SLEEPJ, 2018, 1–6

doi: 10.1093/sleep/zsy164
Advance Access publication Date: 12 September 2018
Original Article

ORIGINAL ARTICLE

Reassessing GWAS findings for the shared genetic basis of insomnia and restless legs syndrome

Maryam El Gewely[1,2], Mélanie Welman[2], Lan Xiong[1,3,4], Sophie Yin[2], Hélène Catoire[4], Guy Rouleau[4], Jacques Y Montplaisir[1,2], Alex Desautels[2,5] and Simon C Warby[1,2,*]

[1]Department of Psychiatry, Université de Montréal, Montréal, Canada, [2]Centre d'études avancées en médecine du sommeil, Montréal, Canada, [3]Centre de recherche, Institut universitaire en santé mentale de Montréal, Montréal, Canada, [4]Montreal Neurological Institute and Hospital, Department of Neurology and Neurosurgery, McGill University, Montreal, Canada and [5]Department of Neurosciences, Université de Montréal, Montréal, Canada

*Corresponding author. Simon C. Warby, Centre d'Études Avancées en Médecine du Sommeil (CÉAMS), 5400 Boul Gouin O, Montréal, Canada, QC H4J 1C5. Email: simon.c.warby@umontreal.ca.

Abstract

Two genome-wide association studies (GWAS) suggest that insomnia and restless legs syndrome (RLS) share a common genetic basis. While the identified genetic variation in the *MEIS1* gene was previously associated with RLS, the two GWAS suggest a novel and independent association with insomnia symptoms. To test the potential pleiotropic effect of *MEIS1*, we genotyped three *MEIS1* variants in 646 chronic insomnia disorder (CID) patients with and without RLS. To confirm our results, we compared the allelic and genotypic distributions of the CID cohort with ethnically matched controls and RLS cases in the French Canadian cohort. The CID cohort was diagnosed by sleep medicine specialists and 26% of the sample received the combined diagnosis of CID+RLS. We find significant differences in allele and genotype distributions between CID-only and CID+RLS groups, suggesting that *MEIS1* is only associated with RLS. Genotype distributions and minor allele frequencies of the three *MEIS1* SNPs of the CID-only and control groups were similar (rs113851554: 5.3% vs. 5.6%; rs2300478: 25.3% vs. 26.5%; rs12469063: 23.6% vs. 24.4%; all $p > 0.05$). Likewise, there were no differences between CID+RLS and RLS-only groups (all $p > 0.05$). In conclusion, our data confirms that *MEIS1* is a genetic risk factor for the development of RLS, but it does not support the pleiotropic effect of *MEIS1* in CID. While a lack of power precluded us from refuting small pleiotropic effects, our findings emphasize the critical importance of isolating CID from other disorders that can cause sleep difficulties, particularly RLS, for future genetic studies.

Statement of Significance

Genetic studies of insomnia are scarce and phenotypic definitions are heterogeneous. In fact, the majority of insomnia genetic studies focus on insomnia symptoms or measure sleep quality rather than the actual disorder. Moreover, few studies reassessed insomnia-related genome-wide association studies (GWAS) findings despite the importance of the independent replication. Hence, our study plays a crucial role in revaluating the latest insomnia-related GWAS findings while using a large and well-phenotyped cohort of chronic insomnia disorder (CID) patients. Our results are not consistent with an independent association between insomnia and *MEIS1* gene, which highlights the importance of using well-phenotyped cohorts and the necessity of isolating CID from confounding disorders such as restless legs syndrome (RLS) in future insomnia genetic studies.

Key Words: sleep disorders; sleep genetics; insomnia; RLS; GWAS; phenotyping; *MEIS1*

Submitted: 12 January, 2018; **Revised:** 4 July, 2018

Introduction

Chronic insomnia disorder (CID) and restless legs syndrome (RLS) are two common sleep disorders in the general population, with a prevalence of 10% [1–6] and 2%–4% [7], respectively. CID is characterized by a subjective complaint of poor sleep quality or quantity that is associated with difficulty initiating or maintaining sleep. The sleep disturbance significantly impacts daytime functioning and occurs at least three times a week for at least 3 months [1]. CID is often comorbid with other sleep disorders. In fact, sleep initiation difficulty resulting from the leg discomfort observed in about 75% of RLS patients makes RLS a confounding factor of insomnia disorder [8–10]. RLS is sensorimotor disorder defined by an urge to move legs that primarily occurs during the evening and/or at night. Symptoms intensify during rest or inactivity states and are relieved by movement [1].

