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BMJ Open

Sleep and circadian biomarkers of postoperative delirium (SLEEP-POD): protocol for a prospective, observational cohort study

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Title: Sleep and circadian biomarkers of postoperative delirium (SLEEP-POD): protocol for a prospective, observational cohort study

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ABSTRACT

Introduction: Surgical patients over 70 experience postoperative delirium (POD) complications in up to 50% of procedures. Sleep/circadian disruption has emerged as a potential risk factor for POD in epidemiological studies. This protocol presents a single-site, prospective observational study designed to examine the relationship between sleep/circadian regulation and POD and how this association is moderated or mediated by AD pathology and genetic risk for AD.

Methods and Analysis: Study staff members will screen for eligible patients (age≥70) seeking joint replacement or spinal surgery at MGH. At the inclusion visit, patients will be asked a series of questionnaires related to sleep and cognition, conduct a 4-lead ECG recording, and be fitted for an actigraphy watch to wear for seven days before surgery. Blood samples will be collected pre- and postoperatively and will be used to gather information about AD variant genes (*APOE-ε4*) and AD-related pathology (total and phosphorylated *tau*). CAM-S and MoCA will be completed twice daily for three days after surgery. Seven-day actigraphy assessments and PROMIS questionnaires will be performed one, three, and twelve months after surgery. Relevant patient clinical data will be monitored and recorded throughout the study.

Ethics and Dissemination: This study is approved by the IRB at MGH, Boston, and it is registered with the US National Institutes of Health on ClinicalTrials.gov (NCT06052397). Plans for dissemination include conference presentations at a variety of scientific institutions. Results from this study are intended to be published in peer-reviewed journals. Relevant updates will be made available on ClinicalTrials.gov.

Strengths and Limitations:

- Limitations of the study include its observational nature and, therefore, the inability to demonstrate causation.
- Treatment effects after elective joint surgery may affect the association between sleep/circadian rhythms and delirium.
- Repeat follow-up 12 months after surgery may increase the likelihood of missing data.
- Despite these pitfalls, older orthopedic patients have similar characteristics, which mitigates variability within this study.
- They also have a high co-morbid burden of sleep disturbances, which can enrich the signal for POD and cognitive decline.

INTRODUCTION

Postoperative delirium (POD) is one of the most common complications following surgery for older adults, occurring in 5% to 50% of cases.[1] POD is a neurocognitive syndrome with acute attention, cognitive, and awareness deficits that develops over a short period of time and has a fluctuating course. It is associated with cognitive decline and Alzheimer's disease (AD) and related dementias (ADRD) but can present in otherwise cognitively intact older adults. Millions of older Americans (≥ 70 years) require major surgery each year and are exposed to the risks of POD and its consequences.[2] Furthermore, there are currently no treatments for POD, but it perpetuates a cascade of poor health outcomes costing approximately \$50 billion annually.[3] Older age, male gender, multiple co-morbidities, sensory impairments, sleep deprivation, use of sedative medications, and baseline cognitive impairment (like pre-existing AD/ADRD) are all major risk factors. POD may also predispose to AD/ADRD, suggesting overlapping pathophysiology.[2,4–6] Yet, minimal attention or cognitive follow-up exists for those who suffer from POD.[7] Given that nearly 40% of POD is preventable with attention to multifactorial preoperative care,[8] the search for modifiable risk factors and novel biomarkers in POD is of great public health importance.

At the same time, there is a silent epidemic of treatable chronic sleep problems and circadian disruption (i.e., shifting of the body's daily biological rhythm), affecting over 70 million older Americans. Such symptoms of these issues include insufficient sleep, irregular timing, unscheduled naps, insomnia, or daytime sleepiness.[9] Sleep/circadian disruption increases with age and cognitive impairment and even predicts preclinical AD pathology[10,11] and onset of AD/ADRD.[12] POD is associated with disruption of melatonin secretion – the key circadian hormone in sleep homeostasis, [13] particularly in older patients. [14] Despite this, there is minimal consideration of sleep/circadian disruption in preoperative medicine. Pre-existing sleep/circadian disruptions are likely to be exacerbated during recovery after major surgery.[15]

Our recent work and that of others suggest that suboptimal sleep/circadian regulation is associated with POD risk [16–20] and progression to dementia, independent of age, sex, education, and cognition.[12] These studies also uncovered that sleep/circadian measures correlated with cerebrospinal fluid amyloid/*tau* burden decades before dementia onset,[10,11] and that plasma *tau* burden was associated with POD in two surgical cohorts.[21,22] Finally, genetic biomarkers, namely the *APOE-ε4* genotype, may moderate the relationship between sleep and AD risk,[23] but the role of sleep/circadian regulation in POD remains controversial.[24,25] While epidemiological evidence points to sleep/circadian disruption as a shared modifiable risk factor for POD and dementia, direct clinical evidence is lacking.

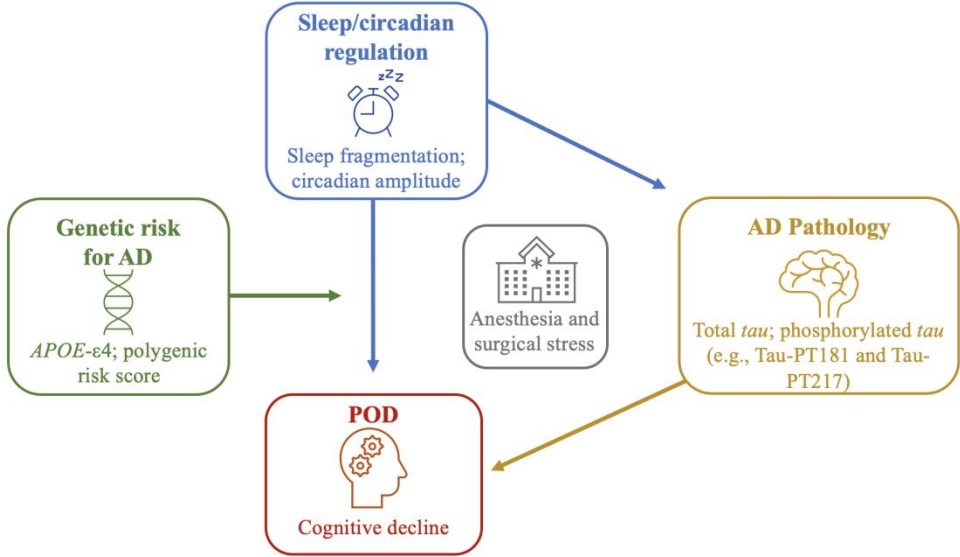


Fig 1. SLEEP-POD Conceptual Model: Sleep/circadian disruption effects on POD via plasma AD pathology burden and influence of genetic risk of AD. *AD*: Alzheimer’s disease, *POD*: postoperative delirium, *APOE-ε4*: apolipoprotein ε4.

This leads us to our central hypothesis that sleep/circadian disruption promotes cognitive vulnerability after anesthesia/surgery via increased AD pathology, and this relationship is exacerbated by genetic risk for AD via *APOE-ε4* or a polygenic risk score (see Fig. 1 for the conceptual model). To test this hypothesis, we propose the SLEEP-POD study – a single-site, prospective, observational study of 150 older patients (≥70y) undergoing elective, major orthopedic surgery at the Massachusetts General Hospital (MGH). This study will assess preoperative sleep and circadian rest/activity rhythms using validated sleep questionnaires and objective week-long actigraphy measures. Cognitive assessments will be performed throughout the study. Blood samples will be retrieved to gather genotyping data and tau measurements.[21,22] The primary outcome will be the presence of POD before discharge or within 3 days of surgery. Additional actigraphy assessments and evaluations for cognition, sleep, and pain will occur periodically during follow-up visits at one, three, and twelve months after surgery.

METHODS AND ANALYSIS

Study design

This protocol presents a single-site, prospective, observational study to examine the relationship between sleep/circadian regulation and POD and how this association is moderated or mediated by tau and genetic risk for AD. Subjects meeting eligibility criteria will be recruited after obtaining written informed consent. Recruitment will take place at MGH. A trained study staff member will carry out the process of consenting patients. The study protocol and assessments to be performed are presented in Table 2.

Study registration

This study is approved by the Institutional Review Board (IRB) at MGH, Boston. This study is registered with the US National Institutes of Health on ClinicalTrials.gov (NCT06052397). The study is expected to be open for recruitment for three years.

Patient and public involvement

Patient advisers were not involved in this research study's design, conduct, reporting, or dissemination plans. However, patient-reported outcome measures from questionnaires will be evaluated as a secondary outcome. The study is registered on ClinicalTrials.gov and updated at regular intervals. Results will be reported in a time-compliant manner on ClinicalTrials.gov.

Inclusion and exclusion criteria

We will include 150 older (≥ 70 years) elective orthopedic surgical patients undergoing joint replacement or spine surgery at MGH with an expected postoperative recovery of greater than 24 hours. Exclusion criteria: known dementia (diagnosis or MMSE < 21) or related treatment, alcohol/drug abuse within two years, inability to wear an actiwatch, need for urgent/emergent surgery or surgery in the prior month, more than 2-day ICU stay in the prior month, or insufficient vision, hearing, or English language for testing completion. A complete listing of trial exclusions and rationale is found in **Table 1**.

Table 1. Inclusion and exclusion criteria with rationale	
Inclusion Criteria	Rationale
≥ 70 years of age	POD most commonly appears in older patients
Scheduled to undergo major orthopedic surgery (total hip replacement, total knee replacement, spine surgery)	Major orthopedic surgeries are associated with an increased risk for POD[21,26]
Exclusion Criteria	Rationale
Inability to give consent due to, but not limited to, preexisting dementia (diagnosis or MMSE < 21), Parkinson's disease or other movement disorders, severe neurocognitive damage, severe psychiatric illness, blindness, or deafness.	Inability to ethically enroll individuals of this population
Expected postoperative recovery for less than 24 hours	We will be unable to assess for POD
Patient with more than two days spent in the ICU during the month prior to surgery	Pre-existing operations/illnesses may temporarily disrupt sleep and confound the study
Patient with inability to wear actiwatch (due to, but not limited to, infection at the site of watch, allergies to the material of watch, limited mobility)	Required for activity-rhythm assessment
Non-English speaking or insufficient vision and/or hearing	Inability to administer cognitive assessments that are primarily validated in English

Clinical data collection

Clinical data, including age, sex, body mass index, comorbidities, length of hospital stay, and surgical details (e.g., surgery duration, anesthesia type, blood loss, complications), will be recorded using passively collected data from the patient's electronic medical record.

DNA and plasma collection

Blood samples will be collected pre- and postoperatively via indwelling lines, and a nutrition assessment will be conducted before all blood collections to assess the nutritional impact on the collected blood samples. The samples will be used to gather information about AD variant genes,

including *APOE-ε4*, and investigate associations with baseline and changes in plasma AD-related pathology (total and phosphorylated *tau*). *Tau* will be measured using a nanotechnology platform.[21,22]

Sleep/circadian regulation measures

In preoperative clinics, patients will complete sleep quality questionnaires (e.g., Pittsburgh Sleep Quality Index [PSQI]) and be instructed to complete an electronic sleep diary while wearing a validated wristwatch (Actiwatch Spectrum Plus) for seven days. We will apply algorithms and quality control best practices[27] to objectively infer rest-activity rhythm measures from the accelerometer data. The watch will collect data that will be used to calculate estimations of sleep duration, regularity, stability, and timing of activity rhythms. Patients will return the watch on the day of surgery and will be asked to wear it for an additional week at their one-, three-, and twelve-month follow-up visits.

Table 2. Study flow chart				
	Screen (<4 weeks)	Preoperative (≥1 week)	Postoperative (1-3 days)	Follow-Up (1, 3, & 12 months)
Informed Consent	X			
eMR Review	X	X	X	X
Eligibility Assessment	X			
Sleep Questionnaires (PSQI, ESS, STOP- BANG, MEQ)		X		
CAM Evaluation			X	X
MoCA/t-MoCA		X	X	X
PROMIS Questionnaires*		X	X	X
Blood Collection (AD genotyping and <i>tau</i> pathology)		X	X	
Nutrition Assessment		X	X	
Actiwatch Data Collection (1-week)		X		X
ECG (PWV, AI)		X		

* Physical function (SF 8b V.1.2) and applied cognition abilities (SF 8a V.1.0) may only be evaluated during follow-up visits.

