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Michelle A. Miller, Ngianga-Bakwin Kandala, Meena Kumari, Michael G. Marmot  
and Francesco P. Cappuccio

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# Relationships Between Sleep Duration and von Willebrand Factor, Factor VII, and Fibrinogen

## Whitehall II Study

Michelle A. Miller, Ngianga-Bakwin Kandala, Meena Kumari,  
Michael G. Marmot, Francesco P. Cappuccio

**Objective**—To examine the relationship between sleep duration and hemostatic factors in a well-characterized cohort.

**Methods and Results**—The relationship between self-reported sleep duration and von Willebrand factor (vWF), fibrinogen, and factor VII was examined in approximately 6400 individuals from the Whitehall II Study. The analysis was stratified by sex (interaction  $P < 0.001$ ). After multiple adjustments, vWF levels were significantly higher in men with both short sleep duration ( $\leq 6$  hours per night; 1.05 [95% CI, 1.01 to 1.08]) and long sleep duration ( $\geq 8$  hours per night; 1.05 [95% CI, 1.02 to 1.08]) compared with those who slept 7 hours ( $P < 0.05$  for both). In women, levels of vWF were significantly higher in individuals who slept 8 hours or longer (1.11 [95% CI, 1.06 to 1.16]) compared with 7 hours ( $P < 0.05$ ). This difference was observed in premenopausal and postmenopausal women. In women, the association was nonlinear ( $P = 0.02$ ), but not in men ( $P = 0.09$ ). No statistically significant associations between sleep duration and fibrinogen or factor VII were observed.

**Conclusion**—Men who slept for short and long durations had higher vWF levels. In women, there was a significant nonlinear association. The highest levels were observed in long sleepers, irrespective of menopausal status. No major associations between sleep and factor VII or fibrinogen were observed. Longitudinal studies are required to investigate causality. (*Arterioscler Thromb Vasc Biol.* 2010;30:00-00.)

**Key Words:** cardiovascular disease prevention ■ epidemiology ■ hemostasis ■ sleep

Studies have demonstrated an association between various health outcomes, including obesity, hypertension, diabetes mellitus, and cardiovascular events (including coronary heart disease [CHD] and stroke) with both sleep quantity and quality.<sup>1–6</sup> A variety of possible pathophysiological mechanisms to support the biological plausibility of an association between sleep deprivation and cardiovascular risk have been proposed.<sup>5,7</sup> Recently, it was demonstrated that in women, but not in men, interleukin (IL) 6 and high-sensitivity C-reactive protein, when adjusted for potential confounders, vary with sleep duration.<sup>8</sup>

Thrombotic factors have also been implicated in the development of cardiovascular disease (CVD). Damage to the lining of the blood vessel walls exposes subendothelium proteins, most notably the glycoprotein, von Willebrand factor (vWF). This leads to the recruitment of factor VIII, collagen, and other clotting factors, including factor VII and factor I (fibrinogen) from the bloodstream; and initiates platelet activation, the clotting cascade, and thrombus formation. Increased circulatory levels of vWF fibrinogen and factor VII have been previously associated with an increased risk of CVD.<sup>9,10</sup> Obstructive sleep apnea is associated with high cardiovascular morbidity and mortality, and a number of activated coagulation factors are

increased in untreated patients with obstructive sleep apnea, potentially contributing to their vascular risk.<sup>11</sup> Shift work has been associated with an increase in indicators of blood coagulation and endothelial injury resulting from the disturbance in the normal circadian rhythm.<sup>12</sup> Furthermore, a recent study<sup>13</sup> demonstrated that polysomnographically verified sleep disruptions are associated with prothrombotic changes. In particular, measures of sleep fragmentation and sleep efficiency were related to vWF in a relatively healthy population. These results suggest that, even in a relatively healthy population, sleep disruptions and disturbances are associated with potential markers of prothrombotic cardiovascular risk.<sup>13</sup>

In the present analysis, we examined the relationship between vWF, factor VII, and fibrinogen and sleep duration in the Whitehall II Study.

