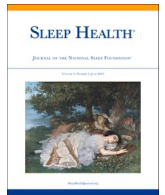




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Is there an association between daytime napping, cognitive function, and brain volume? A Mendelian randomization study in the UK Biobank

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ABSTRACT

Objectives: Daytime napping has been associated with cognitive function and brain health in observational studies. However, it remains elusive whether these associations are causal. Using Mendelian randomization, we studied the relationship between habitual daytime napping and cognition and brain structure.

Methods: Data were from UK Biobank (maximum $n = 378,932$ and mean age = 57 years). Our exposure (daytime napping) was instrumented using 92 previously identified genome-wide, independent genetic variants (single-nucleotide polymorphisms, SNPs). Our outcomes were total brain volume, hippocampal volume, reaction time, and visual memory. Inverse-variance weighted was implemented, with sensitivity analyses (Mendelian randomization-Egger and Weighted Median Estimator) for horizontal pleiotropy. We tested different daytime napping instruments to ensure the robustness of our results.

Results: Using Mendelian randomization, we found an association between habitual daytime napping and larger total brain volume (unstandardized $\beta = 15.80 \text{ cm}^3$ and 95% CI = 0.25; 31.34) but not hippocampal volume ($\beta = -0.03 \text{ cm}^3$ and 95% CI = -0.13; 0.06), reaction time ($\text{exp}\beta = 1.01$ and 95% CI = 1.00; 1.03), or visual memory ($\text{exp}\beta = 0.99$ and 95% CI = 0.94; 1.05). Additional analyses with 47 SNPs (adjusted for excessive daytime sleepiness), 86 SNPs (excluding sleep apnea), and 17 SNPs (no sample overlap with UK Biobank) were largely consistent with our main findings. No evidence of horizontal pleiotropy was found.

Conclusions: Our findings suggest a modest causal association between habitual daytime napping and larger total brain volume. Future studies could focus on the associations between napping and other cognitive or brain outcomes and replication of these findings using other datasets and methods.

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Introduction

Daytime napping, defined as brief daytime bouts of sleep,¹ is a universal² and prevalent behavior.³ Most children under 3-year-olds nap (80% of 1- to 2-year-olds and 65% of 3-year-olds), but napping is less common during school age (12.7% of 6-13-year-olds) and adulthood (13.7% of 26-64-year-olds). Napping rises again in older adults (27% of >65-year-olds), and the impact of this behavior on brain health is of special interest.⁴ Napping seems beneficial to performance on certain cognitive tasks.^{2,5} These benefits arise immediately following a brief nap (eg, 5-15 minutes) and can last

between 1 and 3 hours. After a long nap (> 30 minutes), a temporary deterioration of performance emerges, followed by improvements that can last up to a day.⁵ Some authors argue that individuals who frequently have a nap and those who never nap may differ in the benefits derived from napping, with the latter experiencing no benefits from it.² However, a recent meta-analysis did not find this effect but stated that, in previous studies, this difference was clear for memory tasks, but the effects of napping on other cognitive domains were mixed.⁶

While, recently, more attention has been paid to napping, it remains elusive whether habitual daytime napping could be beneficial or detrimental for cognition.⁷ Given that the most pronounced decline during aging occurs in reaction time and memory,⁸ and the high prevalence of cognitive impairment in the aging population,⁹ the identification of modifiable risk factors, such as sleep habits, is essential. In addition, the association between napping and brain

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volume is not well characterized even though almost a third of older adults nap, and reductions in brain volume are more common in older adults. Moreover, hippocampal and total brain volumes are strong candidates in accounting for variations in memory performance and overall cognition.^{10,11} As most studies on the relationship between napping and cognitive or brain health have been observational, there is uncertainty about whether this is causal in nature.

To overcome this limitation, Mendelian randomization (MR) can be used, which is based on the analysis of genetic markers, found in published genome-wide association studies (GWAS), to examine the possible causal associations between exposures and outcomes.¹² Previous MR studies investigated the causal relationship between sleep and cognitive and structural brain outcomes. These studies reported that both short and long sleep durations are associated with poorer cognitive outcomes,¹³ long sleep duration is associated with increased cortical thickness,¹⁴ and different sleep traits are associated with a greater risk of neurodegenerative diseases.¹⁵⁻¹⁷ Regarding napping, Anderson et al.¹⁸ found suggestive evidence that self-reported habitual daytime napping is associated with lower Alzheimer's disease risk. However, no previous MR studies have investigated the association between daytime napping, cognitive outcomes, and brain volumes. Thus, the present study aimed to use MR to examine whether the relationship between genetic liability to daytime napping, cognitive function, and brain volumes might be causal.

Participants and methods

Sample

The UK Biobank (UKB) cohort has been described in detail elsewhere.¹⁹ Briefly, UKB recruited 500,000 males and females from the general United Kingdom population, aged 40-69 years at baseline (2006-2010). Although UKB recruited participants of distinct ancestries, those included in this study were of white European ancestry and retained if they had relevant (quality-controlled) genotype and phenotype data ($n = 378,932$) (see Table 1 for sample characteristics).

Study design

Our exposure (SNPs_x) sample overlapped with our cognitive function outcome sample (SNPs_y) by 77%, but this was < 10% for the neuroimaging outcomes. This is because the discovery GWAS for the exposure under study was performed in UKB participants, which

was also our analytical sample. However, in the following, we detail, in Sensitivity Analyses, the strategy that we undertook to mitigate this sample overlap.

Genotyping and quality control (QC) in UKB

487,409 UKB participants were genotyped using 1 of 2 customized genome-wide arrays that were imputed to a combination of the UK10K, 1000 Genomes Phase 3, and the Haplotype Reference Consortium reference panels, which resulted in 93,095,623 autosomal variants.²⁰ We then applied additional variant level QC and excluded genetic variants with Fisher's exact test < 0.3, minor allele frequency < 1%, and a missing call rate of $\geq 5\%$. Individual-level QC meant that we excluded participants with excessive or minimal heterozygosity, more than 10 putative third-degree relatives as per the kinship matrix, no consent to extract DNA, sex mismatches between self-reported and genetic sex, missing QC information, and non-European ancestry (based on how individuals had self-reported their ancestry and the similarity with their genetic ancestry, as per a principal component analysis of their genotype).