Abbreviations: eMR= electronic medical record, PSQI = Pittsburg Sleep Quality Index, ESS = Epworth Sleepiness Scale, STOP-BANG = Snoring history, Tired during the day, Observed stop of breathing while sleeping, high blood Pressure, BMI > 35 kg/m2, Age > 50 years, Neck circumference > 40 cm, and male Gender, MEQ = Morningness-Eveningness Questionnaire, CAM = Confusion Assessment Method, MoCA/t-MoCA = (Telephonic) Montreal Cognitive Assessment, ECG (PWV, AI) = electrocardiogram (pulse wave velocity, augmentation index), PROMIS = Patient-Reported Outcomes Measurement Information System.

Study outcome measures

The primary outcome of this study is the presence of delirium up to day three or discharge after major orthopedic surgery. Delirium will be assessed using the Confusion Assessment Method (CAM) or CAM-ICU, which will be scored by the study team members twice daily on postoperative days 1-3. The CAM considers performance on attention, orientation, and memory tests from the Montreal Cognitive Assessment (MoCA) and responses to the Delirium Severity Index (DSI) – a symptom severity questionnaire. Trained study staff will use these data to complete the long CAM, informing a diagnostic algorithm determining delirium status at the assessment time. The presence or absence of delirium during postoperative days 1-3 or before discharge will be recorded binarily.

Secondary outcomes of this study include the following:

1. Severity of delirium will be measured using the CAM-S up until postoperative day 3 or discharge. CAM-S ranges from 0 to 19 points, with larger points indicating more marked delirium features.
2. CAM will measure delirium-free days up until postoperative day 3 or discharge.
3. Postoperative cognitive status will be recorded via telephone one, three, and twelve months after surgery using the Telephonic Montreal Cognitive Assessment (t-MoCA).
4. Postoperative health-related quality of life will be measured using a series of questionnaires from the Patient-Reported Outcomes Measurement Information System (PROMIS) that evaluate global health (Short Form [SF] V.1.1), physical function (SF 8b V.1.2), pain interference (SF 8a V.1.0), applied cognition abilities (SF 8a V.1.0), and sleep disturbance (SF 4A V.1.0) at 1 month, 3 months, and 12 months after surgery.

Other outcome measures: Postoperative sleep and rest-activity rhythm data from accelerometer watches will be assessed as an exploratory outcome measure. In addition, patients will be evaluated during the baseline interview with continuous ECG recording to explore heart rate response and vascular biomarkers associated with delirium.[28,29] A 4-lead ECG with oximetry will be placed on the patients by trained study staff during the inclusion visit. We will explore pulse wave velocity (PWV) and augmentation index (AI) to evaluate artery stiffness and peripheral resistance, given potential relationships to sleep health, aging and pathogenesis of ADRDs that may overlap with delirium.[30,31]

Recruitment

Trained members of the study team will identify potential subjects presenting to MGH for orthopedic surgery. Whenever possible, a study team member (clinical research coordinator, principal investigator, or co-investigator) will meet the patient during the orthopedic surgery clinic visit before surgery. The study procedures will be explained in detail, and consent will be obtained from interested participants. Eligible patients whom we cannot connect with at their preoperative clinic visit will be contacted via telephone. For interested participants, consent will be obtained electronically, and directions for obtaining the actiwatch will be explained for them.

Data analysis

We will construct logistic regression models to test the association between sleep/circadian predictors and POD. We will then use multivariate linear regression to test the association between continuous exposures (e.g., sleep fragmentation and circadian amplitude) and continuous outcomes (e.g., cognition). To test whether *tau* mediates the above relationship, we will bootstrap

the indirect effect CIs, the optimal method from Fritz & MacKinnon’s work,[32] which would provide >80% power to detect the medium-medium-zero effect sizes condition ($\alpha = 0.39$, $\beta = 0.39$, $\tau' = 0$). We will also test whether *tau* burden interacts with sleep/circadian regulation associated with POD. We will introduce interaction terms between *APOE-ε4* status and our two exposure variables into our models. The model fit and outlier assessment will be examined using formal fit criteria and model inspection. We will select covariates based on prior POD or AD studies and the strength of association/extent of confounding while avoiding model overfitting. We will assess a core model with age and sex and build an adjusted model with the optimal number of predictors after evaluating with the Akaike Information Criterion (AIC).

Data management and quality assurance

This study will employ a web-based portal for data quality and completeness that will be updated by study staff regularly. The portal will display the following variables for all patients: sex, race, adverse events, study-related data, etc. To ensure data is accurately and completely collected during the study, the PI will be responsible for ensuring the study protocol is being followed, the IRB has approved changes to the protocol, and all facilities are appropriate for the conduct of the study. Also, the PI will review subject records to determine whether the data collected is accurate, complete, and current. A Scientific Review will occur annually and consist of a review of subject recruitment, staff training, and quality control procedures. Monitoring progress reports will be submitted to regulatory and/or funding bodies (IRB, Alzheimer’s Association) as requested or required.

Limitations

Limitations of the study include its observational nature and, therefore, the inability to demonstrate causation. Treatment effects after elective joint surgery may affect the association between sleep/circadian rhythms and delirium. In addition, repeat follow-up 12 months after surgery may increase the likelihood of missing data. Despite these pitfalls, older orthopedic patients have similar characteristics, which mitigates variability within this study. They also have a high co-morbid burden of sleep disturbances, which can enrich the signal for POD and cognitive decline. To mitigate the likelihood of missing data, we will allow a robust window for assessment completion with repeated contact attempts during the follow-up period.

Patient burden during the study could originate from discomfort while wearing the actiwatch and fatigue during cognitive assessments. When possible, blood collection will be performed while the patient is under general anesthesia and extracted from an indwelling line to minimize patient discomfort. All other efforts will be made to ensure minimal patient discomfort and stress throughout the study.

Ethics and dissemination

The PI of this study will be responsible for final decisions regarding changes to the protocol and will report such changes to the MGB IRB. Electronic patient information will be accessed only as necessary throughout the completion of the study. All data collected from the study will be accessible to the PI. Additional analyses will be performed to formulate predictive models for delirium using sleep/circadian patterns and genetic data. The main papers reporting information about this study will present primary and secondary outcomes, and manuscripts describing the mechanisms for delirium pathophysiology from sub-studies (i.e., circadian/sleep disturbance,

actigraphy, protein biomarkers) will also be published. Plans for dissemination include conference presentations at a variety of scientific institutions. Results from this study are intended to be published in peer-reviewed journals. Relevant updates will be made available on ClinicalTrials.gov.

In conclusion, this study will provide useful data and certain insights into the biomarkers for delirium through analysis of sleep/circadian disruption and AD pathology. We present a novel approach to understanding a modifiable sleep/circadian risk factor for postoperative delirium and cognitive decline after surgery.

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Conflicts of Interest: Dr. Zhongcong Xie provided consulting services to Baxter Pharmaceutical company, Shanghai 9th and 10th Hospital, NanoMosaic Inc., and Anesthesiology and Perioperative Science within the last 36 months. The authors have no other conflicts of interest to declare.

Author Contributions: E.S., P.L., A.M., E.M., O.A., Z.X., K.H., and L.G. were responsible for conceptualizing the trial design. E.S., E.G., A.M., and L.G. are responsible for recruitment, enrollment, and data collection. E.S., E.G., S.N.B., P.L., C.G., A.M., H.D., S.S., E.F.G., R.S., E.M., O.A., Z.X., K.H., and L.G. have critically revised the protocol and approved the final version.

References

1 American Geriatrics Society Expert Panel on Postoperative Delirium in Older Adults. Postoperative delirium in older adults: best practice statement from the American Geriatrics Society. *J Am Coll Surg*. 2015;220:136-148.e1.

2 Witlox J, Eurelings LSM, de Jonghe JFM, *et al*. Delirium in elderly patients and the risk of postdischarge mortality, institutionalization, and dementia: a meta-analysis. *JAMA*. 2010;304:443–51.

3 Gou RY, Hshieh TT, Marcantonio ER, *et al*. One-Year Medicare Costs Associated With Delirium in Older Patients Undergoing Major Elective Surgery. *JAMA Surgery*. Published Online First: 24 February 2021. doi: 10.1001/jamasurg.2020.7260

4 Goldberg TE, Chen C, Wang Y, *et al*. Association of Delirium With Long-term Cognitive Decline: A Meta-analysis. *JAMA Neurology*. 2020;77:1373–81.

5 Inouye SK, Marcantonio ER, Kosar CM, *et al*. The short-term and long-term relationship between delirium and cognitive trajectory in older surgical patients. *Alzheimers Dement*. 2016;12:766–75.

6 Rathmell CS, Akeju O, Inouye SK, *et al*. Estimating the number of cases of dementia that might be prevented by preventing delirium. *British Journal of Anaesthesia*. 2023;0. doi: 10.1016/j.bja.2023.03.001

7 Rahman S, Byatt K. Follow-up services for delirium after COVID-19—where now? *Age Ageing*. 2021;afab014.

8 Fong TG, Tulebaev SR, Inouye SK. Delirium in elderly adults: diagnosis, prevention and treatment. *Nat Rev Neurol*. 2009;5:210–20.

9 Miner B, Kryger MH. Sleep in the Aging Population. *Sleep Med Clin*. 2017;12:31–8.

10 Musiek ES, Bhimasani M, Zangrilli MA, *et al*. Circadian Rest-Activity Pattern Changes in Aging and Preclinical Alzheimer Disease. *JAMA Neurol*. 2018;75:582–90.

11 Gao L, Li P, Gaba A, *et al*. Fractal motor activity regulation and sex differences in preclinical Alzheimer’s disease pathology. *Alzheimers Dement (Amst)*. 2021;13:e12211.

12 Gao L, Li P, Gaykova N, *et al*. Circadian Rest-Activity Rhythms, Delirium Risk, and Progression to Dementia. *Ann Neurol*. 2023;93:1145–57.

13 Dessap AM, Roche-Campo F, Launay J-M, *et al*. Delirium and Circadian Rhythm of Melatonin During Weaning From Mechanical Ventilation: An Ancillary Study of a Weaning Trial. *CHEST*. 2015;148:1231–41.

- 14 Campbell AM, Axon DR, Martin JR, *et al.* Melatonin for the prevention of postoperative delirium in older adults: a systematic review and meta-analysis. *BMC Geriatrics*. 2019;19:272.
- 15 O’Gara BP, Gao L, Marcantonio ER, *et al.* Sleep, Pain, and Cognition: Modifiable Targets for Optimal Perioperative Brain Health. *Anesthesiology*. 2021;135:1132–52.
- 16 Leung JM, Sands LP, Newman S, *et al.* Preoperative Sleep Disruption and Postoperative Delirium. *J Clin Sleep Med*. 2015;11:907–13.
- 17 Leung JM, Tang C, Do Q, *et al.* Sleep Loss the night before surgery and incidence of postoperative delirium in adults 65-95 years of age. *Sleep Med*. 2023;105:61–7.
- 18 Tan C, Saito N, Miyawaki I, *et al.* Preoperative circadian physical activity rhythm and postoperative delirium in cardiovascular surgery patients. *Chronobiology International*. 2020;37:1059–66.
- 19 Ulsa MC, Xi Z, Li P, *et al.* Association of Poor Sleep Burden in Middle Age and Older Adults With Risk for Delirium During Hospitalization. *J Gerontol A Biol Sci Med Sci*. 2022;77:507–16.
- 20 Ibala R, Mekonnen J, Gitlin J, *et al.* A polysomnography study examining the association between sleep and postoperative delirium in older hospitalized cardiac surgical patients. *J Sleep Res*. 2021;30:e13322.
- 21 Liang F, Baldyga K, Quan Q, *et al.* Preoperative Plasma Tau-PT217 and Tau-PT181 Are Associated With Postoperative Delirium. *Ann Surg*. Published Online First: 6 July 2022. doi: 10.1097/SLA.0000000000005487
- 22 McKay TB, Qu J, Liang F, *et al.* Tau as a serum biomarker of delirium after major cardiac surgery: a single centre case-control study. *Br J Anaesth*. 2022;129:e13–6.
- 23 Lim AS, Yu L, Kowgier M, *et al.* Sleep modifies the relation of APOE to the risk of Alzheimer disease and neurofibrillary tangle pathology. *JAMA neurology*. 2013;70.
- 24 Vasunilashorn S, Ngo L, Kosar CM, *et al.* Does Apolipoprotein E Genotype Increase Risk of Postoperative Delirium? *Am J Geriatr Psychiatry*. 2015;23:1029–37.
- 25 Vasunilashorn SM, Ngo LH, Inouye SK, *et al.* Apolipoprotein E genotype and the association between C-reactive protein and postoperative delirium: Importance of gene-protein interactions. *Alzheimer’s & Dementia*. 2020;16:572–80.
- 26 Connolly KP, Kleinman RS, Stevenson KL, *et al.* Delirium Reduced With Intravenous Acetaminophen in Geriatric Hip Fracture Patients. *J Am Acad Orthop Surg*. 2020;28:325–31.
- 27 Li P, Gao L, Gaba A, *et al.* Circadian disturbances in Alzheimer’s disease progression: a prospective observational cohort study of community-based older adults. *The Lancet Healthy Longevity*. 2020;1:e96–105.