Two genome-wide association studies (GWAS) [11, 12] recently reported that a specific genetic variant in Myeloid Ecotropic Viral Insertion Site 1 (*MEIS1*) gene, playing a role in development [13] and previously associated with RLS [14–19], is also independently linked to insomnia complaints. The lead single nucleotide polymorphism (SNP) identified—rs113851554—was associated with RLS by Xiong *et al.* [17] and is within the same linkage disequilibrium block as rs12469063 and rs2300478 [17], two common genetic risk factors for RLS [14, 15, 17, 19]. The key finding of these GWAS [11, 12] argues that insomnia and RLS share a common genetic basis.

To unravel the genetic relationship between these two intertwined disorders, Lane *et al.* [11] emphasized the necessity of conducting further analyses to determine if shared genetic associations are due to causality, partial mediation or pleiotropy. Hammerschlag *et al.* [12] present several lines of compelling evidence to support pleiotropy, among them conditional analyses showing that the previous RLS GWAS leading SNPs [15]—rs6710341, rs12469063 and rs2300478—are not found by the insomnia GWAS. They also conducted a phenotypic analysis to determine the possibility that their findings were influenced by the presence of RLS in the participants. They indeed concluded that RLS contributes to the significant association between *MEIS1* and insomnia; however, this confounding effect is not sufficient to completely account for the association and rs113851554 has an independent effect on insomnia symptoms.

A shared genetic basis for RLS and insomnia would be intriguing for several reasons. A cardinal and common feature between these two disorders is the hyperarousal state that precedes sleep onset. In RLS, leg discomfort occurs (or is maximal) during the evening, at the restful wake state prior to falling asleep [10]. In insomnia, patients engage in somatic, cognitive and cortical activation that prolongs sleep onset [20]. Similar hyperactive phenotype was observed in the animal RLS-model of heterozygous *MEIS1* knockout mice [21, 22]. Further, *MEIS1* knockout mice had slightly less delta power during sleep compared to the wild-type mice [22], which is concordant with some previously reported observations in insomnia patients [20]. Hence, a plausible etiologic hypothesis is that one of the contributing genetic factors of these disorders could be *MEIS1* haploinsufficiency.

However, both GWAS have a major limitation. Despite the methodological differences between the two GWASs [11, 12], both studies used of a single question asking participants if they have trouble falling asleep at night or wake up in the middle of the night to classify subjects as cases or controls for insomnia symptoms. Although a single question may be adequate to identify individuals with general sleep difficulties, it may not be sufficient to isolate insomnia from other self-reported conditions related to sleep quality, such as RLS. Additionally, this question does not clearly differentiate acute insomnia symptoms from the diagnostic criteria of CID [1], such as the duration of sleep difficulties and whether symptoms are accompanied by distress and/or daytime impairment.

Hence, the primary objective of this study is to evaluate the association between CID and *MEIS1* in a clinical setting. If there is an independent association of *MEIS1* with insomnia, we expect to see similar minor allele frequencies between *MEIS1* genetic variants in CID patients with and without RLS.

Methods

Participants

A total of 705 patients from the province of Québec with a diagnosis of primary CID recruited at the sleep clinic at the Centre d'Études Avancées en Médecine du Sommeil were considered for this study. All patients were interviewed by a sleep specialist prior to Cognitive Behavioral Therapy for insomnia and met the diagnostic criteria of primary CID [1] as per the normal practice for clinical care for insomnia. RLS diagnoses were made during the medical consultation based on the International RLS Study Group (IRLSSG) diagnostic criteria [10]. Clinical patients that consented for research were retrospectively assigned to CID-only or CID+RLS (primary insomnia concomitant to RLS) groups by two clinicians specialized in sleep disorders (A.D., J.M.).

