_2R systems are less effective in the aged bladder. Since removal of the mucosa normalizes responses to both VR systems, the deficiency in aged bladders is likely due to changes in the mucosa. Since our previous findings indicate that V_2R expression is increased in the aged bladder [7], age-associated changes in V_2R signaling may be due to changes in receptor sensitivity and/or signaling mechanisms. As an example, in the kidney, V_2R activation can be modulated by adenosine through the A1 receptors [25]. In this regard, we have unpublished findings that show the aged bladder urothelium has shown increased expression of ectonucleotidases, a family of hydrolytic enzymes that are selective for purines, which may increase urothelial adenosine levels [26]. Thus, we speculate that activation of urothelial adenosine receptors could attenuate V_2R-induced changes in detrusor tone which could be more pronounced with increased age. However, a full investigation is necessary to define the relative contribution of each receptor and influencing mechanisms in the aging bladder.

Taken together, these data indicate an endogenous vasopressin signaling mechanism in the urinary bladder which based in part in the mucosa. We theorize that AVP and desmopressin in the urine can act on urothelial VRs to modulate autonomous bladder contractions that reduces wall tension, maintains low intravesical pressure and minimizes sensory outflow [27]. Thus, in healthy adults, activation of urothelial V_2Rs may play an important role to reduce bladder pressure and increase bladder storage during sleep. However, aging associated attenuation in VR signaling (due to decreased AVP release, receptor sensitivity or changes in signaling mechanisms) may prevent diurnal regulation of urine production and bladder capacity leading to nocturnal polyuria and frequency. Thus, VR signaling mechanisms in the kidney and bladder could be considered an integrated physiological system, manipulation of which could concurrently reduce urine production and promote storage, as additive mechanisms in treating nocturia.

5. Conclusion

The findings of the current study are summarized in Table 1. There is an age-dependent, differential sensitivity of mucosal VR activation on basal detrusor activity in the female rat, where V_1R is likely responsible for increasing detrusor tone while V_2R promotes relaxation. Dysregulation of bladder vasopressin signaling in aging may reduce bladder capacity and when faced with increased urine production during sleep, leads to nocturia. This study supports the concept that desmopressin, a V_2R selective agonist, may promote increased capacity during sleep in nocturia patients, which may be particularly more effective in the older adult, through direct action in the urinary bladder.

Acknowledgments

This study was supported by the National Institutes of Health, United States of America (NIH R01 AG056944; NIH R01 HL 109002; NIH P30 DK079307).

Abbreviations

AVP	Arginine vasopressin
ANOVA	Analysis of variance
EC_{50}	50% effective concentration
dAVP	desmopressin
SC	Spontaneous contractions
VR, VRs	Vasopressin receptor(s)
V_1R, V_1Rs	Vasopressin-1 receptor(s)
V_2R, V_2Rs	Vasopressin-2 receptor(s)

References

[1]. Bliwise DL, Wagg A, Sand PK, Nocturia: A highly prevalent disorder with multifaceted consequences, Urology 133S (2019) 3–13. [PubMed: 31310770]

[2]. Hashim H, Blanker MH, Drake MJ, et al. , International continence society (ICS) report on the terminology for nocturia and nocturnal lower urinary tract function, Neurourol. Urodyn 38 (2019) 499–508. [PubMed: 30644584]

[3]. Birder LA, Van Kerrebroeck PEV, Pathophysiological mechanisms of Nocturia and nocturnal Polyuria: The contribution of cellular function, the urinary bladder Urothelium, and circadian rhythm, Urology 133S (2019) 14–23. [PubMed: 31369749]

[4]. Rose GE, Denys MA, Kumps C, et al. , Nocturnal voiding frequency does not describe nocturia-related bother, Neurourol. Urodyn 38 (2019) 1648–1656. [PubMed: 31165518]

[5]. Dani H, Esdaille A, Weiss JP, Nocturia: aetiology and treatment in adults, Nat. Rev. Urol 13 (2016) 573–583. [PubMed: 27455894]