## Methods

### Study Population

The Whitehall II cohort was recruited from 1985 to 1988 (phase 1) from 20 London-based civil service departments (participants were aged 35–55 years). The rationale, design, and methods of the study have been described in detail elsewhere.<sup>14</sup> In this report, we use data from phase 3 (1991–1993), when sleep data were available. Sleep

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duration was determined from the following question: "On an average weekday, how many hours do you spend on the following activities: (a) work, (b) time with family, and (c) sleep?" Response categories were from 1 to 12 hours. These categories were collapsed to form categories of 6 hours or less, 7 hours, and 8 hours or more. Short sleep duration ( $\leq 6$  hours) and long sleep duration ( $\geq 8$  hours) were compared with a reference value of 7 hours, which approximates the average sleep duration in a British population.<sup>15</sup> The participation rate of the original cohort ( $n=10\,308$ ) was 83% during phase 3. We examined only white individuals with complete data on sleep and vWF ( $n=6448$ ), of whom 71.2% were men; on sleep and factor VII ( $n=6689$ ), of whom 71.0% were men; and on sleep and fibrinogen ( $n=6674$ ), of whom 70.8% were men (Table 1).

### Covariates

For the present analyses, age and other covariates were derived from the questionnaires during phase 3. Employment grade was determined from the participant's civil service grade title, and the results were divided into 3 categories in order of decreasing salary: administrative, professional/executive, and clerical/support.

Participants were allocated to 1 of 2 smoking categories: never/ex-smoker or current smoker. Alcohol consumption during the previous week was recorded (units per week). General health status was assessed using the physical and mental health component summaries of the Short Form-36 health survey questionnaire<sup>16</sup>: low scores indicate low functioning. At the screening examination, anthropometric measures were recorded, including height, weight, and waist circumference; body mass index (BMI) was calculated as weight in kilograms divided by height in meters squared. Blood pressure was measured 3 times using a standard mercury sphygmomanometer by trained and certified technicians, and the average of the last 2 readings was taken. Further details of these methods have been published previously.<sup>14</sup>

Individuals were asked to fast overnight if their appointment was before 11:30 AM, and those with appointments between 12:30 AM and 14:30 PM were advised that they could have a light fat-free breakfast of unsweetened tea or coffee and plain bread or toast. Participants sat quietly without reading before having their blood pressure recorded; afterward, venipuncture of the left antecubital vein was performed with a tourniquet. Blood was collected into plain, citrate, or fluoride Sarstedt monovettes. After centrifugation, samples were immediately frozen at  $-80^{\circ}\text{C}$  and stored until assay. Glucose was determined in fluoride plasma by an electrochemical glucose oxidase method. Insulin was measured by radioimmunoassay, and IL-6 was measured by a high-sensitivity ELISA.

### Analysis of vWF

vWF was measured by a double-antibody ELISA with reagents provided by Dako Ltd and standards provided by the National Institute for Biological Standards and Control, as previously described.<sup>17</sup> The coefficient of variation was 16.1%. A spline fit was applied to the results to determine the effect of time of sample collection on mean level. There is no major effect of time on these levels over this period. The proportion of the variability because of "time of sampling" for vWF is 0.15%.

### Analysis of Factor VII

Factor VII activity was determined according to the method described by Brozovic et al,<sup>18</sup> performed without automation. A single batch of factor VII reference plasma, stored at  $-70^{\circ}\text{C}$ , was obtained from a pool of healthy blood donors and was calibrated against an NIBSC standard. The within-batch coefficient of variation was 15%. The proportion of the variability because of time of sampling for factor VII is 0.57%.

### Analysis of Fibrinogen

Fibrinogen was assayed using a modification of the clotting method of Claus. However, using a 1:15 dilution of plasma to 0.9% saline, fibrinogen clotted with a half volume of bovine thrombin, 50 U/mL, in an MDA-180 coagulator (OrganonTeknika) using the manufac-

turer's reagents and the international fibrinogen standard.<sup>19</sup> The within-batch coefficient of variation was 8%. The proportion of the variability because of time of sampling for fibrinogen is 0.39%.

