Healthy Sleep Patterns, Genetic Susceptibility, and Incident of Cirrhosis and Liver Cancer

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Supplementary Methods

Study Design and Population

UK Biobank is a large prospective cohort study containing comprehensive health information of one-half million UK participants from 2006 to 2010. All participants signed the written informed consent following the principles of the Declaration of Helsinki. The North West Multi-Center Research Ethics Committee ethically approved the UK Biobank cohort. Our accession to the UK Biobank data was through application No.51539. In our study, incident liver cancer and cirrhosis were defined as the occurrence after the baseline recruitment according to the National Health Service (NHS) central registries and death registries. The end of the follow-up period was up to the date of diagnosis, loss to follow-up, dropout, the date of death, or complete follow-up (January 2018 for assessment centers in England and Wales and November 2016 for assessment centers in Scotland), whichever occurred first.

Assessment of sleep behaviours

The question is used to determine sleep duration: “About how many hours sleep do you get in every 24 hours? (Please include naps)”. 7–8 h/day sleep duration was categorized as appropriate duration, else were too short (<7 h/day) and too long (≥9 h/day) sleep. Chronotypes were obtained by asking “Do you consider yourself to be a ‘morning’ person or an ‘evening’ person?” with responses of definitely a “morning” person; more a “morning” than “evening” person; ‘more an “evening” than “morning” person’; or definitely an “evening” person’. Daytime sleepiness was assessed with: “How likely are you to doze off or fall asleep during the daytime when you don't mean to? (e.g., when working, reading, or driving)”. The options consisted of never/rarely; sometimes; often; or all of the time. Information on insomnia symptoms was collected by asking “Do you have trouble falling asleep at

night or do you wake up in the middle of the night?". with responses of never/rarely; sometimes; or usually. We classified snoring by asking "Does your partner or a close relative or friend complain about your snoring?" with responses of yes or no. The participants replied with 'do not know' or 'prefer not to answer' in all the above questions were excluded from the analysis.

Assessment of other covariates

The covariates in the study included sex, age at recruitment (continuous), race (white or non-white), family history of cancer (yes/no), body mass index (BMI, kg/m^2, continuous), alcohol intake frequency (never, special occasions only, 1-3 times/ month, 1-2 times/ week, 3-4 time s/ week, daily or almost daily), smoking status (non-smokers, current smokers, ex-smokers), physical activity (quartiles of Metabolic Equivalent Task hours per week, MET-h/week), processed meat consuming frequency (never, <1 time occasion per week, 1 time occasion per week, 2-4 times occasion per week, 5-6 times occasion per week, ≥7 times occasion per week), socioeconomic status (Townsend deprivation index, continuous), menopause status for female participants (yes/no, not sure), aspirin usage (yes/no), the history of diabetes and hypertension (yes/no).

PRS construction

The odds ratio of each SNP from the previous study was extracted to calculate the β (the log-transformed OR) and the genotype dosage of each risk allele (M) was recoded as 0, 1, 2. We categorized participants into low and high genetic risk according to the sum of risk alleles. Participants with less than 3 risk alleles were defined as low risk group, otherwise as high risk group.

Statistical analysis

Person-time was calculated from the date of recruitment until the date of diseases diagnosis, the date of death, or the date of last follow up (January 2018 for assessment centers in England and Wales and November 2016 for assessment centers in Scotland), whichever occurred first. To evaluate the joint and interactive effects between sleep patterns and calculated PRS on liver cancer and cirrhosis incidence, we categorized the participants into six groups jointly subclassified by healthy sleep score (unfavourable, intermediate, and favourable) and PRS (low and high groups). Besides, we categorized the participants into four groups jointly subclassified by sleep behaviours (low risk and high risk sleep behaviours) and PRS (low and high groups). Stratification analyses were performed according to age (<60 or ≥60), sex (female or male), BMI (<25 kg/m^2 or ≥25 kg/m^2), smoking status (never smokers, or smokers), alcohol intake (non-excessive, or excessive alcohol intake), physical activity (< 29.5 MET-h/week, or ≥ 29.5 MET-h/week), family history of cancer (no, or yes), aspirin use (no, or yes), history of diabetes (no, or yes), and history of hypertension (no, or yes).

Supplementary Figures

Figure S1.

