World Journal of Urology (2019) 37:2199–2205
https://doi.org/10.1007/s00345-019-02647-4

ORIGINAL ARTICLE

Liquid chromatography–mass spectrometry identification of serum biomarkers for nocturia in aged men

Satoru Kira[1] · Takahiko Mitsui[1] · Tatsuya Miyamoto[1] · Tatsuya Ihara[1] · Hiroshi Nakagomi[1] · Yuka Hashimoto[2] · Hajime Takamatsu[2] · Masayuki Tanahashi[2] · Masahiro Takeda[2] · Norifumi Sawada[1] · Karl-Erik Andersson[3] · Masayuki Takeda[1]

Received: 19 October 2018 / Accepted: 16 January 2019 / Published online: 23 January 2019

Abstract

Purpose We aimed to investigate the association between nocturia and serum metabolites identified using metabolomics analysis.

Methods This study enrolled 66 men aged 65–80 years, recruited from the outpatient department of a university hospital. The participants were stratified as follows: Nocturia group [45 men with any total international prostate symptom score (IPSS) and an average of 3 nights ≥ 1.5 micturitions/night] and Control group (21 men with total IPSS < 8 and an average of 3 nights < 1.5 micturitions/night). The 24-h frequency–volume chart, IPSS, and Quality-of-Life questionnaire were used to evaluate micturition behavior. Serum metabolite profiles were obtained using liquid chromatography–mass spectrometry (LC–MS)-based metabolomics analysis and compared between the two groups using the unpaired *t* test. The relationship between serum metabolites and nocturia was determined using multivariable logistic regression analysis.

Results There were no differences in background factors between the Nocturia and Control groups. In the IPSS, mean total scores in the Nocturia and Control groups were 12.4 and 4.0, respectively. On frequency–volume chart analysis, nocturnal urine volume and micturition frequency during daytime and nighttime were significantly higher in the Nocturia group. LC–MS highlighted 13 serum metabolites as potential biomarkers of nocturia. On multivariate analysis, increased levels of palmitoylethanolamide, 4-hydroxydocosahexaenoic acid, 9-hydroxyoctadecadienoic acid, 20-hydroxydocosahexaenoic acid, 13-hydroxyoctadecadienoic acid, arachidonoylethanolamide, eicosapentaenoic acid, 12-hydroxy-eicosatetraenoic acid, and arachidonic acid were associated with nocturia.

Conclusions In aged men, the pathogenesis of nocturia involves abnormal metabolism in several signaling pathways involving omega-3 and omega-6 polyunsaturated fatty acids, as well as endocannabinoids.

Keywords Nocturia · Aged · Liquid chromatography mass spectrometry · Metabolomics

Introduction

Nocturia represents an exceedingly widespread lower urinary tract symptom (LUTS) with detrimental effects on quality of life (QOL), quality of sleep, and overall health condition in older people [1]. The etiology of nocturia is multifactorial and may be due to several factors acting alone or in combination; such factors include polyuria, nocturnal polyuria, reduced bladder capacity, sleep disorders, and aging. Nocturia is also associated with numerous morbidities including obesity, obstructive sleep apnea, coronary artery disease, diabetes mellitus (DM), hypertension (HT), and metabolic syndrome (Mets) [1–3]. The exact etiology of nocturia remains unclear, and no single treatment has been proven universally effective and safe in elderly patients with nocturia [4].

Low-molecular-weight biomarkers are compounds that can serve as characteristic features to recognize particular pathologies. Metabolomics analysis can be used to identify novel biomarkers and screen for biomarkers of interest. The metabolomics approach has been successfully applied to profile small-molecule metabolites and reveal insights into the specific metabolic phenotypes linked with several diseases [5, 6]. Liquid chromatography–mass spectrometry

✉ Satoru Kira
skira@yamanashi.ac.jp

Extended author information available on the last page of the article

(LC–MS) represents a sensitive analytical technique that has been included in metabolomics pipelines to produce an effective screening method for the detection of blood biomarkers for Alzheimer's disease and for ovarian, liver, lung, and pancreatic cancer [7, 8]. However, no reports have described the use of LC–MS-based metabolomics analyses for screening serum samples from patients with nocturia.

