12:00 in our patients with COPD are also compatible with aggregated data from healthy subjects and those with asthma, which indicated a peak value around 4:00 and a trough value between 10:00 and 16:00 (7, 8). The high proportion (50%) of patients with COPD crossing GOLD thresholds during the day can be related to previous COPD studies of BEC stability over much longer time periods (4, 9), bearing in mind that crossing thresholds is a relatively crude measure. When examining the same three categories of less than 100, 100–299, and 300 or more eosinophils/μl (as we have done here), only 31% of the patients with COPD were found to cross into another category over 1 year (9). This could suggest that BEC data in Long and colleagues (9) had probably been gathered at the same time of day to avoid the within-day BEC variability we observed here. In any case, future long-term follow-up studies with BEC as a biomarker would surely benefit from same time of day measurement, owing to the diurnal BEC variations now demonstrated in patients with COPD. Such studies should also be standardized for potential technical sources of variability of BEC measurement as outlined by Chipps and colleagues (10).

A study limitation is sample size, as is usually the case in studies requiring the patients to remain available for repeated blood sampling. However, observed patterns were very consistent even in this relatively small group, and despite being obtained in the real-life setting of the outpatient clinic.

We demonstrated significant within-day differences in blood eosinophils in stable patients with COPD with a consistent underlying pattern. Ideally, BEC could be improved as a theragnostic biomarker by reaching a consensus on the time of day of blood sampling. This may not be workable in the reality of the clinic, in which case at least the same time of day should be considered for the follow-up of any given patient. ■

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Obstructive Sleep Apnea Symptom Subtypes and Cardiovascular Risk: Conflicting Evidence to an Important Question

To the Editor:

The recent paper by Trzepizur and colleagues (1) extends evidence that obstructive sleep apnea (OSA) is heterogeneous. The authors studied a clinical cohort of 5,358 patients with OSA and no evidence of overt cardiovascular disease (one-third had hypertension) from seven sleep centers. They found similar symptom subtypes as discovered in the Icelandic Sleep Apnea Cohort and replicated in population-based and clinical cohorts (1–4).

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The investigators did not replicate the increased cardiovascular risk in the excessively sleepy subtype. This was originally shown in the Sleep Heart Health Study (2) and recently replicated in a clinical cohort from Chile (3). In contrast to Trzepizur and colleagues (1), Labarca and colleagues (3) found increased cardiovascular mortality in clinical patients with the excessively sleepy subtype. Studies before the recognition of symptom subtypes also showed increased cardiovascular risk with OSA and excessive sleepiness (5).

These recent articles address an important and challenging question. Studying the impact of symptoms on cardiovascular events in clinical populations carries two major challenges. First, there is potential referral bias. As the authors acknowledge (1), individuals undergoing a sleep study with minimal symptoms may have higher baseline cardiovascular risk. Accounting for this requires careful control for confounders. Although the investigators (1) controlled for important covariates, many unmeasured confounders remain (e.g., lipid levels, use of statins and other medications, diet, exercise, etc.).

A second challenge is accounting for continuous positive airway pressure (CPAP) treatment. Studies demonstrating a relationship between symptom subtypes and cardiovascular outcomes reported CPAP use in a small percentage of participants (2, 3). Trzepizur and colleagues (1) report more than 40% using CPAP, including a higher percentage in subtypes endorsing sleepiness. If CPAP reduces cardiovascular risk, as suggested by epidemiological but not recent randomized studies (5), then the effect of OSA would be stronger in untreated patients. To evaluate whether symptom subtypes independently predict cardiovascular risk, it is important to consider patients not using CPAP.

Two additional points mentioned by the authors (1) warrant further emphasis. First, there are discrepancies in disease severity across samples. Studies showing increased cardiovascular risk among the excessively sleepy subtype (2, 3) included only patients with moderate to severe OSA (apnea–hypopnea index $\geq$ 15 events/h), whereas Trzepizur and colleagues (1) include patients with an apnea–hypopnea index of 5 or more. There were also different hypopnea definitions used by the Sleep Heart Health Study ($\geq$4% desaturation) (2) and Trzepizur and colleagues ($\geq$3% desaturation or arousal) (1). Thus, the sample studied by Trzepizur and colleagues (1) is likely much less hypoxemic. Notably, studies show greater cardiovascular risk in moderate to severe OSA (5), and the investigators confirmed increased cardiovascular risk with higher hypoxic burden (1). Whether the excessively sleepy subtype is particularly important in those with higher hypoxic burden warrants investigation.

Second, by using only seven symptoms to derive clusters, including the reliance on one measure of "excessive daytime sleepiness" (defined as an Epworth Sleepiness Scale $>$ 10) and absence of symptoms like drowsy driving, falling asleep involuntarily, feeling sleepy, or frequent napping, Trzepizur and colleagues (1) may have less accurately identified the excessively sleepy subtype, limiting their ability to observe an effect. A very recent analysis demonstrates optimal prediction of the excessively sleepy subtype with two to five sleepiness-related symptoms (6).

