Galactoglucomannan-rich hemicellulose extract from Norway spruce (*Picea abies*) exerts beneficial effects on chronic prostatic inflammation and lower urinary tract symptoms *in vivo*

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Abstract

Galactoglucomannan (GGM) is the main hemicellulose class in wood of coniferous trees and could be potentially utilized as a possible health-promoting substance for food and pharmaceutical industry. Our aim was to evaluate effects of orally administered GGM-rich extract from Norway spruce in a rat model of chronic prostatitis associated with lower urinary tract symptoms (LUTS). Prostatic inflammation and LUTS was induced in male rats using testosterone and 17β-estradiol exposure for 18 weeks. Rats were treated with 2% GGM dissolved in drinking water during weeks 13 to 18. Pelvic pain response, LUT function and histopathological evaluation of the prostate were assessed. The results show that hormonal exposure induced LUTS seen as decreased urine flow rate, increased bladder pressure, voiding times, bladder capacity and residual urine volumes. GGM had positive effects on urodynamical parameters by decreasing the basal bladder pressure, increasing the urine flow rate and volume, reducing the residual volume and increasing micturition intervals. GGM reduced the extent of the hormone exposure-induced prostatic inflammation. Increase of pelvic pain induced by hormone exposure was only slightly affected by GGM treatment. The results suggest that orally administered GGM may have potential usage for improving lower urinary tract function associated with chronic prostatic inflammation.

Key words: Galactoglucomannan; Lower Urinary Tract Symptoms, Prostatitis

1. Introduction

Hemicelluloses constitute a complex group of heterogeneous polysaccharides embedded in cell walls of trees. Acetylated galactoglucomannan (GGM) is the main water-soluble hemicellulose found abundantly in the wood of coniferous tree species such as Norway spruce *(Picea abies)* [1]. There are interesting findings showing that wood-derived GGM exerts health promoting effects and could possibly be utilized as health-promoting substance for food and pharmaceutical industry. GGM extract has shown to have immunomodulating and radical-scavening [2] activities and prebiotic activities *in vitro* [3]. GGM oligosaccharides has been shown to have prebiotic activities *in vitro* [4] and in dog [5] and effects on increasing fermentation and immune responses in chicks [6]. GGM-derived oligosaccharides have also been showing to improve colonic health in *Salmonella*-infected broiler chicks [7]. Additionally, GGM extracted from *Dendrobium huoshanense* has been also shown to prevent selenium-induced liver damage and fibrosis in rats [8] and sulfated GGM has shown *in vivo* anticoagulant and antithrombotic activities [9].

The nonbacterial chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS) is a common disease affecting men of all ages, and the incidence increases in men with age over 65 [10]. There is increasing evidence supporting the idea that chronic prostatitis can eventually be the etiology to prostate cancer [11]. The symptoms of CP/CPPS are heterogeneous, mostly manifested as pain in the pelvic region as well as lower urinary tract symptoms (LUTS). Histopathologically, chronic inflammation is seen in the prostatic stroma, intraepithelial space and inside the acini [12]. The etiology of the CP/CPPS is unknown and multiple mechanisms have been proposed in the pathogenesis of prostatitis [13]. Increasing interest for treatment and prevention of CP/CPPS is directed on therapeutical usage of naturally occurring phytotherapical

compounds [14]. Studies using plant derived polysaccharides from citrus fruit [15], *Lycium barbarum* (Goji berry) [16] and *Urtica fissa* (Stinging nettle) [17] have shown to exert potential effects on experimental models of prostate cancer and benign prostate hyperplasia. Chronic prostatitis can be studied using preclinical models where gradual development of prostatic inflammation after sex hormone exposure is seen in adult male rats [18-25], resembling human chronic prostatic inflammation CP/CPPS. Additionally, hormonal exposure induces LUTS which has been shown to be associated with prostatic glandular inflammation when the testosterone to 17β- estradiol ratio is high [22] [26] [27].

Since previous studies has shown potential of utilizing plant-derived polysaccharides against prostate-related diseases, our aim was to study whether softwood-derived polysaccharide compounds could be utilized in this concept. We investigated for the first time the *in vivo* effects of the GGM-rich hemicellulose extract from Norway spruce on prostatic inflammation and associated changes on voiding and pain using a non-bacterial prostatic inflammation rat model.

2. Materials and methods

2.1 Extraction of galactoglucomannan-rich hemicellulose extract

The extract (galactoglucomannan-rich hemicellulose extract, abbreviated in the text as GGM) was isolated from Norway spruce (*Picea abies*) using the following methods: spruce wood meal (< 2 mm) was extracted using a flow-through extractor with water at 170°C [28] [29] and further purified by precipitation in 85% ethanol yielding a heteropolysaccharide preparate with a molar mass in the range of 4-20 kD, with an average molar mass of 8.2 kD. The monosaccharide composition determined by acid methanolysis and gas chromatography [30] was galactose 7%,

glucose 15%, mannose 60% (i.e. total GGM 82%); arabinose 1%, 4-O-methylglucuronic acid 2%, xylose 9% (i.e. total xylans 12 %); rhamnose 0.3%, galacturonic acid 3.5% (i.e. total pectins 4%). The product contained 0.6% acetyl groups, corresponding to an acetyl:mannose ratio of about 0.5.

