147

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PLEIOTROPIC EFFECTS OF PUTATIVE STRESS AND URGENCY URINARY INCONTINENCE GENETIC RISK VARIANTS ON OTHER LOWER URINARY TRACT SYMPTOMS

Hypothesis / aims of study

Genetic pleiotropy is said to occur when a single gene, or single genetic variant, influences multiple traits. Identification of pleiotropic effects can provide insight into the shared biological processes leading to different complex conditions[1]. Lower urinary tract symptoms (LUTS) typically demonstrate distinct clusters, of storage, voiding and incontinence LUTS [2], but the underlying reasons for this clustering are poorly understood. In a recent genome-wide association study (GWAS) in women of European descent (n=8,997) we identified 12 genomic loci with strong associations ($p<1x10^{-7}$) for stress incontinence, urgency incontinence, or both. In this follow-up study, we aimed to test for pleiotropic effects of those putative incontinence risk variants on other storage and incontinence LUTS.

Study design, materials and methods

Women attending gynaecology clinics, were recruited to provide either whole blood or saliva for genotyping, and to complete the ICIQ-FLUTS questionnaire, which records the occurrence of individual LUTS on a five point ordinal scale. For each symptom we used a consistent dichotomous case definition of a score ≥2, corresponding to leakage occurring ≥ "sometimes", daytime frequency ≥9, and nocturia ≥2. DNA extractions were performed with an Invitrogen iPrep robot using PureLink gDNA Blood kits. Primers for the top SNP in each of the top 12 loci from our GWAS dataset were prepared using PrimerPicker. Where primers could not be designed for the top SNP in a locus, or where validation assays failed, we picked a proxy known to be in high linkage disequilibrium with the top SNP (using either SNAP or the 1000 Genomes Browser), for which we had already evidence of a strong association with the stress or urgency incontinence. Genotyping was performed using competitive allele specific PCR. For each outcome logistic regression analyses were run (phenotype regressed on genotype, with the assumption of co-dominant/additive inheritance) adjusted for age, parity, and self reported ethnicity. For significant associations, we further adjusted for the original phenotype identified in the GWAS.

Results

After quality control, genotypes were available for 799 women. Participants were of mean age 47.7, mean BMI 25.7kg/m2, and median parity 1. 82.5% were of self-reported European descent. There were highly significant correlations between all storage and incontinence LUTS (p all <0.0001), with Spearman's correlation coefficients ranging from 0.23 (stress incontinence and daytime frequency) up to 0.77 (nocturnal enuresis and insensible incontinence). Despite multiple assay probe redesigns, the rs146033157 SNP was monomorphic across the whole sample, and was therefore excluded from further analysis. Results for the remaining 11 SNPs are shown in the table. We observed nominally significant associations between the rs139329202 SNP, close to the *SULF2* gene, and both nocturnal enuresis (OR 6.72) and insensible incontinence (OR 5.14). We found a nominally significant association between the rs10837192 SNP close to the *LRRC4C* gene, and daytime frequency (OR 1.48). Finally we found nominally significant associations between the rs138724718 SNP close to the *EN1* gene and nocturia (OR 2.46). These associations were each independent after adjustment for the associated incontinence phenotype in the GWAS.

Interpretation of results

Despite the large sample, we lacked power for several uncommon variants, and some uncommon phenotypes. The confidence intervals on estimates were very wide, and none of the observed associations would survive correction for multiple comparisons. These findings should therefore only be considered hypothesis generating. Nonetheless, the results provide some indications of shared genetic susceptibility between urgency incontinence, and daytime frequency, nocturnal enuresis and insensible incontinence. Despite a current conceptual belief in urgency as driver of urgency incontinence, we did not find associations with urgency itself. Despite adjustment for a number of potential confounders, longitudinal mediation analyses would be required to further unpick the causal sequences. We observed very large effect sizes between rs139329202 with both nocturnal enuresis and insensible incontinence. Nocturnal enuresis is sometimes considered an extreme or severe form of urgency incontinence, and is known to persist across the lifecourse with high heritability[3]. The large point estimates observed here, in comparison to a more common, less heritable phenotype such as urgency, suggest that new risk variants might be more easily discovered for nocturnal enuresis in future.

Concluding message

In analysis of pleiotropic effects of incontinence risk variants on other storage and incontinence LUTS, we found significant associations for some variants with nocturnal enuresis, insensible incontinence, nocturia, and frequency, reflecting probable shared genetic susceptibility, and contributing to the clustering these symptoms display. We found strikingly large associations for the top SNP for urgency incontinence with nocturnal enuresis, which may point to enuresis as an extreme manifestation of urgency incontinence, amenable to analysis in future genetic association studies.

	Urinary Urgency		Nocturia		Frequency		Nocturnal Enuresis		Insensible Incontinence	
SNP	**OR**	**p**	**OR**	**p**	**OR**	**p**	**OR**	**p**	**OR**	**p**
rs139329202	0.97	0.968	0.35	0.318	0.70	0.730	**6.72**	**0.023**	**5.14**	**0.047**
rs79077061	0.96	0.867	0.65	0.146	0.88	0.709	1.70	0.212	0.94	0.900
rs78851245	0.96	0.930	1.21	0.686	1.48	0.438	1.83	0.436	n.a.	n.a.
rs78878767	0.71	0.491	0.70	0.533	1.05	0.925	n.a.	n.a.	0.74	0.772
rs13059018	0.49	0.062	0.84	0.594	0.68	0.390	1.25	0.696	0.47	0.317
rs4281556	0.92	0.605	0.89	0.528	0.91	0.675	1.12	0.746	0.70	0.314
rs1218596	1.13	0.669	1.15	0.640	1.58	0.166	0.97	0.958	0.53	0.386
rs10837192	1.24	0.205	1.05	0.785	**1.48**	**0.042**	1.44	0.283	1.44	0.235
rs72738866	1.03	0.843	0.93	0.629	0.85	0.403	0.94	0.850	0.92	0.756
rs138724718	0.86	0.773	**2.46**	**0.047**	0.25	0.174	1.15	0.892	1.90	0.403
rs34998271	1.42	0.244	1.53	0.175	1.34	0.433	1.75	0.293	1.08	0.902

Table: Associations between putative risk SNPs for stress and urgency urinary incontinence with other storage and incontinence LUTS. Significant associations highlighted in bold.

References

1. PLoS One. 2012; 7(5): e34861
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Disclosures

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