[6]. Weiss JP, Everaert K, Management of Nocturia and nocturnal Polyuria, Urology 133S (2019) 24–33. [PubMed: 31586470]

[7]. Monaghan TF, Weiss JP, Everaert K, et al. , Pharmacologic management of nocturnal polyuria: a contemporary assessment of efficacy, safety, and progress toward individualized treatment, Ther. Adv. Urol 13 (2021) 1756287220988438. [PubMed: 33796148]

[8]. Everaert K, Herve F, Bosch R, et al. , International continence society consensus on the diagnosis and treatment of nocturia, Neurourol. Urodyn 38 (2019) 478–498. [PubMed: 30779378]

[9]. Pisipati S, Hashim H, Vasopressin receptors in voiding dysfunction, Handb. Exp. Pharmacol (2011) 453–483. [PubMed: 21290239]

[10]. Moses AM, Steciak E, Urinary and metabolic clearances of arginine vasopressin in normal subjects, Am. J. Physiol 251 (1986) R365–370. [PubMed: 3740319]

[11]. Lee HW, Choo MS, Lee JG, et al. , Desmopressin is an effective treatment for mixed nocturia with nocturnal polyuria and decreased nocturnal bladder capacity, J. Korean Med. Sci 25 (2010) 1792–1797. [PubMed: 21165296]

[12]. Hoverd PA, Fowler CJ, Desmopressin in the treatment of daytime urinary frequency in patients with multiple sclerosis, J. Neurol. Neurosurg. Psychiatry 65 (1998) 778–780. [PubMed: 9810957]

[13]. Birder LA, Wolf-Johnston AS, Jackson EK, et al. , Aging increases the expression of vasopressin receptors in both the kidney and urinary bladder, Neurourol. Urodyn 38 (2019) 393–397. [PubMed: 30311671]

[14]. Andersson KE, Arner A, Urinary bladder contraction and relaxation: physiology and pathophysiology, Physiol. Rev 84 (2004) 935–986. [PubMed: 15269341]

[15]. Innamorati G, Sadeghi H, Eberle AN, et al. , Phosphorylation of the V2 vasopressin receptor, J. Biol. Chem 272 (1997) 2486–2492. [PubMed: 8999963]

[16]. Iwasaki H, Koyama Y, Tanaka Y, et al. , Modulation by desmopressin of neuronal activity in brainstem micturition center, Urology 63 (2004) 994–998. [PubMed: 15135006]

[17]. Cafarchio EM, Auresco LC, da Silva LA, et al. , Unravelling the intravenous and in situ vasopressin effects on the urinary bladder in anesthetized female rats: More than one vasopressin receptor subtype involved? Eur. J. Pharmacol 834 (2018) 109–117. [PubMed: 30025812]

[18]. Jackson EK, Drugs affecting renal excretory function, in: Brunton LL, Knollmann BrC, Hilal-Dandan R (Eds.), Goodman & Gilman’s the Pharmacological Basis of Therapeutics, Thirteenth ed., McGraw Hill Medical, New York 2018.

[19]. Hara M, Minami Y, Ohashi M, et al. , Robust circadian clock oscillation and osmotic rhythms in inner medulla reflecting cortico-medullary osmotic gradient rhythm in rodent kidney, Sci. Rep 7 (2017) 7306. [PubMed: 28779094]

[20]. Uvelius B, Lundin S, Andersson KE, Content and contractile effect of arginine vasopressin in rat urinary bladder, Eur. J. Pharmacol 182 (1990) 549–554. [PubMed: 2226623]

[21]. Berggren T, Andersson KE, Lundin S, et al. , Effect and content of arginine vasopressin in normal and obstructed rat urinary bladder: an in vivo and in vitro investigation, J. Urol 150 (1993) 1540–1543. [PubMed: 8411449]

[22]. Sengiku A, Ueda M, Kono J, et al. , Circadian coordination of ATP release in the urothelium via connexin43 hemichannels, Sci. Rep 8 (2018) 1996. [PubMed: 29386573]