2.2 Experimental animals and study design

Adult male Wistar rats (RccHan:WIST) were obtained from Harlan Laboratories Inc. (the Netherlands) and housed pairwise in the animal facilities of Central Animal Laboratory of University of Turku in Macrolon Type III (800 cm^2) cages. The animals were left to acclimate for 7 days before any procedure, aspen chips (Tapvei Estonia Ltd, Estonia) were provided as bedding material, and soy-free rodent pellets (Harlan Diets 2016 global 16% protein rodent diet) and water was provided *ad libitum*. All animals were housed pairwise under a 12-h light-dark cycle and constant room temperature (± 3°C, humidity 55% ± 15%). The animal experiment was complying the EU Directive 2010/63/EU and ARRIVE guidelines for animal experiments and the study protocol (no: ESAVI/1455/04.10.02/2011) was approved by the National Animal Experiment Board of Finland. The rats were handled in accordance with the institutional animal care policies of the Central Animal Laboratory of University of Turku. The welfare of the animals was monitored daily during the study. The age of the animals was 11-12 weeks (weight 356 ± 14 g) at the beginning of the study. The animals were stratified into groups based on body weight and treated as follows. Group 1: placebo pellet + vehicle (tap water); Group 2: Testosterone +17β- estradiol pellets + vehicle (tap water) and Group 3: Testosterone+17β- estradiol pellets + GGM (2% GGM solution in tap water).

Testosterone (T), 17β- estradiol (E_2) and corresponding placebo hormone implants were obtained from Innovative Research of America (IRA, FL, USA). Implants were 60-day releasing implants with a daily release of 830 μg for T and 83 μg for E_2. Rats were anesthetized with isoflurane (3%, 200 mL/min, Piramal Healthcare Ltd, UK) and the pellets were inserted in subcutaneous pockets formed over the scapular area. Implants were replaced with identical new ones twice during the study (on treatment weeks 6 and 13). The total hormone exposure period was 18 weeks. The period of treatment with GGM was decided to start on week 13 based on preliminary studies showing that the inflammation in the prostate in the Wistar rats begins to develop more after 13 weeks of hormonal exposure. For the treatment period of 5 weeks (between study weeks 13 and 18) GGM was dissolved in tap water as a 2% solution and given to the T+E_2+GGM group. Tap water was given to other two groups (placebo+vehicle and T+E_2+vehicle). Access to drinking and food was *ad libitum* and the consumption was monitored for two weeks during the treatment period.

2.3 Pelvic pain assessment

Referred hyperalgesia reflecting pain in pelvic area was assessed (modified from [31]) at study weeks 6, 13 and 18 using von Frey filaments (North Coast Medical Inc., CA). For the measurements, each animal was placed in a gridiron-floor cage and the area in the vicinity of the prostate was stimulated using the filaments and a positive response was shown as a sharp retraction of the abdomen, immediate licking or scratching of the area of filament stimulation or jumping. Withdrawal thresholds were measured in response to increasing pressure stimuli (7 different filaments with a bending force ranging from 2 to 100 g) applied to the pelvic area. Response to each filament force was measured 10 times beginning from the lowest force

filament. The bending force of the filament to which the animal responded was taken as the baseline threshold to mechanical stimulus. The median response threshold was calculated from the median values of each 10 measurements.

2.4 Urodynamical measurements

At the end of the study (week 18) urodynamic measurements were performed. Rats were anesthetized with chloral hydrate *i.p.* (0.36 g/kg) for a basic anesthetic, and *i.v.* injections of urethane (0.32 g/kg, both Sigma Chemical Co. St. Louis, USA) was given to maintain anesthesia during the urodynamical measurements, if needed. The body temperature was kept constant by a thermostatically controlled animal blanket. The bladder and the distal part of urethra were exposed with a midline incision of the lower abdomen. In transvesical cystometry, a 20G *i.v*. cannula was inserted through the bladder apex into the lumen. The cannula was connected to an infusion pump (World Precision Instruments, Inc., Sarasota, USA) and to a pressure transducer (Statham, Hato Ray, Puerto Rico). Measurements were made with warm saline at an infusion rate of 10 mL/h. An ultrasonic flow probe was used for measurement of the urine flow rate from the distal part of urethra. The flow probe was connected to a flow meter (both Transonic Systems, Inc. Ithaca, NY, USA). The pressure transducer was connected to an amplifier (Grass Instruments Co. Quincy, MA, USA). The pressure and urine flow signals were transferred to a Biopac-system and continuous recording was made with Acq Knowledge 3.5.3 software (Biopac Systems Inc., Santa Barbara, CA, USA). The animals were sacrificed immediately under anesthesia after the urodynamical measurements using CO_2 suffocation and neck dislocation. The following parameters were analyzed (blinded to treatment groups) from data obtained from the measurements: mean bladder pressure during micturition, basal bladder pressure between

micturitions, urine flow rate, bladder capacity, residual urine, voided volume, micturition time and micturition interval.