Outcomes

Cognitive function measures

At baseline, UKB administered a total of 5 cognitive assessments to all participants, via a computerized touch-screen interface, all of which are described in detail elsewhere.²¹ For the purposes of this study and to maximize statistical power, we pragmatically chose *visual memory* and *reaction time*. For the *visual memory* task, respondents were asked to correctly identify matches from 6 pairs of cards after they had memorized their positions. The number of incorrect matches (number of attempts made to correctly identify the pairs) was then recorded, with a greater number reflecting poorer visual memory. Reaction time (in milliseconds) was recorded as the mean time taken by participants to correctly identify matches in a 12-round game of the card game "Snap." A higher score on this test indicated a slower (poorer) reaction time. Both of these variables were positively skewed, and therefore, reaction time scores were transformed using the natural logarithmic function $[\ln(x)]$, while visual memory was transformed using $[\ln(x + 1)]$.

Neuroimaging parameters

Structural brain magnetic resonance imaging (MRI) scans have been performed in a subsample of the UKB using standard protocols²² (Supplementary Note 1). Here, we had complete

Table 1
Sample characteristics by daytime napping groups in UK Biobank.

	Never/rarely ($n = 215,991$)	Sometimes ($n = 143,995$)	Usually ($n = 18,946$)
<i>Covariates</i>			
Age (mean/SD)	55.4 (8.1)	57.6 (7.9)	59.4 (7.5)
Sex (%female)	59%	50%	33%
Education years (mean/SD)	15.4 (4.9)	14.8 (5.1)	14.3 (5.3)
Townsend—most deprived quintile (%)	17%	20%	24%
Body Mass Index—kg/m ² (mean/SD)	26.8 (4.5)	27.9 (4.9)	28.5 (5.2)
Alcohol consumption—times per month-daily (%)	46%	42%	44%
Moderate physical activity—days (mean/SD)	3.6 (2.3)	3.6 (2.3)	3.7 (2.3)
Ever smoking—Current (%)	9%	11%	14%
Type-2 diabetes (%)	3%	6%	10%
Antihypertensives (%)	16%	24%	32%
Cardiovascular disease (%)	5%	8%	14%
<i>Outcome variables</i>			
Reaction time—milliseconds (mean/SD)	548.8 (108.8)	564.1 (116.4)	579.1 (128.5)
Visual memory—number of errors (mean/SD)	4.0 (3.2)	4.4 (3.6)	4.0 (4.6)
Hippocampal volume—cm ³ (mean/SD)	3.8 (0.4)	3.8 (0.5)	3.8 (0.4)
Total brain volume—cm ³ (mean/SD)	1498.7 (72.8)	1488.5 (72.7)	1477.0 (73.5)

^{sd} Standard deviation.

neuroimaging and genotype data for $n = 35,080$ individuals. We analyzed hippocampal volume (average of left + right hippocampal volume, cm^3) and total brain volume (normalized for head size, cm^3).

Selection of genetic instruments

Main daytime napping genetic instrument

Daytime napping was instrumented using 123 genome-wide significant ($P < 5 \times 10^{-8}$) genetic variants discovered in a recent GWAS.²³ These variants were discovered in 452,633 UKB participants, based on the question “do you have a nap during the day?” administered at baseline, with possible responses Never or rarely, Sometimes, and Usually (prefer not to answer coded as missing in the GWAS). The 123 variants explain 1% of the variance in daytime napping. However, here, we selected 92 of the 123 daytime napping SNPs, as we used linkage disequilibrium clumping in PLINK with $r^2 < 0.01$ within 250 kb to exclude correlated variants (Supplementary Table 1). We then calculated the F-statistic that yielded $F = 41$ (indicating a good average strength of our main instrument) using the Cragg–Donald formula²⁴:

$$F = \left(\frac{n - k - 1}{k} \right) \left(\frac{R^2}{1 - R^2} \right)$$

We harmonized the genetic variants between the exposure GWAS and our outcome sample by aligning effect alleles. We also excluded palindromic SNPs (those with the same alleles on the forward and reverse strands) because they can introduce ambiguity in the identification of the effect allele (Supplementary Table 2). Our instrument selection process is detailed in Supplementary Fig. 1.

Additional daytime napping genetic instruments

We additionally partitioned the daytime napping instrument into 2 further subinstruments: i) an 86-SNP instrument that consists of those SNPs that remained genome-wide significant when, in the published GWAS, the authors excluded individuals who had sleep apnea ($n = 5553$) and ii) a 47-SNP instrument that comprised SNPs that remained genome-wide significant on adjustment for excessive daytime sleepiness (Supplementary Table 3). Using the formula $F = (\beta^2/\text{SE}^2)$ to approximate the average instrument strength for these additional instruments in sensitivity analyses, we calculated the F-statistic for each of these additional instruments, which yielded $F = 98.1$ and $F = 47.0$, respectively, indicating good instrument strength.

Statistical analyses

Main analyses

Using PLINK 2.0, we performed linear regressions between each of the daytime napping genetic variants and our outcomes, adjusting for ten principal components to minimize issues of residual confounding by population stratification (ie, confounding of genotype-disease associations by factors related to subpopulation group membership within the overall population). For our MR analyses, inverse-variance weighted (IVW) MR was implemented, with standard sensitivity analyses, including MR-Egger and the weighted median estimator (WME). The IVW, also known as “conventional MR,” estimates the effect of an exposure (eg, daytime napping) on a given outcome (eg, visual memory or reaction time) by taking an average of the genetic variants’ ratio of variant-outcome ($\text{SNP} \rightarrow Y$) to variant-exposure ($\text{SNP} \rightarrow X$) association, which is calculated using the principles of a fixed-effect meta-analysis.²⁵ MR-Egger regression (which yields an intercept term to denote the presence or absence of unbalanced horizontal pleiotropy)²⁶ and the WME can give more robust estimates when up to 50% of the genetic variants are invalid and, thus, do not meet all MR assumptions.²⁷ For the cognitive

function outcomes, results are expressed as exp β -coefficients for log-transformed outcomes, which should be interpreted as % differences in the outcome for every 1-unit increase in daytime napping frequency. For the neuroimaging outcomes, results are expressed as unstandardized beta coefficients to be interpreted as differences in the outcome (in cm^3) for every 1-unit increase in daytime napping.