[23]. Stromberga Z, Chess-Williams R, Moro C, The five primary prostaglandins stimulate contractions and phasic activity of the urinary bladder urothelium, lamina propria and detrusor, BMC Urol. 20 (2020) 48. [PubMed: 32349725]

[24]. Medina P, Segarra G, Vila JM, et al. , V2-receptor-mediated relaxation of human renal arteries in response to desmopressin, Am. J. Hypertens 12 (1999) 188–193. [PubMed: 10090347]

[25]. Rieg T, Vallon V, ATP and adenosine in the local regulation of water transport and homeostasis by the kidney, Am. J. Physiol. Regul. Integr. Comp. Physiol 296 (2009) R419–427. [PubMed: 19020292]

[26]. Wolf-Johnston AS, Menchikova E, Apodaca G, et al. , Desmopressin stimulates urothelial vasopressin receptors and inhibits adenosine metabolism in the aged urinary bladder: novel mechanism for reducing nocturia/nocturnal polyuria, Neurourol. Urodyn 39 (2020) S134–S135.

[27]. Drake MJ, Kanai A, Bijos DA, et al. , The potential role of unregulated autonomous bladder micromotions in urinary storage and voiding dysfunction; overactive bladder and detrusor underactivity, BJU Int. 119 (2017) 22–29. [PubMed: 27444952]

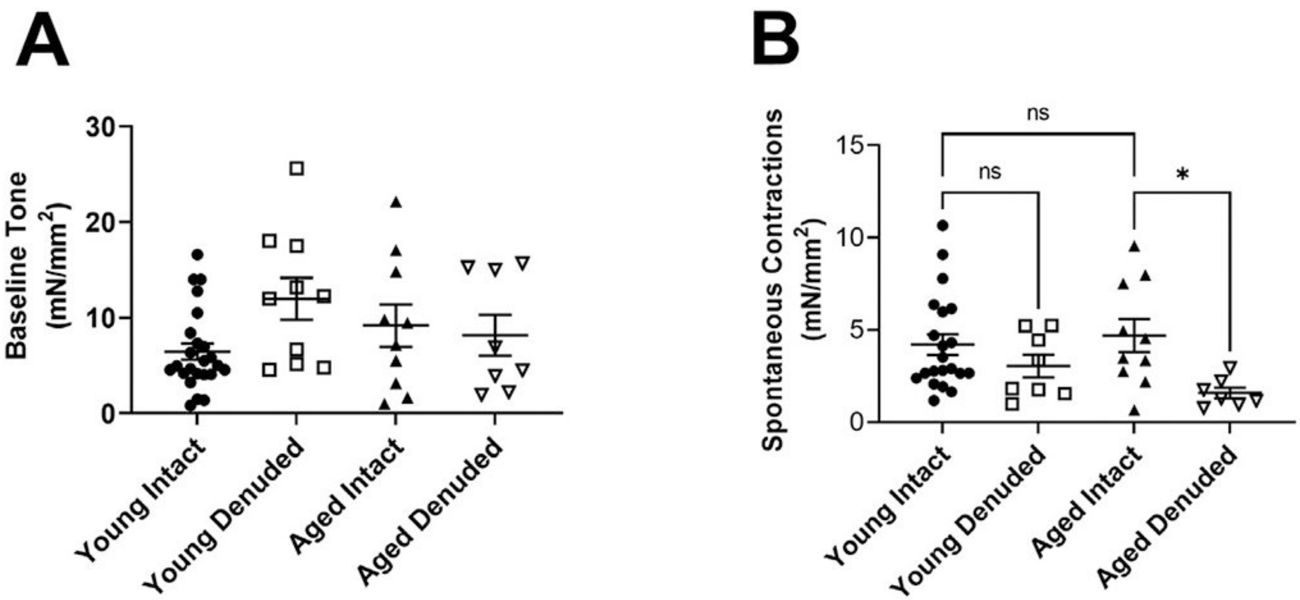

Fig. 1. Baseline tone and amplitude of spontaneous contractions (SCs) from young and aged female rat bladder strips, intact or denuded of mucosa.