28 Gao L, Gaba A, Li P, *et al.* Heart rate response and recovery during exercise predict future delirium risk-A prospective cohort study in middle- to older-aged adults. *J Sport Health Sci.* 2023;12:312–23.

29 Menzenbach J, Frede S, Petras J, *et al.* Perioperative Vascular Biomarker Profiling in Elective Surgery Patients Developing Postoperative Delirium: A Prospective Cohort Study. *Biomedicines.* 2021;9:553. doi: 10.3390/biomedicines9050553

30 Hughes TM, Craft S, Lopez OL. Review of ‘the potential role of arterial stiffness in the pathogenesis of Alzheimer’s disease’. *Neurodegener Dis Manag.* 2015;5:121–35.

31 Iulita MF, Noriega de la Colina A, Girouard H. Arterial stiffness, cognitive impairment and dementia: confounding factor or real risk? *Journal of Neurochemistry.* 2018;144:527–48.

32 Fritz MS, MacKinnon DP. Required Sample Size to Detect the Mediated Effect. *Psychol Sci.* 2007;18:233–9.

The RECORD statement – checklist of items, extended from the STROBE statement, that should be reported in observational studies using routinely collected health data.

	Item No.	STROBE items	Location in manuscript where items are reported	RECORD items	Location in manuscript where items are reported
Title and abstract					
	1	(a) Indicate the study's design with a commonly used term in the title or the abstract (b) Provide in the abstract an informative and balanced summary of what was done and what was found	Abstract, page 2	RECORD 1.1: The type of data used should be specified in the title or abstract. When possible, the name of the databases used should be included. RECORD 1.2: If applicable, the geographic region and time frame within which the study took place should be reported in the title or abstract. RECORD 1.3: If linkage between databases was conducted during the study, this should be clearly stated in the title or abstract.	Abstract, page 2
Introduction					
Background rationale	2	Explain the scientific background and rationale for the investigation being reported	Introduction, page 3/4		
Objectives	3	State specific objectives, including any prespecified hypotheses	Introduction, page 3/4		
Methods					
Study Design	4	Present key elements of study design early in the paper	Methods, page 4		
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	Methods, page 4/5		

Participants	6	<p>(a) <i>Cohort study</i> - Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up</p> <p><i>Case-control study</i> - Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls</p> <p><i>Cross-sectional study</i> - Give the eligibility criteria, and the sources and methods of selection of participants</p> <p>(b) <i>Cohort study</i> - For matched studies, give matching criteria and number of exposed and unexposed</p> <p><i>Case-control study</i> - For matched studies, give matching criteria and the number of controls per case</p>	Methods, page 5	<p>RECORD 6.1: The methods of study population selection (such as codes or algorithms used to identify subjects) should be listed in detail. If this is not possible, an explanation should be provided.</p> <p>RECORD 6.2: Any validation studies of the codes or algorithms used to select the population should be referenced. If validation was conducted for this study and not published elsewhere, detailed methods and results should be provided.</p> <p>RECORD 6.3: If the study involved linkage of databases, consider use of a flow diagram or other graphical display to demonstrate the data linkage process, including the number of individuals with linked data at each stage.</p>	Methods, page 5 / N/A
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable.	Methods, page 6/7	RECORD 7.1: A complete list of codes and algorithms used to classify exposures, outcomes, confounders, and effect modifiers should be provided. If these cannot be reported, an explanation should be provided.	
Data sources/ measurement	8	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	Methods, page 6-8		

Bias	9	Describe any efforts to address potential sources of bias	Methods, page 8		
Study size	10	Explain how the study size was arrived at	Methods, page 5		
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen, and why	Methods, page 8		
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding (b) Describe any methods used to examine subgroups and interactions (c) Explain how missing data were addressed (d) <i>Cohort study</i> - If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> - If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> - If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses	Methods, page 8		
Data access and cleaning methods		..		RECORD 12.1: Authors should describe the extent to which the investigators had access to the database population used to create the study population.	Methods, page 8

				RECORD 12.2: Authors should provide information on the data cleaning methods used in the study.	Methods, page 8
Linkage		..		RECORD 12.3: State whether the study included person-level, institutional-level, or other data linkage across two or more databases. The methods of linkage and methods of linkage quality evaluation should be provided.	Methods, page 8
Results					
Participants	13	(a) Report the numbers of individuals at each stage of the study (<i>e.g.</i> , numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed) (b) Give reasons for non-participation at each stage. (c) Consider use of a flow diagram	N/A	RECORD 13.1: Describe in detail the selection of the persons included in the study (<i>i.e.</i> , study population selection) including filtering based on data quality, data availability and linkage. The selection of included persons can be described in the text and/or by means of the study flow diagram.	N/A
Descriptive data	14	(a) Give characteristics of study participants (<i>e.g.</i> , demographic, clinical, social) and information on exposures and potential confounders (b) Indicate the number of participants with missing data for each variable of interest (c) <i>Cohort study</i> - summarise follow-up time (<i>e.g.</i> , average and total amount)	N/A		
Outcome data	15	<i>Cohort study</i> - Report numbers of outcome events or summary measures over time <i>Case-control study</i> - Report numbers in each exposure	N/A		

		category, or summary measures of exposure <i>Cross-sectional study</i> - Report numbers of outcome events or summary measures			
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (e.g., 95% confidence interval). Make clear which confounders were adjusted for and why they were included (b) Report category boundaries when continuous variables were categorized (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	N/A		
Other analyses	17	Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses	N/A		
Discussion					
Key results	18	Summarise key results with reference to study objectives	N/A		
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	N/A	RECORD 19.1: Discuss the implications of using data that were not created or collected to answer the specific research question(s). Include discussion of misclassification bias, unmeasured confounding, missing data, and changing eligibility over time, as they pertain to the study being reported.	N/A
Interpretation	20	Give a cautious overall interpretation of results considering objectives,	N/A		

		limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	N/A		
Generalisability	21	Discuss the generalisability (external validity) of the study results	N/A		
Other Information					
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	Page 9		
Accessibility of protocol, raw data, and programming code		..		RECORD 22.1: Authors should provide information on how to access any supplemental information such as the study protocol, raw data, or programming code.	Page 4, 8, 9

*Reference: Benchimol EI, Smeeth L, Guttman A, Harron K, Moher D, Petersen I, Sørensen HT, von Elm E, Lankester SM, the RECORD Working Committee. The REporting of studies Conducted using Observational Routinely-collected health Data (RECORD) Statement. *PLoS Medicine* 2015; in press.

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Sleep and circadian biomarkers of postoperative delirium (SLEEP-POD): protocol for a prospective, observational cohort study

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Primary Subject Heading:	Anaesthesia
Secondary Subject Heading:	Neurology

Title: Sleep and circadian biomarkers of postoperative delirium (SLEEP-POD): protocol for a prospective, observational cohort study

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ABSTRACT

Introduction: Surgical patients over 70 experience postoperative delirium (POD) complications in up to 50% of procedures. Sleep/circadian disruption has emerged as a potential risk factor for POD in epidemiological studies. This protocol presents a single-site, prospective observational study designed to examine the relationship between sleep/circadian regulation and POD and how this association could be moderated or mediated by AD pathology and genetic risk for AD.

Methods and Analysis: Study staff members will screen for eligible patients (age≥70) seeking joint replacement or spinal surgery at MGH. At the inclusion visit, patients will be asked a series of questionnaires related to sleep and cognition, conduct a 4-lead ECG recording, and be fitted for an actigraphy watch to wear for seven days before surgery. Blood samples will be collected pre- and postoperatively and will be used to gather information about AD variant genes (*APOE-ε4*) and AD-related pathology (total and phosphorylated *tau*). CAM-S and MoCA will be completed twice daily for three days after surgery. Seven-day actigraphy assessments and PROMIS questionnaires will be performed one, three, and twelve months after surgery. Relevant patient clinical data will be monitored and recorded throughout the study.

Ethics and Dissemination: This study is approved by the IRB at MGH, Boston, and it is registered with the US National Institutes of Health on ClinicalTrials.gov (NCT06052397). Plans for dissemination include conference presentations at a variety of scientific institutions. Results from this study are intended to be published in peer-reviewed journals. Relevant updates will be made available on ClinicalTrials.gov.

Strengths and Limitations:

- Limitations of the study include its observational nature and, therefore, the inability to demonstrate causation.
- Treatment effects after elective joint surgery may affect the association between sleep/circadian rhythms and delirium.
- Repeat follow-up 12 months after surgery may increase the likelihood of missing data.
- Despite these pitfalls, older orthopedic patients have similar characteristics, which mitigates variability within this study.
- They also have a high co-morbid burden of sleep disturbances, which can enrich the signal for POD and cognitive decline.
- Additional limitations include not recording postoperative in-hospital actigraphy measures, however, there are many uncontrollable factors that are presented in the hospital setting that could affect data analysis.
- Excluding those with severe dementia could weaken the signal for POD incidence since this group is more likely to develop POD, but we do not seek to include this cohort at this time, given the study burdens.

INTRODUCTION

Postoperative delirium (POD) is one of the most common complications following surgery for older adults, occurring in 5% to 50% of cases.[1] POD is a neurocognitive syndrome with acute attention, cognitive, and awareness deficits that develops over a short period of time and has a fluctuating course. It is associated with cognitive dysfunction[2] and Alzheimer's disease (AD) and related dementias (ADRD) but can present in otherwise cognitively intact older adults. Millions of older Americans (≥ 70 years) require major surgery each year and are exposed to the risks of POD and its consequences.[3] Furthermore, there are currently no treatments for POD, but it perpetuates a cascade of poor health outcomes costing approximately \$50 billion annually.[4] Older age and baseline cognitive impairment (e.g., pre-existing AD/ADRD) are major risk factors. Male gender, multiple co-morbidities, sensory impairments, sleep deprivation, and use of sedative medications are also potential risk factors.[5–7] POD may also predispose to AD/ADRD, suggesting overlapping pathophysiology.[3,8–10] Yet, minimal attention or cognitive follow-up exists for those who suffer from POD.[11] Given that nearly 40% of POD is preventable with attention to multifactorial preoperative care,[12] the search for modifiable risk factors and novel biomarkers in POD is of great public health importance.