2.5 Histopathological assessment of prostatic inflammation

After the urodynamical measurements and animal sacrifice the hormone-responsive organs prostate-urethra complex, seminal vesicles and pituitary were weighed and excised. Prostate-urethra samples were fixed in 10% neutral formalin solution for 18-20 hours and moved to 70% ethanol for storage. After dehydration, samples were embedded in paraffin and 5 μm sections were cut out and stained with hematoxylin and eosin (H&E). Histopathological assessment was carried out on the H&E-stained prostate sections of each animal. From each block, four serial prostate sections were examined for inflammation: the number of perivascular, stromal/periglandular infiltrates and the number of the inflamed acini of dorsolateral prostate were counted blinded to treatment groups. The inflammation infiltrate was considered to be perivascular when more than ten inflammatory cells were found around the capillary and stromal/periglandular when cells were found in the prostatic stroma and periglandular space. The number of inflamed acini was counted when inflammation infiltrates were found inside the acini.

2.6 Statistical analysis

Statistical analyses were performed with SigmaStat (version 3.5, Systat Software Inc., Richmond, California, USA). Urodynamical data were analyzed using One-way ANOVA and Bonferroni t-test *post-hoc* test or non-parametric data Mann- Whitney Rank Sum Test and Dunn's Method as *post-hoc* test. For analysis of inflammation area counts data from all treatment

groups were first pooled and arranged then into order from lowest to highest values. The data were then divided evenly into three even categories: 1) 0, 2) >0-2 and 3) >2-5 inflammation area counts for perivascular inflammation; 1) 0-<1, 2) 1-6 and 3) >6-17 for stromal inflammation area counts and 1) 0-1, 2) >1-30 and >30-49 for inflamed acini counts. The proportions of different categories between the treatment groups were then analyzed using Chi-Square proportion analysis. The data is represented as difference in proportions of three categories of inflamed counts for each treatment group relative to total animal number (100%) in each treatment group. Data of pelvic pain were analyzed using Two Way Repeated Measures ANOVA and Bonferroni t-test. P-values ≤ 0.05 were considered statistically significant. The final animal number (originally n=12/group, some animal loss due to technical issues) was as following: n= 11 for placebo+ vehicle group, n= 9 for T+E_2+vehicle and n= 12 for T+E_2+GGM group.

3. Results and discussion

In this study, we used a hormonally-induced non-bacterial prostatic inflammation Wistar rat model to investigate the effects of orally administered GGM-rich hemicellulose extract on prostatic inflammation and associated changes on voiding and pain. To our knowledge, this is the first study showing evidence of potential usage of wood-derived GGM-rich hemicellulose extract on attenuating chronic prostatic inflammation conditions.

3.1 Changes in animal and organ weights and food/water consumption

Hormone responsive organ weights were used as indicators for constant hormone release of the implanted pellets. The weight of seminal vesicles, prostate-urethra complex and pituitary gland were increased and the weight of the testicles decreased significantly due to 18-

week hormonal treatment (Table 1). This is in line with previous studies done with rats [21] [32-34] showing that estradiol and testosterone exposure induces similar changes to hormone responsive organs. GGM treatment did not affect the animal body weight or organ weights indicating no direct anti-estrogenic or –androgenic effects (Table 1).

Body weights decreased significantly due to the hormonal exposure. Animals with hormone exposure consumed significantly less nutrition than placebo ones when considering absolute diet values, but in proportion of relative consumption to animal body weights, they actually consumed more nutrition per kilogram per day than the placebo ones. The relative water consumption was also significantly increased due to hormonal exposure (Table 1). The increased consumption of food and water could be explained by the overall changes in energy metabolism induced by sex steroids [35-37].

Interestingly, GGM-treatment did not affect body weight but increased relative food consumption (Table 1). The water (i.e. 2% GGM solution) consumption also increased as well which was evident as both absolute and relative water consumption values. In our preliminary pilot study (unpublished data) there was no difference in the water consumption of GGM-consuming rats compared to their control T+E_2+vehicle group. The cause of the relative increase of food and water consumption due to GGM treatment in this study remains open. Animals showed no signs of change in overall wellbeing during the GGM treatment period or decrease in body weights. It is also notable that consumed diet and water was monitored when the animals were in their normal maintenance cages and not in metabolic cages, which can affect the total measured consumed amounts of diet and drinking water due to daily handling of the cages resulting in less precise results.