A, Bar graphs of baseline tone from young intact (n = 24), young denuded (n = 10), aged intact (n = 10) and aged denuded (n = 8) bladder strips at optimal length. **B,** Mean amplitude of SCs from young intact (n = 21), young denuded (n = 8), aged intact (n = 10) and aged denuded (n = 7) bladder strips at optimal length. Mean ± SD, *p = 0.047, ns = not significant, one-way ANOVA with Tukey’s post-hoc test.

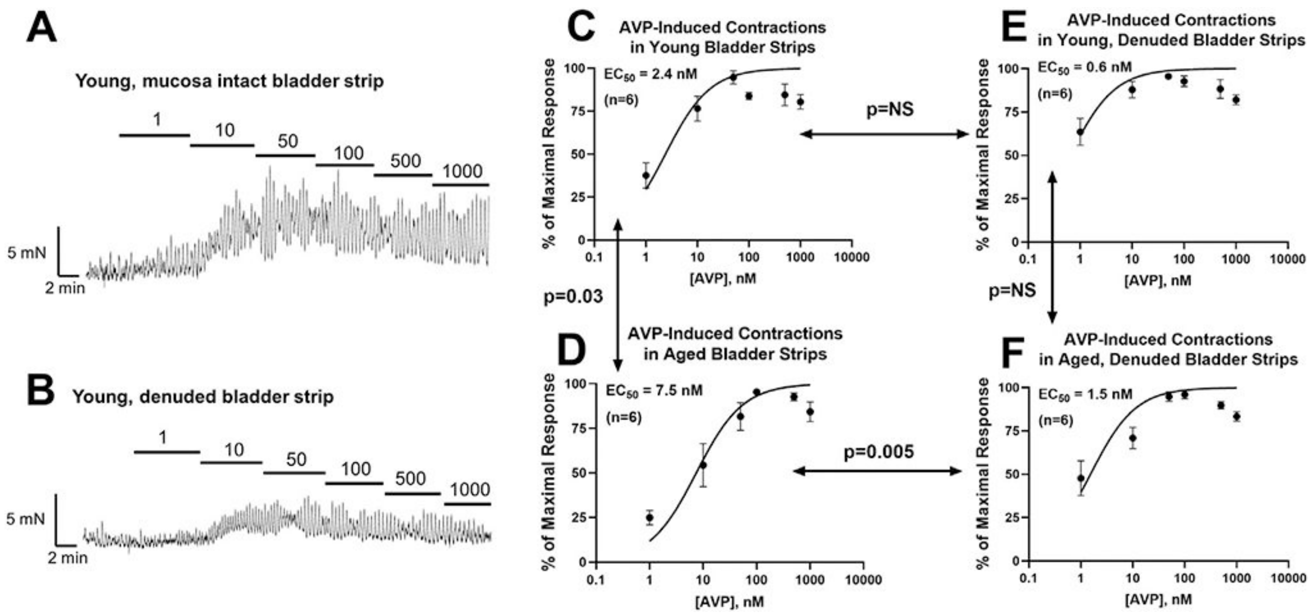

Fig. 2. Concentration–response relationships for AVP-induced contractions of rat bladder strips.
Example AVP dose–response tension trace from **A**, intact and **B**, mucosa denuded young rat bladder strips (1 to 1000 nM AVP). Shown are concentration–response curves for AVP-induced contractions in **C**, young, **D**, young denuded, **E**, aged and **F**, aged denuded bladder strips (n = 6 each). Curves were generated using Prism GraphPad to apply non-linear regression to the standard equation for [agonist] versus normalized response, mean ± SEM. P-values associated with bidirectional arrows (⟺) indicate comparisons between curves to determine whether best-fit unshared EC_{50} values differ between curves (extra sum-of-squares F test).