Here, we performed an LC–MS-based metabolomics analysis to investigate the association between serum metabolites and nocturia in aged men.

Materials and methods

Ethics statement

We conducted a single-center, prospective study from November 2013 to May 2015. The Regional Ethics Committee of our university, which is responsible for the evaluation of epidemiological studies, reviewed and approved this study, which was performed according to the guidelines of the Declaration of Helsinki and its later amendments. All participants provided written informed consent before study enrollment.

Participants

A total of 66 men aged 65–80 years were recruited from the outpatient department of our university. The frequency–volume chart (FVC) for 3 consecutive days, international prostate symptom score (IPSS), and QOL score were used to evaluate LUTS. The presence of comorbidities such as HT, DM, and hyperlipidemia was determined by interviewing the participants and reviewing their medical charts. Men with any total IPSS and an average of 3 nights ≥ 1.5 micturitions per night were assigned to the Nocturia group, whereas those with total IPSS < 8 and an average of 3 nights < 1.5 micturitions per night were assigned to the Control group. The exclusion criteria were cancer, bladder pain syndrome, urinary tract infection, polyuria (40 mL/kg/24 h), nocturnal polyuria index (NPi) $> 70\%$, residual urine volume > 100 mL, or serious systemic complications such as central nervous system disorders, chronic kidney disease, and heart failure. NPi was calculated as urine production during nighttime divided by 24-h urine production. Nocturnal polyuria was defined as NPi > 0.33 [9].

Serum sampling and sample preparation

Blood serum samples from fasted patients were collected at our department between 8:00 and 9:00 am, and stored at -80 °C for metabolic analysis. A total of 3 mL were obtained per patient. After agitation of the sample, 200 µL of serum were added to 800 µL of methanol-containing internal standards, prostaglandin E2 (PGE2-d4), arachidonic acid (AA-d8), leukotriene B4 (LTB4-d4), and 12-hydroxyeicosatetranoic acid (12-HETE-d8). The solution was thoroughly mixed and centrifuged at 3000×g and 4 °C for 10 min. The supernatant was collected and added to 5 mL of hydrochloric acid solution. The mixture was applied to an Oasis® HLB 30 mg cartridge (Waters, Milford, MA, USA) conditioned with 4.5 mL of methanol solution and Milli-Q for solid-phase extraction. After washing the cartridge with 4.5 mL of Milli-Q and 3 mL of hexane, the retained analytes were eluted using 500 µL of methanol. The eluent was dried using nitrogen gas and re-dissolved using 50 µL of 50% acetonitrile for LC–MS analysis.

LC–MS conditions and analysis

Chromatographic separation was performed using Agilent 1200 Series equipment (Agilent Technologies, Palo Alto, CA, USA). Gradient elution was performed on Kinetex C8 columns (2.1 mm × 150 mm, 2.6 µm, kept at 40 °C; Phenomenex, Torrance, CA, USA) using 0.1% (v/v) formic acid in acetonitrile (mobile phase A) and 0.1% (v/v) formic acid in Milli-Q water (mobile phase B) with a flow rate of 0.4 mL/min. The total runtime was 24 min, and the injection volume was 5–10 µL. MS detection was conducted on an Agilent 6460 triple-quadrupole mass spectrometer (Agilent Technologies). Electrospray ionization was performed in both positive- and negative-ion modes. The equipment was controlled using the MassHunter B06.00 software (Agilent Technologies), and the collected data were processed using the MassHunter Quantitative Analysis software, version B. 07.00 (Agilent Technologies). The peaks were extracted to obtain peak information including m/z, LC retention time, and peak area. Peak annotation was performed using an internal database of retention times and m/z values for reference compounds. Lipid species were identified according to a list of retention times and selected reaction monitoring transitions acquired in a separate run for 90 lipid mediators. The peak areas were normalized against those of the internal standards, and the peak area ratios were used for further statistical analyses.