Polysomnography (PSG) was used to quantify periodic leg movements in sleep (PLMS), which supports the diagnosis of RLS, and to screen for apnea-hypopnea. Subjects with uncertain RLS diagnosis and those with apnea-hypopnea index (AHI) greater than 15 were excluded. Based on these criteria, 59 subjects were excluded, leaving 646 patients in the CID cohort. Patients were free of any other neurological disease and provided written informed consent.

Polysomnography

Out of the 646 included subjects who met the inclusion criteria, 591 underwent a nocturnal PSG using a standard montage. Periodic leg movements during sleep (PLMS), as well as apnea and hypopnea, were scored and analyzed according to standard criteria [23]. Electromyography (EMG) on both tibialis anterior was used to calculate the periodic leg movement index (PLMI). Overlapping movements between the two legs within 0.5 seconds are counted as one movement. PLM were defined as movements that lasted 0.5 to 10 seconds, were separated by intervals of 5 to 90 seconds and occurred in a series of at least four consecutive movements. Leg movements were detected with an increase in EMG ≥ 8 μV above the resting baseline for movement onset and a decrease in EMG < 2 μV above the resting level for movement offset.

AHI is the sum of the number of apneas and hypopneas per hour of sleep and was measured from oronasal flow and thoracoabdominal movements. Apnea was defined by the absence (≥90%) of airflow for more than 10 seconds and hypopnea as an airflow reduction (≥30%) that lasted more than 10 seconds and resulted in either arousal (while SaO_2 < 3%) or oxygen desaturation (SaO_2 ≥ 4%).

Genotyping

In the CID cohort, genotyping was performed using a standard Taqman assay. Genomic Deoxyribonucleic acid (gDNA) was isolated from the patient buffy coat using FlexiGene DNA Kit (258) (Qiagen, Canada) and following manufacturer's standard protocol. Three SNPs from *MEIS1* gene located on chromosome 2 were examined, rs113851554 (assay ID:C_154329142_10; context sequence: GTATATGTGGAATTTATATGTTTCA[G/T]TTAGGTTGTTCTTATG), rs12469063 (assay ID: C__31123351_10 and context sequence: CAGCCTGCTTCCAGCTGTGGCAGGC[A/G]TGATGCAGTGAATTGC TTTTGAATG) and rs2300478 (assay ID:C__15754717_10 and context sequence: TAAGCCAGTCTTCTTGTTTTCAGTG[G/T]GTCTGTAAG TATCTGGTCAGAGAA) (Viia7 real time PCR, Thermo Fisher, Scientific, Canada). PCR reactions used 5 ng of gDNA with the following cycling conditions (step 1: 95°C for 10 min; step 2: 95°C for 15 seconds and 60°C for 60 seconds for 50 cycles).

For both groups, CID-only and CID+RLS, genotype distributions were within Hardy-Weinberg equilibrium for the three SNPS (rs113851554: $p = 0.37$ and $p = 1$, rs2300478: $p = 0.18$ and $p = 1$, rs12469063: $p = 0.70$ and $p = 0.49$, respectively). For the CID cohort, genotyping failed in two subjects for rs113851554 and two subjects for rs12469063 (i.e. the genotyping success rate was 99%–100%). Reproducibility of the genotyping (100%) was confirmed by re-genotyping multiple samples within and across assay plates.

Allelic and genotypic distributions of the three SNPs were compared between the CID cohort and French Canadians Cohort (FCC) (486 FCC-controls and 385 FCC-RLS patients) from the province of Québec. The FCC was recruited and diagnosed at the same sleep clinic as the CID-cohort [14, 15, 17]. RLS cases were diagnosed according to standard criteria [24]. Genotyping of the FCC was performed and described previously using the TaqMan SNP assay [14, 15, 17] on Applied Biosystems 7900 Fast Real-Time PCR System. Genotyping success rate in the FCC was >98%; genotyping failed in 16 subjects for rs113851554, 17 subjects for rs2300478, and 15 subjects for rs12469063.