Sensitivity analyses

- To ensure that our results were robust, we performed all of our MR analyses additionally using a 47- and 86-SNP daytime napping instrument, as described earlier. We confirmed a priori before implementing our analyses that these instruments were of adequate strength (via F-statistics).
- To mitigate potential issues with sample overlap between the discovery GWAS for daytime napping and our analytical dataset (both used UKB), we additionally performed our MR analyses using a reduced 17-SNP daytime napping instrument (Supplementary Table 3). This instrument consisted of the SNPs that were replicated (at $P < 5 \times 10^{-8}$)²³ in an independent cohort (23andMe, $n = 541,333$), as an a priori F-statistic confirmed that it was suitable for use in our MR analyses ($F = 67.1$). We only performed these analyses for the cognitive function outcomes, as the overlap in samples between daytime napping and our neuroimaging analytical sample was $< 10\%$, and it is possible that analyses with a 17-SNP instrument in our subsample of $\sim 35,000$ would result in imprecise MR estimates.

Testing of MR assumptions

- Associations between the genetic instrument and exposure instrumented (GWAS robust): this assumption was met, as the daytime napping variants that we instrumented here have been robustly associated with this phenotype in a recent very large-scale GWAS.
- No evidence of horizontal pleiotropy (no association between genetic instruments and the outcome, other than via the exposure under study): we tested this assumption by implementing MR-Egger and WME sensitivity analyses, as detailed above.
- No associations between genetic variants and confounders of the relationships under study: to assess this assumption, we regressed a number of common confounders on our main instrument (92 SNPs) and used a Bonferroni multiple testing correction of $0.05/92 = 0.0005$. The list of confounders that we selected was based on the recent literature²⁸ and included: years of full-time education, deprivation (Townsend deprivation quintiles), smoking (ever/never/ex-smoker), physical activity (days of moderate activity for more than 10 minutes), body mass index (kg/m^2), alcohol consumption (1–8 times per month/16 times per month-daily/rarely or never), prevalent type-2 diabetes (No/Yes), prevalent hypertension (No=not on antihypertensive medication and Yes=on antihypertensive medication), and prevalent cardiovascular disease (No/Yes).

Results

Sample characteristics

57% of our sample reported that they “never/rarely” had a daytime nap, while 38% and 5% reported “sometimes” and “usually” having a daytime nap, respectively. Participants who reported “usually” having a daytime nap were older, less likely to be female, more likely to be deprived, to be a current smoker, on antihypertensives, have a diagnosis of diabetes, and have prevalent cardiovascular disease. This group also had slower reaction times and, on average, a smaller total brain volume compared to those who “never/rarely” or “sometimes” took a daytime nap.

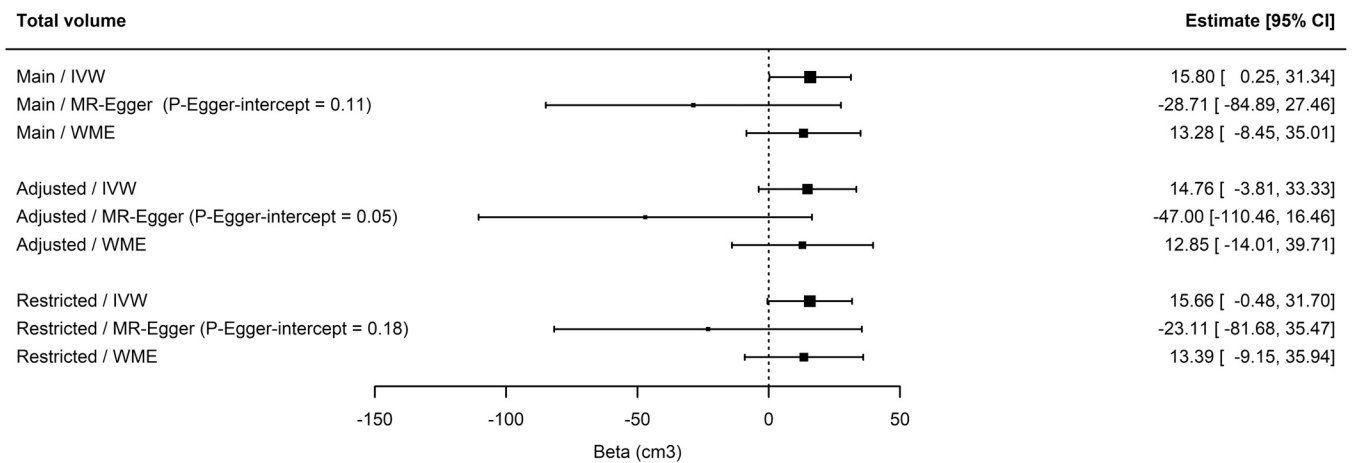


Fig. 1. Associations between daytime napping and total brain volume in UKB, including sensitivity analyses. Note: $n = 35,080$, instrument details: Main = 92-SNP main daytime napping instrument from Dashti et al, 2021, Adjusted = 47-SNP instrument adjusted for excessive daytime sleepiness, Restricted = 86-SNP instrument excluding individuals with self-reported sleep apnea, and 23&Me = 17-SNP instrument used as it has no sample overlap with UKB. IVW, inverse-variance weighted; WME, weighted median estimator; 95% CI, 95% confidence interval.

Main MR results

Associations between daytime napping and total brain, and hippocampal volumes using a 92-SNP genetic instrument

As illustrated in Fig. 1, IVW showed that genetic liability to daytime napping was associated with a 15.80 cm³ larger total brain volume. Both MR-Egger and WME approaches indicated no unbalanced horizontal pleiotropy (MR-Egger intercept P -values > 0.05). The MR-Egger slope was not directionally consistent with the IVW estimate. However, the WME estimate was consistent in terms of direction and size (13.28 cm³) but did not reach conventional levels of statistical significance. Fig. 2 shows that using our main instrument, we found no associations between daytime napping and hippocampal volume. We also found no evidence of horizontal pleiotropy using MR-Egger and WME approaches (MR-Egger intercept P -values > 0.05). We present these associations in Supplementary Table 4.