3.2 Alterations in LUT function

Urodynamical measurement were performed to assess lower urinary tract symptoms (LUTS). The results show that T+E_2- treatment in male Wistar rats induced altered voiding resembling LUTS. LUTS can be divided into storage symptoms such as altered bladder sensation and increased daytime frequency, voiding symptoms such as intermitted and slow urine stream, and postmicturition symptoms such as feeling of incomplete bladder emptying [38]. Common urodynamical symptoms associated with CPPS are increased bladder pressure, reduced urine flow rate and increased postvoid residual urine volume [39]. The mean bladder pressures (placebo+vehicle: 30.8 ±2.87 cmH_2O; T+E_2+vehicle: 35.1 ±3.52 cmH_2O; T+E_2+ GGM: 33.1 ±2.08 cmH_2O) during micturition did not statistically significantly differ between groups: (P= 0.58). The basal bladder pressure during micturition and urine flow rate measured as both maximal and mean values were significantly lower and the micturition times were significantly prolonged in T+E_2+vehicle group compared to placebo group (Fig. 1A-D). GGM significantly decreased the basal bladder pressure (Fig. 1A) and urine flow rates compared to vehicles (Fig. 1B-C). T+E_2+vehicle treatment increased significantly bladder capacity and residual volume compared with placebo group. GGM did not affect bladder capacity but reduced significantly residual urine volumes (Fig. 1E-F). The micturition intervals and voided volumes were not significantly affected by the hormonal treatment (Fig. 1G-H). There was a trend (P= 0.065) of longer micturition intervals in GGM-treated animals. Voided volumes were significantly increased in GGM-treated animals compared with T+E_2+vehicle group (Fig 1H). Increase in the bladder weight i.e. bladder hypertrophy is an indication of obstructive voiding in rats [21] [22] [40]. Bladder weights were slightly increased in both T+E_2+ vehicle -treated and T+E_2+ GGM -treated animals compared to placebo group (Table 1). Taken together, GGM had positive effects on

LUTS by decreasing the basal bladder pressure, emptying the bladder more efficiently by increasing the urine stream rate and volume, thus also reducing the residual volume remaining in the bladder after voiding, which was reflected also as increased micturition intervals.

3.3 Impact on abdominal pain

Pain response to stimuli using von Frey filaments was assessed to study abdominal pain. Chronic abdominal pain is often associated with prostatitis. It has been postulated that chronic prostate inflammation can cause irreversible changes in neurotransmission through various mechanisms leading to chronic pain [31] [41]. The pain response measurements showed that after six weeks of hormonal exposure the threshold of the animals for pain response was significantly decreased in both hormone-treated groups compared to placebo group (Fig. 1I). At the 13-week prior to the five-week treatment period the pain response of both hormone-exposed groups did not significantly differ from placebo anymore. At the end of the study at week 18 the pain threshold was significantly lower in the T+E_2+vehicle but not in the T+E_2+GGM group compared to the placebo + vehicle group. Thus, the pain response of GGM-treated group could be considered to be improved closer to the pain response situation of the placebo group. On the other hand, GGM treatment did not significantly improve the pain response compared neither to the 18 week time point against T+E_2+vehicle or when comparing of the 13 week and 18 week time points inside the T+E_2+GGM group (Fig. 1I). It is known that men with CP/CPPS including pelvic pain can have significant fluctuations in symptoms over time [42] and our results indicate that a similar situation in pain response of the rats were similarly fluctuating during the study.

3.4 Changes is prostate inflammation

Histopathologically, chronic inflammation is often seen in the prostatic stroma, intraepithelial space and inside the acini [12]. Our results show that hormone exposure induced significant prostatic inflammation seen as perivascular, stromal/glandular inflammation and as an increased number of inflamed acini (Fig. 2A, B and D-F). GGM significantly reduced the amounts of inflamed areas in stroma compared to T+E_2+vehicle group (Fig. 2E). Only 27% of the counts of stromal inflammation in the T + E_2 +GGM group belonged to the high score group (>6-17 inflamed area counts) and 73% to the middle score category (>1-6 inflamed area counts). The proportions of T+E_2+vehicle were 87% and 13%, respectively. A similar change in the proportions of inflammation severity was seen in prostatic acini, i.e. 18% of the group counts were in the severe category (>30-49 area counts) and 81% in the middle score category (>1-30 area counts), whereas T + E_2 +vehicle proportions were 75% and 25%, respectively (Fig. 2F).