Statistical analysis

Statistical analyses were performed using R software. Independent *t*-tests were conducted to examine the differences between CID-only and CID+RLS groups in age, AHI, body mass index (BMI) and psychometric scores (Beck Anxiety Inventory [BAI], Beck Depression Inventory [BDI], and Insomnia Severity Index [ISI]) (Table 1). Nonparametric test (Man-Whitney-Wilcoxon) was used to compare PLMI between the two groups. Chi-square analyses tested sex, genotype, and allele distributions differences between groups. Finally, unadjusted and adjusted logistic regression models were used to predict the presence of RLS while using genotype, age, and sex as predictors. Associations were presented as odds ratios (OR) and their 95% confidence intervals (CIs). Power calculations were computed using Quanto (http://biostats.usc.edu/Quanto.html).

Results

Twenty-six percent of the CID cohort was diagnosed with CID+RLS (Table 1). This group was significantly older by an average of 4 years and as expected, had higher periodic leg movement index (PLMI) than the CID-only group (27 vs. 13 events/hour, respectively). Considering the CID-only group mean age (of 49 years), the average PLMI is equivalent to what was previously reported of healthy subjects of equivalent age group [25–27]. On average, subjects with CID+RLS reported more anxiety and depressive symptoms than the CID-only group but BAI and BDI scores remained within the mild range for both groups [28, 29]. There were no between-group differences in sex distribution, BMI, insomnia severity index scores or AHI.

In the CID cohort, rs113851554, rs2300478, and rs12469063 minor allele frequencies (MAF) are significantly higher in the presence of RLS compared to its absence (Table 2). Importantly, MAF of the three *MEIS1* SNPs we obtained for the CID-only group are comparable to the MAF found in Canadian (FCC) and European controls [30]. In fact, the risk allele frequency of the three SNPs in the two RLS-free groups (CID-only [CID cohort], FCC-controls [FCC]) and population-based European controls [30] (1000 Genomes project) are almost equivalent rs113851554: 5.3% vs. 5.6% vs. 5.3%; rs2300478: 25.3% vs. 26.5% vs. 24.8%; rs12469063: 23.6% vs. 24.4% vs. 23.9% respectively; all p-value > 0.05) (Table 3). In other words, the differences in MAF of rs113851554 between CID-only group (5.3%) and FCC-controls (5.6%) were very small and in a different direction from the effect on RLS. This concordance is also observed at the genotype distribution level (all p-value > 0.05) (Table 4). These results were confirmed with the logistic regression (Table 5); adjusted dominant model showed that the presence of at least one minor allele of rs113851554 increases the risk of RLS by 2.72 times (95% CI = 1.83–4.03, $p < 0.001$). Similarly, adjusted dominant models of the two other SNPs were significantly linked to the presence of RLS (Table 5, top panel). Unadjusted models had similar findings for the three

Table 1. Descriptive statistics of CID cohort

	CID-only				CID+RLS				
	Avg.	*SD*	Range	*N*	Avg.	*SD*	Range	*N*	*p*
Age	49	14	14–83	476	53	12	14–83	170	**0.0008**
Sex, % female	64	NA	NA	476	68	NA	NA	170	0.3299
BMI	26	5	18–50	421	27	5	18–51	163	0.1314
BAI	10	8	0–47	428	13	10	0–52	151	**0.002**
BDI	13	10	0–60	427	15	10	0–51	153	**0.023**
ISI	17	5	5–28	170	18	5	8–28	34	0.5167
PLMI	13[a]	19	0–130	424	27	31	0–182	167	**1.664e-07**
AHI	1	2	0–14	405	1	2	0–12	157	0.6718

Avg. = average; *N* = sample size. *p*-values < 0.05 are indicated in bold.
[a]The average PLMI in the CID-only is equal to the PLMI of healthy subjects within the same age group [25].