Associations between daytime napping and cognitive function using a 92-SNP genetic instrument

Figs. 3 and 4 show that, using our main instrument, we found no associations between daytime napping and reaction time or visual

memory. We also found no evidence of horizontal pleiotropy using MR-Egger and WME approaches (all MR-Egger intercept P -values > 0.05). We present these associations in Supplementary Table 4.

Sensitivity analyses

Associations between daytime napping and total brain, and hippocampal volumes using 47- and 86-SNP genetic instruments

When we used a 47-SNP daytime napping instrument (adjusted for excessive daytime sleepiness), the associations with total brain volume were consistent in terms of size and direction with our main results (Fig. 1). This was very similar for associations between the 86-SNP daytime napping and total brain volume (Fig. 1). However, potentially, due to lower total power (particularly in terms of the variance explained (R^2) in daytime napping by these reduced instruments), these estimates had wider 95% CIs around them. In line with our main results above, we observed no association between a 47-SNP daytime napping instrument (excluding individuals with self-reported sleep apnea) and hippocampal volume or an 86-SNP instrument and hippocampal volume (Fig. 2). MR-Egger detected the presence of unbalanced horizontal pleiotropy using the 47-SNP

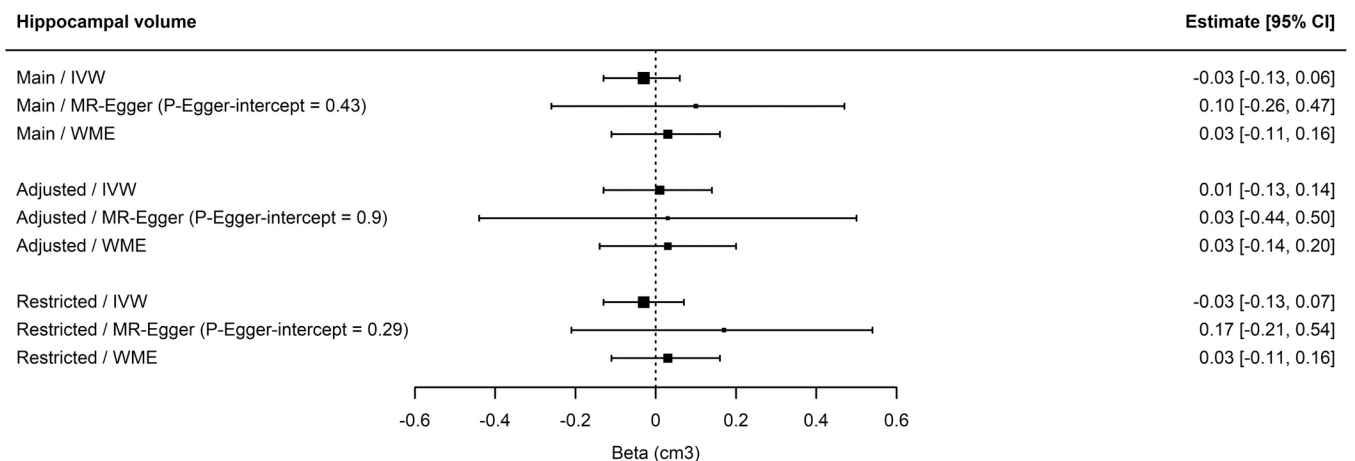


Fig. 2. Associations between daytime napping and hippocampal volume in UKB including sensitivity analyses. Note: $n = 35,080$, instrument details: Main = 92-SNP main daytime napping instrument from Dashti et al, 2021, Adjusted = 47-SNP instrument adjusted for excessive daytime sleepiness, Restricted = 86-SNP instrument excluding individuals with self-reported sleep apnea, 23&Me = 17-SNP instrument used as it has no sample overlap with UKB. IVW, inverse-variance weighted; WME, weighted median estimator; 95% CI, 95% confidence interval.

instrument. Therefore, we excluded the SNP that was most strongly associated with total brain volume (rs301817) and reran our MR analyses, and the MR-Egger intercept P -value was > 0.05 . The IVW and WME estimates, as well as the MR-Egger slope, remained very similar (and all estimates still crossed the null), and we have not presented them here. There were no other issues with unbalanced horizontal pleiotropy, as per the MR-Egger and WME results. We present these associations in [Supplementary Tables 5 and 6](#).

Associations between daytime napping and cognitive function using 47- and 86-SNP genetic instruments

As results presented in [Figs. 3 and 4](#) suggest, sensitivity analyses using the 47-SNP instrument also showed no associations with reaction time or visual memory. Similar results emerged for the 86-SNP instrument with no evidence of associations with either of the 2 cognitive function measures. For reaction time, the MR-Egger intercept P -value indicated the presence of unbalanced horizontal pleiotropy using both the 47- and 86-SNP instruments. Thus, we excluded one SNP that was the most strongly associated with reaction time (rs2099810) and reran our MR analyses, and the MR-Egger intercept had $P > 0.05$. The MR-Egger slopes, as well as the IVW and WME results, remained unchanged and are, therefore, not presented. However, we did not detect any issues with horizontal pleiotropy for visual memory, with both MR-Egger intercept P -values > 0.05 . We present these associations in [Supplementary Tables 5 and 6](#).

Association between daytime napping and cognitive function using a 17-SNP instrument with no sample overlap

Using this restricted instrument to ensure no overlap between our exposure and outcome samples, across all 3 MR approaches, we observed no associations with reaction time or visual memory. MR-Egger detected no issues with unbalanced horizontal pleiotropy ($P > 0.05$). Results are presented in [Figs. 3 and 4](#).

Testing MR Assumption III

Associations between our main 92-SNP daytime napping genetic instrument and common confounders

After a Bonferroni correction, we observed that 12 variants were associated with education, 2 with deprivation, 4 with smoking, 2 with physical activity, 19 with body mass index, 1 with alcohol consumption, 3 with diabetes, 8 with hypertension, and 1 with

cardiovascular disease. We present these associations in [Supplementary Table 7](#).