At the same time, there is a silent epidemic of treatable chronic sleep problems and circadian disruption (i.e., shifting of the body's daily biological rhythm) affecting over 70 million older Americans. Such symptoms of these issues include insufficient sleep, irregular timing, unscheduled naps, insomnia, or daytime sleepiness.[13] Sleep/circadian disruption increases with age and cognitive impairment and is associated with frailty,[14] pain and opioid use,[15] preclinical AD pathology[16,17] and the onset of AD/ADRD.[18] POD is associated with disruption of melatonin secretion – the key circadian hormone in sleep homeostasis, [19] particularly in older patients. [20] Despite this, there is minimal consideration of sleep/circadian disruption in preoperative medicine. Pre-existing sleep/circadian disruptions are likely to be exacerbated during recovery after major surgery.[21]

Our recent work and that of others suggest that suboptimal sleep/circadian regulation is associated with POD risk [22–26] and progression to dementia, independent of age, sex, education, and cognition.[18] These studies also uncovered that sleep/circadian measures correlated with cerebrospinal fluid amyloid/*tau* burden decades before dementia onset,[16,17] and that plasma *tau* burden was associated with POD in two surgical cohorts.[27,28] Finally, genetic biomarkers, namely the *APOE*- $\epsilon 4$ genotype, may moderate the relationship between sleep and AD risk,[29] but its role in POD remains controversial.[30,31] While epidemiological evidence points to sleep/circadian disruption as a shared modifiable risk factor for POD and dementia, direct clinical evidence is lacking.

This leads us to our central hypothesis that sleep/circadian disruption promotes cognitive vulnerability after anesthesia/surgery via increased AD pathology, and this relationship is exacerbated by genetic risk for AD via *APOE*- $\epsilon 4$ or a polygenic risk score (see **Fig. 1** for the conceptual model). To test this hypothesis, we propose the SLEEP-POD study – a single-site, prospective, observational study of 150 older patients (≥ 70 y) undergoing elective, major orthopedic surgery at the Massachusetts General Hospital (MGH). This study will assess preoperative sleep and circadian rest/activity rhythms using validated sleep questionnaires and

objective week-long actigraphy measures. Cognitive assessments will be performed throughout the study. Blood samples will be retrieved to gather genotyping data and tau measurements.[27,28] The primary outcome will be the presence of POD before discharge or within 3 days of surgery. Additional actigraphy assessments and evaluations for cognition, sleep, and pain will occur periodically during follow-up visits at one, three, and twelve months after surgery.

METHODS AND ANALYSIS

Study design

This protocol presents a single-site, prospective, observational study to examine the relationship between sleep/circadian regulation and POD and how this association is moderated or mediated by *tau* and genetic risk for AD. Subjects meeting eligibility criteria will be recruited after obtaining written informed consent. Recruitment will take place at MGH. A trained study staff member will carry out the process of consenting patients. The study protocol and assessments to be performed are presented in **Table 2**.

Study registration

This study is approved by the Institutional Review Board (IRB) at MGH, Boston. This study is registered with the US National Institutes of Health on ClinicalTrials.gov (NCT06052397). The study is expected to be open for recruitment for three years.

Inclusion and exclusion criteria

We will include 150 older (≥70 years) elective orthopedic surgical patients undergoing joint replacement or spine surgery at MGH with an expected postoperative recovery of greater than 24 hours. Exclusion criteria: known severe dementia (diagnosis or MMSE<18) or related treatment, alcohol/drug abuse within two years, inability to wear an actiwatch, need for urgent/emergent surgery or surgery in the prior month, more than 2-day ICU stay in the prior month, or insufficient vision, hearing, or English language for testing completion. A complete listing of trial exclusions and rationale is found in **Table 1**.

Table 1. Inclusion and exclusion criteria with rationale	
Inclusion Criteria	Rationale
≥70 years of age	POD most commonly appears in older patients
Scheduled to undergo major orthopedic surgery (total hip replacement, total knee replacement, spine surgery)	Major orthopedic surgeries are associated with an increased risk for POD[27,32]
Exclusion Criteria	Rationale
Inability to give consent due to, but not limited to, preexisting severe dementia (diagnosis or MMSE <18), Parkinson’s disease or other movement disorders, severe neurocognitive damage, severe psychiatric illness, blindness, or deafness.	Inability to ethically enroll individuals of this population due to study burden
Expected postoperative recovery for less than 24 hours	We will be unable to assess for POD
Patient with more than two days spent in the ICU during the month prior to surgery	Pre-existing operations/illnesses may temporarily disrupt sleep and confound the study

Patient with inability to wear actiwatch (due to, but not limited to, infection at the site of watch, allergies to the material of watch, limited mobility)	Required for activity-rhythm assessment
Non-English speaking or insufficient vision and/or hearing	Inability to administer cognitive assessments that are primarily validated in English

Recruitment

Trained members of the study team will identify potential subjects presenting to MGH for orthopedic surgery. Whenever possible, a study team member (clinical research coordinator, principal investigator, or co-investigator) will meet the patient during the orthopedic surgery clinic visit before surgery. The study procedures will be explained in detail, and consent will be obtained from interested participants. Eligible patients whom we cannot connect with at their preoperative clinic visit will be contacted via telephone. For interested participants, consent will be obtained electronically, and directions for obtaining the actiwatch will be explained for them.

Sleep/circadian instruments (actigraphy, sleep diary, and questionnaires)

In preoperative clinics, patients will complete sleep quality questionnaires (e.g., Pittsburgh Sleep Quality Index [PSQI]) and be instructed to complete an electronic sleep diary while wearing a validated wristwatch (Actiwatch Spectrum Plus) for seven days (**Table 2**). We will apply algorithms and quality control best practices[33] to objectively infer rest-activity rhythm measures from the accelerometer data. The watch will collect data that will be used to calculate estimations of sleep duration, regularity, stability, and timing of activity rhythms. Patients will return the watch on the day of surgery and will be asked to wear it for an additional week at their one-, three-, and twelve-month follow-up visits.

Sleep/circadian measures

The data collection methods described above inform how we will test our hypothesis. The exposures enlisted in this study can be broken down into co-primary exposures relating to sleep and circadian disruption and subsequent exploratory exposures associated with sleep/circadian disruption.

Sleep efficiency (SE) will test how sleep disruption materializes upon testing our hypothesis. The data collected from the sleep diary and actigraphy watch will inform the ratio for SE (SE = total time asleep/time spent in bed).[34] Secondary/exploratory exposures for sleep analysis include sleep duration (total sleep time), sleep latency (time taken to fall asleep), wake-after-sleep-onset (WASO), sleep fragmentation (kRA), and daytime sleepiness or napping.[34–36]

Relative amplitude (RA) will act as the exposure for identifying how circadian disruption influences the relationship tested in our hypothesis. The actigraphy-derived measures M10 (the most active 10-hour period per day) and L5 (the least active 5-hour period per day) will inform RA (M10-L5/M10+L5).[18] Secondary/exploratory exposures for circadian analysis include phase (peak activity time in hours during a 24-hour period), MESOR (midline-estimating statistic of rhythm, or rhythm-adjusted mean of a 24-hour period), interdaily stability (IS; stability of between-day rhythms), and intradaily variability (IV; within-day fragmentation of rhythms).[37–39]

DNA and plasma collection

Blood samples will be collected pre- and postoperatively via indwelling lines, and a nutrition assessment will be conducted before all blood collections to assess the nutritional impact on the collected blood samples. The samples will be used to gather information about AD variant genes, including *APOE-ε4*, and investigate associations with baseline and changes in plasma AD-related pathology (total and phosphorylated *tau*). *Tau* will be measured using a nanotechnology platform,[27,28] which employs 20,000 nanoneedles to quantify phosphorylated *tau* at a very low abundance in blood samples. Phospho-specific antibodies identify *tau* (e.g., Tau-PT217, Tau-PT181) and produce a color shift that is recognized by digital software. The software uses a threshold for color shift to determine if a binding event is positive (i.e., Poisson statistics).

Clinical data collection

Clinical data, including age, sex, body mass index, comorbidities, length of hospital stay, and surgical details (e.g., surgery duration, anesthesia type, blood loss, complications), will be recorded using passively collected data from the patient’s electronic medical record.

Table 2. Study flow chart				
	Screen (<4 weeks)	Preoperative (≥1 week)	Postoperative (1-3 days)	Follow-Up (1, 3, & 12 months)
Informed Consent	X			
eMR Review	X	X	X	X
Eligibility Assessment	X			
Sleep Questionnaires (PSQI, ESS, STOP- BANG, MEQ)		X		
CAM Evaluation			X	X
MoCA/t-MoCA		X	X	X
PROMIS Questionnaires*		X	X	X
Blood Collection (AD genotyping and <i>tau</i> pathology)		X	X	
Nutrition Assessment		X	X	
Actiwatch Data Collection (1-week)		X		X
ECG (PWV, AI)		X		

* *Physical function (SF 8b V.1.2) and applied cognition abilities (SF 8a V.1.0) may only be evaluated during follow-up visits. Abbreviations:* eMR= electronic medical record, PSQI = Pittsburgh Sleep Quality Index, ESS = Epworth Sleepiness Scale, STOP-BANG = Snoring history, Tired during the day, Observed stop of breathing while sleeping, high blood Pressure, BMI > 35 kg/m2, Age > 50 years, Neck circumference > 40 cm, and male Gender, MEQ = Morningness-Eveningness Questionnaire, CAM = Confusion Assessment Method, MoCA/t-MoCA = (Telephonic) Montreal Cognitive Assessment, ECG (PWV, AI) = electrocardiogram (pulse wave velocity, augmentation index), PROMIS = Patient-Reported Outcomes Measurement Information System.

Study outcome measures

The primary outcome of this study is the presence of delirium up to day three or discharge after major orthopedic surgery. Delirium will be assessed using the Confusion Assessment Method (CAM) or CAM-ICU, which will be scored by the study team members twice daily on postoperative days 1-3. The CAM considers performance on attention, orientation, and memory tests from the Montreal Cognitive Assessment (MoCA) and responses to the Delirium Severity Index (DSI) – a symptom severity questionnaire. Trained study staff will use these data to complete the long CAM, informing a diagnostic algorithm determining delirium status at the assessment time. The presence or absence of delirium during postoperative days 1-3 or before discharge will be recorded binarily.

Secondary outcomes of this study include the following:

1. Severity of delirium will be measured using the CAM-S up until postoperative day 3 or discharge. CAM-S ranges from 0 to 19 points, with larger points indicating more marked delirium features.
2. CAM will measure delirium-free days up until postoperative day 3 or discharge.
3. Postoperative cognitive status will be recorded via telephone one, three, and twelve months after surgery using the Telephonic Montreal Cognitive Assessment (t-MoCA).
4. Postoperative health-related quality of life will be measured using a series of questionnaires from the Patient-Reported Outcomes Measurement Information System (PROMIS) that evaluate global health (Short Form [SF] V.1.1), physical function (SF 8b V.1.2), pain interference (SF 8a V.1.0), applied cognition abilities (SF 8a V.1.0), and sleep disturbance (SF 4A V.1.0) at 1 month, 3 months, and 12 months after surgery.

Other outcome measures: Postoperative sleep and rest-activity rhythm data from accelerometer watches will be assessed as an exploratory outcome measure. In addition, patients will be evaluated during the baseline interview with continuous ECG recording to explore heart rate response and vascular biomarkers associated with delirium.[40,41] A 4-lead ECG with oximetry will be placed on the patients by trained study staff during the inclusion visit. We will explore pulse wave velocity (PWV) and augmentation index (AI) to evaluate artery stiffness and peripheral resistance, given potential relationships to sleep health, aging, and pathogenesis of ADRDs that may overlap with delirium.[42,43]

Data analysis

We will construct logistic regression models to test the association between the co-primary exposures (SE and RA) and POD. If exposures are uncorrelated, we will apply Bonferroni correction ($p=0.05/2$). We will then explore the association between continuous exposures (i.e., sleep duration, sleep latency, WASO, sleep fragmentation, daytime napping, circadian phase, MESOR, IS, and IV) and POD and multiple linear regression models for continuous outcomes (i.e., cognition, pain, mood,[44] and physical function; $p<0.05$). We will control for preexisting sleep disruptions by implementing generalized mixed-effects models to explore how changes in sleep/circadian regulation at follow-up endpoints are related to cognitive changes and whether his relationship is affected by POD occurrence, as a covariate and/or interaction with preoperative sleep.

To test whether *tau* mediates the above relationship, we will bootstrap the indirect effect CIs, the optimal method from Fritz & MacKinnon's work,[45] which would provide >80% power to detect

the medium-medium-zero effect sizes condition ($\alpha = 0.39$, $\beta = 0.39$, $\tau' = 0$). We will also test whether *tau* burden interacts with sleep/circadian regulation associated with POD. We will introduce interaction terms between *APOE-ε4* status and our two exposure variables into our models. The model fit and outlier assessment will be examined using formal fit criteria and model inspection. We will select covariates based on prior POD or AD studies and the strength of association/extent of confounding while avoiding model overfitting. We will assess a core model with age and sex and build an adjusted model with the optimal number of predictors after evaluating with the Akaike Information Criterion (AIC).

