Genome-wide scan of job-related exhaustion with three replication studies implicate a susceptibility variant at the UST gene locus

Sonja Sulkava\textsuperscript{1,4,6}, Hanna M. Ollila\textsuperscript{1,4,6}, Kirsi Ahola\textsuperscript{7}, Timo Partonen\textsuperscript{2}, Katriina Viitasalo\textsuperscript{8}, Johannes Kettunen\textsuperscript{4}, Maarit Lappalainen\textsuperscript{5}, Mika Kvimäki\textsuperscript{7,9}, Jussi Vahtera\textsuperscript{7,10}, Jaana Lindström\textsuperscript{3}, Mikko Härmä\textsuperscript{7}, Sampsa Puttonen\textsuperscript{7}, Veikko Salomaa\textsuperscript{1} and Tiina Paunio\textsuperscript{1,2,6,7,∗}

\textsuperscript{1}Public Health Genomics Unit, \textsuperscript{2}Department of Mental Health and Substance Abuse Services and, \textsuperscript{3}Diabetes Prevention Unit, National Institute for Health and Welfare, Helsinki, Finland, \textsuperscript{4}Institute for Molecular Medicine Finland FIMM and, \textsuperscript{5}Institute for Molecular Medicine Finland FIMM Technology Centre, University of Helsinki, Helsinki, Finland, \textsuperscript{6}Department of Psychiatry, Helsinki University Central Hospital, Helsinki, Finland, \textsuperscript{7}Development of Work and Work Organizations, Finnish Institute of Occupational Health, Helsinki, Finland, \textsuperscript{8}Finnair Health Services, Finnair, Vantaa, Finland, \textsuperscript{9}Department of Epidemiology and Public Health, University College London, London, UK and \textsuperscript{10}Department of Public Health, University of Turku, Turku, Finland

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Job-related exhaustion is the core dimension of burnout, a work-related stress syndrome that has several negative health consequences. In this study, we explored the molecular genetic background of job-related exhaustion. A genome-wide analysis of job-related exhaustion was performed in the GENMETS subcohort (n = 1256) of the Finnish population-based Health 2000 study. Replication analyses included an analysis of the strongest associations in the rest of the Health 2000 sample (n = 1660 workers) and in three independent populations (the FINRISK population cohort, n = 10 753; two occupational cohorts, total n = 1451). Job-related exhaustion was ascertained using a standard self-administered questionnaire (the Maslach Burnout Inventory (MBI)-GS exhaustion scale in the Health 2000 sample and the occupational cohorts) or a single question (FINRISK). A variant located in an intron of UST, uronyl-2-sulfotransferase (rs13219957), gave the strongest statistical evidence in the initial genome-wide study (P = 1.55 \times 10^{-7}), and was associated with job-related exhaustion in all the replication sets (P < 0.05; P = 6.75 \times 10^{-7} from the meta-analysis). Consistent with studies of mood disorders, individual common genetic variants did not have any strong effect on job-related exhaustion. However, the nominally significant signals from the allelic variant of UST in four separate samples suggest that this variant might be a weak risk factor for job-related exhaustion. Together with the previously reported associations of other dermatan/chondroitin sulfate genes with mood disorders, these results indicate a potential molecular pathway for stress-related traits and mark a candidate region for further studies of job-related and general exhaustion.

INTRODUCTION

Job-related exhaustion is the core symptom of burnout, a work-related stress syndrome that consists of three dimensions: exhaustion, cynicism, and diminished professional efficacy. (1). The burnout syndrome is associated with several health problems, including depressive disorders (2), physical illnesses (3), sleep problems (4) and complaints of cognitive impairment (5). The prevalence of severe burnout in the Finnish working population has varied from 2 to 7% (2,6), while mild symptoms have been reported by up to 25% of workers. As severe burnout is linked to an increased risk of medically certified sickness absence, work disability pensions (7), and even all-cause mortality among workers aged under 45 years (8), it imposes an
Burnout is not regarded as a disease in the medical classification systems, Classification of diseases and Related Health Problems (ICD-10) or Diagnostic and Statistical Manual of Mental Disorders (DSM-IV), but it correlates moderately with a diagnosis of depression: in the Finnish population, for example, half of the individuals with severe burnout had suffered from major depressive disorder, minor depressive disorder (two to four symptoms) or dysthymia during the previous year. Depressive disorder is also more common in individuals with mild burnout (20.3%) than in those with no symptoms of burnout (7.3%) (2). Job-related exhaustion predicts depressive symptoms 3–4 years later, but not vice versa (9). Importantly, the concepts of burnout and depression are not identical, as both burnout syndrome and job-related exhaustion symptoms are also seen without clinical depression (2). Burnout and, especially, job-related exhaustion both correlate strongly with the personality trait of neuroticism (10). It is possible that burnout is part of the same continuum with internalizing disorders which share part of their genetic background with neuroticism and partly have separate neuroticism-independent genetic risk factors (11).