Statistical analyses

Subject characteristics and FVC parameters were analyzed using the unpaired Welch t test, while Fisher's exact test was used for categorical variables. Metabolite profiles were compared between the Nocturia group and the Control group using the Welch t test as a non-adjusted analysis. To determine the relationship between metabolites and nocturia, we also performed multivariable logistic regression analysis and obtained odds ratios and 95% confidence intervals.

The following covariates and adjustments were included: age (continuous), body mass index (continuous), 24-h urine production (continuous), use of drugs for LUTS treatment (yes/no), and presence of metabolic comorbidities including HT, DM, or hyperlipidemia (yes/no). p values < 0.05 were considered to indicate statistical significance. Statistical analyses were performed using SPSS version 22 (IBM Corp., Armonk, NY, USA).

Results

Of the 66 men enrolled in the study, 45 were assigned to the Nocturia group and 21 to the Control group. The two groups did not differ regarding age, body mass index, prevalence of any specific lifestyle-related disease, or use of most drugs for LUTS, but the numbers of participants receiving anticholinergic drugs or having a lifestyle-related disease (any of the three evaluated here) were significantly higher in the Nocturia group (Table 1).

FVC analysis revealed that micturition frequency during daytime and nighttime, urine production during nighttime, NPi, and prevalence of nocturnal polyuria were significantly higher in the Nocturia group than in the Control group, whereas voided volume per micturition during daytime and nighttime was significantly smaller in the Nocturia group. There were no significant differences between the two groups regarding urine production during daytime or 24-h total urine production (Table 2).

Of the 90 lipid metabolites investigated in this LC–MS-based metabolomics study, 39 were detected in the serum samples. After excluding lipids with levels below 20%, 19 lipids were included in the analysis. According to the unpaired *t* test, the levels of 13 serum metabolites were significantly higher in the Nocturia group than in the Control group (Table 3).

Multivariable logistic regression analysis revealed that increased levels of palmitoylethanolamide (PEA), 4-hydroxydocosahexaenoic acid (4-HDoHE), 9-hydroxyoctadecadienoic acid (9-HODE), 20-HDoHE, 13-HODE, arachidonoylethanolamide (AEA), eicosapentaenoic acid (EPA), 12-HETE, and arachidonic acid were significantly associated with nocturia in aged men (Table 4).

Table 1 Baseline characteristics of study participants stratified into the Control and Nocturia groups

Characteristic	Control	Nocturia	p value
Age, years	70.5 ± 4.0	72.3 ± 4.0	0.1119
Body mass index, kg/m^2	22.9 ± 2.4	23.0 ± 3.0	0.8802
Lifestyle-related disease	Number	Number	
Hypertension	2	14	0.0697
Diabetes mellitus	1	3	> 0.99
Hyperlipidemia	0	5	0.1691
Either of above diseases	2	17	0.0209
Drug for LUTS	Number	Number	
α(1)-Receptor antagonist	12	36	0.0753
5α-Reductase inhibitors	5	8	0.7406
Anticholinergic drug	0	8	0.0478
β(3)-Receptor agonist	2	9	0.4802
PDE(5)-inhibitor	1	1	0.5385

Values are mean ± SD

LUTS lower urinary tract symptoms, *PDE* phosphodiesterase

Discussion

This LC–MS-based metabolomics study identified several serum biomarkers for nocturia in aged men. While LC–MS revealed 13 serum metabolites potentially associated with nocturia, logistic multivariable regression analysis suggested that increased levels of nine metabolites are significantly associated with nocturia. Because these metabolites are n-3 or n-6 polyunsaturated fatty acids (PUFAs) or endocannabinoids, our findings suggest that abnormalities

Table 2 Frequency–volume chart parameters describing the micturition behavior of patients in the Nocturia and Control groups