3.5 Discussion of possible actions of GGM

The causal relationships between the urodynamical measurements, pelvic pain and prostatic inflammation parameters remain open in light of the possible mechanism of action of GGM. There are however significant correlations between the measured parameters (Table 2). The observed decrease of severity of the inflammation in the prostatic lobe does unlikely explain *per se* the improvement of the urodynamical changes seen in this study. The mechanism of action remains to be discovered, but intriguing possibilities could be related to immunomodulating properties [2] or possible also probiotic-modifying properties [3] [4] [5] of galactoglucomannan. Fermentable carbohydrates have the ability to improve colonic health of both humans and animals. These carbohydrates are able to resist hydrolytic digestion and are fermented in the large bowel [43]. GGM could be considered as a fermentable carbohydrate and

is unlikely absorbed into the bloodstream as such because of the large molar mass on galactoglucomannan. Supporting our suggestion of an indirect mechanism of action is given by Faber et al. [4] who showed using an *in vitro* digestion model (to simulate gastric and small intestinal hydrolytic digestion) that GGM oligosaccharides extracted from softwood-derived molasses resisted hydrolytic digestion and were fermented as indicated by a decrease in pH, increased SCFA (short-chain fatty acids) production and beneficial microbial changes. In addition to the main hemicellulose fraction of the extract galactoglucomannan, which compromises 82% of the total fraction, the two other compartments of this extract, pectic polysaccharides (4%) and xylans (12%), may have a role on the biological effects of the extract in this study. Xylans are common hemicellulose compartments in plant cell walls among other hemicelluloses and pectins. Xylo-oligosaccharides has been shown to have antioxidative and prebiotic properties [44][45] and xylan-derived oligosaccharides from commonly consumed food stuff are generally considered as health-promoting dietary fibres [46]. Additionally, monosaccharide xylose from hardwood, the main sugar building block for xylan, has been used for bioproduction of xylitol, a natural alternative sweetener, from many plant sources including wood-derived xylans and exerts many health-promoting properties [47]. Additionally, pectin is a natural part of the human diet that also bypasses enzymatic digestion of the small intestine but is degraded by the microflora of the colon. Many commonly consumed fruits and vegetables such as apple, pear and citrus fruits contain notable amounts of pectin. Pectin has been linked to exert beneficial health effects on cholesterol and lipid metabolism, diabetes, intestinal infections, diarrhoea and even cancer [48]. In particularly, pectic polysaccharides has been shown to induce proliferation of B cells and secretion of cytokines and chemokines [49] [50] and possess immunomodulating activity against intestinal Peyer's patch cells and macrophages [51]

[52]. Thus, it is an intriguing idea wherever modulation of immune response through the gut-associated lymphoid tissues or other immune-related responses in an indirect manner would have a role on the prostatic inflammation and associated function of the lower urinary tract. Interestingly, there is some clinical evidence showing that changes in microbiota of the gut is associated with symptoms of Interstitial Cystitis / Bladder Pain Syndrome (IC/PBS) [53], a condition belonging together with CP/CPPS to a syndrome family Urologic Pelvic Pain Syndrome (UCPPS). It is also known that bladder and gut interact through neural links between pelvic organs modulating organs physiological function [54] and share common neuronal pathways [55] and there is evidence of bidirectional cross-sensitization of the colon and lower urinary tract [56]. Thus, it is an intriguing possibility that the beneficial actions of GGM in this study on prostate inflammation, LUT function and pelvic pain could be an indirect effect through colonic modulation. Further studies are needed to enlighten the mechanism of action of GGM.

4. Conclusions

Orally administered Norway spruce-derived galactoglucomannan-rich hemicellulose extract showed beneficial effects on lower urinary tract function and inflammation severity associated with nonbacterial chronic prostatic inflammation in the rat.