Table 2. *MEIS1* SNPs genotypic and allelic frequencies

MEIS1 SNPs	Genotypes/alleles	CID-only *n* (%)	CID+RLS *n* (%)	Chi-square *p*-value[a]
rs113851554[T]	GG	428 (90)	128 (76.2)	**4.84e-05**
	GT	46 (9.6)	38 (22.6)	
	TT	2 (0.4)	2 (1.2)	
	G	902 (94.7)	294 (87.5)	**1.62e-05**
	T	50 (5.3)	50 (12.5)	
rs2300478[G]	TT	271 (56.9)	76 (44.7)	**0.021772**
	GT	169 (35.5)	76 (44.7)	
	GG	36 (7.6)	18 (10.6)	
	T	711 (74.7)	228 (67.1)	**0.008343**
	G	241 (25.3)	112 (32.9)	
rs12469063[G]	AA	279 (58.7)	73 (43.2)	**0.001956**
	AG	168 (35.4)	80 (47.3)	
	GG	28 (5.9)	16 (9.5)	
	A	726 (76.4)	226 (66.9)	**0.000767**
	G	224 (23.6)	112 (33.1)	

[a]Including Yate's correction. *p*-values < 0.05 are indicated in bold.

Table 3. Comparison of the minor allele frequencies (%) of *MEIS1* SNPs between CID cohort (CID-only; CID+RLS), French Canadian Cohort (FCC-controls and FCC-RLS cases) [14, 15, 17] and 1000 Genomes Project [30] European population

			CID cohort		French Canadian Cohort		1000 Genomes Project		Chi-square *p* values
	RLS status	Allele	*N*	%	*N*	%	*N*	%	
rs113851554	Negative RLS	G	902	94.7	918	94.4	953	94.7	0.9456
		T	50	5.3	54	5.6	53	5.3	
	Positive RLS	G	294	87.5	612	82.9	NA	NA	0.292104
		T	50	12.5	126	17.1			
rs2300478	Negative RLS	T	711	74.7	703	73.5	757	75.2	0.6764
		G	241	25.3	253	26.5	249	24.8	
	Positive RLS	T	228	67.1	481	63.9	NA	NA	0.320825
		G	112	32.9	271	36.1			
rs12469063	Negative RLS	A	726	76.4	729	75.6	766	76.1	0.917039
		G	224	23.6	235	24.4	240	23.9	
	Positive RLS	A	226	66.9	485	64.8	NA	NA	0.515941
		G	112	33.1	263	35.2			

Table 4. Comparison of *MEIS1* SNPs genotype distributions (*n* (%)) between CID cohort (number of CID-only and CID+RLS patients of each SNP is as follows: rs113851554: 476 and 168; rs2300478:476 and 170; rs1249063: 475 and 169, respectively) and French Canadian Cohort [14, 15, 17] (number of FCC-controls and FCC-RLS cases of each SNP is as follows: rs113851554:486 and 369; rs2300478: 478 and 376; rs12469063: 482 and 374, respectively)

	RLS status	Genotypes	CID cohort	French Canadian Cohort	Chi-square *p*-value
rs113851554[T]	Negative RLS	GG	428 (90.0)	433 (89.0)	0.7313
		GT	46 (9.6)	52 (11.0)	
		TT	2 (0.4)	1 (0.0)	
	Positive RLS	GG	128 (76.2)	253 (66.0)	0.1537
		GT	38 (22.6)	106 (28.0)	
		TT	2 (1.2)	10 (3.0)	
rs2300478[G]	Negative RLS	TT	271 (56.9)	252 (53.0)	0.1098
		GT	169 (35.5)	199 (41.5)	
		GG	36 (7.6)	27 (5.5)	
	Positive RLS	TT	76 (44.7)	155 (41.2)	0.5942
		GT	76 (44.7)	171 (45.5)	
		GG	18 (10.6)	50 (13.3)	
rs12469063[G]	Negative RLS	AA	279 (58.7)	266 (55.2)	0.1173
		AG	168 (35.4)	197 (40.9)	
		GG	28 (5.9)	19 (3.9)	
	Positive RLS	AA	73 (43.2)	157 (42)	0.6300
		AG	80 (47.3)	171 (45.7)	
		GG	16 (9.5)	46 (12.3)	

Table 5. Logistic regression predicting the risk for RLS or chronic insomnia disorder (CID-only vs. CID+RLS, top; CID-only vs. FCC-controls, bottom) using dominant genetic models for *MEIS1* SNPs