### Ethics Committee Approval and Statistical Analysis

Ethics committee approval for the Whitehall II Study was obtained from the University College London Medical School, London, England, committee on the ethics of human research. Subjects provided written informed consent.

In the univariate analysis, for continuous and categorical variables, Kruskal-Wallis and  $\chi^2$  tests, respectively, were used to determine the statistical significance of any difference in the distribution of baseline variables during phase 3 across categories of sleep duration.

Skewed variables (eg, insulin, glucose, vWF, fibrinogen, and factor VII) were natural-log transformed to approach a normal distribution, and results are presented as geometric means and 95% CIs.

The statistical significance between coagulation at phase 3 and categories of sleep duration, adjusted for other baseline variables, was tested in multivariate linear regression models. Results are expressed as geometric means ratios separately for the measured hemostatic factors by category of sleep duration, with 7 hours of sleep as the reference category (geometric means ratio, 1.00) and stratified by sex. All analyses were conducted using computer software (STATA 10.0). A spline regression analysis was performed to investigate the effect of time (using restricted cubic spline) of blood sampling on the mean levels of the individual coagulation factors. The models include a spline time adjustment.  $P < 0.05$  was considered statistically significant.

## Results

### Characteristics of the Population by Categories of Sleep Duration

The interaction between sleep duration and sex on the 3 coagulation factors was significant ( $P < 0.001$ ); therefore, all analyses were stratified by sex. Characteristics for the participants at phase 3 in men and women are reported in Table 1 by categories of sleep duration. Variation across sleep duration groups was significant for a number of the characteristics (Table 1). Men who were short sleepers were more likely to be in the lowest employment grade and unmarried (Table 1). Women who were short sleepers were also more likely to be unmarried (Table 1).

No significant effect of time of blood sampling was observed on the mean level of each of the coagulation factors (supplemental Figure; available online at <http://atvb.ahajournals.org>). In men, there was no significant variation in the unadjusted levels of vWF, fibrinogen, or factor VII across the sleep categories (Table 1). In contrast, in women, there was a significant variation in factor VII (Table 1,  $P = 0.04$ ).

### Multiple Regression Analysis

Each of these hemostatic factors is known to be associated with potential cardiovascular risk factors. Therefore, a correlation matrix between these markers and risk factors, and data from previously published studies, helped to identify confounders that were used in the multiple regression analysis. The level of the hemostatic factors at 7 hours of sleep was used as a reference. Results are shown in Table 2 and Table 3. Age and smoking, which vary significantly with categories of sleep in both men and women (Tables 1), along with BMI and sampling time, were used as covariates in the basic model (model 1) to which other variables were added. Models were

**Table 1. Baseline Characteristics During Phase 3 (1991–1993) in Men and Women Across Categories of Sleep Duration: The Whitehall II Study\***

