論文の内容の要旨

論文題目　Identification of genetic factors of sleep disorders

（睡眠障害の遺伝要因の探索）

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In humans, narcolepsy is a sleep disorder that is characterized by sleepiness, cataplexy, and rapid eye movement (REM) sleep abnormalities. Narcolepsy is caused by a reduction in the number of neurons that produce hypocretin (orexin) neuropeptide. Both genetic and environmental factors contribute to the development of narcolepsy. A main genetic factor of narcolepsy is a human leukocyte antigen (HLA) class II allele, specifically the *HLA-DQB1*06:02*. This *HLA* allele is thought to be necessary, but not sufficient, for the development of narcolepsy.

Essential hypersomnia (EHS) is another type of sleep disorder characterized by excessive daytime sleepiness without cataplexy. Previous studies have reported that EHS and narcolepsy are associated with the same susceptibility genes. Especially, about 30–50% of EHS patients carry *HLA-DQB1*06:02* allele (EHS with/without *HLA-DQB1*06:02*). The pathogenesis of EHS is thought to be partially similar to that of narcolepsy.

In the first section of the study, I tried to detect rare susceptible variants for sleep disorders, especially copy number variations (CNVs). DNA microarray data and a CNV detection software application, PennCNV, were used in order to detect CNVs in 327 narcoleptic patients, 33 EHS with *HLA-DQB1*06:02* patients, 104 EHS without *HLA-DQB1*06:02* patients and 459 healthy individuals. Overall, a significant

enrichment of rare and large CNVs (frequency ≤ 1%, size ≥ 100 kb) in the narcoleptic patients was observed (case-control ratio of CNV count = 1.54, $P = 5.00 \times 10^{-4}$). Both EHS with *HLA-DQB1*06:02* and EHS without *HLA-DQB1*06:02* were also found to possess higher burden of rare and large CNVs (case-control ratio of CNV count = 1.46, $P = 3.24 \times 10^{-2}$, case-control ratio of CNV count = 1.43, $P = 2.17 \times 10^{-3}$, respectively). Next a region-based association analysis was performed, including CNVs with its size ≥ 30kb. Rare and large CNVs in *PARK2* region showed a significant association with narcolepsy. Four narcoleptic patients were discovered to carry duplications of the gene region, while no controls carried the duplication, which was further confirmed by quantitative PCR assay ($P = 3.07 \times 10^{-2}$). This duplication was also found in two EHS without *HLA-DQB1*06:02* out of 104 patients ($P = 2.91 \times 10^{-2}$). Furthermore, a pathway analysis on narcoleptic patients revealed enrichments of gene disruptions by rare and large CNVs in immune response, acetyltransferase activity, cell cycle regulation and regulation of cell development. This study constitutes the first report on the risk association between multiple rare and large CNVs and the pathogenesis of sleep disorders.

In the second section, I evaluated contribution of common variants on the onset of narcolepsy. In addition, shared genetic background of narcolepsy and EHS with/without *HLA-DQB1*06:02*, based on CDCV hypothesis was assessed. A polygenic analysis, using 393 narcoleptic patients, 38 EHS with *HLA-DQB1*06:02* patients, 119 EHS without *HLA-DQB1*06:02* patients, other neuropsychiatric disorders including 376 panic disorders, 213 autism and 56 schizophrenia, and 1,582 healthy

individuals, were performed. Narcolepsy heritability was estimated as 58.1% ($P_{HLA\text{-}DQB1*06:02}$ = 2.30×10^{-48}, P_{others} = 6.73×10^{-2}). Heritability of narcolepsy explained by the region other than *HLA* was estimated as 1.3% (P_{others} = 2.43×10^{-2}). The results also indicated that common small-effect SNPs contributed to the development of narcolepsy. Reported susceptible genes for narcolepsy in Japanese population, *CPT1B*, *TRA@*, *P2RY11*, were found to explain 0.8% of heritability (P_{others} = 9.74 × 10^{-2}), suggesting that a certain proportion of narcolepsy susceptible genetic factors was not yet discovered. Next, the shared genetic background among narcolepsy and EHS with/without *HLA-DQB1*06:02* were assessed. Common genetic background between narcolepsy and EHS with *HLA-DQB1*06:02* was estimated including *HLA-DQB1*06:02* effects (narcolepsy: 58.1%, $P_{HLA\text{-}DQB1*06:02}$ = 2.30 × 10^{-48}, P_{others} = 6.73 × 10^{-2}, EHS with *HLA-DQB1*06:02*: 40.4%, $P_{HLA\text{-}DQB1*06:02}$ = 7.02 × 10^{-14}, P_{others} = 1.34 × 10^{-1}). This suggested that substantial proportion of heritability was shared between narcolepsy and EHS with *HLA-DQB1*06:02*. Narcolepsy polygenic risk, excluding *HLA-DQB1*06:02* effects, did not explain the onset of EHS either with or without *HLA-DQB1*06:02*. However, EHS with *HLA-DQB1*06:02* patients tended to have larger polygenic risk of "narcolepsy" than EHS without *HLA-DQB1*06:02* patients (EHS with *HLA-DQB1*06:02*: 1.4%, P_{others} = 1.56 × 10^{-1}, EHS without *HLA-DQB1*06:02*: 0.4%, P_{others} = 3.06 × 10^{-1}). Neither narcolepsy polygenic risks on other neuropsychiatric diseases or polygenic risks among healthy controls were estimated to be significant, indicating that the analysis was enough reliable. This study could be regarded as the first polygenic risks among sleep disorders.