Discussion

Using a comprehensive Mendelian randomization design, we found an association between genetic liability to self-reported habitual daytime napping and larger total brain volume but not hippocampal volume, reaction time, or visual memory in the UK Biobank. To our knowledge, no prior studies have used MR to try to disentangle the relationship between daytime napping, cognitive, and structural brain outcomes.

Measures of brain volume have been used as proxies of neurodegeneration.²⁹ Reductions in brain volume are expected throughout the lifespan, but this process is accelerated in people with cognitive decline and neurodegenerative diseases.³⁰ Crucially, it is proposed that sleep deficits could be related to these structural changes. For example, several neuroimaging studies have found lower brain volume in people with sleep problems, such as insomnia^{31,32} and poor sleep quality.³³ Moreover, it has been suggested that sleep disturbances may be risk factors for neurodegenerative disorders by promoting processes, such as inflammation and synaptic damage.³⁴ Following this, recent MR studies found that daytime sleepiness was associated with higher Amyotrophic Lateral Sclerosis risk^{16,17} and suggestive evidence that reduced daytime napping is associated with higher Alzheimer's disease risk.¹⁸ In line with these studies, we found an association between habitual daytime napping and larger total brain volume, which could suggest that napping regularly provides some protection against neurodegeneration by compensating for poor sleep.

As previously mentioned, declines in brain volume are expected with aging. In this regard, a meta-analysis of 56 longitudinal MRI studies on healthy individuals found that, after 35 years old, a steady decline in whole brain volume occurs (0.2% per year), which accelerates to 0.5% per year at the age of 60 and greater than 0.5% after the age of 60.³⁵ Assuming a linear decline between 0.2% and 0.5% per year, our finding of a larger total brain volume (ie, 15.8 cm³ \approx 1.3% difference) in those who habitually nap is approximately equivalent to 2.6-6.5 years of difference in aging. In addition, this difference approximately equates to the difference in brain volume between people with normal cognitive function and mild cognitive impairment.^{36,37} Understanding this difference has important clinical

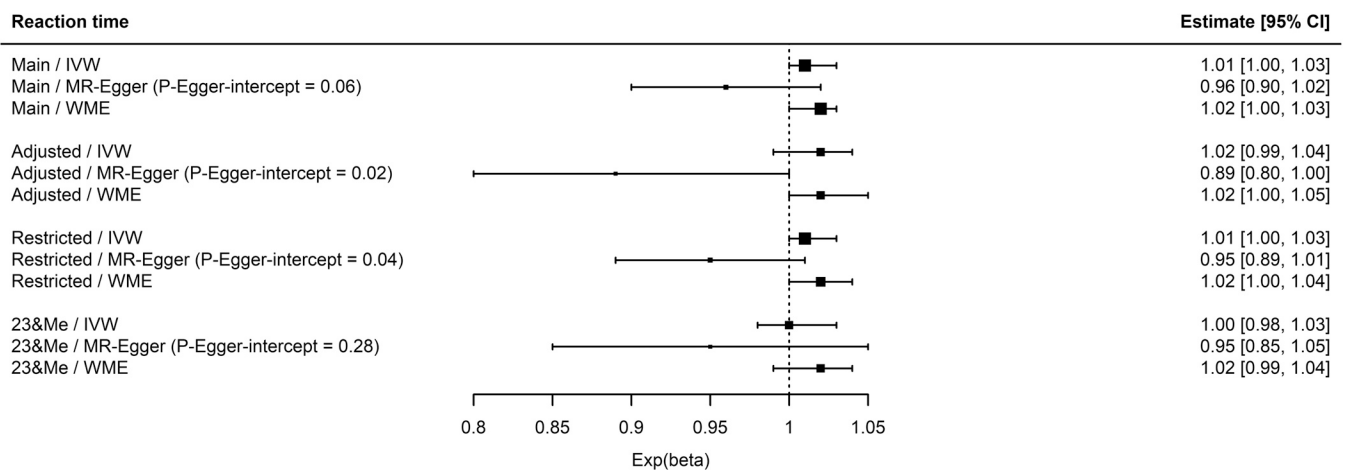


Fig. 3. Associations between daytime napping and reaction time in UKB, including sensitivity analyses. Note: $n = 378,932$, instrument details: Main = 92-SNP main daytime napping instrument from Dashti et al, 2021, Adjusted = 47-SNP instrument adjusted for excessive daytime sleepiness, Restricted = 86-SNP instrument excluding individuals with self-reported sleep apnea, and 23&Me = 17-SNP instrument used as it has no sample overlap with UKB. IVW, inverse-variance weighted; WME, weighted median estimator; 95% CI, 95% confidence interval. Exp(beta): exponentiated beta (eg, an exponentiated beta of 1.01 in reaction time represents an estimated 1% increased or slower reaction time for every 1-unit increase in daytime napping frequency).