The heritability of job-related exhaustion was estimated to be 33% for both males and females in a large Swedish study (12), i.e. corresponding to that reported for depression (13). In a smaller Dutch study, the genetic component explained 30% of the variance in males, but only 13% in females (14). To date, the molecular genetic background of job-related exhaustion remains unexplored.

Work stressors, such as excessive work demands, have a fundamental role in the development of burnout (15,16), and job-related exhaustion is based on the concept that the symptoms are a reaction to the work environment. Therefore, we hypothesized that the interaction of genetic vulnerability factors with a stressful work environment is a key element in job-related exhaustion. In order to search for genetic variants contributing to the individual vulnerability to job-related exhaustion we performed a genome-wide association study (GWAS) of 555 388 single-nucleotide polymorphisms (SNPs), using the quantitative exhaustion scale of the Maslach Burnout Inventory-General Survey (MBI-GS) in the GENMETS cohort of the Finnish Health 2000 sample (n = 1256) and performed replication analyses for the strongest findings in the rest of the workers in that cohort (n = 1660), in two occupational cohorts (n = 1451), and in a further Finnish population cohort, FINRISK (n = 10 753) (Fig. 1). To examine the possible interaction of genetic vulnerability and work stress in the etiology of job-related exhaustion, we performed an interaction analysis at the genome-wide level. Given the requirement of large sample sizes for detecting interaction in genetic studies by hypothesis-free analysis, we focused on our gene-environment interaction (G × E) testing mainly on the variants with a main effect on job-related exhaustion.

RESULTS

Genome-wide analysis

We performed a genome-wide association analysis for the exhaustion score of MBI-GS in 1256 individuals from the population-based Health 2000 sample. Figure 2A illustrates the quantile–quantile (Q–Q) plot for the observed versus expected −2log(P-value) and Supplementary Material, Fig. SF1 the Manhattan plot. Some more significant results were seen than expected by chance, but none of these reached the genome-wide significance threshold of P < 5 × 10^{-8}. The complete list of P-values is contained in the Supplementary Material (ftp://ftp.techset.co.uk/Sulkava_GWAS/). The most significant association was obtained for rs13219957 (P = 1.55 × 10^{-7}, β = 0.306, R^2 = 2.1%), which is situated on chromosome 6 in an intron of the gene encoding for uronyl-2-sulfotransferase (UST). The results for SNPs with P < 5 × 10^{-5} for the association in the genome-wide analysis (the level corresponding to the power of 62% when QTL variance was estimated to be 1.5%) are displayed in Table 1. Of the 31 reported SNPs, only two (rs7677237, rs4237913) showed a nominally significant interaction with the subjects’ sex (0.01 < P < 0.05).

Gene–environment interaction analysis

In order to evaluate our hypothesis of a gene-environment interaction in the etiology of job-related burnout, we used the GENMETS sample to search for evidence of an interaction between work stress as defined by job demands and genetic variants at a genome-wide level, and separately for variants with a suggestive main effect on job-related exhaustion. In the genome-wide interaction analysis, none of the associations were significant (P < 5 × 10^{-8}), and the results did not differ from those expected by chance alone (Supplementary Material, Fig. SF2). SNPs with P < 5 × 10^{-5} in the interaction analysis are reported in Supplementary Material, Table ST2.

Table 1 shows that 8 of 31 SNPs with a suggestive main effect on job-related exhaustion had a significant interaction with work stress as defined by work demands, and four tests were
significant after correction for multiple testing (rs10502587, \( P_{\text{Bonferroni}} = 0.015 \); rs13381088, \( P_{\text{Bonferroni}} = 0.006 \); rs3786310, \( P_{\text{Bonferroni}} = 0.006 \); rs3826611, \( P_{\text{Bonferroni}} = 0.0062 \)). These four SNPs were situated in the intron of \( FAM59A \) and were in a high LD in GENMETS (D' > 0.8). The work demand score had a moderate correlation with the exhaustion score (Pearson's correlation = 0.363, \( P < 0.05 \)).

