

Circadian Clock and Gene NPAS2 Polymorphism in the General Population

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Abstract

The article describes the results of the study the number of hours of sleep per night, and their association with the candidate gene NPAS2 rs4851377 in the open population of men 25 - 44 years old. Most men needed 7 hours of sleep per day - 46.7% were male with an 8-hour sleep (24.4%) in second place. Genotype C/C gene NPAS2 rs4851377 is more common in those who slept in the day at least 8 hours (33.3%) and 9 o'clock (33.3%), and genotype C/T and T/T were in persons with 7-hour sleep (50% and 53.3%, respectively). It was found that 6 hours of sleep a factor of 4.5 was observed significantly more frequently among carriers of the allele T, than the C allele, whose sleep was 9 hours; and 7 hours of sleep in comparison with 9-hour sleep was 4 times more often carriers of allele T, allele carriers than C.

Keywords: Epidemiology; NPAS2 rs4851377 Gene; Continuance Sleep

Introduction

The circadian clock affects many physiological and behavioral aspects, such as body temperature, hormone release, and the sleep-wake cycle. Consolidation of sleep and synchronization of sleep-wake cycles are regulated by a complex interaction between circadian processes and homeostatic processes [1]. The central driver of the circadian rhythm is located in the suprachiasmatic nucleus of the hypothalamus and controls the peripheral clock in the extra-suprachiasmatic region of the brain, regulating the sleep-wake cycles [2].

Usually the cycle of biochemical reactions in neurons of suprachiasmatic nuclei does not fit exactly in 24 hours and in most people it is approximately 25 hours. The molecular clock of the organism "adapts" to the local light cycle. Activation of certain hourly genes caused by light, and the subsequent synthesis of de novo corresponding proteins, for a long time, suppresses the activity of other watch genes by

protein complexes [3,4]. Many problems with sleep are caused by the inability to correctly “set” the circadian clock in accordance with the daily alternation of day and night. Moreover, sleep disturbances (e.g. jet lag syndrome, called jet lag) caused by transmeridional flights or shift work are also associated with the synchronization of circadian clock of the organism [5]. If the work of the biological clock is broken, either the connections between local oscillators in different tissues or between the central oscillator (SCN of the hypothalamus) and the rest of the body are disconnected. These disorders are the basis for further failure of neuroendocrine rhythms and behavior, which is manifested in healthy people, but is especially characteristic of psychiatric and neurological patients [6].

Molecular mechanisms of circadian oscillations are based on positive/negative transcriptional-translational feedback, generated by at least nine major circadian genes [7]. Among them, the gene NPAS2, which is located on chromosome 2 in 2q11.2, is one of the most important circadian genes. NPAS2 forms heterodimers with BMAL1, and then activates the circadian genes PER and CRY, which are necessary to maintain biological rhythms in many organisms. Animal studies have shown that loss of normal function of the NPAS2 gene can cause defects in several aspects of the circadian system, such as patterns of sleep and behavior [8].

Aim of the Study

Therefore, the aim of our study was to study the prevalence and association of the gene NPAS2 rs4851377 with the duration of sleep per day in men in the open population of 45 - 69 years.

Materials and Methods

As part of the epidemiological study of 2014 - 2016 gg. a random representative sample of the male population of 25 - 44 years of one of the districts of Novosibirsk was surveyed. A random method was used to select 200 men, the average age of 35.5 years, who underwent psychosocial testing. The testing was carried out using the CD Jenkins, *et al.* “4-item Jenkins Sleep Questionnaire” (JSQ) is a violation of the quality and duration of sleep [9] validated to the Russian population during a large-scale epidemiological study carried out under the WHO “MONICA” program (Multinational Monitoring of Trends and Determinants of Cardiovascular Disease) and the MONICA-Psychosocial Optional Study (MOPSY) routines [10,11]. The respondents were asked to answer the test questions independently. The respondents answered the questions of the test, the way they feel at the moment.

In men included in the study, the frequency distribution of genotypes rs4851377 of the NPAS2 gene was studied. With the help of molecular genetic methods polymorphic variants of rs4851377 gene NPAS2 were studied in 179 respondents. Genomic DNA was isolated from venous blood by the phenol-chloroform extraction method. The gene polymorphism was tested by real-time PCR according to the manufacturer’s protocol (TaqMan probes, Applied Biosystems, USA) on an ABI 7900HT instrument.