Parameter	Control	Nocturia	p value
Micturition frequency during daytime	6.28 ± 2.2	7.96 ± 2.3	0.0055
Micturition frequency during nighttime	0.69 ± 0.5	2.47 ± 0.7	< 0.0001
Voided volume per micturition during daytime, mL	208 ± 83	158 ± 63	0.0084
Voided volume per micturition during nighttime, mL	307 ± 132	204 ± 57	< 0.0001
24-h total urine production, mL	1530 ± 480	1715 ± 413	0.11
Urine production during daytime, mL	1055 ± 425	1022 ± 337	0.74
Urine production during nighttime, mL	475 ± 150	694 ± 177	< 0.0001
NPi	0.325 ± 0.1	0.41 ± 0.09	0.0005
No. of nocturnal polyuria pts.	8	35	0.002

Values are mean ± SD

NPi nocturnal polyuria index

Table 3 Metabolomic profiles of serum samples from subjects with nocturia and those without nocturia

Compound	Ratio	Non-adjusted *p* value
Palmitoylethanolamide	1.35	< 0.0001
4-HDoHE	1.87	0.00013
9-KODE	1.66	0.0056
13-KODE	1.71	0.0059
9-HODE	1.37	0.0061
20-HDoHE	1.50	0.0071
13-HODE	1.36	0.0076
Arachidonoyl ethanolamide	1.34	0.012
Eicosapentaenoic acid	1.57	0.013
Docosahexaenoic acid	1.30	0.026
19,20-diHDoPE	1.33	0.028
12-HETE	1.26	0.046
Arachidonic acid	1.26	0.048

The ratio refers to the value noted in subjects with nocturia, divided by the value noted in subjects without nocturia

p values refer to the between-group differences

HDoHE hydroxydocosahexaenoic acid, *KODE* oxo-octadecadienoic acid, *HODE* hydroxyoctadecadienoic acid, *HETE* hydroxy-eicosatetraenoic acid

Table 4 Metabolomic biomarkers of nocturia

Biomarker	Odds ratio	95% CI	*p* value
Palmitoylethanolamide	2.262	1.342, 3.813	0.002
4-HDoHE	2.111	1.161, 3.839	0.014
9-KODE	2.365	0.903, 6.199	0.08
13-KODE	1.291	0.996, 1.673	0.054
9-HODE	3.006	1.267, 7.131	0.013
20-HDoHE	1.561	1.024, 2.380	0.039
13-HODE	2.006	1.097, 3.667	0.024
Arachidonoyl ethanolamide	10.934	1.519, 75.155	0.015
Eicosapentaenoic acid	1.897	1.010, 3.564	0.047
Docosahexaenoic acid	1.282	0.982, 1.674	0.068
19,20-diHDoPE	1.239	0.960, 1.600	0.099
12-HETE	1.823	1.060, 3.135	0.030
Arachidonic acid	1.650	1.046, 2.601	0.031

95% CI 95% confidence interval, *HDoHE* hydroxydocosahexaenoic acid, *KODE* oxo-octadecadienoic acid, *HODE* hydroxyoctadecadienoic acid, *diHDoPE* dihydroxy-docosapentaenoic acid, *HETE* hydroxy-eicosatetraenoic acid

in signaling pathways involving these metabolites play a role in the pathogenesis of nocturia in aged men. Relevant metabolites detected on the LC–MS analysis include: the n-3 PUFAs EPA, 4-HDoHE, 20-HDoHE, docosahexaenoic acid (DHA), and 19,20-dihydroxy-docosapentaenoic acid (19,20-diHDoPE); the n-6 PUFAs arachidonic acid, 12-HETE, 9-HODE, 13-HODE, 9-oxo-octadecadienoic acid (9-KODE), and 13-KODE; the endocannabinoids AEA and PEA.