### Replication analyses and meta-analysis

A replication analysis was performed for five SNPs with a \( P \)-value of \( < 1 \times 10^{-5} \) in the rest of the workers (\( n = 1660 \)) from the Health 2000 study. In addition, we selected one SNP with \( P < 1 \times 10^{-4} \) (rs10488596 in \( CHRM4 \), \( P_{\text{GWAS}} = 7.01 \times 10^{-5} \)) because of its location in the intron of the functionally interesting cholinergic receptor gene. The statistically strongest signal of the genome-wide analysis, from SNP rs13219957 in the \( UST \) intron, showed also a suggestively significant association with job-related exhaustion in the replication dataset (\( P = 0.038, \beta = 0.097 \)) (Table 2), which, though, did not survive correction for multiple testing (\( P_{\text{Bonferroni}} = 0.207 \)). No other variant yielded a suggestive evidence (pointwise \( P < 0.05 \)) in the analysis (Table 2). The gene–environment interaction of rs10502587 in \( FAM59A \) was not replicated in the Health 2000 replication dataset (\( P_{G \times E} = 0.782 \)).

Rs13219957 was genotyped in the Finnish FINRISK population cohort where a one-item measure assessing job-related exhaustion was available for 10 753 workers. We observed an allelic replication for job-related exhaustion at a population level (\( P = 0.010, \beta = 0.050 \)). The variant was also associated with the question on the inconvenience caused by time pressure at work (\( n = 10 140, P = 0.036, \beta = 0.042 \)) and a question on general exhaustion (not limited to the work context) in the complete FINRISK sample (\( n = 20 813 P = 0.049, \beta = 0.027 \)).
effect on general exhaustion was slightly stronger among workers \((n = 12,812, P = 0.085, \beta = 0.029)\) than among non-workers \((n = 8001, P = 0.394, \beta = 0.020)\), even though the association in workers did not reach the nominal level of significance.

The effect of rs13219957 on job-related exhaustion was further tested in two occupational cohorts: a cohort of employees of an airline company \((n = 1378)\) and a cohort of nurses \((n = 73)\). These two samples were combined for the analysis because of the small sample size in the nurse cohort. Their meta-analysis showed a significant association of rs13219957 with the MBI-GS exhaustion score \((P = 0.047, \beta = 0.102)\).

Finally, we performed a fixed-effect meta-analysis across all the study samples \((n = 15,120)\), which yielded a \(P\)-value of \(6.75 \times 10^{-7}\) and a normalized \(\beta\)-value of 0.081. The results of the replication studies and the meta-analysis for rs13219957 are reported in Table 3.

### Association of rs13219957 with the different domains of burnout symptoms and with depressive symptoms

The Burnout syndrome consists of three dimensions: exhaustion, cynicism and diminished professional efficacy, which are reflected by three subscales of MBI-GS. We calculated the associations of rs13219957 with other symptoms of the burnout syndrome in the cohorts which had primarily been analysed for job-related exhaustion. In the Health 2000 population samples, the strongest association was seen with the MBI’s exhaustion subscale (GENMETS: exhaustion \(P = 1.55 \times 10^{-7}, \beta = 0.306\), cynicism \(P = 0.005, \beta = 0.165\), diminished professional efficacy \(P = 0.014, \beta = 0.143\) (Table 4). In the occupational cohorts combined, the signal was stronger for diminished professional efficacy \((P = 0.025, \beta = 0.102)\) when compared with exhaustion \((P = 0.047, \beta = 0.102)\), but in a separate analysis of the nurse cohort, the signal was strongest for cynicism \((P = 0.015, \beta = 0.513)\), possibly because of the specific natures of the occupational groups.

Considering the co-morbid pattern of job-related exhaustion and depression, and the risk of job-related exhaustion on subsequent depressive disorder, we also assessed the association of rs13219957 with symptoms of depression, including anhedonia, somatic symptoms and appetite. Similarly weighted principal components were found in every cohort, even though the measures for depressive symptoms slightly varied between them (see Materials and methods). No significant association of rs13219957 with depressive symptoms was seen in the Health 2000 replication sample or in the FINNRISK sample \((P > 0.05)\). In the Finnair sample, a significant association was observed only with the somatic symptoms of depression \((P = 0.014, \beta = 0.129)\). In the GENMETS dataset, a less significant signal compared with the job-related exhaustion signal was
Table 2. Results of the replication analysis of job-related exhaustion in the Health 2000 replication sample and the pooled Health 2000 sample