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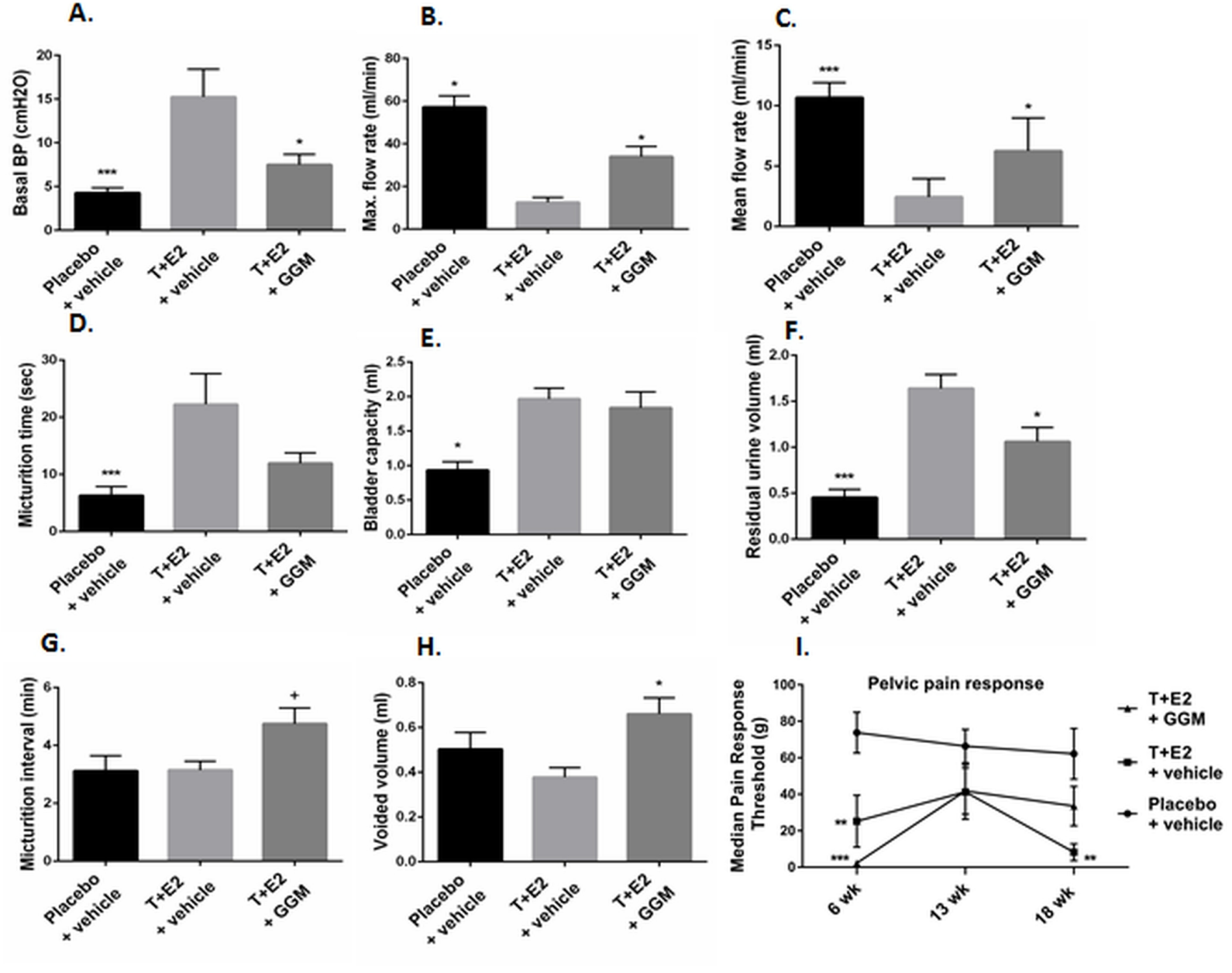
Captions to illustrations

Fig. 1. Urodynamical parameters resembling function of the lower urinary tract: A) basal bladder pressure, **B)** maximal flow rate, **C)** mean flow rate, **D)** micturition time, **E)** bladder capacity, **F)** residual urine volume , **G)** micturition interval and **H)** voided volumes . Statistical analyses were performed using One Way ANOVA for A, C, D and F-H. Kruskal-Walls ANOVA on Ranks for B and E against T+E_2 + vehicle group. *** P= <0.001, *P<0.05, +P=0.065. **Median pain response threshold I)** measured on study weeks 6, 13 and 18. Statistical significant differences ***= P<0.001, **=P<0.01 are against placebo + vehicle group. Data are represented as average values and SEM.

Fig. 2. Inflammation in the dorsolateral prostate lobes: Representative images showing prostatic acini and the stroma around the acini in **A)** placebo + vehicle group **B)** T+E_2 + vehicle group and **C)** T+E_2 + GGM group. No visible inflammation areas are present in the placebo-group whereas infiltration of inflammatory cells are present in the T+E_2 + vehicle group seen as (arrows in the picture) perivascular inflammation (PI), stromal inflammation (SI) and inflamed acini (IA). The severity of the prostatic inflammation seen as reduced inflammation areas were evident in T+E_2 + GGM group (C). **Proportions of inflamed areas** in **D)** perivascular, **E)** stromal/periglandular and **F)** prostate acini in the dorsolateral prostate lobe. The data is shown as difference in proportions of three categories of inflammation area counts for each treatment group relative to total animal samples (100%) in each treatment group. Statistical significant differences shown in figures are against T+E_2 + vehicle group.