Characteristics	Sleep Duration, h			P Value†
	≤6	7	≥8	
<b>Men (n=5312)</b>				
No. of subjects	1279	2525	1508	NA
Age, y‡	48.7 (5.7)	49.0 (6.0)	49.7 (6.2)	0.001
Not married§	290 (22.7)	409 (16.2)	243 (16.1)	<0.001
Lowest employment¶	112 (8.8)	94 (3.7)	72 (4.8)	<0.001
BMI	25.6 (3.4)	24.9 (3.0)	25.0 (3.2)	<0.001
Waist circumference, cm	88.6 (9.9)	86.9 (9.0)	87.4 (9.4)	<0.001
Weekly alcohol consumption, U	13.1 (14.7)	12.7 (113.7)	12.5 (14.1)	0.30
Current smoking	220 (17.2)	314 (12.4)	189 (12.5)	0.001
SF-36 score				
Mental	50.2 (9.2)	51.6 (8.0)	51.7 (8.2)	<0.001
Physical	53.0 (6.5)	53.5 (5.5)	52.7 (6.6)	0.007
Blood pressure, mm Hg				
Diastolic	81.1 (9.3)	80.5 (9.0)	81.6 (9.2)	0.004
Systolic	122.1 (13.6)	121.4 (12.7)	122.5 (13.4)	0.08
Fasting glucose, mmol/L	5.3 (5.2–5.3)	5.3 (5.2–5.3)	5.3 (5.2–5.3)	0.81
Fasting insulin, mmol/L	5.6 (5.4–5.8)	5.3 (5.2–5.5)	5.3 (5.1–5.6)	0.10
Factor VII activity, % standard	86.2 (84.9–87.6)	85.9 (85.0–86.9)	84.9 (83.6–86.2)	0.31
Fibrinogen, g/L	2.29 (2.26–2.32)	2.26 (2.24–2.28)	2.29 (2.26–2.32)	0.14
vWF, IU/dL	99.9 (97.8–102.1)	98.3 (96.8–99.8)	101.0 (99.5–103.1)	0.09
<b>Women (n=2205)</b>				
No. of subjects	598	962	645	NA
Age, y‡	50.9 (6.2)	50.1 (6.2)	50.2 (6.4)	0.02
Not married§	280 (46.8)	353 (36.7)	198 (30.7)	<0.001
Lowest employment¶	225 (37.6)	318 (33.1)	209 (32.4)	0.19
BMI	25.7 (4.8)	25.2 (4.5)	25.4 (4.4)	0.14
Waist circumference, cm	75.0 (11.7)	74.5 (11.4)	75.0 (11.3)	0.54
Weekly alcohol consumption, U	5.7 (8.3)	6.0 (7.4)	5.8 (7.3)	0.60
Current smoking	153 (25.6)	202 (21.0)	123 (19.1)	0.04
SF-36 score				
Mental	49.1 (9.8)	50.5 (9.2)	50.4 (9.4)	0.006
Physical	50.1 (8.6)	50.9 (8.3)	50.0 (9.07)	0.09
Blood pressure, mm Hg				
Diastolic	76.5 (9.0)	76.2 (9.2)	76.3 (8.8)	0.69
Systolic	118.0 (13.9)	116.6 (13.9)	117.4 (13.4)	0.16
Fasting glucose, mmol/L	5.1 (5.0–5.1)	5.0 (5.0–5.1)	5.1 (5.0–5.1)	0.80
Fasting insulin, mmol/L	4.8 (4.5–5.1)	5.0 (4.8–5.2)	5.1 (4.8–5.4)	0.32
Factor VII activity, % standard	92.2 (90.1–94.3)	90.6 (88.9–92.4)	89.0 (86.9–91.2)	0.04
Fibrinogen, g/L	2.52 (2.47–2.57)	2.50 (2.46–2.54)	2.50 (2.45–2.55)	0.92
vWF, IU/dL	101.4 (98.5–104.4)	98.6 (96.3–101.0)	103.2 (99.9–106.5)	0.09

NA indicates data not applicable; SF-36, Short-Form 36 Health Survey.

\*Data are given as mean (SD) unless otherwise indicated.

†For comparison across sleep duration groups using  $\chi^2$  analysis for categorical variables and the Kruskal-Wallis test for continuous variables.

‡Ranges from 39 to 63 years (median, 48 years).

§Data are given as number (percentage) of each group.

¶Clerical/support.

||Data are given as geometric mean (95% CI).

