ORIGINAL RESEARCH

# Associations Between Sleep Duration and Cardiometabolic Diseases Among Residents in Southwest China

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**Introduction:** The burden of cardiometabolic diseases (CMDs), defined as stroke, coronary artery disease, and diabetes mellitus, continues to pose a global challenge. Sleep duration has been linked to cardiovascular health. However, there is a lack of focused investigations on CMDs in underdeveloped areas of China.

**Purpose:** This study aimed to examine the relationship between sleep duration and CMDs among residents from southwest China. **Patients and Methods:** This large cross-sectional study screened data from the National Key Research and Development Program (2018YFC1311400). Based on sleep duration reported through a standardized questionnaire, encompassing the sleep patterns of the past five years, participants were classified into three groups: <6, 6–8, >8 hours. Baseline characteristics were compared, and Poisson regression models were used to assess the relationship between sleep duration and CMDs. Subgroup analysis was conducted based on age and gender.

**Results:** This study included 28,908 participants with an average age of  $65.6 \pm 10.0$  years, of whom 57.6% were female. The overall prevalence of CMDs was 22.6%. After multivariate adjustments, the prevalence ratios (PR) (95% CI) for CMDs across the three groups (6–8h, <6h and >8h) were: reference, 1.140 (1.068–1.218), 1.060 (0.961–1.169) (*P* for trend =0.003), respectively. The subgroup analysis revealed that among older females, a longer sleep duration (>8h) was also associated with an increased prevalence risk of CMDs, with PR 1.169 (1.001–1.365) (*p*=0.049).

**Conclusion:** A shorter sleep duration (<6 hours) was associated with an increased risk of CMDs in the general population, while a longer sleep duration (>8 hours) also raised the prevalence risk among older females.

Keywords: cardiometabolic disease, sleep duration, Southwest China

#### Introduction

Cardiometabolic diseases (CMDs), including stroke, coronary artery disease (CAD), and diabetes mellitus (DM), have significantly contributed to the escalating global burden of disease and disability in recent years.<sup>1</sup> In China, stroke has emerged as the leading cause of both mortality and morbidity, with a pooled annual prevalence rate of 1.33%.<sup>2</sup> Moreover, CAD had a prevalence rate exceeding 0.81% in 2022, playing a prominent role in cardiovascular-related mortality.<sup>3</sup> DM, driven by the surge in obesity rates, stands out as another notable chronic condition projected to affect approximately 45.86% of individuals by 2040.<sup>4</sup>

In recent decades, there has been a decline in individuals' sleep duration attributed to shifts in personal lifestyles, socio-economic factors, work patterns, and global development.<sup>5–7</sup> Existing research has indicated that reduced sleep duration is associated with an increased incidence of stroke, cardiovascular diseases, and DM.<sup>5,8–10</sup>

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Some studies have demonstrated a U-shaped relationship between sleep duration and the occurrence of DM and cardiovascular events, suggesting that both insufficient and excessive sleep can lead to adverse health outcomes.<sup>8,10–12</sup> However, no association between sleep duration and cardiometabolic outcomes in adolescents was also reported.<sup>13,14</sup>

China is currently facing a formidable challenge in terms of managing cardiovascular disease, as the incidence and mortality rates continue to rise annually. Furthermore, since 2012, rural areas have experienced significantly higher mortality rates from cardiovascular disease compared to urban areas.<sup>15</sup>

Despite numerous studies having explored the impact of sleep duration on specific diseases globally, especially in developed country, investigations focusing on the concept of CMDs remain limited, especially among Chinese population and in low-income area of the country. Therefore, this study aims to investigate associations between sleep duration and CMDs among residents in southwest China.

# Material and Methods

#### Study Design

This project is a sub-project of the National Key Research and Development Program (2018YFC1311400) aimed at establishing a solid foundation for the prevention and treatment of chronic cardiovascular diseases. Under the guidance of Chongqing Municipal Health Commission and local Centers for Disease Control, from August 2018 to December 2020, the Cardiovascular Department of the First Affiliated Hospital of Chongqing Medical University conducted a comprehensive cross-sectional investigation by employing phased stratified random sampling on community residents in 16 districts or counties located in southwest China.