A.
Basal BP (cmH2O)
20
15
10
5
0
***
*
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
B.
Max. flow rate (ml/min)
80
60
40
20
0
*
*
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
C.
Mean flow rate (ml/min)
15
10
5
0
***
*
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
D.
Micturition time (sec)
30
20
10
0
***
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
E.
Bladder capacity (ml)
2.5
2.0
1.5
1.0
0.5
0.0
*
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
F.
Residual urine volume (ml)
2.0
1.5
1.0
0.5
0.0
***
*
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
G.
Micturition interval (min)
6
4
2
0
+
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
H.
Voided volume (ml)
0.8
0.6
0.4
0.2
0.0
*
Placebo + vehicle
T+E2 + vehicle
T+E2 + GGM
I.
Pelvic pain response
Median Pain Response Threshold (g)
100
80
60
40
20
0
**
***
**
6 wk
13 wk
18 wk
T+E2 + GGM
T+E2 + vehicle
Placebo + vehicle

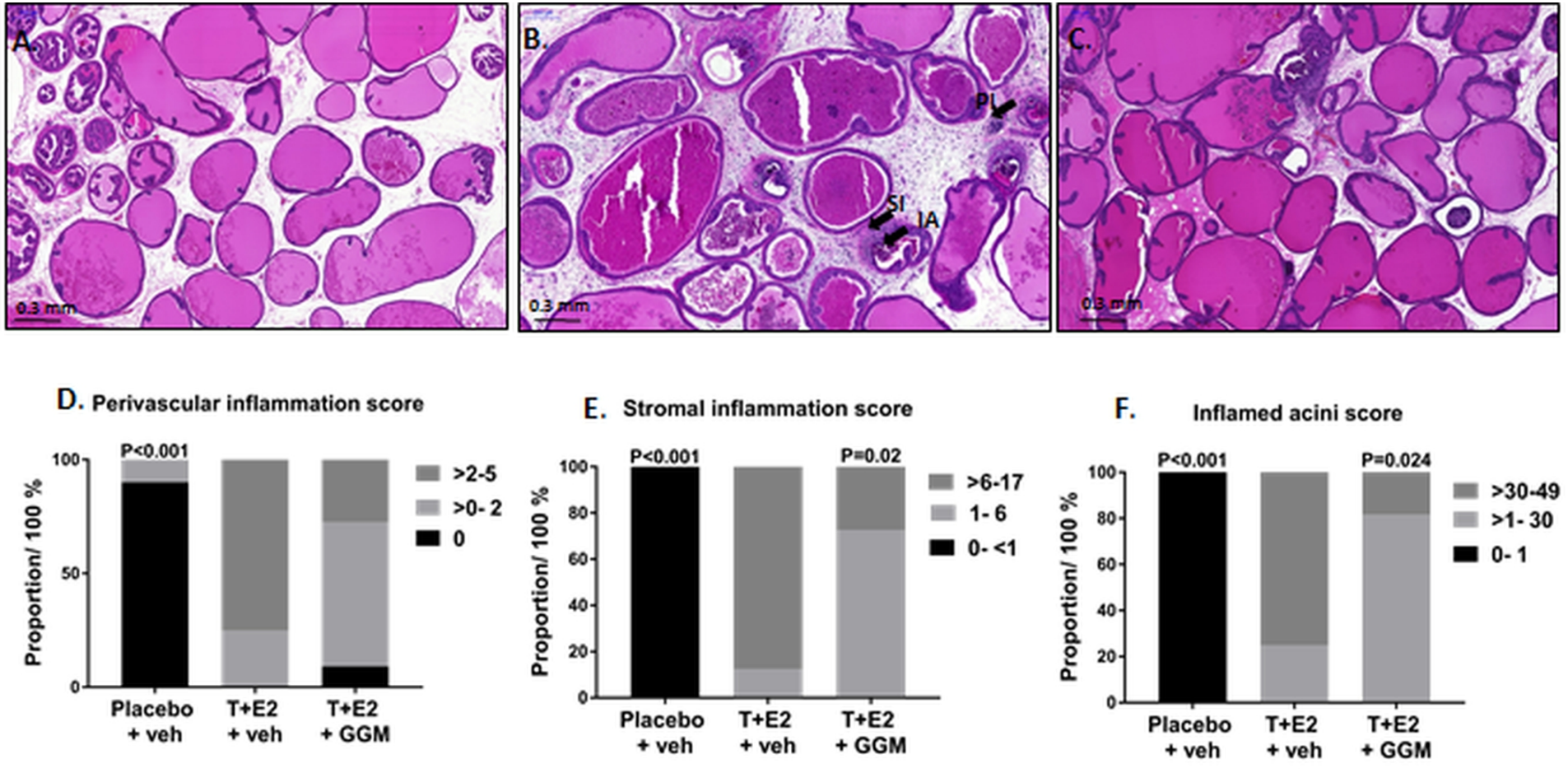

A.
0.3 mm
B.
PL
SI
IA
0.3 mm
C.
0.3 mm
D. Perivascular inflammation score
P<0.001
Proportion/ 100 %
100
50
0
>2-5
>0- 2
0
Placebo + veh
T+E2 + veh
T+E2 + GGM
E. Stromal inflammation score
P<0.001
P=0.02
Proportion/ 100 %
100
80
60
40
20
0
>6-17
1- 6
0- <1
Placebo + veh
T+E2 + veh
T+E2 + GGM
F.
Inflamed acini score
P<0.001
P=0.024
Proportion/ 100 %
100
80
60
40
20
0
>30-49
>1- 30
0- 1
Placebo + veh
T+E2 + veh
T+E2 + GGM