**Table 2. Cross-Sectional Relationships Between vWF and Duration of Sleep at Phase 3 (1991–1993): The Whitehall II Study\***

Model†	Employment					Marital Status				
	Coefficient (95% CI) ≤6 h	Reference 7 h	Coefficient (95% CI) ≥8 h	P Value		Coefficient (95% CI) ≤6 h	Reference 7 h	Coefficient (95% CI) ≥8 h	P Value	
				Linear Trend	Nonlinear Trend				Linear Trend	Nonlinear Trend
<b>Men</b>										
1	1.03 (0.99–1.07)	1.00	1.10 (1.06–1.14)‡	0.48	0.18	NA	NA	NA	NA	NA
2	1.01 (0.91–1.06)	1.00	1.10 (1.06–1.14)‡	0.37	0.20	1.02 (0.98–1.07)	1.00	1.10 (1.06–1.14)‡	0.43	0.23
3	1.02 (0.98–1.05)	1.00	1.04 (1.01–1.08)‡	0.40	0.19	1.02 (0.99–1.06)	1.00	1.04 (1.01–1.08)‡	0.45	0.22
4	1.03 (1.00–1.06)‡	1.00	1.05 (1.02–1.08)	0.33	0.16	1.03 (1.00–1.07)‡	1.00	1.05 (1.02–1.08)‡	0.38	0.19
5	1.05 (1.01–1.08)‡	1.00	1.05 (1.02–1.08)‡	0.55	0.09	1.05 (1.02–1.08)‡	1.00	1.05 (1.02–1.08)‡	0.63	0.10
<b>Women</b>										
1	1.12 (1.05–1.20)‡	1.00	1.18 (1.10–1.26)‡	0.08	0.05	NA	NA	NA	NA	NA
2	1.10 (1.03–1.18)‡	1.00	1.17 (1.09–1.25)‡	0.07	0.05	1.10 (1.03–1.18)‡	1.00	1.19 (1.11–1.27)‡	0.03	0.03
3	1.03 (0.98–1.09)	1.00	1.12 (1.06–1.18)‡	0.07	0.05	1.02 (0.97–1.08)	1.00	1.12 (1.06–1.19)‡	0.03	0.03
4	1.03 (0.99–1.08)	1.00	1.09 (1.04–1.14)‡	0.07	0.05	1.03 (0.98–1.07)	1.00	1.10 (1.05–1.15)‡	0.04	0.03
5	1.04 (0.99–1.09)	1.00	1.11 (1.06–1.16)‡	0.06	0.02	1.03 (0.98–1.08)	1.00	1.11 (1.05–1.17)‡	0.03	0.01

NA indicates not applicable.

\*The duration of sleep was measured in hours. Data are given as the geometric mean ratio for vWF by category of sleep duration (with 7 hours as the reference). For men, n=1126 for those with 6 or fewer hours of sleep, n=2184 for those with 7 hours of sleep, and n=1280 for those with 8 or more hours of sleep. For women, n=508 for those with 6 or fewer hours of sleep, n=831 for those with 7 hours of sleep, and n=519 for those with 8 or more hours of sleep.

†Model 1 was adjusted for age, BMI, smoking, and time sample was taken; model 2, adjusted for age, employment/marital status, BMI, smoking, and time sample was taken; model 3, adjusted for age, employment/marital status, BMI, smoking, alcohol, and time sample was taken; model 4, adjusted for age, employment/marital status, BMI, smoking, alcohol, systolic blood pressure, physical score, mental score, glucose level, and time sample was taken; and model 5, adjusted for age, employment/marital status, BMI, smoking, alcohol, systolic blood pressure, physical score, mental score, insulin, and time sample was taken.

‡P=0.05 for the contrast between sleep categories and the reference (7 hours).

also run with adjustment for either employment status, which is a known determinant of vWF; or for marital status, which shows significant variation across categories of sleep in both men and women (Tables 1).