Among the 13 serum metabolites potentially associated with nocturia in aged men, arachidonic acid, 12-HETE, 9-HODE, 13-HODE, 9-KODE, and 13-KODE are part of the n-6 PUFA metabolism. Among the contributors to n-6 PUFA metabolism, cyclooxygenase-1 (COX-1) and COX-2, which regulate arachidonic acid metabolism pathways, play a key role in cell inflammation. COX-2 was found to be highly expressed in human prostate tissue [10], and inflammation is known to play a role in benign prostatic hyperplasia and associated LUTS. Prostanoids are believed to be involved in the pathophysiology of LUTS, and several nonsteroidal anti-inflammatory drugs have shown efficacy in the treatment of nocturia. Thus, the effects of COX inhibitors such as diclofenac [11] and celecoxib [12] have been evaluated in benign prostatic hyperplasia patients with nocturia. Although COX inhibitors may induce several toxic side effects, our results suggest that the COX pathway may, indeed, be a suitable treatment target in nocturia.

Mets is comprised of several cardiovascular risk factors such as obesity, HT, DM, and dyslipidemia. A previous epidemiological survey revealed that Mets is also associated with nocturia [2]. Another survey from Japan confirmed that the risk for nocturia increases significantly with the number of Mets components [13]. This is in agreement with our present observations, namely that the number of lifestyle-related diseases was significantly higher in the Nocturia group. Moreover, logistic multivariable regression analysis indicated an association of nocturia with 9-HODE and 13-HODE, which are metabolites of linoleic acid (an n-6 PUFA). It has been suggested that both these HODEs are involved in the development of interleukin-1b-mediated vascular dysfunction and atherosclerosis and may become components of atheromatous plaques [14, 15]. Furthermore, the previous reports showed that, in aged men, atherosclerosis was associated with erectile dysfunction and LUTS, including nocturia [16, 17]. Thus, we speculate that 9-HODE or 13-HODE could be involved in nocturia due to the role of such compounds in atherosclerosis and vascular dysfunction.

We found that elevated serum levels of the endocannabinoids PEA and AEA were associated with nocturia in aged men. The endocannabinoid system consists of the cannabinoid receptors (cannabinoid receptor type 1, type 2, and the G-protein-coupled receptor 55), ligands such as AEA, PEA, and 2-arachidonoylglycerol, and related enzymes for biosynthesis or degradation. Evidence from many animal studies suggests that components of the endocannabinoid system are involved in the regulation of lower urinary tract function [18, 19]. For example, using fatty acid amide hydrolase inhibitors to prevent the degradation of endocannabinoids and related amidated signaling lipids results in the amplification of endocannabinoid activity and appears to be an attractive approach for treating lower urinary tract dysfunction [18, 19]. However, clinical studies on the effects of interfering with the cannabinoid system in LUTS are scarce and essentially restricted to actions of cannabinoid extracts in patients with multiple sclerosis, and the results have so far not been convincing [20]. It has been suggested that dysfunction of the endocannabinoid system might be associated with obesity and Mets, because overproduction of endocannabinoids may induce reward system dysfunction [21]. In this context, our findings support the hypothesis that abnormal levels of serum cannabinoids might be involved in the etiology of nocturia due to the relationship of such compounds with the development of Mets.

Our LC–MS analysis identified n-3 and n-6 PUFAs, which are metabolites of essential fatty acids, as relevant in nocturia. Essential fatty acids cannot be synthesized by mammals and must, therefore, be obtained from food. While an increased n-6/3 PUFA ratio, which is characteristic of current Western dietary patterns, was reported to be associated with increased risk of obesity, type 2 DM, and cardiovascular disease, diets with a good n-6/3 PUFA ratio are considered to provide health benefits [22]. Since obesity, type 2 DM, and cardiovascular disease are associated with nocturia, it may be hypothesized that an increased n-6/3 PUFA ratio might also be associated with nocturia. In this context, our present findings suggest that participants categorized in the Nocturia group may have consumed a diet with an inappropriate n-6/3 PUFA ratio, contributing to nocturia. However, the n-3 PUFAs EPA and DHA are known to have neuroprotective, anti-inflammatory, cardioprotective, and antioxidative effects [23, 24]. Considering these positive effects, one may speculate that elevated serum levels of EPA and DHA would be associated with reduced nocturia symptoms. However, our present findings contradict this expectation. Furthermore, a previous prospective cohort study reported that increased intake of EPA and DHA was associated with higher risk of benign prostatic hyperplasia [25].