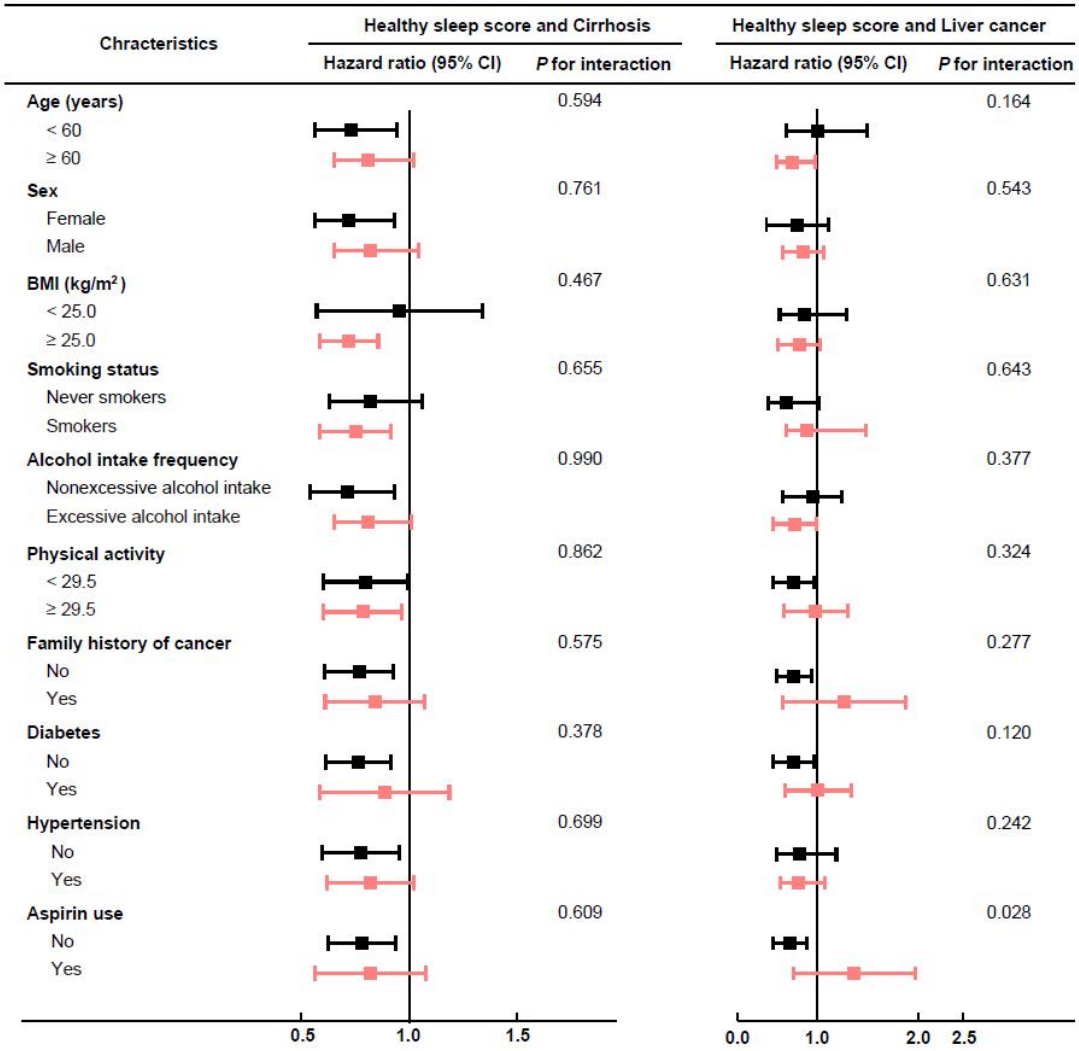

Figure S1. Stratified hazard ratio of cirrhosis and liver cancer in relation to healthy sleep score among participants in the UK Biobank.

5 sleep behaviours (morning person, sleep duration, 7-8 h/d, (7-8 hours per day), no frequent insomnia, no snoring and no excessive daytime sleepiness) were summed to calculated the healthy sleep score ranging from 0 to 5, and the score was categorized into 2 subgroups: 3, 4 and 5 versus 0,1 and 2 (HR=1.0). HRs were adjusted for sex (males or females), age at recruitment (years, continuous), race (white or non-white), family history of cancer (yes or no), body mass index (kg/m^2, continuous), alcohol intake frequency (never,

special occasions only, one to three times a month, once or twice a week, three or four times a week, daily or almost daily), smoking status (non-smokers, ex-smokers, current smokers), physical activity (quartiles), Townsend deprivation index (continuous), menopause status (yes, no, not sure), aspirin use (yes or no), history of diabetes (yes or no), and history of hypertension (yes or no).

Figure S2.