**von Willebrand Factor**

In men, linear and nonlinear trends, when adjusted for age, BMI, smoking, and sampling time, were not significant (P=0.48 and P=0.18, respectively) (Table 2). In the fully

**Table 3. Cross-Sectional Relationships Between vWF, Factor VII, and Fibrinogen and Duration of Sleep at Phase 3 (1991–1993): Fully Adjusted Model: The Whitehall II Study\***

Model 5†	Employment					Marital Status				
	Coefficient (95% CI) ≤6 h	Reference 7 h	Coefficient (95% CI) ≥8 h	P Value		Coefficient (95% CI) ≤6 h	Reference 7 h	Coefficient (95% CI) ≥8 h	P Value	
				Linear Trend	Nonlinear Trend				Linear Trend	Nonlinear Trend
<b>Men</b>										
vWF	1.05 (1.01–1.08)‡	1.00	1.05 (1.02–1.08)‡	0.55	0.09	1.05 (1.02–1.08)‡	1.00	1.05 (1.02–1.08)‡	0.63	0.10
Factor VII	1.02 (1.00–1.05)‡	1.00	1.01 (0.98–1.03)	0.35	0.15	1.02 (1.00–1.05)‡	1.00	1.01 (0.98–1.03)	0.32	0.14
Fibrinogen	1.00 (0.99–1.02)	1.00	1.00 (0.99–1.02)	0.50	0.78	1.00 (0.99–1.02)	1.00	1.01 (0.99–1.02)	0.52	0.79
<b>Women</b>										
vWF	1.04 (0.99–1.09)	1.00	1.11 (1.06–1.16)‡	0.06	0.02	1.03 (0.98–1.08)	1.00	1.11 (1.05–1.17)‡	0.03	0.01
Factor VII	1.02 (0.97–1.06)	1.00	1.02 (0.98–1.07)	0.42	0.46	1.01 (0.97–1.05)	1.00	1.03 (0.99–1.07)	0.53	0.50
Fibrinogen	1.00 (0.98–1.03)	1.00	1.00 (0.98–1.03)	0.98	0.99	1.00 (0.97–1.02)	1.00	1.01 (0.98–1.03)	0.72	0.92

\*The duration of sleep was measured in hours. Data are given as the geometric mean ratio for vWF by category of sleep duration (with 7 hours as the reference). For men, the number by duration of sleep and analyte was as follows: vWF, 6 or fewer, n=1126; 7, n=2184; and 8 or more, n=1280; factor VII, 6 or fewer, n=1157; 7, n=2268; and 8 or more, n=1324; and fibrinogen, 6 or fewer, n=1151; 7, n=2248; and 8 or more, n=1324. For women, the number by duration of sleep and analyte was as follows: vWF, 6 or fewer, n=508; 7, n=831; and 8 or more, n=519; factor VII, 6 or fewer, n=536; 7, n=848; and 8 or more, n=556; and fibrinogen, 6 or fewer, n=538; 7, n=863; and 8 or more, n=550.

†Adjusted for age, employment/marital status, BMI, smoking, alcohol, systolic blood pressure, physical score, mental score, insulin, and time sample was taken. ‡P=0.05 for the contrast between sleep categories and the reference (7 hours).

adjusted model (model 5) with adjustment for employment grade, linear ( $P=0.55$ ) and nonlinear ( $P=0.09$ ) trends were not significant. However, when compared with 7 hours, vWF was significantly increased in both short and long sleepers ( $P<0.05$ ) (Table 2). This difference was maintained when further adjustment for serum IL-6 was made for short and long sleepers (1.04 [95% CI, 1.00 to 1.07] and 1.05 [95% CI, 1.02 to 1.08], respectively;  $P<0.05$ ) compared with 7 hours.

In women, although the linear trend was not significant ( $P=0.08$ ), there was a borderline significant nonlinear relationship with sleep duration and vWF when adjusted for age, BMI, smoking, and sampling time ( $P=0.05$ ). This remained borderline significant when adjusting for either employment grade ( $P=0.05$ ) and significant when adjusting for marital status ( $P=0.03$ ). The observed relationship with sleep was maintained after further adjustments for alcohol intake, blood pressure, physical and mental scores, and glucose level ( $P=0.05$ ) or with the previous adjustment and insulin instead of glucose ( $P=0.02$ ). Similarly, the relationship was maintained after multiple adjustment if marital, instead of employment, status was used ( $P=0.01$ ). However, in this case, a significant linear association was also observed ( $P=0.01$ ) (Table 2). In women, as in men, there was a consistently higher level of vWF in those individuals who slept 8 hours per night or more compared with those who slept 7 hours ( $P<0.05$ ). After adjustment for serum IL-6, there was still a significant nonlinear relationship ( $P<0.02$ ); the values in long sleepers were significantly higher than in those who slept 7 hours (1.11 [95% CI, 1.05 to 1.07];  $P<0.05$  for model 5 [employment] with IL-6 adjustment). Similar results were observed when substituting marital status for employment status ( $P<0.02$  for model 5 only and  $P<0.02$  for model 5 with IL-6 adjustment).