<table>
<thead>
<tr>
<th>SNP</th>
<th>CHR</th>
<th>Gene</th>
<th>A1</th>
<th>MAF</th>
<th>(\beta_{\text{repl}})</th>
<th>SE_{\text{repl}}</th>
<th>(P_{\text{repl}})</th>
<th>(\beta_{\text{comb}})</th>
<th>SE_{\text{comb}}</th>
<th>(P_{\text{comb}})</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs13219957</td>
<td>6</td>
<td>UST</td>
<td>A</td>
<td>0.150</td>
<td>0.097</td>
<td>0.047</td>
<td>0.038</td>
<td>0.178</td>
<td>0.036</td>
<td>9.68E-07</td>
</tr>
<tr>
<td>rs7677237</td>
<td>4</td>
<td>HERC6</td>
<td>C</td>
<td>0.040</td>
<td>0.042</td>
<td>0.088</td>
<td>0.633</td>
<td>0.237</td>
<td>0.063</td>
<td>0.0002</td>
</tr>
<tr>
<td>rs10502587</td>
<td>18</td>
<td>FAM59A</td>
<td>T</td>
<td>0.105</td>
<td>-0.007</td>
<td>0.054</td>
<td>0.892</td>
<td>-0.134</td>
<td>0.041</td>
<td>0.0013</td>
</tr>
<tr>
<td>rs11121720</td>
<td>1</td>
<td>UBIAD1</td>
<td>C</td>
<td>0.064</td>
<td>0.057</td>
<td>0.070</td>
<td>0.417</td>
<td>0.196</td>
<td>0.053</td>
<td>0.0002</td>
</tr>
<tr>
<td>rs2402382</td>
<td>2</td>
<td>PAPOLG</td>
<td>C</td>
<td>0.218</td>
<td>0.003</td>
<td>0.042</td>
<td>0.944</td>
<td>0.095</td>
<td>0.032</td>
<td>0.0033</td>
</tr>
<tr>
<td>rs10488596</td>
<td>7</td>
<td>CHR2M</td>
<td>T</td>
<td>0.152</td>
<td>0.018</td>
<td>0.047</td>
<td>0.701</td>
<td>0.110</td>
<td>0.036</td>
<td>0.0021</td>
</tr>
</tbody>
</table>

SNP, single-nucleotide polymorphism; CHR, chromosome; A1, minor allele; \(\beta_{\text{repl}}\), beta for the analysis of the replication dataset; \(P_{\text{repl}}\), \(P\)-value for the analysis of the replication dataset; \(\beta_{\text{comb}}\), \(\beta\) for the analysis of the combined dataset; SE_{\text{comb}}, SE for the analysis of the combined Health 2000 dataset; \(P_{\text{comb}}\), \(P\)-value for the analysis of the combined Health 2000 dataset.

Nominal \(P\)-values are reported. Of the seven SNPs with \(P < 1 \times 10^{-5}\) in GENMETS two were not included in the replication analysis: rs17036631 because of the strong linkage (\(D^2 = 1.00\)) with rs11121720, and rs2034705 due to technical problems in genotyping.

Table 3. Association of rs13219957 with job-related exhaustion in the Health 2000 samples, the FINRISK sample, and the occupational samples, and the meta-analysis of the studies

<table>
<thead>
<tr>
<th>Sample</th>
<th>(P)</th>
<th>(\beta)</th>
<th>SE</th>
<th>MAF</th>
<th>(n)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Health 2000</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GENMETS</td>
<td>1.55E-07</td>
<td>0.306</td>
<td>0.058</td>
<td>0.139</td>
<td>1256</td>
</tr>
<tr>
<td>Health 2000 replication</td>
<td>0.037</td>
<td>0.097</td>
<td>0.047</td>
<td>0.150</td>
<td>1660</td>
</tr>
<tr>
<td>FINRISK</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FINRISK</td>
<td>0.010</td>
<td>0.050</td>
<td>0.019</td>
<td>0.143</td>
<td>10753</td>
</tr>
<tr>
<td>Occupational cohorts</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Finnair</td>
<td>0.093</td>
<td>0.089</td>
<td>0.053</td>
<td>0.152</td>
<td>1378</td>
</tr>
<tr>
<td>Nurses</td>
<td>0.151</td>
<td>0.309</td>
<td>0.213</td>
<td>0.169</td>
<td>73</td>
</tr>
<tr>
<td>Occupational cohorts meta-analysis</td>
<td>0.047</td>
<td>0.102</td>
<td>0.051</td>
<td>0.153</td>
<td>1451</td>
</tr>
<tr>
<td>All</td>
<td>6.73E-07</td>
<td>0.081</td>
<td>0.016</td>
<td>0.144</td>
<td>15120</td>
</tr>
</tbody>
</table>

MAF, minor allele frequency.
Nominal \(P\)-values are reported.

\(^a\)Health 2000 GWAS and the replication sample are pooled together and combined with the other cohorts, using meta-analysis.

\(^b\)\(Q\) test \(P\)-value = 0.012, \(I^2 = 73\%\), suggesting some heterogeneity which is mostly caused by the FINRISK sample with its different measures of job-related exhaustion.

detected (Table 4). In the combined Health 2000 sample, a suggestive association with Beck Depression Inventory (BDI) components disappeared (\(P > 0.05\)) after adjustment for the job-related exhaustion score, whereas the association with job-related exhaustion was only marginally diminished (\(P < 0.05\)) when adjusting for BDI components (Supplementary Material, Tables ST3 and ST4).