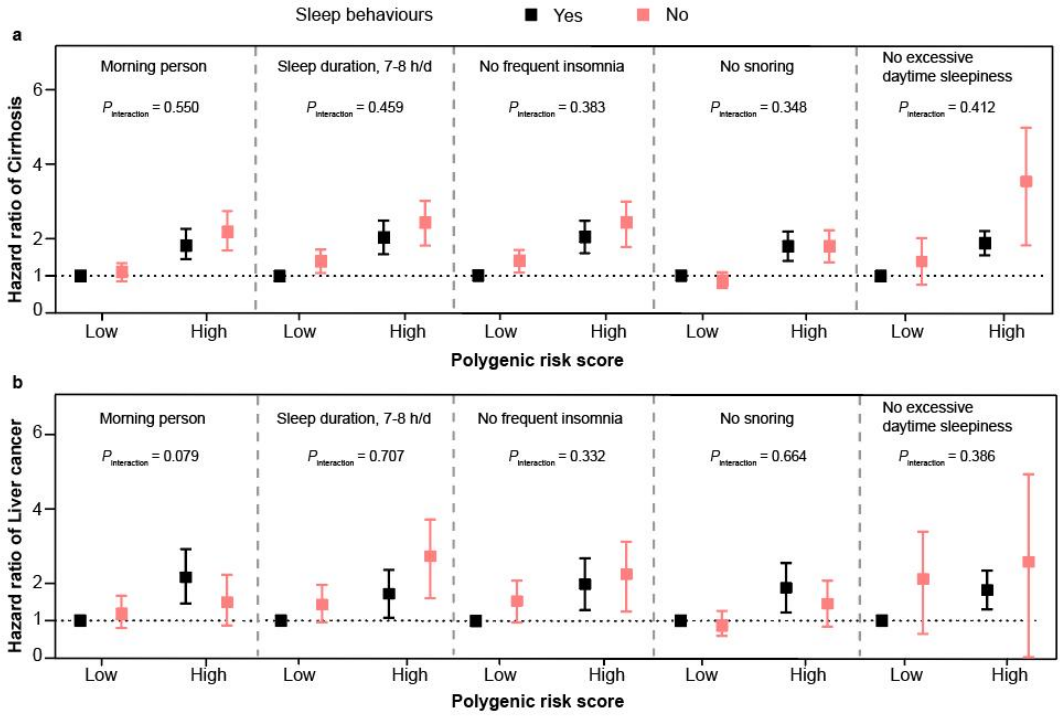

Figure S2. Hazard ratio of cirrhosis and liver cancer in relation to 5 sleep behaviours and polygenic risk score among participants in the UK Biobank.

PNPLA3 (rs738409) G allele, *TM6SF2* (rs58542926) T allele, and *HSD17B13* (rs72613567) T allele were combined into polygenic risk score ranging from 0 to 5 for risk-increasing alleles, and 0,1 and 2 as low polygenic risk score, 3, 4 and 5 as high polygenic risk score. HRs were adjusted for sex (males or females), age at recruitment (years, continuous), race (white or non-white), family history of cancer (yes or no), body mass index (kg/m^2,

continuous), alcohol intake frequency (never, special occasions only, one to three times a month, once or twice a week, three or four times a week, daily or almost daily), smoking status (non-smokers, ex-smokers, current smokers), physical activity (quartiles), Townsend deprivation index (continuous), menopause status (yes, no, not sure), aspirin use (yes or no), history of diabetes (yes or no), history of hypertension (yes or no), and the other 4 sleep behaviours. The reference value for sleep behaviours and polygenic risk score was no and low (HR=1.0). 7-8 h/d, 7-8 hours per day. The $P_{interaction}$ were for multiplicative interaction between 5 sleep behaviours and polygenic risk score.

Figure S3.