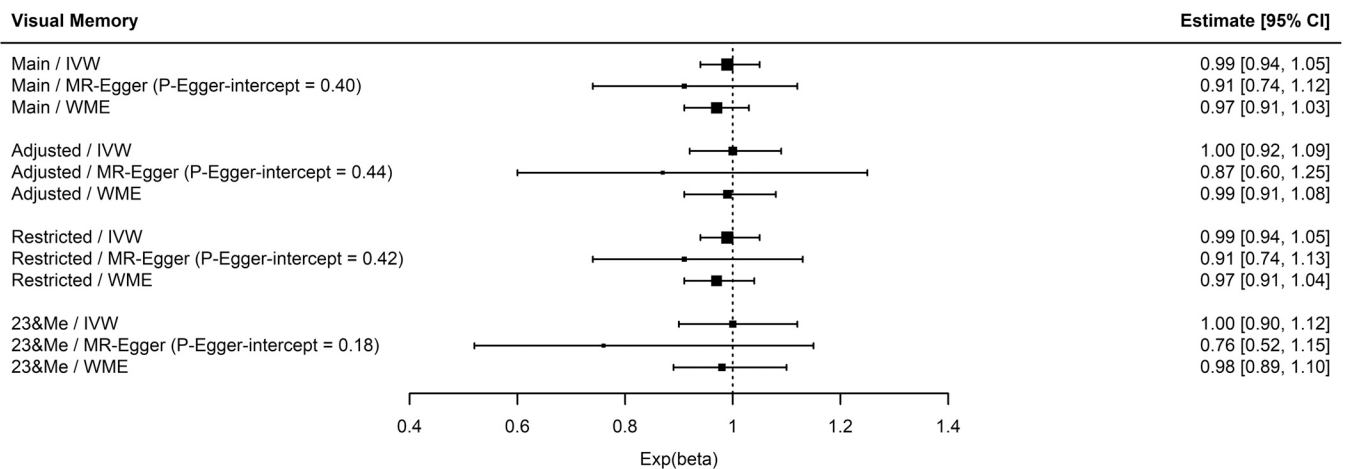


Fig. 4. Associations between daytime napping and visual memory in UKB, including sensitivity analyses. Note: $n = 378,932$, instrument details: Main = 92-SNP main daytime napping instrument from Dashti et al, 2021, Adjusted = 47-SNP instrument adjusted for excessive daytime sleepiness, Restricted = 86-SNP instrument excluding individuals with self-reported sleep apnea, and 23&Me = 17-SNP instrument used as it has no sample overlap with UKB. IVW, inverse-variance weighted; WME, weighted median estimator; 95% CI, 95% confidence interval. Exp(beta): exponentiated beta.

implications for preventing aging-related cognitive impairments, especially if generalizable to the whole population.

The finding of larger total brain volume in relation to habitual daytime napping was found only using the IVW estimate with our main genetic instrument (92 SNPs). However, we wish to emphasize that the IVW estimate in the adjusted (47 SNPs; 14.76 cm^3) and the restricted (86 SNPs; 15.66 cm^3) instruments were almost identical to the estimate using our main instrument (15.80 cm^3). These additional instruments were also consistent in terms of direction. We predict that more precise estimates, with narrower confidence intervals, may be observed if we replicate these analyses with the entire MRI sample when it becomes available ($\approx 100,000$). Moreover, we need to emphasize that, even though we found that participants who “never/rarely” had a daytime nap had a larger total brain volume, this does not imply causation; thus, our Mendelian randomization helped elucidate whether this association is causal.

We also expected to find that habitual daytime napping would be associated with hippocampal volume. Our hypothesis was based on the fact that the hippocampus, as a brain structure that plays a crucial role in memory,³⁸ could be a useful proxy of the variations in memory performance reported to be associated with daytime napping.⁶ However, we did not find this association, nor an association between genetic liability to habitual daytime napping and visual memory performance. Previous studies have reported mixed findings for sleep phenotypes and hippocampal volume, with a number of studies revealing that people with sleep problems have reduced hippocampal volume,^{39–42} while other studies report no associations.^{43–45} However, in contrast to our study, most of these studies were conducted in people with sleep disorders, such as insomnia, rapid eye movement (REM)-sleep behavior disorder, or sleep-disordered breathing, and in samples with less than 1 hundred participants. In line with our results, a recent cross-sectional analysis in the UKB revealed that napping was not related to hippocampal volume.⁴⁶

We were surprised by the lack of a causal link between daytime napping and our cognitive outcomes, especially visual memory, given the evidence of cross-sectional, observational associations between daytime napping and memory,⁶ and the relationship between cognitive function and AD.⁴⁷ However, we found no evidence to support this hypothesis. More reliable cognitive measures may be required to identify these effects. In this regard, our results may be influenced by test characteristics (eg, task sensitivity and difficulty, timing, or instructions).² Furthermore, UKB cognitive assessments are not standardized and were designed specifically for this cohort.

Nonetheless, it is worth mentioning that we examined the association between genetic liability to habitual daytime napping and cognitive function, and not the effect of taking a nap before performing a cognitive test. In addition, it is important to establish that, despite these limitations, UKB cognitive data are valuable resources for researchers seeking determinants of cognitive function.²¹

Moreover, individual differences in the experiences with napping, for example, the presence of sleep apnea⁴⁸ and daytime sleepiness,² may affect the degree of cognitive benefit generated by naps. In this regard, we partitioned the daytime napping instrument into 2 subinstruments (1 excluding individuals who had sleep apnea and the other adjusting for excessive daytime sleepiness). Still, no evidence of associations between self-reported daytime napping and reaction time or visual memory was found. However, other factors, such as slow waves' production, the quality of the prior sleep period, or the presence of sleep inertia, could also influence napping restoration,² which could lead to different effects on cognition. The association between napping and cognitive function may also be influenced by depression, as the frequency of napping has been associated with depressive symptoms.^{49,50} Also, the relationship between depression and cognition is well established.⁵¹

In addition, we only analyzed the frequency of napping. However, observational studies have shown that the length and timing of naps could also affect cognitive function. Unfortunately, information on these dimensions is not available in UKB. Regarding length, previous studies reported that, unlike long naps, the beneficial effects of brief naps are evident almost immediately after waking but last for a shorter period of time.⁵ Nap's timing also determines its effect on cognition, with the post-lunch dip period being the most favorable time to take a nap to overcome the temporary drop in alertness and performance evidence during this period.⁵²

To validate our MR findings, it was checked that the 3 core assumptions that underlie MR were met. Assumption I was met as we instrumented the best available genetic variants as they have been robustly associated with daytime napping in a recent large-scale GWAS.²³ MR-Egger and WME sensitivity analyses were implemented to check assumption II. No evidence of horizontal pleiotropy was found, which corroborates that the association between our genetic variants (for the exposure) and outcomes was only via the exposure under study. Finally, assumption III was tested by performing regressions between our genetic instruments and unobserved confounders, and we found that some of the variants were associated with common confounders. These associations should be further investigated, as they may constitute vertical, rather than horizontal pleiotropy.

Limitations

Limitations of the study should be noted. First, our exposure and cognitive outcome samples overlapped by 77%. However, sensitivity analyses using a reduced 17-SNP daytime napping instrument, replicated by the GWAS authors²³ in an independent cohort (23andMe), confirmed that it was suitable for use in our MR analyses. Using this reduced instrument, we observed no associations with reaction time or visual memory. Second, participants were only white European; future work should examine if these findings are replicated in other ancestries. Third, future instruments for the length and timing of daytime napping are necessary. Fourth, another limitation of our study was the self-report nature of the exposure under study, but napping is notoriously difficult to measure using objective methods. However, in UKB, there was consistency between self-reported sleep measures and accelerometer-derived daytime inactivity duration, which increases confidence in the SNPs for daytime napping. Finally, volunteers from UKB were 40–69 years at baseline; when large cohorts, such as UKB, provide data spanning different generations, it is of interest for future studies to investigate whether the present results are replicated in other age groups.