**DISCUSSION**

To our knowledge, this is the first study to examine the genetic basis of job-related exhaustion with molecular genetic methods. We performed the GWAS for the quantitative exhaustion dimension of burnout in a sample of 1256 individuals. The strongest association found, rs13219957 (\(P = 1.55 \times 10^{-7}\), \(\beta = 0.306\)) in the intron of UST, was replicated at the \(P < 0.05\) level in the replication sample from the same study and in two occupational cohorts. The genetic variant was also associated with a single question concerning job-related exhaustion in a further population-based sample of 10 753 individuals (\(P = 0.01\), \(\beta = 0.050\)). Finally, a meta-analysis including all 15 120 Finnish individuals yielded a genome-wide suggestively significant association (\(P = 6.75 \times 10^{-7}\), \(\beta = 0.081\)). Even though there is a lack of strict genome-wide significance, nominally significant replications in three replication samples suggest that our results are unlikely to be a false-positive finding. Rs13219957, which is situated in the intronic region of the UST, marks a candidate region for further studies of job-related and general exhaustion.

Our results show no risk variants of a large or intermediate effect size and genome-wide significance in the association analysis, which is consistent with previous genome-wide studies and a meta-analysis on depression (17,18). However, a surplus of low detected \(P\)-values compared with expected ones (Fig. 2A) suggests the presence of variants with weak genetic effects that cannot be detected with adequate significance levels by the GWAS with our sample size. This is further supported by the replication of the association of the GWAS top finding, rs13219957, at \(P < 0.05\), when using large sufficient population samples.

The marker rs13219957 is situated in the intronic region of the UST (uronyl-2-sulfotransferase) gene. Uronyl-2-sulfotransferase is an enzyme that catalyzes the transfer of a sulfate group to dermanin or chondroitin sulfate (19). Dermanin and chondroitin sulfate form the polysaccharide part of chondroitin sulfate proteoglycans (CSPGs), which are an abundant subtype of proteoglycans in the extracellular matrix of nervous system (20). Highly negative sulfate groups of proteoglycans have been proposed to play a part in the binding of growth factors (21) and also to enhance the Alzheimer-like changes in tau protein (22) and amyloid \(\beta\) peptide (23). The importance of CSPGs and chondroitin/dermanin sulfate sulfotransferases for the brain has been illustrated by findings that show their high expression in neurogenic regions of the central nervous system mice, while an inhibition of sulfation was demonstrated to decrease the proliferation of cultured neural stem cells (24). Mice with inhibited UST showed a
Table 4. Association of rs13219957 with symptoms of burnout and depression in all the cohorts

<table>
<thead>
<tr>
<th>Burnout syndrome</th>
<th>Job-related exhaustion</th>
<th>Cynicism</th>
<th>Diminished professional efficacy</th>
<th>Depression</th>
<th>Anhedonia</th>
<th>Somatic symptoms</th>
<th>Appetite</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$P$</td>
<td>$\beta$</td>
<td>$P$</td>
<td>$\beta$</td>
<td>$P$</td>
<td>$\beta$</td>
<td>$P$</td>
</tr>
<tr>
<td>GENMETS</td>
<td>$1.55 \times 10^{-7}$</td>
<td>0.306</td>
<td>$0.005$</td>
<td>0.165</td>
<td>$0.014$</td>
<td>0.143</td>
<td>$0.004$</td>
</tr>
<tr>
<td>Health 2000 replication</td>
<td>$0.037$</td>
<td>0.097</td>
<td>0.118</td>
<td>0.073</td>
<td>0.508</td>
<td>0.031</td>
<td>0.856</td>
</tr>
<tr>
<td>FINNRISK</td>
<td>$0.010$</td>
<td>0.050</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>0.426</td>
</tr>
<tr>
<td>Finnair</td>
<td>0.093</td>
<td>0.089</td>
<td>0.257</td>
<td>0.054</td>
<td>$0.032$</td>
<td>0.100</td>
<td>0.216</td>
</tr>
<tr>
<td>Nurses</td>
<td>0.151</td>
<td>0.309</td>
<td>$0.015$</td>
<td>0.513</td>
<td>0.520</td>
<td>0.137</td>
<td>NA</td>
</tr>
<tr>
<td>Occupational cohorts meta-analysis</td>
<td>$0.047$</td>
<td>0.102</td>
<td>0.096</td>
<td>0.078</td>
<td>$0.025$</td>
<td>0.102</td>
<td>NA</td>
</tr>
</tbody>
</table>

Nominal $P$-values are reported.
$P$-values <0.05 are shown in boldface.