The study flow chart was presented in Figure 1, with local community doctors serving as the investigators. The survey primarily consisted of face-to-face questionnaires and a few telephone interviews. Data were entered into the research system by skilled data entry clerks, who conducted stratified sampling checks to ensure the authenticity and reliability of the data.

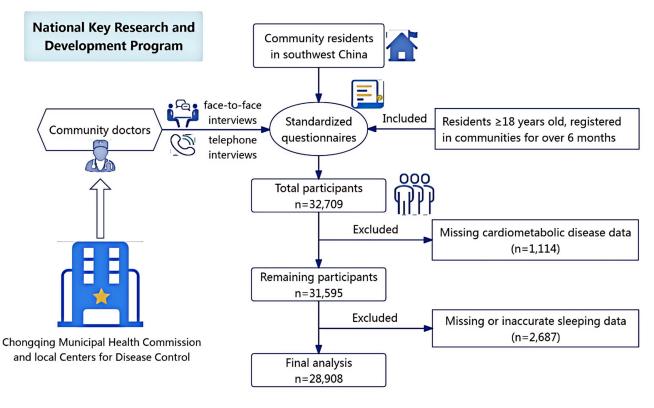


Figure I The study flow chart.

Residents aged 18 years or older who had been registered residents in their respective communities for a minimum of 6 months were enrolled. A total of 32,709 participants were assessed, with 3,801 excluded due to (1) missing or inaccurate sleeping data (n=2687), and (2) missing CMDs data (n=1114). Ultimately, the analysis was conducted on a cohort of 28,908 residents.

#### **Definition of Terms**

The dataset comprised demographic information, family medical history, dietary and lifestyle factors, sleep duration, physical examinations, and serum biochemical tests. Participants were asked to recall their living conditions over the past five years.

Both alcohol consumption and smoking were habitual practices that occurred at least three times per week. The term "well-educated" referred to individuals who had completed at least a high school education or higher. "High income" was defined as an average personal income exceeding 5000 RMB per month. "Physical inactivity" was characterized by engaging in outdoor activities less than 20 hours per week. "Vegetables insufficiency" indicated a weekly intake of vegetables less than three days. "Fruits consumption habit" was defined as a weekly intake of fruits for more than five days. Finally, the term "cardiac-cerebral vascular disease (CCVD) family history" encompassed any documented occurrence of cardiovascular or cerebrovascular diseases in a first-degree relative.

The investigators conducted physical examinations and performed serum biochemical tests. Height and weight measurements were taken with lightweight clothing, while heart rate and blood pressure were recorded as an average from three readings after a five-minute rest. The heart rate was measured using a stethoscope to record the number of heart beats per minute, while the blood pressure was measured using an electronic or mercury sphygmomanometer. Both blood lipid and glucose tests were carried out following a minimum fasting period of seven hours. All measuring instruments and equipment were provided by the community doctors' units.

DM was defined as the presence of at least one of the following criteria:<sup>16</sup> (a) Fasting plasma glucose (FPG) level of  $\geq$ 7.0 mmol/L, (b) 2 hours- post challenge plasma glucose level of  $\geq$ 11.1 mmol/L, (c) pharmacologically treated with glucose-lowering medications.

The definition of CAD included coronary artery stenosis exceeding 50%, which was confirmed by documented medical records through computed tomography angiography (CTA) or percutaneous coronary angiography (CAG), regardless of the presence of ischemic symptoms.<sup>17</sup>

Stroke includes ischemic or hemorrhagic stroke as demonstrated by cranial imaging.<sup>18</sup>

CMDs were defined as any one of DM, CAD or stroke.<sup>1</sup> The three conditions must be supported by clear medical records from a secondary or higher-level medical institutions.

Sleep duration refers to the total amount of time obtained during the nocturnal sleep episode.<sup>19</sup> Participants were categorized into three groups based on their self-reported average daily sleep duration pattern over the past five years: <6 hours, 6-8 hours, and >8 hours. The primary outcomes examined were CMDs and its components.