Overall, Trzepizur and colleagues (1) address a very important question and add new evidence. However, more questions arise than answers. Larger studies with more comprehensive control for covariates and more attention to CPAP treatment as a confounder are needed. Including patients with mild OSA and limited sleepiness-related symptoms may contribute to their result. We assert that claiming "subtypes based on OSA symptoms have no independent predictive value in clinical-based samples" (1) is premature and not supported by the overall evidence. ■

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Reply to Keenan *et al.*

From the Authors:

We appreciate the correspondence of Dr. Keenan and colleagues about our recently published study in the *Journal* addressing the independent association of sleep apnea specific hypoxic burden and symptom subtypes with the risk of major adverse cardiac event (MACE) (a composite outcome including all-cause mortality, acute myocardial infarction, stroke, and unplanned coronary revascularization) (1).

Contrary to recent data from the Sleep Heart Health Study (2), we found no independent association between obstructive sleep apnea (OSA) symptom subtypes and MACE after adjustment for confounders. In particular, patients with OSA from the excessively sleepy subtype were not at higher risk of MACE. A number of studies have reported that excessive daytime sleepiness (EDS) might represent *per se* an independent cardiovascular (CV) risk factor. However, most of those studies relied on population-based cohorts or on highly selected populations including older patients (3) and patients with a past history of hypertension (4) or myocardial infarction (5). Furthermore, recent data from the Sleep Heart Health Study cohort reported limited independent impact of EDS and no combined effect of EDS and OSA on incident CV events (6).

We agree with Keenan and colleagues that evaluating the relevance of symptom subtypes for early identification of patients with OSA at high CV risk in clinical practice is challenging owing to unmeasured confounders and potential referral bias. Within a monocentric clinic-based cohort study in Santiago, Labarca and colleagues (7) demonstrated an increased CV mortality in patients with OSA belonging to the excessively sleepy subtype. However, compared with our study ($N = 5,358$), the sample size was much smaller ($N = 780$), the number of events was limited, and most importantly, the prevalent metabolic and CV comorbidities including coronary heart disease were more common in the excessively sleepy group in the Labarca study. Therefore, a contribution of prevalent metabolic and CV diseases to the association of EDS with CV mortality cannot be formally excluded.

As pointed out by Keenan and colleagues, the risk of bias related to unmeasured confounders is a major issue in observational cohort studies. Although we controlled for important covariates, the presence of potential unmeasured confounding factors such as exercise and diet cannot be excluded. As shown in Table 1, further adjustment for the use of cardioprotective medications including statins, antihypertensive drugs, and antiplatelet agents did not alter the magnitude of the association between symptom subtypes and MACE.

As expected within a clinic-based cohort, a large proportion of our patients were successfully treated by continuous positive airway pressure, which might have reduced CV risk. However, as shown in Table 1, we found no significant association between symptom subtypes and MACE when the analysis was restricted to untreated patients ($n = 3,156$). Furthermore, as mentioned by Keenan and colleagues, previous data showing increased CV risk among the excessively sleepy subtype included only patients with moderate to severe OSA (2, 7) and used the 4% desaturation hypopnea definition (2) rather than the American Academy of Sleep Medicine 2012

Table 1. Cox Proportional Hazard Model Assessing the Association of Incident Cardiovascular Events and All-Cause Mortality with Symptom Subtypes in the Whole Population and in Specific Subgroups

	Fully Adjusted Model [*HR (95% CI)*]*	*P* Value
All patients ($N = 5,358$)		
Minimally symptomatic (ref)	—	—
Disturbed sleep	1.12 (0.74–1.69)	0.5997
Excessively sleepy	1.00 (0.78–1.28)	0.9985
Moderately sleepy	0.99 (0.78–1.27)	0.9645
Moderate to severe obstructive sleep apnea ($n = 3,819$)		
Minimally symptomatic (ref)	—	—
Disturbed sleep	1.07 (0.65–1.77)	0.7826
Excessively sleepy	1.02 (0.77–1.36)	0.8887
Moderately sleepy	1.02 (0.77–1.34)	0.9014
Nontreated obstructive sleep apnea ($n = 3,156$)		
Minimally symptomatic (ref)	—	—
Disturbed sleep	0.87 (0.49–1.54)	0.6357
Excessively sleepy	0.85 (0.61–1.19)	0.3409
Moderately sleepy	1.03 (0.75–1.42)	0.8392

Definition of abbreviations: CI = confidence interval; HR = hazard ratio; ref = reference.
*Adjusted for age, gender, body mass index, smoking status, presence of prevalent disease (diabetes, chronic obstructive pulmonary disease, and hypertension), type of sleep study, study site, β-blocker, statins, antihypertensive drugs, and antiplatelet agent medications.

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