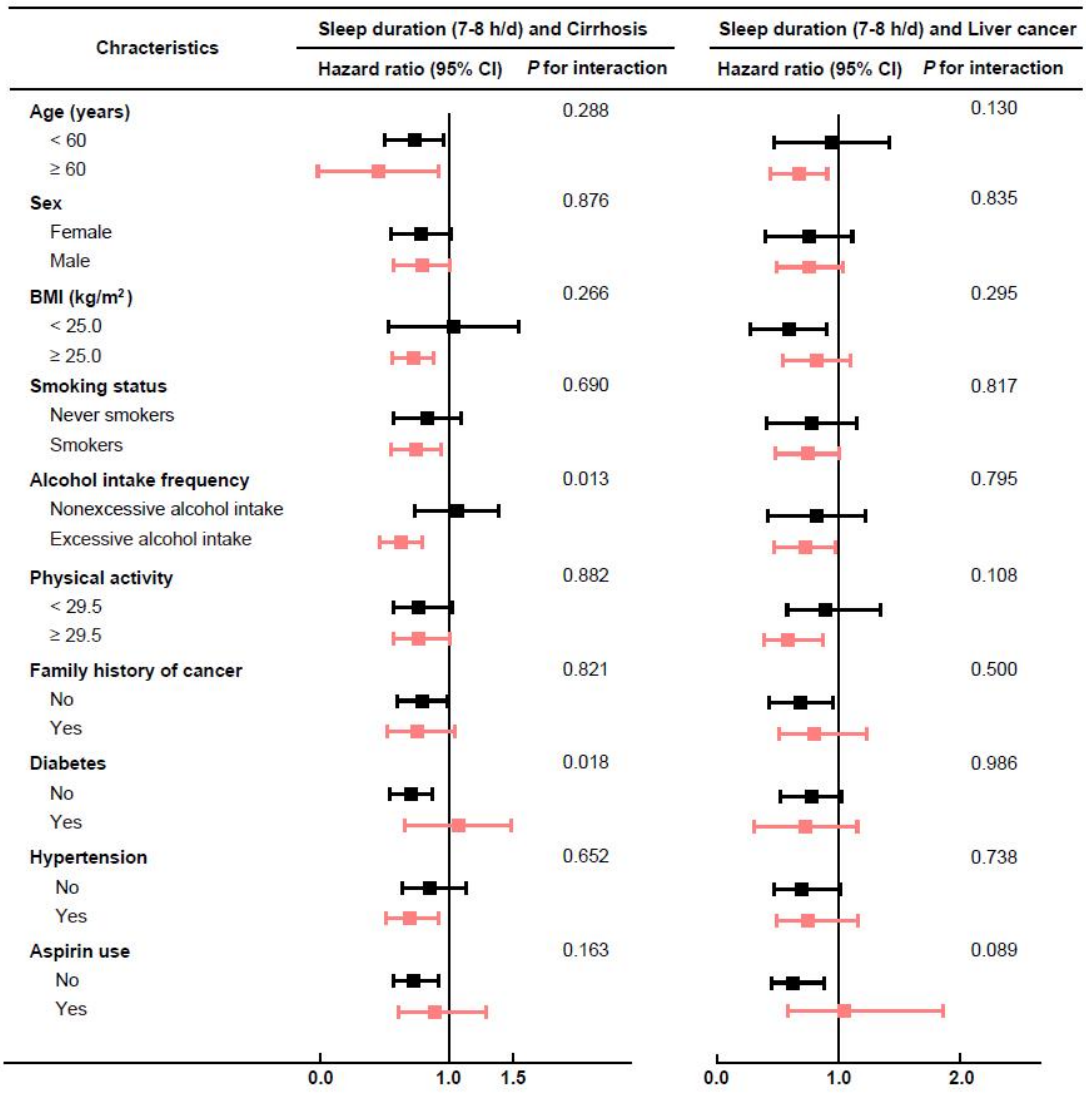

Figure S3. Stratified hazard ratio of cirrhosis and liver cancer in relation to sleep duration among participants in the UK Biobank.

HR of sleep duration (7-8 h/d) yes versus no (HR=1.0). HRs were adjusted for sex (males or females), age at recruitment (years, continuous), race (white or non-white), family history of cancer (yes or no), body mass index (kg/m^2, continuous), alcohol intake frequency (never, special occasions only, one to three times a month, once or twice a week, three or four times a week, daily or almost daily), smoking status (non-smokers, ex-smokers, current smokers), physical activity (quartiles), Townsend deprivation index (continuous), menopause status (yes, no, not sure), aspirin use (yes or no),

history of diabetes (yes or no), history of hypertension (yes or no), and the other 4 sleep behaviours. 7-8 h/d, 7-8 hours per day.

Supplementary Table

Table S1 Hazard ratios (95% CI) of Cirrhosis and Liver cancer incidence in relation to polygenic risk score

Outcome	Polygenic risk score		*P* for trend
	Low	High	
Cirrhosis incidence			
Model 1	1.00 (ref)	1.86 (1.58-2.21)	<0.001
Model 2	1.00 (ref)	1.89 (1.60-2.24)	<0.001
Liver cancer incidence			
Model 1	1.00 (ref)	1.70 (1.28-2.26)	<0.001
Model 2	1.00 (ref)	1.72 (1.30-2.28)	<0.001

Abbreviations: CI, confidence interval.

Model 1 adjusted for sex (males or females), age at recruitment (years, continuous).

Model 2 additionally adjusted for race (white or non-white), family history of cancer (yes or no), body mass index (kg/m^2, continuous), alcohol intake frequency (never, special occasions only, one to three times a month, once or twice a week, three or four times a week, daily or almost daily), smoking status (non-smokers, ex-smokers, current smokers), physical activity (quartiles), Townsend deprivation index (continuous), menopause status (yes, no, not sure), aspirin use (yes or no), history of diabetes (yes or no), and history of hypertension (yes or no).