#### Statistical Analysis

Continuous variables following a normal distribution were expressed as mean  $\pm$  standard deviation (SD). Differences between groups were compared using one-way analysis of variance (ANOVA) if the variances were uniformed. Otherwise, skewed distributions were expressed as the median and quartile ranges (25%-75%), and the Kruskal–Wallis test was utilized. Categorical variables were represented by cases with percentages, and differences between groups were compared using the Pearson chi-square test.

The Poisson regression models were utilized to ascertain the association between sleep duration and CMDs as well as its components. These models were adjusted for disease-related variables, and each prevalence ratio (PR) value along with its 95% confidence interval (CI) was computed. We initially performed univariate regression analysis to identify the factors influencing the outcome variables in the baseline data (p < 0.05), and subsequently included them individually for multivariate regression analysis. Furthermore, considering the close association of CMDs with age and gender, subgroup analyses based on age and sex were conducted to validate the robustness of our findings. The demarcation between the younger and older age was set at 65 years.

The data were analyzed using SPSS version 25.0 (IBM, USA), and GraphPad Prism 8.4.3 was utilized for generating the figures. All statistical tests were two-tailed, and a significance level of p < 0.05 was applied.

#### Results

The present study was a retrospective cross-sectional analysis that included 28,908 participants ranging in age from 20 to 98 years old, with a mean age of  $65.6 \pm 10.0$  years. Among the participants, 57.6% were female. The data presented in Figure 2 indicates that only 29.0% of the general population slept for a duration of 7 hours per day, with proportions of the three groups (6–8h, <6h, >8h) being 74.9%, 18.6%, and 6.5% respectively.

According to Table 1, individuals with a sleep duration of less than 6 hours tended to be older, more likely to have a family history of CCVD, lower income, physical inactivity, and insufficient vegetable intake. They also exhibited higher systolic and diastolic blood pressure levels but had lower weight and body mass index. Additionally, they had slower heart rates and lower levels of triglycerides and fasting glucose.

On the other hand, residents with a sleep duration exceeding 8 hours were slightly more educated, less likely to smoke, and more inclined towards consuming fruits. They also had greater height but lower cholesterol levels.

The overall prevalence rates of CMDs, DM, stroke, and CAD were 22.6%, 18.6%, 1.6%, and 3.7% respectively (displayed in Table 2). Residents with a sleep duration of less than 6 hours had the highest prevalence rate. After multivariate adjustments (Table 3), the PR (95% CI) for CMDs across the three sleep duration groups (6–8h, <6h, and >8h) were: reference, 1.140 (1.068–1.218), 1.060 (0.961–1.169) ( $P_{for trend} = 0.003$ ), respectively.

As displayed in Figure 3, except for the older male subgroup, the association between shorter sleep duration (<6h) and an increased risk of CMDs remained consistent across all subgroup analyses based on age and gender. In the gender subgroup analysis, it was found that among females, particularly older females, a longer sleep duration (>8h) also significantly elevated the prevalence risk of CMDs, with PR 1.169 (1.001–1.365) (p=0.049) (refer to Supplementary Tables 1 and 2)

In analyses of CMD subcomponents, a shorter sleep duration (<6 h) was associated with an increased risk of DM across all subgroups, including women who slept for longer durations (>8 h). Additionally, among women, a shorter sleep duration (<6 h) was also linked to an elevated risk of CAD. On the other hand, a longer sleep duration (>8 h) was associated with an increased risk of hemorrhagic stroke in the elderly population (aged  $\geq$ 65 years old). Conversely, no statistically significant correlation between stroke or CAD and sleep duration was observed in the subgroups of individuals aged younger than 65 years or among males (refer to Supplementary Figure 1).

#### Discussion

This study examined the association between self-reported sleep duration and CMDs among adults in southwest China. Epidemiological data from the sample revealed that 74.9% of the population in southwest China reported sleeping for 6 to 8 hours, while the overall prevalence of CMDs was found to be 22.6%, which closely aligns with the prevalence of 24.2% observed in the UK Biobank population.<sup>1</sup> In comparison to residents who slept for 6–8 hours, those who slept less than 6 hours exhibited an increased risk of CMDs. Additionally, this elevated CMDs risk was also observed specifically in older females who slept over 8 hours.