It is generally considered that Mets comorbidities, which include lifestyle-related diseases such as HT, DM, and hyperlipidemia, play a key role in nocturia. Therefore, lifestyle intervention or education regarding Mets comorbidities might improve nocturia. A questionnaire-based study reported that a self-management program including fluid management and avoidance of caffeine and alcohol in addition to the standard care resulted in a significant improvement of nocturia in men [26]. An FVC-based study found that, following 1 year of intervention, there were no differences in nocturia between lifestyle intervention focused on weight loss and diabetes support and education in men with type 2 DM [26, 27]. Thus, whether lifestyle intervention to improve Mets comorbidities can also improve nocturia remains unclear. Considering the results of the present study, future research is warranted to elucidate the role of lifestyle interventions, including those focused on the dietary intake of n-3 and n-6 PUFAs.

This study has several limitations. First, the sample size was small and all participants were aged men. In addition, this study had a cross-sectional design. Finally, we did not have enough data about diet, fluid consumption, or caffeine intake. Because the serum metabolites of n-3 and n-6 PUFAs depend on dietary food intake, future investigations should have a longitudinal study design containing lifestyle habit and a larger sample, including both men and women.

Conclusion

Our findings suggest that abnormal metabolism in the n-3, 6 PUFA, and endocannabinoid signaling pathways are associated with nocturia in aged men. Future studies with longitudinal setting and employing metabolomics analysis are warranted to identify potential targets for new treatments of nocturia.

Acknowledgements This study was funded by Astellas Pharma, Inc. and designed by Astellas Pharma, Inc. in collaboration with the authors. We thank Ms. Sachiko Tsuchiya for her assistance with data management.

Author contribution SK: data collection, data analysis, and manuscript writing. TM: data analysis and manuscript editing. TM: data collection and data analysis. TI: data analysis. HN: data collection and data analysis. YH: data analysis. HT: data analysis. MT: data analysis. MT: data analysis and other (supervision). NS: data analysis. K-EA: manuscript editing and other (supervision). MT: data analysis, manuscript editing, and other (supervision)

Funding This study was funded by Astellas Pharma, Inc.

Compliance with ethical standards

Conflict of interest The study was designed by Astellas Pharma, Inc. in collaboration with the authors. Several of the authors are employees of Astellas Pharma, Inc. The other authors have no potential competing interests to declare.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Informed consent Informed consent was obtained from all individual participants included in the study.

References

1. Van Kerrebroeck P, Andersson KE (2014) Terminology, epidemiology, etiology, and pathophysiology of nocturia. Neurourol Urodyn 33(S1):S2–S5. https://doi.org/10.1002/nau.22595