Table 1. Animal weights, organ weights and food and water consumption

	Placebo + veh	**T+E_2 +veh**	**T+ E_2 + GGM**	**P-value T+E_2+veh vs. placebo**	**P-value T+E_2+veh vs. T+E_2+GGM**
Start animal weight (g)	354 ±4.2	359 ±5.2	354 ±2.7	NS (KW)	NS (KW)
End animal weight (g)	522 ±6.7	328 ±8.5	324 ±3.0	<0.05 (KW)	NS (KW)
Kidney weight (10^{-1} g)	12.2 ±0.30	16.4 ±0.78	14.9 ±0.57	<0.001(A)	NS (A)
Testis weight (10^{-1} g)	18.9 ±0.72	8.7 ±0.72	8.5 ±0.48	<0.001(A)	NS (A)
Seminal Vesicles weight (10^{-1} g)	2.6 ±0.11	5.9 ±0.28	5.4 ±0.24	<0.05 (KW)	NS (KW)
Prostate-urethra Complex weight (10^{-1} g)	11.6 ±0.71	22.5 ±1.22	22.1 ±0.90	<0.0001 (A)	NS (A)
Pituitary gland weight (10^{-2} g)	1.1 ±0.04	4.4 ±0.50	3.8 ±0.35	<0.05 (KW)	NS (KW)
Bladder weight (10^{-1} g)	1.5 ±0.11	1.8 ±0.06	1.8 ±0.08	NS 0.069 (A)	NS (A)
Diet consumption, absolute values (g)	23.4 ±0.36	16.4 ±0.24	17.3 ±0.12	<0.05 (KW)	NS (KW)
Relative diet consumption/ animal weight (10^{-3} g/g)	46.3 ±0.91	50.7 ±1.34	53.8 ±0.41	<0.01 (A)	0.05 (A)
Absolut water consumption (mL	32.5 ±1.39	32.0 ±1.95	40.2 ±1.80	NS (A)	<0.01 (A)
Relative water consumption/ animal weight (10^{-3}mL/g)	64.2 ±2.89	97.3 ±4.60	125.0 ±5.80	<0.001 (A)	<0.001 (A)

Values are represented as mean and SEM. Statistical analysis using either A= One Way ANOVA or KW= Kruskall Wallis ANOVA on Ranks.

Table 2. Correlations between urodynamical, inflammation and pelvic pain parameters.

	18 wk pelvic pain threshold	Perivascular inflammation	Stromal/ periglandular inflammation	No of inflamed acini
18 wk pelvic pain		-0,477 0,009	-0,570 0,001	-0,517 0,004
Mean bladder pressure	NS	NS	NS	NS
Basal bladder pressure	-0.436 0.01	0.550 0.002	0.652 0.002	0.649 <0.001
Max. flow rate	0.661 <0.001	-0.691 <0.001	-0.747 <0.001	-0.721 <0.001
Mean flow rate	0.544 0.001	-0.648 <0.001	-0.673 <0.001	-0.662 <0.001
Micturition time	-0.402 0.02	0.582 <0.001	0.553 0.002	0.531 0.003
Bladder capacity	NS	0.682 <0.001	0.602 <0.001	0.600 <0.001
Residual urine volume	-0.392 0.03	0.617 <0.001	0.654 <0.001	0.689 <0.001
Micturition interval	NS	NS	NS	NS
Voided volume	-0.409 0.02	NS	NS	NS

Spearman Rank Order Correlation analysis. Upper values in each cell: Correlation coefficient; lower values in each cell: P-values.

Abstract

Galactoglucomannan (GGM) is the main hemicellulose class in wood of coniferous trees and could be potentially utilized as a possible health-promoting substance for food and pharmaceutical industry. Our aim was to evaluate effects of orally administered GGM-rich extract from Norway spruce in a rat model of chronic prostatitis associated with lower urinary tract symptoms (LUTS). Prostatic inflammation and LUTS was induced in male rats using testosterone and 17β-estradiol exposure for 18 weeks. Rats were treated with 2% GGM dissolved in drinking water during weeks 13 to 18. Pelvic pain response, LUT function and histopathological evaluation of the prostate were assessed. The results show that hormonal exposure induced LUTS seen as decreased urine flow rate, increased bladder pressure, voiding times, bladder capacity and residual urine volumes. GGM had positive effects on urodynamical parameters by decreasing the basal bladder pressure, increasing the urine flow rate and volume, reducing the residual volume and increasing micturition intervals. GGM reduced the extent of the hormone exposure-induced prostatic inflammation. Increase of pelvic pain induced by hormone exposure was only slightly affected by GGM treatment. The results suggest that orally administered GGM may have potential usage for improving lower urinary tract function associated with chronic prostatic inflammation.