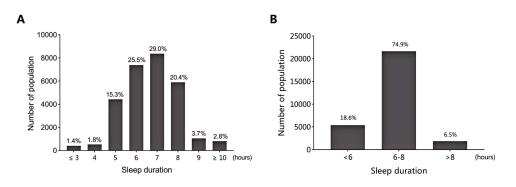


Figure 2 The distribution of sleep duration among the general population (A) and three groups (B).

Characteristics at enrollment	Overall	Slee	P value			
	N=28,908	6–8 h n=21,662	< 6 h n=5375	> 8h n=1871		
Age, yrs	65.6 ± 10.0	64.8 ± 10.2	69.3 ± 7.9	64.6 ± 11.0	< 0.001	
Female gender, n%	16,660 (57.6)	12,585 (58.1)	2994 (55.7)	1081(57.8)	0.006	
Rural residents, n%	8378 (29.0)	5792 (26.7)	2052 (38.2)	534 (39.9)	< 0.001	
Well educated, n%	2773 (9.6)	2330 (10.8)	196 (3.6)	247 (13.2)	< 0.001	
CCVD family history, n%	1874 (6.5)	775 (3.6)	1059 (19.7)	40 (2.1)	< 0.001	
High income, n%	3598 (12.4)	3160 (14.6)	167 (3.1)	271 (14.5)	< 0.001	
Lifestyles						
Alcohol consumption, n%	3704 (12.8)	2804 (12.9)	690 (12.8)	210 (11.2)	0.102	
Smoking, n%	4345 (15.0)	3304 (15.3)	803 (14.9)	238 (12.7)	0.013	
Sleeping duration, hrs	7.0 (6.0-8.0)	7.0 (6.0-8.0)	5.0 (5.0-5.0)	9.0 (9.0-10.0)	< 0.001	
Physical inactivity, n%	3639 (12.6)	2395 (11.1)	1072 (19.9)	172 (9.2)	< 0.001	
Vegetables insufficiency, n%	1451 (5.0)	1103 (5.1)	301 (5.6)	47 (2.5)	< 0.001	
Fruits consumption habit, n%	12,649 (43.8)	10,210 (47.1)	1402 (26.1)	1037 (55.4)	< 0.001	
Physical examinations						
Height, cm	155.4 ± 8.4	155.7 ± 8.4	154.1 ± 8.1	155.9 ± 8.4	< 0.001	
Weight, kg	58.9 ± 10.1	59.3 ± 10.2	56.9 ± 9.5	59.3 ± 10.0	< 0.001	
BMI, kg/m <sup>2</sup>	24.3 ± 3.3	24.4 ± 3.4	23.9 ± 3.3	24.3 ± 3.3	< 0.001	
Heart rate, bpm	73.4 ± 10.8	73.5 ± 10.8	73.0 ± 10.7	74.0 ± 10.5	0.001	
SBP, mmHg	138.9 ± 19.1	138.3 ± 19.0	140.9 ± 18.9	138.9 ± 19.1	< 0.001	
DBP, mmHg	82.0 ± 11.2	81.6 ± 11.2	83.3 ± 11.0	82.8 ± 11.5	< 0.001	
Serum biochemical tests						
TG, mmol/L	1.3 (0.9–1.9)	1.3 (0.9–1.9)	1.2 (0.90-1.8)	1.3 (0.9–2.0)	< 0.001	
TC, mmol/L	5.0 (4.3–5.7)	5.0 (4.3–5.7)	5.1 (4.4–5.9)	4.8 (4.1–5.6)	< 0.001	
HDL-C, mmol/L	1.5 (1.3–1.8)	1.5 (1.3–1.8)	1.6 (1.3–1.8)	1.4 (1.2–1.7)	< 0.001	
LDL-C, mmol/L	2.6 (2.0-3.2)	2.6 (2.0-3.2)	2.7 (2.1–3.1)	2.4 (1.9–3.0)	< 0.001	
Fasting glucose, mmol/L	5.7 ± 1.2	5.8 ± 1.2	5.6 ± 1.3	5.7 ± 1.3	< 0.001	