2. Kupelian V, McVary KT, Kaplan SA, Hall SA, Link CL, Aiyer LP, Mollon P, Tamimi N, Rosen RC, McKinlay JB (2009) Association of lower urinary tract symptoms and the metabolic syndrome: results from the Boston Area Community Health Survey. J Urol 182(2):616–624. https://doi.org/10.1016/j.juro.2012.11.026
3. Fitzgerald MP, Litman HJ, Link CL, McKinlay JB, BACH Survey Investigators (2007) The association of nocturia with cardiac disease, diabetes, body mass index, age and diuretic use: results from the BACH survey. J Urol 177(4):1385–1389. https://doi.org/10.1016/j.juro.2006.11.057
4. Fine ND, Weiss JP, Wein AJ (2017) Nocturia: consequences, classification, and management. F1000Res 6:1627. https://doi.org/10.12688/f1000research.11979.1
5. Yoshimi N, Futamura T, Kakumoto K, Salehi AM, Sellgren CM, Holmén-Larsson J, Jakobsson J, Pålsson E, Landén M, Hashimoto K (2016) Blood metabolomics analysis identifies abnormalities in the citric acid cycle, urea cycle, and amino acid metabolism in bipolar disorder. BBA Clin 5:151–158. https://doi.org/10.1016/j.bbacli.2016.03.008
6. White L, Ma J, Liang S, Sanchez-Espiridion B, Liang D (2017) LC–MS/MS determination of D-mannose in human serum as a potential cancer biomarker. J Pharm Biomed Anal 137:54–59. https://doi.org/10.1016/j.jpba.2016.12.017
7. Liu Y, Qing H, Deng Y (2014) Biomarkers in Alzheimer's disease analysis by mass spectrometry-based proteomics. Int J Mol Sci 15(5):7865–7882. https://doi.org/10.3390/ijms15057865
8. Tessitore A, Gaggiano A, Cicciarelli G, Verzella D, Capece D, Fischietti M, Zazzeroni F, Alesse E (2013) Serum biomarkers identification by mass spectrometry in high-mortality tumors. Int J Proteom 2013:125858. https://doi.org/10.1155/2013/125858
9. Weiss JP, Blaivas JG, Blanker MH, Bliwise DL, Dmochowski RR, Drake M, DuBeau CE, Hijaz A, Rosen RC, Van Kerrebroeck PE, Wein AJ (2013) The New England Research Institutes, Inc. (NERI) Nocturia advisory conference 2012: focus on outcomes of therapy. BJU Int 111(5):700–716. https://doi.org/10.1111/j.1464-410X.2012
10. Nickel JC, Roehrborn CG, O'Leary MP, Bostwick DG, Somerville MC, Rittmaster RS (2008) The relationship between prostate inflammation and lower urinary tract symptoms: examination of baseline data from the REDUCE trial. Eur Urol 54(6):1379–1384. https://doi.org/10.1016/j.eururo.2007.11.026
11. Addla SK, Adeyoju AB, Neilson D, O'Reilly P (2006) Diclofenac for treatment of nocturia caused by nocturnal polyuria: a prospective, randomised, double-blind, placebo-controlled crossover study. Eur Urol 49(4):720–725. https://doi.org/10.1016/j.eururo.2005.11.026
12. Falahatkar S, Mokhtari G, Pourreza F, Asgari SA, Kamran AN (2008) Celecoxib for treatment of nocturia caused by benign prostatic hyperplasia: a prospective, randomized, double-blind, placebo-controlled study. Urology 72(4):813–816. https://doi.org/10.1016/j.urology.2008.04.069
13. Aoki Y, Yokoyama O (2012) Metabolic syndrome and nocturia. Low Urin Tract Symptoms 4(S1):11–15. https://doi.org/10.1111/j.1757-5672.2011.00118.x
14. Waddington EI, Croft KD, Sienuarine K, Latham B, Puddey IB (2003) Fatty acid oxidation products in human atherosclerotic plaque: an analysis of clinical and histopathological correlates. Atherosclerosis 167(1):111–120. https://doi.org/10.1016/S0021-9150(02)00391-X
15. Ku G, Thomas CE, Akeson AL, Jackson RL (1992) Induction of interleukin 1 beta expression from human peripheral blood monocyte-derived macrophages by 9-hydroxyoctadecadienoic acid. J Biol Chem 267(20):14183–14188
16. Tsujimura A, Hiramatsu I, Aoki Y, Shimoyama H, Mizuno T, Nozaki T, Shirai M, Kobayashi K, Kumamoto Y, Horie S (2017) Atherosclerosis is associated with erectile function and lower urinary tract symptoms, especially nocturia, in middle-aged men. Prostate Int 5(2):65–69. https://doi.org/10.1016/j.prnil.2017.01.006
17. Inci M, Sarli B, Davarci M, Yalcinkaya FR, Rifaioğlu MM, Davran R, Arica S, Motor S, Demirbaş O (2013) Relationship between endothelial dysfunction and nocturia with benign prostatic hyperplasia. Scand J Urol 47(5):384–389. https://doi.org/10.3109/21681805.2012.762038
18. Andersson KE (2016) Potential future pharmacological treatment of bladder dysfunction. Basic Clin Pharmacol Toxicol 119(S3):75–85. https://doi.org/10.1111/bcpt.12577
19. Hedlund P (2014) Cannabinoids and the endocannabinoid system in lower urinary tract function and dysfunction. Neurourol Urodyn 33(1):46–53. https://doi.org/10.1002/nau.22442
20. Kavia RB, De Ridder D, Constantinescu CS, Stott CG, Fowler CJ (2010) Randomized controlled trial of Sativex to treat detrusor overactivity in multiple sclerosis. Mult Scler 16(11):1349–1359. https://doi.org/10.1177/1352458510378020
21. Vemuri VK, Janero DR, Makriyannis A (2008) Pharmacotherapeutic targeting of the endocannabinoid signaling system: drugs for obesity and the metabolic syndrome. Physiol Behav 93(4–5):671–686. https://doi.org/10.1016/j.physbeh.2007.11.012
22. Simopoulos AP (2011) Importance of the omega-6/omega-3 balance in health and disease: evolutionary aspects of diet. World Rev Nutr Diet 102:10–21. https://doi.org/10.1159/000327785
23. Lauritzen I, Blondeau N, Heurteaux C, Widmann C, Romey G, Lazdunski M (2000) Polyunsaturated fatty acids are potent neuroprotectors. EMBO J 19(8):1784–1793. https://doi.org/10.1093/emboj/19.8.1784
24. Saravanan P, Davidson NC, Schmidt EB, Calder PC (2010) Cardiovascular effects of marine omega-3 fatty acids. Lancet 376(9740):540–550. https://doi.org/10.1016/S0140-6736(10)60445-X
25. Suzuki S, Platz EA, Kawachi I, Willett WC, Giovannucci E (2002) Intakes of energy and macronutrients and the risk of benign prostatic hyperplasia. Am J Clin Nutr 75(4):689–697. https://doi.org/10.1093/ajcn/75.4.689
26. Yap TL, Brown C, Cromwell DA, van der Meulen J, Emberton M (2009) The impact of self-management of lower urinary tract symptoms on frequency–volume chart measures. BJU Int 104(8):1104–1108. https://doi.org/10.1111/j.1464-410X.2009.08497.x
27. Breyer BN, Phelan S, Hogan PE, Rosen RC, Kitabchi AE, Wing RR, Brown JS, Look AHEAD Research Group (2014) Intensive lifestyle intervention reduces urinary incontinence in overweight/obese men with type 2 diabetes: results from the look AHEAD trial. J Urol 192(1):144–149. https://doi.org/10.1016/j.juro.2014.02.036