The gene-environment-wide analysis did not reveal an interaction between job demands and selected genetic variants, but considering the large sample sizes needed for studying $G \times E$ (35), it is quite likely that we were underpowered to detect such phenomena. Gene–environment analysis of the strongest findings of the original GWAS did not show any replicable interaction. Interestingly, when using samples derived from workers of an airline company or nurses rather than those from the general population, rs13219957 showed stronger associations with diminished professional efficacy and cynicism symptoms than with job-related exhaustion. This could reflect the combined effect of the genetic risk factors and the work environment on the manifestation of burnout. The $G \times E$ part of our study must be seen as an initial explorative trial to open the discussion on the topic.

The moderate size of the original sample in the GWAS is a limitation of our study, which was partially balanced by the use of substantially larger replication samples. This did enable us to confirm the association of rs13219957 with job-related exhaustion, but the moderate original sample size decreased the power, increased the rate of type II errors and made it very likely that we did not detect all of the common genetic variation linked to job-related exhaustion (Power calculation, Supplementary Material). Job-related exhaustion was evaluated with a subscale of a burnout questionnaire (MBI-GS), which is a standard procedure to assess burnout (36), because there are at present no biological markers or other validated objective measures of burnout available. In the FINRISK sample, the job-related exhaustion was evaluated with a single question concerning workers’ experience regarding high work load. A single unvalidated question is not equivalent to the exhaustion subscale in the MBI-GS questionnaire, which can be the reason for the lowest effect size seen in this cohort and for the heterogeneity in the meta-analysis when the FINRISK cohort is included (Table 3).

However, the effect noticed in relation to also the other work-stress-related question suggests that the question indeed relates to the job-related exhaustion phenomenon (Results). In the $G \times E$ study, the measurement of job demands is likely to be affected by the mental state of the responders causing a common method problem with job-related exhaustion questions.

In sum, we report here the first attempt to explore the genetic background of job-related exhaustion. The variant of $UST$ with the strongest association in our genome-wide analysis showed disturbed neuronal migration during brain development (25), which could also affect the functioning of the mature brain.

In a twin study (14), genetic factors explained most of the correlation of job-related exhaustion and anxious depression. In our study, the association of rs13219957 with depressive symptoms was significant in the GENMETS dataset and one of the replication cohorts, the Finnair sample, in which it was associated with the somatic symptoms component of depression (Table 4). The component weights questions of fatigue and loss of energy which are also key elements in the burnout syndrome. This can be interpreted as a certain specificity of the rs13219957 for exhaustion-like symptoms that can be assessed by job-related exhaustion measures, but also by measures that weight somatic symptoms of depression among workers. Homogenization of the sample when restricting the analysis to workers only in the study of job-related exhaustion can affect opportunities to detect an association. More studies are needed to reveal the genetic overlap of job-related exhaustion and depression and to determine whether the genetic factors differ when high work stress is present.

The association of the UST variant with depression symptoms was in this study not as strong as with exhaustion symptoms. However, an intronic variant of $UST$ (rs2500535) has been reported to be associated with antidepressive treatment response at a genome-wide significance level (26) (not in linkage disequilibrium with rs13219957 in the Finnish population). In addition, $UST$ shows a functional connection with the genes presented in the GWASs of depression. In one study (27), the strongest signal for association ($P = 1.8 \times 10^{-5}$) was observed for an SNP located in the vicinity of $DSEL$ (dermatan-sulfate epimerase-like), which, like $UST$, is considered to be involved in the posttranslational modification of dermatan sulfate (28).

However, in a meta-analysis of depression, there was no further support for this result (29). In several earlier studies (30–33), the genomic region containing $DSEL$ has also been found to be linked to bipolar mood disorder. The close functional similarity of $UST$ and $DSEL$ may be an indication of the common biological mechanisms for job-related exhaustion and mood disorders. Further support for the role of the dermatan sulfate pathway in the regulation of mood was obtained in a recent meta-analysis of major depressive disorder (34), where a variant of the dermatan chondroitin/sulfate proteoglycan gene, VCAN, emerged as the second-strongest finding.
a significant association also in three replication samples. The results give quite strong support for the idea that the variant marks an area with a weak regulating effect on such exhaustion symptoms that are seen in the context of professional work. Fine mapping of the genomic area and functional studies are still needed to locate the effective variant. Association studies in populations outside Finland are needed to assess how our findings can be generalized. Considering previous reports about associations of the other dermatan/chondroitin sulfate-related genes with mood disorders, the whole molecular pathway should be regarded as a candidate pathway for studies of stress-related traits. The hypothesis that there are interactions between genes and stressful work environment in job-related exhaustion must be examined further using longitudinal study settings and larger samples.

**MATERIALS AND METHODS**

**Population and sampling**

The subjects for this study were obtained from two Finnish population-based studies and two Finnish occupational samples: the Health 2000 study including the GENMETS GWAS dataset and the replication dataset, the FINRISK cohorts, the Finnair airline company sample and the nurse sample.