	Unadjusted model		Adjusted model		
MEIS1 SNPs	OR (95% CI)	*p*	OR (95% CI)	*p*	80% power
Chronic insomnia disorder, without vs. with RLS (CID-only vs. CID+RLS)					
rs113851554	2.79 (1.88–4.11)	1.48e-05	2.72 (1.83–4.03)	2.78e-05	1.9
rs2300478	1.64 (1.22–2.20)	0.00628	1.58 (1.17–2.13)	0.01212	1.6
rs12469063	1.87 (1.39–2.53)	0.000536	1.81 (1.34–2.45)	0.00118	1.6
Chronic insomnia disorder vs. unaffected controls (CID-only vs. FCC-controls)					
rs113851554	0.92 (0.65–1.29)	0.68	0.93 (0.65–1.33)	0.75	1.6
rs2300478	0.84 (0.680–1.04)	0.19	0.81 (0.65–1.00)	0.11	1.4
rs12469063	0.87 (0.697–1.07)	0.27	0.83 (0.66–1.03)	0.16	1.4

Sex and age are used as predictors in the adjusted model. The 80% power is the OR at which this study has 80% power to detect a statistical difference based on the sample size.

SNPs. When comparing CID-only to the unaffected FCC controls for the three *MEIS1* SNPs, the MAF of the CID-only was lower than the MAF of unaffected controls, producing non-significant OR scores less than one. However, our study was not sufficiently powered to find small effect sizes (Table 5, bottom panel).

Discussion

This is the first study to examine the role of *MEIS1* in patients diagnosed with CID. Our data demonstrate that rs113851554, previously associated with RLS [14–19] and recently associated with insomnia symptoms in the general population [11, 12], is not associated with CID in the absence of RLS in our insomnia patients. Further, we did not find an association with two other RLS associated *MEIS1* SNPs (rs12469063 and rs2300478) [14] in CID. This lack of association between *MEIS1* and CID is consistent with a recent finding reported by Salminen *et al.* [22], which also did not find evidence to support a role for *MEIS1* deficiency in causing sleep disturbances such as sleep initiation or sleep maintenance difficulties in mice. Since our data is not consistent with a pleiotropic effect of *MEIS1* as suggested by the recent insomnia-related GWASs [11, 12], it is possible that these studies were confounded with the presence of RLS. The possibility of such confounding was directly addressed by Hammerschlag *et al.* [12] using a variety of methods such as a conditional phenotypic analysis. They estimated that the UK Biobank insomnia trait was confounded by RLS in 12% of cases and 6% of controls which partially contributed to the significant association they observed. However, there remained a significant effect of *MEIS1* on insomnia that could not be accounted for by this confounding, and they, therefore, concluded that *MEIS1* is likely to have pleiotropic effects on both RLS and insomnia. We attempted to verify this finding, but our data is not consistent with this conclusion, as we cannot find evidence for an association between *MEIS1* and CID independent of RLS. The consistency of the results between the two studies [11, 12] is likely explained by the fact that both studies used subjects drawn from the same general population cohort (UK Biobank). Therefore, our study further illustrates that the heterogeneity in the phenotyping definitions of insomnia is a major concern in the methodology of the previous genetic studies of insomnia [31, 32].

Limitations also need to be considered while interpreting our results. First, our study is a retrospective study, and the collection of phenotypic data is not completely uniform between all subjects; for example, 9% of our cohort did not have a PSG and not all patients answered all questionnaires. Second, although we had sufficient statistical power to show an association between rs113851554 and RLS (Table 5, top panel), we did not have sufficient statistical power (Table 5, bottom panel) to replicate the effect sizes reported previously from the general population cohort for insomnia symptoms (OR = 1.19 [12] or 1.26 [11]). We hypothesized that if there exists an association between rs113851554 and insomnia, the effect size should be stronger in a cohort of well-characterized patients with CID. However, the effect size we observed was smaller and in the opposite direction (OR = 0.932) relative to the previous findings. While it still may be statistically possible that there is an association between rs113851554 and insomnia, our data argues that the effect size is likely smaller and less biologically meaningful than previously reported.