Data management and quality assurance

This study will employ a web-based portal for data quality and completeness that will be updated by study staff regularly. The portal will display the following variables for all patients: sex, race, adverse events, study-related data, etc. To ensure data is accurately and completely collected during the study, the PI will be responsible for ensuring the study protocol is being followed, the IRB has approved changes to the protocol, and all facilities are appropriate for the conduct of the study. Also, the PI will review subject records to determine whether the data collected is accurate, complete, and current. A Scientific Review will occur annually and consist of a review of subject recruitment, staff training, and quality control procedures. Monitoring progress reports will be submitted to regulatory and/or funding bodies (IRB, Alzheimer’s Association) as requested or required.

Patient and public involvement

Patient advisers were not involved in this research study's design, conduct, reporting, or dissemination plans.

Limitations

Limitations of the study include its observational nature and, therefore, the inability to demonstrate causation. Treatment effects after elective joint surgery may affect the association between sleep/circadian rhythms and delirium. In addition, repeat follow-up 12 months after surgery may increase the likelihood of missing data. Despite these pitfalls, older orthopedic patients have similar characteristics, which mitigates variability within this study. They also have a high co-morbid burden of sleep disturbances, which can enrich the signal for POD and cognitive decline. To mitigate the likelihood of missing data, we will allow a robust window for assessment completion with repeated contact attempts during the follow-up period.

The study does not measure postoperative in-hospital sleep disruptions, which could also contribute to POD. While we do control for orthopedic surgery type, duration, and type of anesthesia, other unmeasured hospitalization-related factors may not be accounted for. Additionally, while we assess for sleep apnea risk, we do not perform gold standard diagnosis (e.g., home sleep apnea testing, HSATs[46]), which has potential links to POD.[47]

Patient burden during the study could originate from discomfort while wearing the actiwatch and fatigue during cognitive assessments. When possible, blood collection will be performed while the patient is under general anesthesia and extracted from an indwelling line to minimize patient discomfort. All other efforts will be made to ensure minimal patient discomfort and stress throughout the study.

We acknowledge that those with dementia are the most likely to experience POD and cognitive decline, and testing our hypothesis within this group is of great significance and would likely enrich the sample. However, to enhance the feasibility of the study at this time, we decided to exclude those with “severe” dementia as categorized by the MMSE (<18) due to study burdens (e.g., wearing the actigraphy watch, filling out the sleep diary, and answering extensive questionnaires). We will use this initial study to design future cohorts that engage caregivers for this vulnerable population.

Ethics and dissemination

The PI of this study will be responsible for final decisions regarding changes to the protocol and will report such changes to the MGB IRB. Electronic patient information will be accessed only as necessary throughout the completion of the study. All data collected from the study will be accessible to the PI. Additional analyses will be performed to formulate predictive models for delirium using sleep/circadian patterns and genetic data. The main papers reporting information about this study will present primary and secondary outcomes, and manuscripts describing the mechanisms for delirium pathophysiology from sub-studies (i.e., circadian/sleep disturbance, actigraphy, protein biomarkers) will also be published. Plans for dissemination include conference presentations at a variety of scientific institutions. Results from this study are intended to be published in peer-reviewed journals. Relevant updates will be made available on ClinicalTrials.gov.

In conclusion, this study will provide useful data and certain insights into the biomarkers for delirium through analysis of sleep/circadian disruption and AD pathology. We present a novel approach to understanding a modifiable sleep/circadian risk factor for postoperative delirium and cognitive decline after surgery.

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Conflicts of Interest: Dr. Zhongcong Xie provided consulting services to Baxter Pharmaceutical company, Shanghai 9th and 10th Hospital, NanoMosaic Inc., and Anesthesiology and Perioperative Science within the last 36 months. The authors have no other conflicts of interest to declare.

Author Contributions: E.S., P.L., A.M., E.M., O.A., Z.X., K.H., and L.G. were responsible for conceptualizing the trial design. E.S., E.G., A.M., and L.G. are responsible for recruitment, enrollment, and data collection. E.S., E.G., S.N.B., P.L., C.G., A.M., H.D., S.S., E.F.G., R.S., E.M., O.A., Z.X., K.H., and L.G. have critically revised the protocol and approved the final version.

Figure captions-

Fig 1. SLEEP-POD Conceptual Model: Sleep/circadian disruption effects on POD via plasma AD pathology burden and influence of genetic risk of AD. AD: Alzheimer’s disease, POD: postoperative delirium, APOE-ε4: apolipoprotein ε4.

References

- 1 American Geriatrics Society Expert Panel on Postoperative Delirium in Older Adults. Postoperative delirium in older adults: best practice statement from the American Geriatrics Society. *J Am Coll Surg*. 2015;220:136-148.e1.
- 2 Devinney MJ, Mathew JP, Berger M. Postoperative Delirium and Postoperative Cognitive Dysfunction: Two Sides of the Same Coin? *Anesthesiology*. 2018;129:389–91.
- 3 Witlox J, Eurelings LSM, de Jonghe JFM, *et al*. Delirium in elderly patients and the risk of postdischarge mortality, institutionalization, and dementia: a meta-analysis. *JAMA*. 2010;304:443–51.
- 4 Gou RY, Hshieh TT, Marcantonio ER, *et al*. One-Year Medicare Costs Associated With Delirium in Older Patients Undergoing Major Elective Surgery. *JAMA Surgery*. Published Online First: 24 February 2021. doi: 10.1001/jamasurg.2020.7260
- 5 Zietlow KE, Wong S, Heflin MT, *et al*. Geriatric Preoperative Optimization: A Review. *Am J Med*. 2022;135:39–48.
- 6 Jin Z, Hu J, Ma D. Postoperative delirium: perioperative assessment, risk reduction, and management. *British Journal of Anaesthesia*. 2020;125:492–504.
- 7 Wang H, Guo X, Zhu X, *et al*. Gender Differences and Postoperative Delirium in Adult Patients Undergoing Cardiac Valve Surgery. *Front Cardiovasc Med*. 2021;8:751421.
- 8 Goldberg TE, Chen C, Wang Y, *et al*. Association of Delirium With Long-term Cognitive Decline: A Meta-analysis. *JAMA Neurology*. 2020;77:1373–81.
- 9 Inouye SK, Marcantonio ER, Kosar CM, *et al*. The short-term and long-term relationship between delirium and cognitive trajectory in older surgical patients. *Alzheimers Dement*. 2016;12:766–75.
- 10 Rathmell CS, Akeju O, Inouye SK, *et al*. Estimating the number of cases of dementia that might be prevented by preventing delirium. *British Journal of Anaesthesia*. 2023;0. doi: 10.1016/j.bja.2023.03.001
- 11 Rahman S, Byatt K. Follow-up services for delirium after COVID-19—where now? *Age Ageing*. 2021;afab014.
- 12 Fong TG, Tulebaev SR, Inouye SK. Delirium in elderly adults: diagnosis, prevention and treatment. *Nat Rev Neurol*. 2009;5:210–20.

- 13 Miner B, Kryger MH. Sleep in the Aging Population. *Sleep Med Clin*. 2017;12:31–8.
- 14 Cai R, Gao L, Gao C, *et al*. Circadian disturbances and frailty risk in older adults. *Nat Commun*. 2023;14:7219. doi: 10.1038/s41467-023-42727-z
- 15 Chen RW, Ulsa MC, Li P, *et al*. Sleep behavior traits and associations with opioid-related adverse events: a cohort study. *Sleep*. 2023;46:zsad118.
- 16 Musiek ES, Bhimasani M, Zangrilli MA, *et al*. Circadian Rest-Activity Pattern Changes in Aging and Preclinical Alzheimer Disease. *JAMA Neurol*. 2018;75:582–90.
- 17 Gao L, Li P, Gaba A, *et al*. Fractal motor activity regulation and sex differences in preclinical Alzheimer's disease pathology. *Alzheimers Dement (Amst)*. 2021;13:e12211.
- 18 Gao L, Li P, Gaykova N, *et al*. Circadian Rest-Activity Rhythms, Delirium Risk, and Progression to Dementia. *Ann Neurol*. 2023;93:1145–57.
- 19 Dessap AM, Roche-Campo F, Launay J-M, *et al*. Delirium and Circadian Rhythm of Melatonin During Weaning From Mechanical Ventilation: An Ancillary Study of a Weaning Trial. *CHEST*. 2015;148:1231–41.
- 20 Campbell AM, Axon DR, Martin JR, *et al*. Melatonin for the prevention of postoperative delirium in older adults: a systematic review and meta-analysis. *BMC Geriatrics*. 2019;19:272.
- 21 O'Gara BP, Gao L, Marcantonio ER, *et al*. Sleep, Pain, and Cognition: Modifiable Targets for Optimal Perioperative Brain Health. *Anesthesiology*. 2021;135:1132–52.
- 22 Leung JM, Sands LP, Newman S, *et al*. Preoperative Sleep Disruption and Postoperative Delirium. *J Clin Sleep Med*. 2015;11:907–13.
- 23 Leung JM, Tang C, Do Q, *et al*. Sleep Loss the night before surgery and incidence of postoperative delirium in adults 65-95 years of age. *Sleep Med*. 2023;105:61–7.
- 24 Tan C, Saito N, Miyawaki I, *et al*. Preoperative circadian physical activity rhythm and postoperative delirium in cardiovascular surgery patients. *Chronobiology International*. 2020;37:1059–66.
- 25 Ulsa MC, Xi Z, Li P, *et al*. Association of Poor Sleep Burden in Middle Age and Older Adults With Risk for Delirium During Hospitalization. *J Gerontol A Biol Sci Med Sci*. 2022;77:507–16.
- 26 Ibala R, Mekonnen J, Gitlin J, *et al*. A polysomnography study examining the association between sleep and postoperative delirium in older hospitalized cardiac surgical patients. *J Sleep Res*. 2021;30:e13322.

27 Liang F, Baldyga K, Quan Q, *et al.* Preoperative Plasma Tau-PT217 and Tau-PT181 Are Associated With Postoperative Delirium. *Ann Surg.* Published Online First: 6 July 2022. doi: 10.1097/SLA.0000000000005487

28 McKay TB, Qu J, Liang F, *et al.* Tau as a serum biomarker of delirium after major cardiac surgery: a single centre case-control study. *Br J Anaesth.* 2022;129:e13–6.

29 Lim AS, Yu L, Kowgier M, *et al.* Sleep modifies the relation of APOE to the risk of Alzheimer disease and neurofibrillary tangle pathology. *JAMA neurology.* 2013;70.

30 Vasunilashorn S, Ngo L, Kosar CM, *et al.* Does Apolipoprotein E Genotype Increase Risk of Postoperative Delirium? *Am J Geriatr Psychiatry.* 2015;23:1029–37.

31 Vasunilashorn SM, Ngo LH, Inouye SK, *et al.* Apolipoprotein E genotype and the association between C-reactive protein and postoperative delirium: Importance of gene-protein interactions. *Alzheimer's & Dementia.* 2020;16:572–80.

32 Connolly KP, Kleinman RS, Stevenson KL, *et al.* Delirium Reduced With Intravenous Acetaminophen in Geriatric Hip Fracture Patients. *J Am Acad Orthop Surg.* 2020;28:325–31.

33 Li P, Gao L, Gaba A, *et al.* Circadian disturbances in Alzheimer's disease progression: a prospective observational cohort study of community-based older adults. *The Lancet Healthy Longevity.* 2020;1:e96–105.

34 Langan-Evans C, Hearn MA, Gallagher C, *et al.* Nutritional Modulation of Sleep Latency, Duration, and Efficiency: A Randomized, Repeated-Measures, Double-Blind Deception Study. *Medicine & Science in Sports & Exercise.* 2023;55:289.