The statistical analysis was carried out using the SPSS 11.5 software package [12]. To test the statistical significance of the differences between the groups, Chi-square (χ^2) was used [13]. Values of $p \leq 0.05$ were considered statistically significant.

The study passed the expertise of the local committee on biomedical ethics (Minutes No. 4 of 15.10.2009).

Results

In the open population of men 25 - 44 years, the duration of sleep per day was as follows: “5 hours or more” in 2.2% of individuals, “6 hours” - 17.8%, “7 hours” - 46.7%, “8 hours” - 24.4%, “9 hours” - 6.7% and “10 hours or more” - 2.2%.

The prevalence of polymorphic variants of candidate gene NPAS2 rs4851377 in the population was as follows: homozygous genotype C/C in 13.3%, heterozygous genotype C/T -53.3% and homozygous genotype T/T -33.3%.

Circadian Clock and Gene NPAS2 Polymorphism in the General Population

In the population, the genotype C/C of the gene NPAS2 rs4851377 was found more often in those who slept at least 8 hours (33.3%) and 9 hours (33.3%), and the C/T and T genotypes/T were in persons with a 7-hour sleep (50% and 53.3%, respectively) ($\chi^2 = 18,425$ df = 10 p < 0,05).

Allele from the candidate gene NPAS2 was found in 43.3% of men, and the allele of T - in 56.7% of men.

It was found that a 6-hour sleep 4.5-fold (95% CI 0.735 - 27.536) was significantly more often observed among persons bearing the T allele than the C allele, whose sleep was 9 hours ($\chi^2 = 6,142$ df = 1 p < 0.05); also 7-hour sleep compared with 9-hour sleep was 4 times more often (95% CI 0.66 - 24.245) in carriers of T allele than in carriers of C allele ($\chi^2 = 5.488$ df = 1 p < 0.05).

Discussion

In the studied male population of 25 - 44 years, the majority of subjects answered that they had 7 hours of sleep per day - 46.7%, men with 8-hour sleep (24.4%) took the second place, 6 hours of sleep per day were required - 17.8% of men. Our results are in agreement with the conclusions we found in the available literature - the older the age, the less sleep is required for proper rest. Czeisler CA and co-authors, after analyzing more than 300 publications, found that a person of active working age should sleep an average of 7 - 9 hours a day [14,15].

The most common in the population was the heterozygous genotype of the candidate gene NPAS2 C/T -53.3%, followed by the homozygous T/T genotype, with both variants of the candidate gene being more common in men who had only 7 hours per day. The genotype C/C of the candidate NPAS2 gene was significantly more frequent among those who slept at least 8 hours (33.3%) and 9 hours (33.3%).

The major allele T of the NPAS2 candidate gene was 4.5 times more common in men whose sleep was 6 hours a day and 4 times more common in men with a 7-hour sleep.

Thus, the obtained data testify to the determination by the site of rs4851377 of candidate gene NPAS2 "suckiness" and "larkiness" in men.

Conclusions

1. Among men 25 - 44 years, the majority slept 7 hours of sleep per day - 46.7%, in second place were men with an 8-hour sleep (24.4%), 6 hours of sleep per day was required - 17.8% of men.
2. The prevalence of polymorphic variants of the candidate gene NPAS2 rs4851377 in the population was as follows: the C/C genotype in 13.3%, the C/T genotype of 53.3% and the T/T genotype of 33.3%.
3. In the population, the genotype C/C of the gene NPAS2 rs4851377 was found more often in those who slept at least 8 hours (33.3%) and 9 hours (33.3%), and C/T genotypes and T/T were in persons with a 7-hour sleep (50% and 53.3%, respectively).
4. It was found that a 6-hour sleep 4.5 times more reliably was observed among persons bearing the T allele than the C allele, in which the sleep was 9 hours; also a 7-hour sleep compared with a 9-hour sleep was 4 times more often in carriers of the T allele than in the carriers of the allele C.

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Conflict of Interest

There is no conflict of interest.

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