It is important to emphasize that GWASs have made significant contributions in identifying genes and pathways involved in sleep and sleep disorders. An important part of this progress, however, is the independent replication of GWAS findings in smaller, but more precisely phenotyped cohorts [31]; the independent replication should be considered an integral part of the GWAS methodology. While our well-phenotyped data does not conform to the GWASs finding, it emphasizes the critical importance of isolating CID from other disorders that can cause sleep difficulties, particularly RLS, for future genetic studies.

In summary, we did not find an association between *MEIS1* and CID in our clinical cohort. However, we did replicate a clear association between *MEIS1* SNPs and RLS, even in patients with comorbid CID. Our findings also suggest that population-based cohorts should include psychometric tools with higher sensitivity and specificity in order to better characterize insomnia phenotypes, such as the insomnia severity index (ISI) [33] and the Cambridge–Hopkins diagnostic questionnaire for RLS [10, 34].

Acknowledgments

The authors would like to acknowledge the contributions of the patients to this study and thank them for their participation. We would also like to thank Régine Desnesle, Mélanie Vendette, Jean Paquet, Claude Berthiaume and Marie-Josée Quinn for their assistance and expertise. Patient samples were collected through the Sleep Research Bioank at CÉAMS.

Funding

Funding for this work was provided by the Funding for this work was provided by the 'Chaire Pfizer, Bristol-Myers Squibb, SmithKline Beecham, Eli Lilly en psychopharmacologie de l'Université de Montréal', 'Faculté des études supérieures et postdoctorales de l'Université de Montréal', and the 'Centre de Recherche de l'Hôpital du Sacré-Coeur de Montréal'.

Conflict of interest statement. None declared.