Conclusions

In summary, our Mendelian randomization study of daytime napping and cognitive/structural brain outcomes suggests an association between genetically instrumented daytime napping and larger total brain volume but not hippocampal volume, reaction time, or visual memory. This study improves our knowledge of the impact of habitual daytime napping on brain health, which is essential to understanding cognitive impairment in the aging population. The lack of evidence for an association between napping, hippocampal volume, and cognitive outcomes in the present study may indicate that other brain areas and cognitive outcomes (e.g., alertness) may be affected by habitual daytime napping and should be studied in the future. These findings further our understanding of the relationship between daytime napping frequency, cognitive function, and structural brain outcomes and elucidate the importance of using different measures to better understand how sleep relates to brain health. Future studies, such as randomized controlled trials, should further explore these relationships.

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Declaration of conflicts of interest

The authors declare that they have no conflict of interest.

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Ethics approval

Ethics approval is not needed as this work was conducted under the approved UK Biobank project number 71702.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi: 10.1016/j.sleh.2023.05.002.

References

- Dhand R, Sohal H. Good sleep, bad sleep! The role of daytime naps in healthy adults. *Curr Opin Pulm Med*. 2006;12:379–382.
- Milner CE, Cote KA. Benefits of napping in healthy adults: impact of nap length, time of day, age, and experience with napping. *J Sleep Res*. 2009;18:272–281.
- Zhang Z, Xiao X, Ma W, Li J. Napping in older adults: a review of current literature. *Curr Sleep Med Rep*. 2020;6:129–135.
- Kocevska D, Lysen TS, Dotinga A, Koopman-Verhoeff ME, Luijk MPCM, Antypa N, et al. Sleep characteristics across the lifespan in 1.1 million people from the Netherlands, United Kingdom and United States: a systematic review and meta-analysis. *Nat Hum Behav*. 2021;5:113–122.
- Lovato N, Lack L. The effects of napping on cognitive functioning. In: Kerkhof GA, Dongen HPA van, eds. *Progress in Brain Research*. Elsevier; 2010:155–166.
- Leong RLF, Lo JC, Chee MWL. Systematic review and meta-analyses on the effects of afternoon napping on cognition. *Sleep Med Rev*. 2022;65:101666.
- Cai H, Su N, Li W, Li X, Xiao S, Sun L. Relationship between afternoon napping and cognitive function in the ageing Chinese population. *Gen Psychiatry*. 2021;34:e100361.
- Blazer DG, Yaffe K, Karlawish J. Cognitive aging: a report from the institute of medicine. *JAMA*. 2015;313:2121–2122.
- Hu C, Yu D, Sun X, Zhang M, Wang L, Qin H. The prevalence and progression of mild cognitive impairment among clinic and community populations: a systematic review and meta-analysis. *Int Psychogeriatr*. 2017;29:1595–1608.
- Ritchie SJ, Dickie DA, Cox SR, et al. Brain volumetric changes and cognitive ageing during the eighth decade of life. *Hum Brain Mapp*. 2015;36:4910–4925.
- Vibha D, Tiemeier H, Mirza SS, et al. Brain volumes and longitudinal cognitive change: a population-based study. *Alzheimer Dis Assoc Disord*. 2018;32:43–49.
- Smith GD, Ebrahim S. 'Mendelian randomization': can genetic epidemiology contribute to understanding environmental determinants of disease? *Int J Epidemiol*. 2003;32:1–22.
- Henry A, Katsoulis M, Masi S, et al. The relationship between sleep duration, cognition and dementia: a Mendelian randomization study. *Int J Epidemiol*. 2019;48:849–860.
- Andrews SJ, Fulton-Howard B, O'Reilly P, Marcora E, Goate AM. Causal associations between modifiable risk factors and the Alzheimer's phenotype. *Ann Neurol*. 2021;89:54–65.
- Grover S, Sharma, M. Sleep, pain, and neurodegeneration: a Mendelian Randomization Study. *Frontiers in Neurology*. 2022; 13. <https://doi.org/10.3389/fneur.2022.765321>.
- Cullell N, Cárceles-Márquez J, Gallego-Fábrega C, et al. Sleep/wake cycle alterations as a cause of neurodegenerative diseases: a Mendelian randomization study. *Neurobiol Aging*. 2021;106:320.e1–320.e12.
- Zhang G, Zhang L, Xia K, Zhuang Z, Huang T, Fan D. Daytime sleepiness might increase the risk of ALS: a 2-sample Mendelian randomization study. *J Neurol*. 2021;268:4332–4339.
- Anderson EL, Richmond RC, Jones SE, et al. Is disrupted sleep a risk factor for Alzheimer's disease? Evidence from a two-sample Mendelian randomization analysis. *Int J Epidemiol*. 2021;50:817–828.
- Sudlow G, Gallacher J, Allen N, et al. UK Biobank: an open access resource for identifying the causes of a wide range of complex diseases of middle and old age. *PLoS Med*. 2015;12:e1001779.
- Bycroft C, Freeman C, Petkova D, et al. The UK Biobank resource with deep phenotyping and genomic data. *Nature*. 2018;562:203–209.
- Lyall DM, Cullen B, Allerhand M, et al. Cognitive test scores in UK Biobank: data reduction in 480,416 participants and longitudinal stability in 20,346 participants. *PLoS One*. 2016;11:e0154222.
- Littlejohns TJ, Holliday J, Gibson LM, et al. The UK Biobank imaging enhancement of 100,000 participants: rationale, data collection, management and future directions. *Nat Commun*. 2020;11:2624.
- Dashti HS, Daghlas I, Lane JM, et al. Genetic determinants of daytime napping and effects on cardiometabolic health. *Nat Commun*. 2021;12:900.
- Burgess S, Thompson SG, CHD CRP. Genetics Collaboration. Avoiding bias from weak instruments in Mendelian randomization studies. *Int J Epidemiol*. 2011;40:755–764.
- Burgess S, Bowden J. Integrating summarized data from multiple genetic variants in Mendelian randomization: bias and coverage properties of inverse-variance weighted methods ArXiv151204486 Stat 2015. <http://arxiv.org/abs/1512.04486>. Accessed August 31, 2021.