Affiliations

Satoru Kira[1] · Takahiko Mitsui[1] · Tatsuya Miyamoto[1] · Tatsuya Ihara[1] · Hiroshi Nakagomi[1] · Yuka Hashimoto[2] · Hajime Takamatsu[2] · Masayuki Tanahashi[2] · Masahiro Takeda[2] · Norifumi Sawada[1] · Karl-Erik Andersson[3] · Masayuki Takeda[1]

Takahiko Mitsui
tmitsui@yamanashi.ac.jp

Tatsuya Miyamoto
tatsuyamiyamoto.urology@gmail.com

Tatsuya Ihara
tihara@yamanashi.ac.jp

Hiroshi Nakagomi
hnakagom@yamanashi.ac.jp

Yuka Hashimoto
yuka.hashimoto@astellas.com

Hajime Takamatsu
hajime.takamatsu@astellas.com

Masayuki Tanahashi
masayuki.tanahashi@astellas.com

Masahiro Takeda
masahiro.takeda@astellas.com

Norifumi Sawada
nsawada@yamanashi.ac.jp

Karl-Erik Andersson
keanders@wakehealth.edu

Masayuki Takeda
matakeda@yamanashi.ac.jp

1 Department of Urology, Interdisciplinary Graduate School of Medicine and Engineering, University of Yamanashi, Shimokato 1110, Chuo, Yamanashi 409-3898, Japan

2 Pharmacology Research Labs, Astellas Pharma Inc, Tsukuba, Japan

3 Institute for Regenerative Medicine, Wake Forest University School of Medicine, Winston-Salem, NC, USA