Health 2000 is a large epidemiological health study carried out in 2000–2001 (http://www.terveys2000.fi/doc/methodologyrep. pdf). The sample collection was performed in two stages with stratified cluster sampling, and it was representative of the Finnish population aged 30 years and over. The study design has been described in detail earlier (37). Characteristics of the sample for this genetic study are shown in Supplementary Material, Table ST1. The Ethics Committee of the Helsinki and Uusimaa Hospital District approved the study protocol, and a written informed consent was obtained from all participants after providing a description of the study.

The Health 2000 sample comprised 8028 adults aged 30 years and over; 93% had participated in at least some phase of the study. Inclusion criteria for our genetic study comprised an age ≤ 64 years, currently working or on sick leave, and no more than one missing value per dimension of the MBI-GS. These criteria were met by 3441 individuals, and DNA samples had been obtained from 3253 of them. Of the 3253 individuals, 1456 were genotyped at a genome-wide level in the context of the GENMETS substudy of the Health 2000 survey. The GENMETS sample comprised of the metabolic syndrome cases and the controls (38). Selection criteria for the GENMETS study are shown in the Supplementary Material. To minimize the selection effect, the case–control status in the GENMETS study was used as a covariate in the genetic analyses. To minimize the selection effect, the case–control status in the FINRISK study was used as a covariate in the genetic analyses.

In secondary analyses, the FINRISK sample, job-related exhaustion was assessed with a single question: how often are you troubled by having to stretch your strength to the extreme to be able to cope with your present work or work load? The five-stage response scale ranged from almost always to never. Only workers responded to the question. In addition we asked: how often are you troubled by continuous busyness/stress at your work? This question was similarly scaled. General exhaustion was measured by the simple question assessing feelings during the past month: ‘Do you feel exhausted or overworked?’ The response scale ranged from 1 to 3 (1 = often, 2 = sometimes, 3 = not at all). All the scales were inverted for the analyses to make the comparison with other results easier. These variables followed the normal distribution.

In secondary analyses of Health 2000 and the occupational cohorts, we used two other subscales of MBI-GS, which included five questions concerning cynicism and six concerning professional efficacy. Also the best measure of depressive symptoms was selected for each cohort. Three principal components from the 21-item BDI (46) were formed for the Health 2000 sample, using the principal component analysis (PCA). The first component focused on anhedonia, the second on somatic symptoms and the third on appetite (Supplementary Material, Tables ST6 and ST7). To achieve a normal distribution of the principal components of BDI, we used the 1/× transformation of the principal components for 1997 and 2002, questionnaires and a question on general exhaustion in all of the years. A written informed consent was obtained from all participants. The appropriate ethics committees approved the surveys.

The FINRISK cohorts in 1997, 2002 and 2007 comprised, together, 32 549 individuals (1997: n = 7159, response rate = 71.6%; 2002: n = 13 437, response rate = 71.3%; 2007: n = 11 953, response rate = 66.9%). An answer to the question about job-related exhaustion was available for 12 493 workers, and about general exhaustion for 25 424 subjects, both workers and non-workers.

The Finnair airline company sample included workers who were occupied in different ground service functions at the Helsinki Vantaa airport, or as pilots or cabin attendants (40). The nurse sample was derived from a bigger study of female health care professionals, which was described in detail earlier (41). All of the nurses were working in shifts and whether in high stress or low stress environments. Altogether, we had for 1518 employees of the Finnair airline company and for 77 nurses information from MBI-GS. The occupational samples were analysed together using fixed-effect meta-analysis.

**Measures**

Job-related exhaustion was assessed in the Health 2000 study and in the occupational cohorts using the exhaustion subscale of the Finnish version of the MBI-GS (42). MBI-GS is a widely used questionnaire that covers three dimensions of burnout with separate subscales (43). The method is applicable to any profession, and its reliability and validity have been confirmed in different samples (44,45). All the subscales consist of five symptom statements scored on a seven-item frequency scale ranging from 0 (never) to 6 (daily). One missing value per subscale was accepted and replaced with a mean value.

In the FINRISK sample, job-related exhaustion was assessed with a single question: how often are you troubled by having to stretch your strength to the extreme to be able to cope with your present work or work load? The five-stage response scale ranged from almost always to never. Only workers responded to the question. In addition we asked: how often are you troubled by continuous busyness/stress at your work? This question was similarly scaled. General exhaustion was measured by the simple question assessing feelings during the past month: ‘Do you feel exhausted or overworked?’. The response scale ranged from 1 to 3 (1 = often, 2 = sometimes, 3 = not at all). All the scales were inverted for the analyses to make the comparison with other results easier. These variables followed the normal distribution.