 Table I Baseline Data at Enrollment Grouped by Sleeping Duration

Abbreviations: CCVD, cardiac-cerebral vascular disease; BMI, body mass index; SBP: systolic blood pressure; DBP, diastolic blood pressure; TG, triglycerides; TC, total cholesterol; HDL-C, high density lipoprotein cholesterol; LDL-C, low density lipoprotein cholesterol.

Table 2 Prevalence of Cardiometabolic Diseases Among Different Groups

Comorbidity	Overall	II Sleeping duration, hrs/day			P value	
	N=28,908	6–8 h n=21,662	< 6 h n=5375	> 8h n=1871		
CMDs, n%	6521 (22.6)	4730 (21.8)	1352 (25.2)	439 (23.5)	< 0.001	
DM, n%	5365 (18.6)	3917 (18.1)	1081 (20.1)	367 (19.6)	0.001	
Stroke, n%	475 (1.6)	320 (1.5)	126 (2.3)	29 (1.5)	< 0.001	
lschemic stroke, n%	408 ((1.4)	276 (1.3)	109 (2.0)	23 (1.2)	< 0.001	
Hemorrhagic stroke, n%	93 (0.3)	63 (0.3)	21 (0.4)	9 (0.5)	0.232	
CAD, n%	1077 (3.7)	763 (3.5)	240 (4.5)	74 (4.0)	0.004	

Abbreviations: CMDs, cardiometabolic diseases; DM, diabetes mellitus; CAD, coronary artery disease.

# Sleep Duration and CMDs

Previous research conducted in various global contexts consistently reveals a U-shaped relationship, whereby both short and long sleep durations are associated with an increased risk of cardiovascular diseases, stroke, and DM.<sup>5,8,10–12</sup> The

Models	6–8h	<6h PR (95% CI)	P value	>8h PR (95% CI)	P value	<b>P</b> for trend
Crude	Reference	1.152 (1.084–1.224)	< 0.001	1.075 (0.974–1.185)	0.150	< 0.001
Model I	Reference	1.103 (1.038–1.173)	0.002	1.077 (0.976–1.187)	0.139	0.004
Model 2	Reference	1.123 (1.056–1.194)	< 0.001	1.073 (0.973–1.183)	0.159	0.002
Model 3	Reference	1.140 (1.068–1.218)	< 0.001	1.060 (0.961–1.169)	0.244	0.003

Table	3	Associations	Between	Sleep	Duration	with	Cardiometabolic	Diseases	After
Adiustr	ner	nts							

**Notes**: The Poisson regression was employed. Crude: univariate adjustment by group of sleeping duration. Model 1: adjusted by crude + age (continuous variable). Model 2: adjusted by model 1 + female gender + height (continuous variable). Model 3: adjusted by model 2 + rural residents + well educated + CCVD family history + high income + smoking + physical inactivity + vegetables insufficiency + fruits consumption habit.

Abbreviations: PR, prevalence ratio; CCVD, cardiac-cerebral vascular disease.

impact of sleep duration on the risk factors and outcomes of these CMDs remains consistent.<sup>5</sup> Furthermore, there exists a mutual influence and reciprocal relationship among CMDs that renders them interrelated risk factors to one another.

The complex interrelation is exemplified by instances such as cardiovascular diseases and DM, collectively constituting risk factors for stroke, as evidenced in existing literature.<sup>20</sup> In the context of cardiovascular diseases, age plays a crucial role as the U-shaped curve becomes more pronounced with advancing age.<sup>10</sup> However, certain studies have not verified the impact of prolonged sleep duration on the risk of CAD and hypertension, which are key factors for CMDs.<sup>6,9</sup>

This research demonstrated that in southwestern China, a sleep duration of less than 6 hours was associated with an increased risk of CMDs across the general population, consistent with findings from other investigations.<sup>5,9,12</sup> Furthermore, it was also observed that a sleep duration exceeding 8 hours was associated with an elevated risk of CMDs in women, as well as an increased susceptibility to hemorrhagic stroke in the elderly population. This finding suggests the possible presence of a U-shaped curve relationship between sleep duration and CMDs.