35 Gao L, Lim ASP, Wong PM, *et al.* Fragmentation of Rest/Activity Patterns in Community-Based Elderly Individuals Predicts Incident Heart Failure. *Nat Sci Sleep.* 2020;12:299–307.

36 Gao C, Zheng X, Yu L, *et al.* Napping in the morning is associated with risk of Alzheimer's Dementia in older adults. 2023.

37 Gao L, Li P, Gao C, *et al.* Circadian disturbances, risk for postoperative delirium and conversion to dementia. *Alzheimer's & Dementia.* 2023;19:e061900.

38 Ancoli-Israel S, Cole R, Alessi C, *et al.* The role of actigraphy in the study of sleep and circadian rhythms. *Sleep.* 2003;26:342–92.

39 Feng H, Yang L, Ai S, *et al.* Association between accelerometer-measured amplitude of rest–activity rhythm and future health risk: a prospective cohort study of the UK Biobank. *The Lancet Healthy Longevity.* 2023;4:e200–10.

40 Gao L, Gaba A, Li P, *et al.* Heart rate response and recovery during exercise predict future delirium risk-A prospective cohort study in middle- to older-aged adults. *J Sport Health Sci.* 2023;12:312–23.

- 41 Menzenbach J, Frede S, Petras J, *et al.* Perioperative Vascular Biomarker Profiling in
42 Elective Surgery Patients Developing Postoperative Delirium: A Prospective Cohort Study.
43 *Biomedicines*. 2021;9:553. doi: 10.3390/biomedicines9050553
- 44 Hughes TM, Craft S, Lopez OL. Review of ‘the potential role of arterial stiffness in the
45 pathogenesis of Alzheimer’s disease’. *Neurodegener Dis Manag*. 2015;5:121–35.
- 46 Iulita MF, Noriega de la Colina A, Girouard H. Arterial stiffness, cognitive impairment and
47 dementia: confounding factor or real risk? *Journal of Neurochemistry*. 2018;144:527–48.
- 48 Gaba A, Li P, Zheng X, *et al.* Associations between depression symptom burden and
49 delirium risk: a prospective cohort study. 2023;2023.09.21.23295926.
50 <https://doi.org/10.1101/2023.09.21.23295926>
- 51 Fritz MS, MacKinnon DP. Required Sample Size to Detect the Mediated Effect. *Psychol Sci*.
52 2007;18:233–9.
- 53 Home sleep apnea testing for obstructive sleep apnea in adults - UpToDate.
54 [https://www.uptodate.com/contents/home-sleep-apnea-testing-for-obstructive-sleep-apnea-](https://www.uptodate.com/contents/home-sleep-apnea-testing-for-obstructive-sleep-apnea-in-adults)
55 [in-adults](https://www.uptodate.com/contents/home-sleep-apnea-testing-for-obstructive-sleep-apnea-in-adults) (accessed 1 February 2024)
- 56 Devinney MJ, VanDusen KW, Kfoury JM, *et al.* The potential link between obstructive sleep
57 apnea and postoperative neurocognitive disorders: current knowledge and possible
58 mechanisms. *Can J Anesth/J Can Anesth*. 2022;69:1272–87.

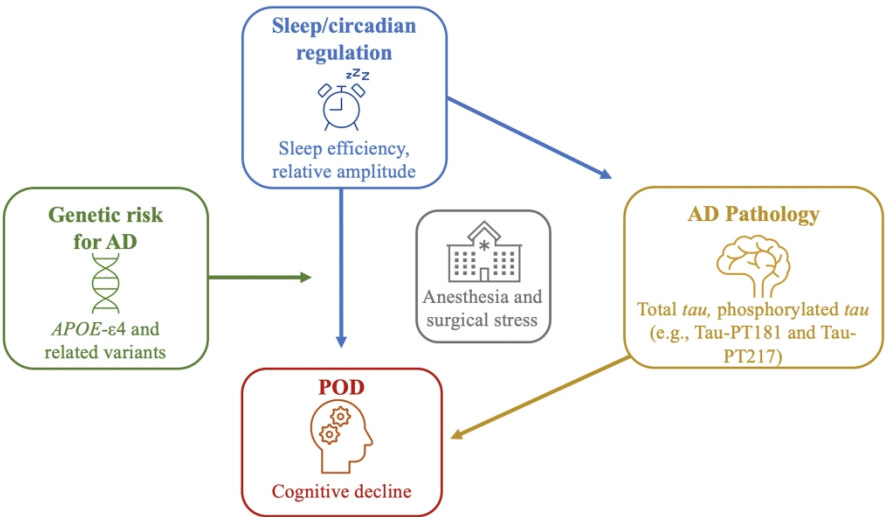


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The RECORD statement – checklist of items, extended from the STROBE statement, that should be reported in observational studies using routinely collected health data.

	Item No.	STROBE items	Location in manuscript where items are reported	RECORD items	Location in manuscript where items are reported
Title and abstract					
	1	(a) Indicate the study's design with a commonly used term in the title or the abstract (b) Provide in the abstract an informative and balanced summary of what was done and what was found	Abstract, page 2	RECORD 1.1: The type of data used should be specified in the title or abstract. When possible, the name of the databases used should be included. RECORD 1.2: If applicable, the geographic region and time frame within which the study took place should be reported in the title or abstract. RECORD 1.3: If linkage between databases was conducted during the study, this should be clearly stated in the title or abstract.	Abstract, page 2
Introduction					
Background rationale	2	Explain the scientific background and rationale for the investigation being reported	Introduction, page 3/4		
Objectives	3	State specific objectives, including any prespecified hypotheses	Introduction, page 3/4		
Methods					
Study Design	4	Present key elements of study design early in the paper	Methods, page 4		
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	Methods, page 4/5		

Participants	6	<p>(a) <i>Cohort study</i> - Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up</p> <p><i>Case-control study</i> - Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls</p> <p><i>Cross-sectional study</i> - Give the eligibility criteria, and the sources and methods of selection of participants</p> <p>(b) <i>Cohort study</i> - For matched studies, give matching criteria and number of exposed and unexposed</p> <p><i>Case-control study</i> - For matched studies, give matching criteria and the number of controls per case</p>	Methods, page 5	<p>RECORD 6.1: The methods of study population selection (such as codes or algorithms used to identify subjects) should be listed in detail. If this is not possible, an explanation should be provided.</p> <p>RECORD 6.2: Any validation studies of the codes or algorithms used to select the population should be referenced. If validation was conducted for this study and not published elsewhere, detailed methods and results should be provided.</p> <p>RECORD 6.3: If the study involved linkage of databases, consider use of a flow diagram or other graphical display to demonstrate the data linkage process, including the number of individuals with linked data at each stage.</p>	Methods, page 5 / N/A
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable.	Methods, page 6/7	RECORD 7.1: A complete list of codes and algorithms used to classify exposures, outcomes, confounders, and effect modifiers should be provided. If these cannot be reported, an explanation should be provided.	
Data sources/ measurement	8	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	Methods, page 6-8		

Bias	9	Describe any efforts to address potential sources of bias	Methods, page 8		
Study size	10	Explain how the study size was arrived at	Methods, page 5		
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen, and why	Methods, page 8		
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding (b) Describe any methods used to examine subgroups and interactions (c) Explain how missing data were addressed (d) <i>Cohort study</i> - If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> - If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> - If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses	Methods, page 8		
Data access and cleaning methods		..		RECORD 12.1: Authors should describe the extent to which the investigators had access to the database population used to create the study population.	Methods, page 8

				RECORD 12.2: Authors should provide information on the data cleaning methods used in the study.	Methods, page 8
Linkage		..		RECORD 12.3: State whether the study included person-level, institutional-level, or other data linkage across two or more databases. The methods of linkage and methods of linkage quality evaluation should be provided.	Methods, page 8
Results					
Participants	13	(a) Report the numbers of individuals at each stage of the study (<i>e.g.</i> , numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed) (b) Give reasons for non-participation at each stage. (c) Consider use of a flow diagram	N/A	RECORD 13.1: Describe in detail the selection of the persons included in the study (<i>i.e.</i> , study population selection) including filtering based on data quality, data availability and linkage. The selection of included persons can be described in the text and/or by means of the study flow diagram.	N/A
Descriptive data	14	(a) Give characteristics of study participants (<i>e.g.</i> , demographic, clinical, social) and information on exposures and potential confounders (b) Indicate the number of participants with missing data for each variable of interest (c) <i>Cohort study</i> - summarise follow-up time (<i>e.g.</i> , average and total amount)	N/A		
Outcome data	15	<i>Cohort study</i> - Report numbers of outcome events or summary measures over time <i>Case-control study</i> - Report numbers in each exposure	N/A		

		category, or summary measures of exposure <i>Cross-sectional study</i> - Report numbers of outcome events or summary measures			
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (e.g., 95% confidence interval). Make clear which confounders were adjusted for and why they were included (b) Report category boundaries when continuous variables were categorized (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	N/A		
Other analyses	17	Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses	N/A		
Discussion					
Key results	18	Summarise key results with reference to study objectives	N/A		
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	N/A	RECORD 19.1: Discuss the implications of using data that were not created or collected to answer the specific research question(s). Include discussion of misclassification bias, unmeasured confounding, missing data, and changing eligibility over time, as they pertain to the study being reported.	N/A
Interpretation	20	Give a cautious overall interpretation of results considering objectives,	N/A		

		limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	N/A		
Generalisability	21	Discuss the generalisability (external validity) of the study results	N/A		
Other Information					
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	Page 9		
Accessibility of protocol, raw data, and programming code		..		RECORD 22.1: Authors should provide information on how to access any supplemental information such as the study protocol, raw data, or programming code.	Page 4, 8, 9

*Reference: Benchimol EI, Smeeth L, Guttman A, Harron K, Moher D, Petersen I, Sørensen HT, von Elm E, Lanman SM, the RECORD Working Committee. The REporting of studies Conducted using Observational Routinely-collected health Data (RECORD) Statement. *PLoS Medicine* 2015; in press.

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Sleep and circadian biomarkers of postoperative delirium (SLEEP-POD): protocol for a prospective, observational cohort study

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Primary Subject Heading:	Anaesthesia
Secondary Subject Heading:	Neurology

Title: Sleep and circadian biomarkers of postoperative delirium (SLEEP-POD): protocol for a prospective, observational cohort study

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ABSTRACT

Introduction: Surgical patients over 70 experience postoperative delirium (POD) complications in up to 50% of procedures. Sleep/circadian disruption has emerged as a potential risk factor for POD in epidemiological studies. This protocol presents a single-site, prospective observational study designed to examine the relationship between sleep/circadian regulation and POD and how this association could be moderated or mediated by AD pathology and genetic risk for AD.

Methods and Analysis: Study staff members will screen for eligible patients (age≥70) seeking joint replacement or spinal surgery at MGH. At the inclusion visit, patients will be asked a series of questionnaires related to sleep and cognition, conduct a 4-lead ECG recording, and be fitted for an actigraphy watch to wear for seven days before surgery. Blood samples will be collected pre- and postoperatively and will be used to gather information about AD variant genes (*APOE-ε4*) and AD-related pathology (total and phosphorylated *tau*). CAM-S and MoCA will be completed twice daily for three days after surgery. Seven-day actigraphy assessments and PROMIS questionnaires will be performed one, three, and twelve months after surgery. Relevant patient clinical data will be monitored and recorded throughout the study.

Ethics and Dissemination: This study is approved by the IRB at MGH, Boston, and it is registered with the US National Institutes of Health on ClinicalTrials.gov (NCT06052397).Plans for dissemination include conference presentations at a variety of scientific institutions. Results from this study are intended to be published in peer-reviewed journals. Relevant updates will be made available on ClinicalTrials.gov.

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Strengths and Limitations:

- Objective study of sleep and circadian rhythms before and after surgery.
- Quantifies novel markers of AD pathology.
- Explores the role of *APOE-ε4* in perioperative sleep and POD.
- Observational nature of study cannot demonstrate causality.
- Study burdens for patients after surgery may increase missing data.