In secondary analyses of Health 2000 and the occupational cohorts, we used two other subscales of MBI-GS, which included five questions concerning cynicism and six concerning professional efficacy. Also the best measure of depressive symptoms was selected for each cohort. Three principal components from the 21-item BDI (46) were formed for the Health 2000 sample, using the principal component analysis (PCA). The first component focused on anhedonia, the second on somatic symptoms and the third on appetite (Supplementary Material, Tables ST6 and ST7). To achieve a normal distribution of the principal components of BDI, we used the 1/× transformation of the principal components for
anhedonia and appetite and the log transformation of the principal component for somatic symptoms. In the FINRISK sample, the 13-item BDI was factorized with PCA. Three components corresponding to the components of the 21-item BDI were detected (Supplementary Material, Tables ST7 and ST8). Also in this sample, the $1/\times$ transformation was made for the anhedonia and appetite components. In the Finnair cohort, PCA was made for the 10-item depression scale (DEPS) (47), which yielded two components corresponding to the anhedonia and somatic symptoms components of BDI (Supplementary Material, Tables ST9 and ST10). Logarithmic transformation was performed on the anhedonia component. Independent of the transformation, all the scores are reported so that higher score means worse symptoms.

The level of demands at work was used as an indicator of the environment’s stressfulness. It was evaluated by using a mean score for the five questions in the Job Content Questionnaire (48). The demand score was normally distributed in the sample.

DNA extraction

DNA was extracted from peripheral blood leukocytes using the commercially available Puregene DNA isolation kit (Puregene, Gentra Systems, Minneapolis, MN, USA). The extraction method is described in more detail by Blin and Stafford (49).

Genotyping and quality control

The GENMETS samples were genotyped with the Illumina 610K platform (Illumina Inc., San Diego, CA, USA) at the Wellcome Trust Sanger Institute (Hinxton, UK). The call rate was >95% for both individuals and markers. Markers with a minor allele frequency (MAF) <1% or a Hardy–Weinberg $P < 1 \times 10^{-6}$ were excluded from the analyses. Heterozygosity and gender checks were performed, and individuals with any discrepancies were removed. Cryptic relatedness was detected for six individuals, and also they were removed. The genotyping was successful for 1256 individuals with job-related exhaustion information and 555,388 SNPs. In order to control for population stratification in GENMETS, 20 principal components were created and used as covariates in the genome-wide analysis. The replication sets were genotyped at the Institute for Molecular Medicine Finland using iPLEX Gold chemistry (Sequenom, San Diego, CA, USA). In the initial health 2000 replication set, for all six SNPs, the success rate was >95%, the MAF >0.01 and the Hardy–Weinberg equilibrium, $P > 0.001$. The call rate for genotyping of individuals was 95%, and this was passed by 1660 individuals with a non-missing phenotype. The same quality standards for genotyping were required in the occupational cohorts.

Rs13219957 was genotyped in the FINRISK 1997, 2002 and 2007 cohorts using iPLEX Gold chemistry (Sequenom, San Diego, CA, USA). Individuals with a call rate exceeding 80% for the iplex were kept in the analysis ($n = 21390$). Genotyping of rs13219957 was successful for 21,256 individuals (99.4%), 10,753 of whom had answered to the question concerning job-related exhaustion and 20,813 to the question about general exhaustion. Genotyping was performed at the Institute for Molecular Medicine Finland. The same genotyping technique was performed in the occupational cohorts and genotyping was successful for 73 individuals from the nurse cohort and 1378 individuals from the Finnair cohort.

Statistical analysis

The exhaustion score was logarithmically transformed in order to follow normal distribution and then treated as a quantitative trait. The association between genotyped SNPs and the exhaustion score was tested using the linear regression analysis of the PLINK software. We expected the additive model to be valid, meaning that one copy of an allele would have less of an effect than two copies, but more of an effect than no copies. Z-score normalization was performed for all the analyses, meaning that distribution was standardized for a mean of 0 and a SD of 1. Beta ($\beta$) stands in this article for the effect of one minor allele in units of standard deviation. Age, sex, principal components for GWAS data, and metabolic syndrome status for GENMETS were used as covariates in the GWAS. Linear regression analysis with age and sex as covariates was performed in the rest of the analyses. $R^2$ stands for a coefficient of determination, indicating how much the genetic variant explains about the variance in job-related exhaustion after removal of the variance caused by age and sex. The Bonferroni correction for the Health 2000 replication sample was calculated according to the equation $P_{\text{Bonferroni}} = 1 - (1 - P)^{\text{number of tests}}$. The programs used in this article are listed in detail in the Supplementary Material.

SUPPLEMENTARY MATERIAL

Supplementary Material is available at HMG online.

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