26. Bowden J, Davey Smith G, Burgess S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. *Int J Epidemiol.* 2015;44:512–525.
27. Bowden J, Smith GD, Haycock PC, Burgess S. Consistent estimation in Mendelian randomization with some invalid instruments using a weighted median estimator. *Genet Epidemiol.* 2016;40:304–314.
28. Leng Y, Redline S, Stone KL, Ancoli-Israel S, Yaffe K. Objective napping, cognitive decline, and risk of cognitive impairment in older men. *Alzheimers Dement.* 2019;15:1039–1047.
29. Owen JE, Veasey SC. Impact of sleep disturbances on neurodegeneration: Insight from studies in animal models. *Neurobiol Dis.* 2020;139:104820.
30. Anderton BH. Ageing of the brain. *Mech Ageing Dev.* 2002;123:811–817.
31. Altna E, Vrenken H, Van Der Werf YD, van den Heuvel OA, Van Someren EJW. Reduced orbitofrontal and parietal gray matter in chronic insomnia: a voxel-based morphometric study. *Biol Psychiatry.* 2010;67:182–185.
32. Li M, Yan J, Li S, et al. Altered gray matter volume in primary insomnia patients: a DARTEL-VBM study. *Brain Imaging Behav.* 2018;12:1759–1767.
33. Alperin N, Wiltshire J, Lee SH, et al. Effect of sleep quality on amnesic mild cognitive impairment vulnerable brain regions in cognitively normal elderly individuals. *Sleep.* 2019;42:10. <https://doi.org/10.1093/sleep/zsy254> 1.
34. Musiek ES, Holtzman DM. Mechanisms linking circadian clocks sleep, and neurodegeneration. *Science.* 2016;354:1004–1008.
35. Hedman AM, van Haren NEM, Schnack HG, Kahn RS, Hulshoff Pol HE. Human brain changes across the life span: a review of 56 longitudinal magnetic resonance imaging studies. *Hum Brain Mapp.* 2012;33:1987–2002.
36. He J, Farias S, Martinez O, Reed B, Mungas D, DeCarli C. Differences in Brain Volume, Hippocampal Volume, Cerebrovascular Risk Factors, and Apolipoprotein E4 Among Mild Cognitive Impairment Subtypes | Dementia and Cognitive Impairment | JAMA Neurology | JAMA Network. 2009; 66. <https://jamanetwork.com/journals/jamaneurology/article-abstract/798403>. Accessed February 28, 2023.
37. Ryu D-W, Hong YJ, Cho JH, et al. Automated brain volumetric program measuring regional brain atrophy in diagnosis of mild cognitive impairment and Alzheimer's disease dementia. *Brain Imaging Behav.* 2022;16:2086–2096.
38. Eichenbaum H, Otto T, Cohen NJ. The hippocampus—what does it do? *Behav Neural Biol.* 1992;57:2–36.
39. Campabadal A, Segura B, Junque C, et al. Cortical gray matter and hippocampal atrophy in idiopathic rapid eye movement sleep behavior disorder. *Front Neurol.* 2019;10:312.
40. Sforza E, Celle S, Saint-Martin M, Barthélémy JC, Roche F. Hippocampus volume and subjective sleepiness in older people with sleep-disordered breathing: a preliminary report. *J Sleep Res.* 2016;25:190–193.
41. Joo EY, Kim H, Suh S, Hong SB. Hippocampal substructural vulnerability to sleep disturbance and cognitive impairment in patients with chronic primary insomnia: magnetic resonance imaging morphometry. *Sleep.* 2014;37:1189–1198.
42. Koo DL, Shin J-H, Lim J-S, Seong J-K, Joo EY. Changes in subcortical shape and cognitive function in patients with chronic insomnia. *Sleep Med.* 2017;35:23–26.
43. Noh HJ, Joo EY, Kim ST, et al. The relationship between hippocampal volume and cognition in patients with chronic primary insomnia. *J Clin Neurol.* 2012;8:130.
44. Winkelman JW, Benson KL, Buxton OM, et al. Lack of hippocampal volume differences in primary insomnia and good sleeper controls: an MRI volumetric study at 3Tesla. *Sleep Med.* 2010;11:576–582.
45. Spiegelhalter K, Regen W, Baglioni C, et al. Insomnia Does Not Appear to be Associated With Substantial Structural Brain Changes. *Sleep.* 2013;36:731–737.
46. Fjell AM, Sørensen Ø, Amlie IK, et al. Self-reported sleep relates to hippocampal atrophy across the adult lifespan: results from the Lifebrian consortium. *Sleep.* 2020;43:zsz280.
47. Silva MVF, Loures Cd.MG, Alves LCV, de Souza LC, Borges KBG, Carvalho Md.G. Alzheimer's disease: risk factors and potentially protective measures. *J Biomed Sci.* 2019;26:33.
48. Masa JF, Rubio M, Pérez P, Mota M, de Cos JS, Montserrat JM. Association between habitual naps and sleep apnea. *Sleep.* 2006;29:6.
49. Cross N, Terpening Z, Rogers NL, et al. Napping in older people 'at risk' of dementia: relationships with depression, cognition, medical burden and sleep quality. *J Sleep Res.* 2015;24:494–502.
50. Liu Y, Peng T, Zhang S, Tang K. The relationship between depression, daytime napping, daytime dysfunction, and snoring in 0.5 million Chinese populations: exploring the effects of socio-economic status and age. *BMC Public Health.* 2018;18:759.
51. Rock P, Roiser J, Riedel W, et al. Cognitive impairment in depression: a systematic review and meta-analysis. *Psychol Med.* 2013;44:1–12.
52. Slama H, Deliens G, Schmitz R, Peigneux P, Leproult R. Afternoon nap and bright light exposure improve cognitive flexibility post lunch. *PLoS One.* 2015;10:e0125359.