Interestingly, in the subgroup analysis, no statistically significant correlation between stroke or CAD and sleep duration was observed in the subgroups of young and male individuals. This finding is consistent with some research on cardiometabolic risks in adolescents before.<sup>13,14</sup> However, although sleep duration was not shown to directly affect cardiovascular health when people were young, some risk factors such as sleep quality, life and diet behaviors can be influenced. Research with longer follow-up is needed to explore whether there is future impact.

#### Potential Mechanisms and Intervention

The mechanisms by which sleep duration influences CMDs are intricate and diverse. Short sleep duration is implicated in the alteration of leptin and ghrelin levels, subsequently affecting appetite and energy balance within the human body, leading to obesity, which is a risk factor for CMDs.<sup>5</sup> Conversely, respiratory disorders and inflammatory markers associated with obesity can also impact both the duration and quality of sleep.<sup>5</sup> Furthermore, there is evidence suggesting a causal relationship between stroke and the development of sleep disorders, accompanied by changes in circadian rhythm dynamics.<sup>20</sup> As a result, CMDs and sleep duration mutually influence each other.

In terms of blood pressure regulation, insufficient sleep duration can contribute to hypertension by affecting muscle relaxation and sympathetic tone modulation.<sup>5</sup> Additionally, inadequate sleep duration can lead to abnormal lipid metabolism, glucose intolerance, insulin resistance, and vascular calcification—significant factors contributing to stroke, CAD, and DM.<sup>5</sup> Disease such as obstructive sleep apnea, which has an impact on both sleep duration and quality, was reported as an independent risk factor of CMDs. The underlying mechanism including hypoxia, generation of reactive oxygen species, inflammation, and sympathetic activation.<sup>21–23</sup>

In the intervention and prevention of CMDs and their associated risk factors, evidence suggests that systematically screening and managing sleep disorders can significantly contribute to primary and secondary prevention of CMDs, improve prognosis, and reduce overall mortality rates.<sup>20</sup> Improving sleep duration and quality reduces the threat of CMDs risk factors by enhancing the effectiveness of weight loss interventions and promoting healthy blood pressure

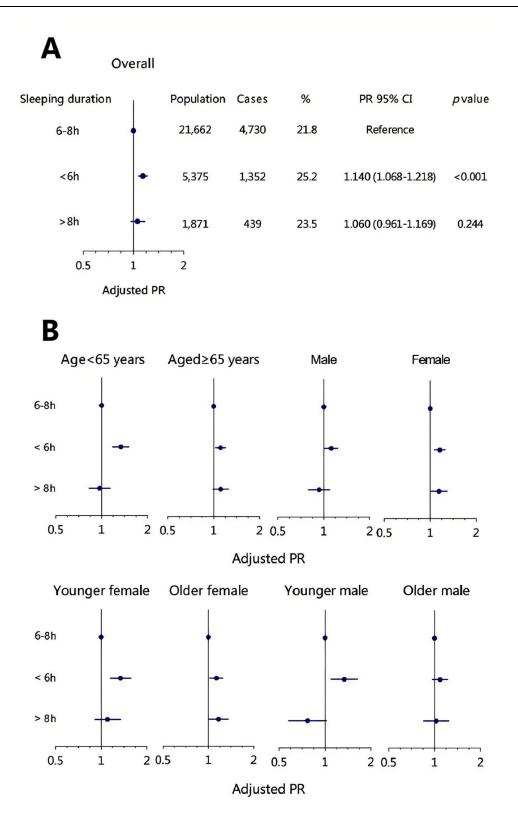


Figure 3 The relationship between sleep duration with CMD among general population (A) and different subgroups (B).

conditions.<sup>24,25</sup> Therefore, strategically addressing sleep conditions and targeting diseases such as sleep apnea through improving daily habits and clinical therapy is crucial for both the general population and patients with CMDs in terms of prevention and treatment.