### Factor VII and Fibrinogen

In men, both the linear and nonlinear trends, when adjusted for age, BMI, smoking, and sampling time, were not significant ( $P=0.43$  and  $P=0.31$ , respectively). Likewise, no significant effect was observed in women ( $P=0.16$  and  $P=0.28$ , respectively). These associations were unchanged after multiple adjustments, and no significant associations were observed when adjustment was made for marital status instead of employment grade (Table 3). A small, but significant, increase (2%) in factor VII in the male short sleepers was observed in the fully adjusted model (Table 3) and was maintained when adjusted for IL-6 (1.02 [95% CI, 1.00 to 1.05];  $P<0.05$ ).

In men and women, there were no significant linear or nonlinear trends observed with any of the models; and no difference in levels was observed in short or long sleepers compared with those sleeping 7 hours, after making multiple adjustments (Table 3).

### Premenopausal and Postmenopausal Women

The levels of each of the hemostatic markers was significantly higher in postmenopausal women compared with premenopausal women (vWF: 105.6 [95% CI, 103.2 to 108.1] IU/dL versus 95.5 [95% CI, 93.4 to 97.6] IU/dL; factor VII: 97.6 [95% CI, 96.1 to 99.2] percentage standard versus

83.6 [95% CI, 82.0 to 85.2] percentage standard; fibrinogen: 2.62 [95% CI, 2.59 to 2.66] g/L versus 2.38 [95% CI, 2.35 to 2.42] g/L;  $P<0.001$  for each).

With the fully adjusted employment model, the increased level of vWF was observed in both long sleeping premenopausal women (1.10 [95% CI, 1.03 to 1.18]) and postmenopausal women (1.08 [95% CI, 1.01 to 1.17]) ( $P<0.05$  versus 7 hours). In premenopausal, but not postmenopausal, women (as for the men), a significant increase was also observed in short sleeping women (1.07 [95% CI, 1.00 to 1.15];  $P<0.05$  versus 7 hours). The nonlinear trend was significant in premenopausal, but not postmenopausal, women ( $P=0.04$  and  $P=0.15$ , respectively).

For factor VII and fibrinogen, as for the total group of women, in the adjusted models, there were no observed associations between these factors in sleep in either premenopausal or postmenopausal women (data not shown).

## Discussion

Our findings from more than 6400 British white-collar civil servants suggest that the associations of sleep duration with different hemostatic factors are, in part, sex dependent and vary according to the marker studied.

In the simple model, there were no linear or nonlinear trends observed between sleep and the measured markers in men; for each of the markers, the levels were significantly higher in long sleepers compared with those sleeping 7 hours. However, in women, there was a significant nonlinear association between sleep and vWF but not between sleep and factor VII or fibrinogen. The levels of vWF and factor VII were also significantly increased in the short and long sleepers compared with those women who slept 7 hours. After multiple adjustments for potential confounding factors, the significant nonlinear association between sleep and vWF in women was maintained. However, although the level in short sleepers was no longer significantly higher than in the reference group (7 hours), the level in long sleepers was significantly higher; this difference was observed in both premenopausal and postmenopausal women and after adjustment for IL-6. With adjustment for marital status instead of employment grade, both linear and nonlinear trends were significant. No significant linear or nonlinear relationships with sleep were observed with the other hemostatic factors in these fully adjusted models.