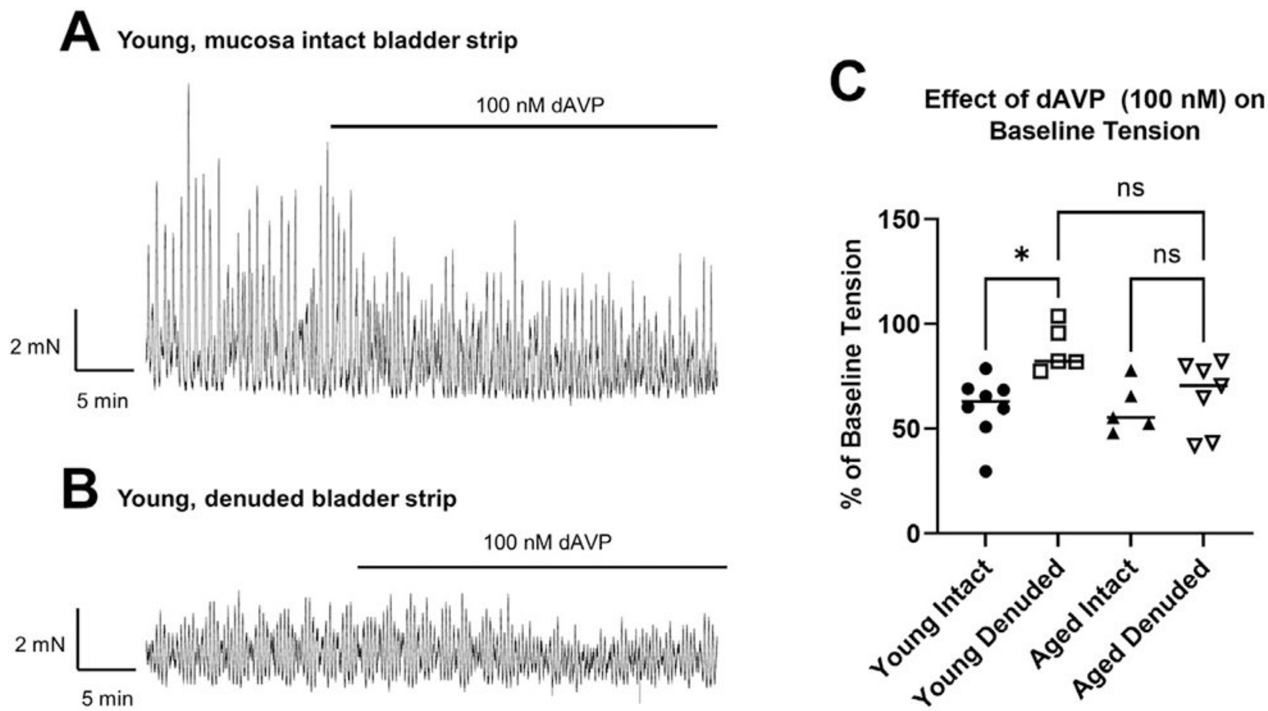

Fig. 3. Change in baseline tone in response to single dose challenge with desmopressin in young and aged rat bladder strips.

Example tension traces from **A**, mucosa intact and **B**, denuded young rat bladder strips in the presence of 100 nM desmopressin (dAVP). **C**. Bar graph of percentage change in baseline tone from young intact (n = 8), young denuded (n = 5), aged intact (n = 5) and aged denuded (n = 7). Mean ± SD, *p = 0.01, ns = not significant, one-way ANOVA with Tukey's post-hoc test.

Table 1

Role of vasopressin receptors in young and aged urinary bladders.

• Activation of V_2Rs with desmopressin reduces bladder tone.
• Non-selective activation of V_1Rs and V_2Rs with AVP increases bladder tone, suggesting that V_1Rs mediate bladder contraction that overrides V_2R induced relaxation.
• In the aged bladder, both V_1R- and V_2R-mediated responses are reduced relative to responses in the young bladder.
• The VR signaling malfunction in the aged bladder resides in the bladder mucosa (urothelium).
• Data is consistent with the hypothesis that administration of a V_2R agonist (e.g., desmopressin) may be an effective option for nocturia.