References

1. American Academy of Sleep Medicine. *International Classification of Sleep Disorders*, 3rd ed. Darien, IL: International Classification of Sleep Disorders (ICSD-3); 2014.
2. American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders*, 5th ed. Arlington, VA: American Psychiatric Association; 2013.
3. Aikens JE, *et al*. Help-seeking for insomnia among adult patients in primary care. *J Am Board Fam Pract*. 2005;**18**(4):257–261.
4. Morin CM, *et al*. Prevalence of insomnia and its treatment in Canada. *Can J Psychiatry*. 2011;**56**(9):540–548.
5. Simon GE, *et al*. Prevalence, burden, and treatment of insomnia in primary care. *Am J Psychiatry*. 1997;**154**(10):1417–1423.
6. Morin CM, *et al*. Epidemiology of insomnia: prevalence, self-help treatments, consultations, and determinants of help-seeking behaviors. *Sleep Med*. 2006;**7**(2):123–130.
7. Allen RP, *et al*. Physician-diagnosed restless legs syndrome in a large sample of primary medical care patients in western Europe: Prevalence and characteristics. *Sleep Med*. 2010;**11**(1):31–37.
8. Montplaisir J, *et al*. Clinical, polysomnographic, and genetic characteristics of restless legs syndrome: a study of 133 patients diagnosed with new standard criteria. *Mov Disord*. 1997;**12**(1):61–65.
9. Ulfberg J, *et al*.; Nordic RLS Study Group. Comorbidity in restless legs syndrome among a sample of Swedish adults. *Sleep Med*. 2007;**8**(7-8):768–772.
10. Allen RP, *et al*.; International Restless Legs Syndrome Study Group. Restless legs syndrome/Willis-Ekbom disease diagnostic criteria: updated International Restless Legs Syndrome Study Group (IRLSSG) consensus criteria–history, rationale, description, and significance. *Sleep Med*. 2014;**15**(8):860–873.
11. Lane JM, *et al*. Genome-wide association analyses of sleep disturbance traits identify new loci and highlight shared genetics with neuropsychiatric and metabolic traits. *Nat Genet*. 2017;**49**(2):274–281.
12. Hammerschlag AR, *et al*. Genome-wide association analysis of insomnia complaints identifies risk genes and genetic overlap with psychiatric and metabolic traits. *Nat Genet*. 2017;**49**(11):1584–1592.
13. Toresson H, *et al*. Expression of Meis and Pbx genes and their protein products in the developing telencephalon: implications for regional differentiation. *Mech Dev*. 2000;**94**(1–2):183–187.
14. Winkelmann J, *et al*. Genome-wide association study identifies novel restless legs syndrome susceptibility loci on 2p14 and 16q12.1. *PLoS Genet*. 2011;**7**(7):e1002171.
15. Winkelmann J, *et al*. Genome-wide association study of restless legs syndrome identifies common variants in three genomic regions. *Nat Genet*. 2007;**39**(8):1000–1006.
16. Winkelmann J, *et al*. Genetics of restless legs syndrome. *Sleep Med*. 2017;**31**:18–22.
17. Xiong L, *et al*. MEIS1 intronic risk haplotype associated with restless legs syndrome affects its mRNA and protein expression levels. *Hum Mol Genet*. 2009;**18**(6):1065–1074.
18. Schormair B, *et al*.; 23andMe Research Team; DESIR study group. Identification of novel risk loci for restless legs syndrome in genome-wide association studies in individuals of European ancestry: a meta-analysis. *Lancet Neurol*. 2017;**16**(11):898–907.
19. Yang Q, *et al*. Association studies of variants in MEIS1, BTBD9, and MAP2K5/SKOR1 with restless legs syndrome in a US population. *Sleep Med*. 2011;**12**(8):800–804.
20. Riemann D, *et al*. The hyperarousal model of insomnia: a review of the concept and its evidence. *Sleep Med Rev*. 2010;**14**(1):19–31.
21. Spieler D, *et al*. Restless legs syndrome-associated intronic common variant in MEIS1 alters enhancer function in the developing telencephalon. *Genome Res*. 2014;**24**(4):592–603.
22. Salminen AV, *et al*. Sleep disturbance by pramipexole is modified by MEIS1 in mice. *J Sleep Res*. 2018;**27**(4):e12557.
23. Iber C, Ancoli-Israel S, Chesson A, Quan S. *The AASM Manual for the Scoring of Sleep and Associated Events: Rules, Terminology and Technical Specifications*. Westchester, IL: The AASM manual for the scoring of sleep and associated events: rules, terminology and technical specifications; 2007.
24. Allen RP, *et al*.; Restless Legs Syndrome Diagnosis and Epidemiology workshop at the National Institutes of Health; International Restless Legs Syndrome Study Group. Restless legs syndrome: diagnostic criteria, special considerations, and epidemiology. A report from the restless legs syndrome diagnosis and epidemiology workshop at the National Institutes of Health. *Sleep Med*. 2003;**4**(2):101–119.
25. Pennestri MH, *et al*. PLMS and PLMW in healthy subjects as a function of age: prevalence and interval distribution. *Sleep*. 2006;**29**(9):1183–1187.
26. Youngstedt SD, *et al*. Periodic leg movements during sleep and sleep disturbances in elders. *J Gerontol A Biol Sci Med Sci*. 1998;**53**(5):M391–M394.
27. Montplaisir J, Michaud M, Denesle R, Gosselin A. Periodic leg movements are not more prevalent in insomnia or hypersomnia but are specifically associated with sleep disorders involving a dopaminergic impairment. *Sleep Medicine*; 2000;1(2):163–167.
28. Beck AT, *et al*. An inventory for measuring depression. *Arch Gen Psychiatry*. 1961;**4**:561–571.
29. Beck AT, *et al*. An inventory for measuring clinical anxiety: psychometric properties. *J Consult Clin Psychol*. 1988;**56**(6):893–897.
30. Auton A, *et al*.; 1000 Genomes Project Consortium A global reference for human genetic variation. *Nature*. 2015;**526**(7571):68–74.
31. Gehrman PR, *et al*. The role of genes in the insomnia phenotype. *Sleep Med Clin*. 2013;**8**(3):323–331.
32. Lind MJ, *et al*. Genetic pathways to insomnia. *Brain Sci*. 2016;**6**(4):64.
33. Morin CM, *et al*. The Insomnia Severity Index: psychometric indicators to detect insomnia cases and evaluate treatment response. *Sleep*. 2011;**34**(5):601–608.
34. Allen RP, *et al*. Validation of the self-completed Cambridge-Hopkins questionnaire (CH-RLSq) for ascertainment of restless legs syndrome (RLS) in a population survey. *Sleep Med*. 2009;**10**(10):1097–1100.