To our knowledge, this is the first large-scale study to describe the associations between different hemostatic measures and sleep duration in both men and women. Although our findings are restricted to a white population, the study has the benefit that the population has been extensively characterized and, thus, allows for the adjustment of well-known confounders, including measures of obesity, glucose metabolism, blood pressure, and measures of social position.

Hemostatic markers are associated with CVD and are important in fibrinolysis and clotting. Data from the Whitehall II and other studies have indicated a role for these factors in the underlying association between social position and CHD. There is a grade gradient in vWF, and the levels of vWF are associated with other confounding factors, including alcohol intake and fasting glucose level.<sup>17</sup> Data from this and

other studies have also indicated that individuals and in particular men who were short sleepers were more<sup>6</sup> likely to be in the lowest employment grades and unmarried. To investigate the association between sleep and hemostatic factors, we adjusted for either employment grade or marital status (the latter showing a greater variation with sleep) and other potential confounding factors. However, we did not find any major effect of employment grade on the observed sleep-related effects (Table 2, models 1 and 2).

Previously, long sleep has been associated with an increase in carotid intima-media thickness<sup>20</sup> and an increase in all-cause mortality,<sup>3</sup> and short sleep has been associated with an increase in CVD.<sup>3</sup> A recent meta-analysis of prospective studies has indicated that, although short sleep may be associated with CHD and stroke, long sleep is associated with CHD, stroke, and total CVD.<sup>21</sup> Previously, we demonstrated that sleep duration is associated with changes in markers of inflammation<sup>8</sup> and that it is possible that sleep may have its effect on hemostatic and inflammatory pathways by activation of the innate immune system, nuclear factor  $\kappa$ B, and toll-like receptor pathways.<sup>7</sup> The results from this study indicate that long sleep is associated with an increase in vWF but not fibrinogen or factor VII. This may indicate the presence of endothelial damage in individuals with a long sleep duration, which has led to increased exposure of vWF to the bloodstream. In turn, this would normally lead to the initiation of the clotting cascade and recruitment of clotting factors, including factor VII and factor I (fibrinogen) from the bloodstream. Only vWF levels are increased in these individuals, which may indicate that vWF is an early marker of disease. Adjustment for IL-6, which upregulates vWF, had no effect on the results. Because this analysis is cross-sectional, it cannot demonstrate causality. Further longitudinal studies and analysis of prospective data are required to establish if there is a temporal association between sleep duration and level of vWF.

The present study has several limitations. (1) Only self-reported measures of sleep duration were used, which did not explicitly ask participants to differentiate between time asleep and time in bed. It is not usually feasible to obtain more detailed assessment of sleep duration in such large epidemiological studies. An association between self-reported duration of sleep and diary, actigraphy, or polysomnography estimates has been previously reported.<sup>22–24</sup> (2) Our samples were taken at only 1 point but were adjusted for sampling time. Moreover, no major difference was observed between time-adjusted and time-unadjusted modeling results (data not shown). (3) The possibility that the individuals with a long sleep duration may have some as yet undiagnosed underlying disease pathology cannot be excluded. However, our findings are still consistent with the idea that sleeping approximately 7 hours per night appears to be optimal for health; prospective studies in large epidemiological cohorts are still required to further investigate these underlying mechanisms. (4) Only fibrinogen, factor VII, and vWF were measured when this phase of the study was conducted. The measurement of further factors, such as plasminogen activator inhibitor-1, thrombomodulin, tissue factor pathway inhibitor, or sE-selectin, may enable clarification of the underlying mecha-

nism and determine, for example, if there is generalized endothelial activation.

This study adds to the growing evidence that suggests that there is a nonlinear relationship between cardiovascular risk factors and duration of sleep<sup>25</sup> and that the association between sleep duration and cardiovascular risk or risk factors may be different in men and women.<sup>8,26</sup> Variations in duration of sleep are associated with vWF levels, providing a potential contributing mechanism to the high CVD risk seen in short and long sleepers.

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