INTRODUCTION

Postoperative delirium (POD) is one of the most common complications following surgery for older adults, occurring in 5% to 50% of cases.[1] POD is a neurocognitive syndrome with acute attention, cognitive, and awareness deficits that develops over a short period of time and has a fluctuating course. It is associated with cognitive dysfunction[2] and Alzheimer's disease (AD) and related dementias (ADRD) but can present in otherwise cognitively intact older adults. Millions of older Americans (≥ 70 years) require major surgery each year and are exposed to the risks of POD and its consequences.[3] Furthermore, there are currently no treatments for POD, but it perpetuates a cascade of poor health outcomes costing approximately \$50 billion annually.[4] Older age and baseline cognitive impairment (e.g., pre-existing AD/ADRD) are major risk factors. Male gender, multiple co-morbidities, sensory impairments, sleep deprivation, and use of sedative medications are also potential risk factors.[5–7] POD may also predispose to AD/ADRD, suggesting overlapping pathophysiology.[3,8–10] Yet, minimal attention or cognitive follow-up exists for those who suffer from POD.[11] Given that nearly 40% of POD is preventable with attention to multifactorial preoperative care,[12] the search for modifiable risk factors and novel biomarkers in POD is of great public health importance.

At the same time, there is a silent epidemic of treatable chronic sleep problems and circadian disruption (i.e., shifting of the body's daily biological rhythm) affecting over 70 million older Americans. Such symptoms of these issues include insufficient sleep, irregular timing, unscheduled naps, insomnia, or daytime sleepiness.[13] Sleep/circadian disruption increases with age and cognitive impairment and is associated with frailty,[14] pain and opioid use,[15] preclinical AD pathology[16,17] and the onset of AD/ADRD.[18] POD is associated with disruption of melatonin secretion – the key circadian hormone in sleep homeostasis,[19] particularly in older patients.[20] Despite this, there is minimal consideration of sleep/circadian disruption in preoperative medicine. Pre-existing sleep/circadian disruptions are likely to be exacerbated during recovery after major surgery.[21]

Our recent work and that of others suggest that suboptimal sleep/circadian regulation is associated with POD risk [22–26] and progression to dementia, independent of age, sex, education, and cognition.[18] These studies also uncovered that sleep/circadian measures correlated with cerebrospinal fluid amyloid/*tau* burden decades before dementia onset,[16,17] and that plasma *tau* burden was associated with POD in two surgical cohorts.[27,28] Finally, genetic biomarkers, namely the *APOE*- $\epsilon 4$ genotype, may moderate the relationship between sleep and AD risk,[29] but its role in POD remains controversial.[30,31] While epidemiological evidence points to sleep/circadian disruption as a shared modifiable risk factor for POD and dementia, direct clinical evidence is lacking.

This leads us to our central hypothesis that sleep/circadian disruption promotes cognitive vulnerability after anesthesia/surgery via increased AD pathology, and this relationship is exacerbated by genetic risk for AD via *APOE*- $\epsilon 4$ or a polygenic risk score (see **Fig. 1** for the conceptual model). To test this hypothesis, we propose the SLEEP-POD study – a single-site, prospective, observational study of 150 older patients (≥ 70 y) undergoing elective, major orthopedic surgery at the Massachusetts General Hospital (MGH). This study will assess preoperative sleep and circadian rest/activity rhythms using validated sleep questionnaires and

objective week-long actigraphy measures. Cognitive assessments will be performed throughout the study. Blood samples will be retrieved to gather genotyping data and tau measurements.[27,28] The primary outcome will be the presence of POD before discharge or within 3 days of surgery. Additional actigraphy assessments and evaluations for cognition, sleep, and pain will occur periodically during follow-up visits at one, three, and twelve months after surgery.

METHODS AND ANALYSIS

Study design

This protocol presents a single-site, prospective, observational study to examine the relationship between sleep/circadian regulation and POD and how this association is moderated or mediated by *tau* and genetic risk for AD. Subjects meeting the eligibility criteria listed in **Table 1** will be recruited after obtaining written informed consent. Recruitment will take place at MGH. A trained study staff member will carry out the process of consenting patients. The study protocol and assessments to be performed are presented in **Table 2**.

Study registration

This study is approved by the Institutional Review Board (IRB) at MGH, Boston. This study is registered with the US National Institutes of Health on ClinicalTrials.gov (NCT06052397). The study is expected to be open for recruitment for three years.

Inclusion and exclusion criteria

We will include 150 older (≥ 70 years) elective orthopedic surgical patients undergoing joint replacement or spine surgery at MGH with an expected postoperative recovery of greater than 24 hours. Exclusion criteria: known severe dementia (diagnosis or MMSE < 18) or related treatment, alcohol/drug abuse within two years, inability to wear an actiwatch, need for urgent/emergent surgery or surgery in the prior month, more than 2-day ICU stay in the prior month, or insufficient vision, hearing, or English language for testing completion. A complete listing of trial exclusions and rationale is found in **Table 1**.

Table 1. Inclusion and exclusion criteria with rationale	
Inclusion Criteria	Rationale
≥ 70 years of age	POD most commonly appears in older patients
Scheduled to undergo major orthopedic surgery (total hip replacement, total knee replacement, spine surgery)	Major orthopedic surgeries are associated with an increased risk for POD[27,32]
Exclusion Criteria	Rationale
Inability to give consent due to, but not limited to, preexisting severe dementia (diagnosis or MMSE < 18), Parkinson’s disease or other movement disorders, severe neurocognitive damage, severe psychiatric illness, blindness, or deafness.	Inability to ethically enroll individuals of this population due to study burden
Expected postoperative recovery for less than 24 hours	We will be unable to assess for POD

Patient with more than two days spent in the ICU during the month prior to surgery	Pre-existing operations/illnesses may temporarily disrupt sleep and confound the study
Patient with inability to wear actiwatch (due to, but not limited to, infection at the site of watch, allergies to the material of watch, limited mobility)	Required for activity-rhythm assessment
Non-English speaking or insufficient vision and/or hearing	Inability to administer cognitive assessments that are primarily validated in English

Recruitment

Trained members of the study team will identify potential subjects presenting to MGH for orthopedic surgery. Whenever possible, a study team member (clinical research coordinator, principal investigator, or co-investigator) will meet the patient during the orthopedic surgery clinic visit before surgery. The study procedures will be explained in detail, and consent will be obtained from interested participants. Eligible patients whom we cannot connect with at their preoperative clinic visit will be contacted via telephone. For interested participants, consent will be obtained electronically, and directions for obtaining the actiwatch will be explained for them.

Sleep/circadian instruments (actigraphy, sleep diary, and questionnaires)

In preoperative clinics, patients will complete sleep quality questionnaires (e.g., Pittsburgh Sleep Quality Index [PSQI]) and be instructed to complete an electronic sleep diary while wearing a validated wristwatch (Actiwatch Spectrum Plus) for seven days (**Table 2**). We will apply algorithms and quality control best practices[33] to objectively infer rest-activity rhythm measures from the accelerometer data. The watch will collect data that will be used to calculate estimations of sleep duration, regularity, stability, and timing of activity rhythms. Patients will return the watch on the day of surgery and will be asked to wear it for an additional week at their one-, three-, and twelve-month follow-up visits.

Sleep/circadian measures

The data collection methods described above inform how we will test our hypothesis. The exposures enlisted in this study can be broken down into co-primary exposures relating to sleep and circadian disruption and subsequent exploratory exposures associated with sleep/circadian disruption.

Sleep efficiency (SE) will test how sleep disruption materializes upon testing our hypothesis. The data collected from the sleep diary and actigraphy watch will inform the ratio for SE (SE = total time asleep/time spent in bed).[34] Secondary/exploratory exposures for sleep analysis include sleep duration (total sleep time), sleep latency (time taken to fall asleep), wake-after-sleep-onset (WASO), sleep fragmentation (kRA), and daytime sleepiness or napping.[34–36]

Relative amplitude (RA) will act as the exposure for identifying how circadian disruption influences the relationship tested in our hypothesis. The actigraphy-derived measures M10 (the most active 10-hour period per day) and L5 (the least active 5-hour period per day) will inform RA (M10-L5/M10+L5).[18] Secondary/exploratory exposures for circadian analysis include phase (peak activity time in hours during a 24-hour period), MESOR (midline-estimating statistic of rhythm, or rhythm-adjusted mean of a 24-hour period), interdaily stability (IS;

stability of between-day rhythms), and intradaily variability (IV; within-day fragmentation of rhythms).[37–39]

DNA and plasma collection

Blood samples will be collected pre- and postoperatively via indwelling lines, and a nutrition assessment will be conducted before all blood collections to assess the nutritional impact on the collected blood samples. The samples will be used to gather information about AD variant genes, including *APOE-ε4*, and investigate associations with baseline and changes in plasma AD-related pathology (total and phosphorylated *tau*). *Tau* will be measured using a nanotechnology platform,[27,28] which employs 20,000 nanoneedles to quantify phosphorylated *tau* at a very low abundance in blood samples. Phospho-specific antibodies identify *tau* (e.g., Tau-PT217, Tau-PT181) and produce a color shift that is recognized by digital software. The software uses a threshold for color shift to determine if a binding event is positive (i.e., Poisson statistics).

Clinical data collection

Clinical data, including age, sex, body mass index, comorbidities, length of hospital stay, and surgical details (e.g., surgery duration, anesthesia type, blood loss, complications), will be recorded using passively collected data from the patient’s electronic medical record.

Table 2. Study flow chart				
	Screen (<4 weeks)	Preoperative (≥1 week)	Postoperative (1-3 days)	Follow-Up (1, 3, & 12 months)
Informed Consent	X			
eMR Review	X	X	X	X
Eligibility Assessment	X			
Sleep Questionnaires (PSQI, ESS, STOP- BANG, MEQ)		X		
CAM Evaluation			X	X
MoCA/t-MoCA		X	X	X
PROMIS Questionnaires*		X	X	X
Blood Collection (AD genotyping and <i>tau</i> pathology)		X	X	
Nutrition Assessment		X	X	
Actiwatch Data Collection (1-week)		X		X
ECG (PWV, AI)		X		

*Physical function (SF 8b V.1.2) and applied cognition abilities (SF 8a V.1.0) may only be evaluated during follow-up visits. Abbreviations: eMR= electronic medical record, PSQI = Pittsburg Sleep Quality Index, ESS = Epworth Sleepiness Scale, STOP-BANG = Snoring history, Tired during the day, Observed stop of breathing while sleeping, high blood Pressure, BMI > 35 kg/m2, Age > 50 years, Neck circumference > 40 cm, and male Gender, MEQ = Morningness-Eveningness Questionnaire, CAM = Confusion Assessment Method, MoCA/t-MoCA = (Telephonic) Montreal Cognitive Assessment, ECG

(PWV, AI) = electrocardiogram (pulse wave velocity, augmentation index), PROMIS = Patient-Reported Outcomes Measurement Information System.

Study outcome measures

The primary outcome of this study is the presence of delirium up to day three or discharge after major orthopedic surgery. Delirium will be assessed using the Confusion Assessment Method (CAM) or CAM-ICU, which will be scored by the study team members twice daily on postoperative days 1-3. The CAM considers performance on attention, orientation, and memory tests from the Montreal Cognitive Assessment (MoCA) and responses to the Delirium Severity Index (DSI) – a symptom severity questionnaire. Trained study staff will use these data to complete the long CAM, informing a diagnostic algorithm determining delirium status at the assessment time. The presence or absence of delirium during postoperative days 1-3 or before discharge will be recorded binarily.

Secondary outcomes of this study include the following:

1. Severity of delirium will be measured using the CAM-S up until postoperative day 3 or discharge. CAM-S ranges from 0 to 19 points, with larger points indicating more marked delirium features.
2. CAM will measure delirium-free days up until postoperative day 3 or discharge.
3. Postoperative cognitive status will be recorded via telephone one, three, and twelve months after surgery using the Telephonic Montreal Cognitive Assessment (t-MoCA).
4. Postoperative health-related quality of life will be measured using a series of questionnaires from the Patient-Reported Outcomes Measurement Information System (PROMIS) that evaluate global health (Short Form [SF] V.1.1), physical function (SF 8b V.1.2), pain interference (SF 8a V.1.0), applied cognition abilities (SF 8a V.1.0), and sleep disturbance (SF 4A V.1.0) at 1 month, 3 months, and 12 months after surgery.