## Sleep Status Among Chinese Population

The average sleep duration among the Chinese population in 2022 was recorded at 7.4 hours per night.<sup>26</sup> A significant majority of Chinese adults, over 64.8%, reported sleeping less than 8 hours.<sup>27</sup> Findings from a study conducted between 2008 and 2018 revealed an increase in both poor sleep quality and short sleep duration among the elderly population.<sup>28</sup> In 2022, it was demonstrated that approximately 41.3% of older individuals in China slept less than 6 hours per day, with an overall average of 6.5 hours.<sup>29</sup> Research conducted specifically in rural areas of China identified that 52.4% of older people slept less than seven hours.<sup>30</sup>

Sleep duration variation exhibited correlations with various factors including gender, educational attainment, socioeconomic status, familial composition, presence of chronic illnesses, and overall quality of life.<sup>28,30</sup> Furthermore, social issues arising from recent socioeconomic development in China such as higher divorce rates, increase smoking and alcohol abuse, weakened family bonds, and widening socioeconomic disparity between rich and poor also have a notable impact on sleep duration.<sup>7</sup>

The findings of this study revealed a higher prevalence of individuals sleeping for less than 6 hours per night compared to the national average, while a lower prevalence was observed for those sleeping more than 8 hours per night. These results indicated regional disparities in sleep patterns within southwestern China when compared to the national level.

The duration of sleep has been found to be associated with various diseases affecting the neurological, cardiovascular, and endocrine systems.<sup>5,31</sup> Furthermore, decreased sleep duration has been linked to certain risk factors such as obesity, hypertension, and high blood lipid level.<sup>5</sup> The prevalence of obesity in 2018 in China was 51.2% and was predicted to be 70.5% in 2030, while the prevalence of hypertension in China was predicted to increase 7.9% from 2018 to 2030.<sup>32</sup>

In China, the prevalence of CAD in rural area has surpassed urban area since 2016. The mortality rates of cardiovascular and cerebrovascular disease in rural area were higher than in urban region.<sup>15</sup> The observed regional disparities in sleep duration may contribute to variations in disease prevalence across distinct regions, which shows the importance of paying attention to the sleep duration of residents in economically underdeveloped areas.

# Limitation

This study has several limitations. Firstly, it should be noted that this was a cross-sectional retrospective study, meaning that follow-up data was not available. Secondly, the reliance on self-reported sleep duration data introduces a potential source of bias as participants may exhibit variability and subjectivity in reporting their sleep habits.<sup>33</sup> Also, various factors such as illness and social-psychological state may impact sleep duration. Thirdly, the identification of patients with stroke and CAD relied solely on examining existing medical records which inherently limits the accuracy of determining prevalence and incidence rates. Latent or asymptomatic cases may have been overlooked, leading to an underestimation of true prevalence and incidence rates within the study population.

To address these limitations, large-scale prospective studies on a selected population with documented evidence of CMDs and objective measurements of sleep durations can be conducted in the future.

# Conclusion

Our study is among the few that directly investigate the association between sleep duration and CMDs, thereby updating the existing literature on limited Asian data, particularly in economically underdeveloped Southwestern China. Additionally, we perform subgroup analyses based on age and gender to enhance the credibility of our findings.

In conclusion, this study demonstrates that a sleep duration (<6 hours) is associated with an increased risk of CMDs in the general population, while a longer sleep duration (>8 hours) also raises the prevalence risk specifically among older females. Considering the crucial role of sleep duration in lifestyle, it is imperative to implement health education and interventions targeting optimal sleep duration for primary prevention of CMDs.

# **Data Sharing Statement**

The data that support the findings of this study are available from the corresponding author upon reasonable request.

#### **Ethics Approval and Consent to Participate**

This study complies with the Declaration of Helsinki and was approved by the ethics committee of the First Affiliated Hospital of Chongqing Medical University, No. 2020-23. All the participants were informed of the study content and signed informed consent.

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# Disclosure

All authors have no conflicts of interest to declare for this work.

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