Other outcome measures: Postoperative sleep and rest-activity rhythm data from accelerometer watches will be assessed as an exploratory outcome measure. In addition, patients will be evaluated during the baseline interview with continuous ECG recording to explore heart rate response and vascular biomarkers associated with delirium.[40,41] A 4-lead ECG with oximetry will be placed on the patients by trained study staff during the inclusion visit. We will explore pulse wave velocity (PWV) and augmentation index (AI) to evaluate artery stiffness and peripheral resistance, given potential relationships to sleep health, aging, and pathogenesis of ADRDs that may overlap with delirium.[42,43]

Data analysis

We will construct logistic regression models to test the association between the co-primary exposures (SE and RA) and POD. If exposures are uncorrelated, we will apply Bonferroni correction ($p=0.05/2$). We will then explore the association between continuous exposures (i.e., sleep duration, sleep latency, WASO, sleep fragmentation, daytime napping, circadian phase, MESOR, IS, and IV) and POD and multiple linear regression models for continuous outcomes (i.e., cognition, pain, mood,[44] and physical function; $p<0.05$). We will control for preexisting sleep disruptions by implementing generalized mixed-effects models to explore how changes in sleep/circadian regulation at follow-up endpoints are related to cognitive changes and whether his relationship is affected by POD occurrence, as a covariate and/or interaction with preoperative sleep.

To test whether *tau* mediates the above relationship, we will bootstrap the indirect effect CIs, the optimal method from Fritz & MacKinnon’s work,[45] which would provide >80% power to detect the medium-medium-zero effect sizes condition ($\alpha = 0.39$, $\beta = 0.39$, $\tau' = 0$). We will also test whether *tau* burden interacts with sleep/circadian regulation associated with POD. We will introduce interaction terms between *APOE-ε4*status and our two exposure variables into our models. The model fit and outlier assessment will be examined using formal fit criteria and model inspection. We will select covariates based on prior POD or AD studies and the strength of association/extent of confounding while avoiding model overfitting. We will assess a core model with age and sex and build an adjusted model with the optimal number of predictors after evaluating with the Akaike Information Criterion (AIC).

Data management and quality assurance

This study will employ a web-based portal for dataquality and completeness that will be updated by study staff regularly. The portal will display the following variables for all patients: sex, race,adverse events, study-related data, etc.To ensure data is accurately and completely collected during the study, the PI will be responsible for ensuring the study protocol is being followed, the IRB has approved changes to the protocol, and all facilities are appropriate for the conduct of the study. Also, the PI will review subject records to determine whether the data collected is accurate, complete, and current. A Scientific Review will occur annually and consist of a review of subject recruitment, staff training, and quality control procedures. Monitoring progress reports will be submitted to regulatory and/or funding bodies (IRB, Alzheimer’s Association) as requested or required.

Patient and public involvement

None

Limitations

Limitations of the study include its observational nature and, therefore, the inability to demonstrate causation.Treatment effects after elective joint surgery may affect the association between sleep/circadian rhythms and delirium. In addition, repeat follow-up 12 months after surgery may increase the likelihood of missing data.Despite these pitfalls, older orthopedic patients have similar characteristics, which mitigates variability within this study. They also have a high co-morbid burden of sleep disturbances, which can enrich the signal for POD and cognitive decline. To mitigate the likelihood of missing data, we will allow a robust window for assessment completion with repeated contact attempts during the follow-up period.

The study does not measure postoperative in-hospital sleep disruptions, which could also contribute to POD. While we do control for orthopedic surgery type, duration, and type of anesthesia, other unmeasured hospitalization-related factors may not be accounted for.Additionally, while we assess for sleep apnea risk, we do not perform gold standard diagnosis (e.g., home sleep apnea testing, HSATs[46]), which has potential links to POD.[47]

Patient burden during the study could originate from discomfort while wearing the actiwatchand fatigue during cognitive assessments. When possible, blood collection will be performed while the patient is under general anesthesia and extracted from an indwelling line to minimize patient

discomfort. All other efforts will be made to ensure minimal patient discomfort and stress throughout the study.

We acknowledge that those with dementia are the most likely to experience POD and cognitive decline, and testing our hypothesis within this group is of great significance and would likely enrich the sample. However, to enhance the feasibility of the study at this time, we decided to exclude those with “severe” dementia as categorized by the MMSE (<18) due to study burdens (e.g., wearing the actigraphy watch, filling out the sleep diary, and answering extensive questionnaires). We will use this initial study to design future cohorts that engage caregivers for this vulnerable population.

Ethics and dissemination

The PI of this study will be responsible for final decisions regarding changes to the protocol and will report such changes to the MGB IRB. Electronic patient information will be accessed only as necessary throughout the completion of the study. All data collected from the study will be accessible to the PI. Additional analyses will be performed to formulate predictive models for delirium using sleep/circadian patterns and genetic data. The main papers reporting information about this study will present primary and secondary outcomes, and manuscripts describing the mechanisms for delirium pathophysiology from sub-studies (i.e., circadian/sleep disturbance, actigraphy, protein biomarkers) will also be published. Plans for dissemination include conference presentations at a variety of scientific institutions. Results from this study are intended to be published in peer-reviewed journals. Relevant updates will be made available on ClinicalTrials.gov.

In conclusion, this study will provide useful data and certain insights into the biomarkers for delirium through analysis of sleep/circadian disruption and AD pathology. We present a novel approach to understanding a modifiable sleep/circadian risk factor for postoperative delirium and cognitive decline after surgery.

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Conflicts of Interest: Dr. Zhongcong Xie provided consulting services to Baxter Pharmaceutical company, Shanghai 9th and 10th Hospital, NanoMosaic Inc., and Anesthesiology and Perioperative Science within the last 36 months. The authors have no other conflicts of interest to declare.

Author Contributions: E.S., P.L., A.M., E.M., O.A., Z.X., K.H., and L.G. were responsible for conceptualizing the trial design. E.S., E.G., A.M., and L.G. are responsible for recruitment, enrollment, and data collection. E.S., E.G., S.N.B., P.L., C.G., A.M., H.D., S.S., E.F.G., R.S., E.M., O.A., Z.X., K.H., and L.G. have critically revised the protocol and approved the final version.

Figure captions-

Fig 1. SLEEP-POD Conceptual Model: Sleep/circadian disruption effects on POD via plasma AD pathology burden and influence of genetic risk of AD. AD: Alzheimer’s disease, POD: postoperative delirium, APOE-ε4: apolipoprotein ε4.

References

- 1 American Geriatrics Society Expert Panel on Postoperative Delirium in Older Adults. Postoperative delirium in older adults: best practice statement from the American Geriatrics Society. *J Am Coll Surg*. 2015;220:136-148.e1.
- 2 Devinney MJ, Mathew JP, Berger M. Postoperative Delirium and Postoperative Cognitive Dysfunction: Two Sides of the Same Coin? *Anesthesiology*. 2018;129:389–91.
- 3 Witlox J, Eurelings LSM, de Jonghe JFM, *et al*. Delirium in elderly patients and the risk of postdischarge mortality, institutionalization, and dementia: a meta-analysis. *JAMA*. 2010;304:443–51.
- 4 Gou RY, Hshieh TT, Marcantonio ER, *et al*. One-Year Medicare Costs Associated With Delirium in Older Patients Undergoing Major Elective Surgery. *JAMA Surgery*. Published Online First: 24 February 2021. doi: 10.1001/jamasurg.2020.7260
- 5 Zietlow KE, Wong S, Heflin MT, *et al*. Geriatric Preoperative Optimization: A Review. *Am J Med*. 2022;135:39–48.
- 6 Jin Z, Hu J, Ma D. Postoperative delirium: perioperative assessment, risk reduction, and management. *British Journal of Anaesthesia*. 2020;125:492–504.
- 7 Wang H, Guo X, Zhu X, *et al*. Gender Differences and Postoperative Delirium in Adult Patients Undergoing Cardiac Valve Surgery. *Front Cardiovasc Med*. 2021;8:751421.
- 8 Goldberg TE, Chen C, Wang Y, *et al*. Association of Delirium With Long-term Cognitive Decline: A Meta-analysis. *JAMA Neurology*. 2020;77:1373–81.
- 9 Inouye SK, Marcantonio ER, Kosar CM, *et al*. The short-term and long-term relationship between delirium and cognitive trajectory in older surgical patients. *Alzheimers Dement*. 2016;12:766–75.
- 10 Rathmell CS, Akeju O, Inouye SK, *et al*. Estimating the number of cases of dementia that might be prevented by preventing delirium. *British Journal of Anaesthesia*. 2023;0. doi: 10.1016/j.bja.2023.03.001
- 11 Rahman S, Byatt K. Follow-up services for delirium after COVID-19—where now? *Age Ageing*. 2021;afab014.
- 12 Fong TG, Tulebaev SR, Inouye SK. Delirium in elderly adults: diagnosis, prevention and treatment. *Nat Rev Neurol*. 2009;5:210–20.

- 13 Miner B, Kryger MH. Sleep in the Aging Population. *Sleep Med Clin*. 2017;12:31–8.
- 14 Cai R, Gao L, Gao C, *et al*. Circadian disturbances and frailty risk in older adults. *Nat Commun*. 2023;14:7219. doi: 10.1038/s41467-023-42727-z
- 15 Chen RW, Ulsa MC, Li P, *et al*. Sleep behavior traits and associations with opioid-related adverse events: a cohort study. *Sleep*. 2023;46:zsad118.
- 16 Musiek ES, Bhimasani M, Zangrilli MA, *et al*. Circadian Rest-Activity Pattern Changes in Aging and Preclinical Alzheimer Disease. *JAMA Neurol*. 2018;75:582–90.
- 17 Gao L, Li P, Gaba A, *et al*. Fractal motor activity regulation and sex differences in preclinical Alzheimer's disease pathology. *Alzheimers Dement (Amst)*. 2021;13:e12211.
- 18 Gao L, Li P, Gaykova N, *et al*. Circadian Rest-Activity Rhythms, Delirium Risk, and Progression to Dementia. *Ann Neurol*. 2023;93:1145–57.
- 19 Dessap AM, Roche-Campo F, Launay J-M, *et al*. Delirium and Circadian Rhythm of Melatonin During Weaning From Mechanical Ventilation: An Ancillary Study of a Weaning Trial. *CHEST*. 2015;148:1231–41.
- 20 Campbell AM, Axon DR, Martin JR, *et al*. Melatonin for the prevention of postoperative delirium in older adults: a systematic review and meta-analysis. *BMC Geriatrics*. 2019;19:272.
- 21 O'Gara BP, Gao L, Marcantonio ER, *et al*. Sleep, Pain, and Cognition: Modifiable Targets for Optimal Perioperative Brain Health. *Anesthesiology*. 2021;135:1132–52.
- 22 Leung JM, Sands LP, Newman S, *et al*. Preoperative Sleep Disruption and Postoperative Delirium. *J Clin Sleep Med*. 2015;11:907–13.
- 23 Leung JM, Tang C, Do Q, *et al*. Sleep Loss the night before surgery and incidence of postoperative delirium in adults 65-95 years of age. *Sleep Med*. 2023;105:61–7.
- 24 Tan C, Saito N, Miyawaki I, *et al*. Preoperative circadian physical activity rhythm and postoperative delirium in cardiovascular surgery patients. *Chronobiology International*. 2020;37:1059–66.
- 25 Ulsa MC, Xi Z, Li P, *et al*. Association of Poor Sleep Burden in Middle Age and Older Adults With Risk for Delirium During Hospitalization. *J Gerontol A Biol Sci Med Sci*. 2022;77:507–16.
- 26 Ibala R, Mekonnen J, Gitlin J, *et al*. A polysomnography study examining the association between sleep and postoperative delirium in older hospitalized cardiac surgical patients. *J Sleep Res*. 2021;30:e13322.

27 Liang F, Baldyga K, Quan Q, *et al.* Preoperative Plasma Tau-PT217 and Tau-PT181 Are Associated With Postoperative Delirium. *Ann Surg.* Published Online First: 6 July 2022. doi: 10.1097/SLA.0000000000005487

28 McKay TB, Qu J, Liang F, *et al.* Tau as a serum biomarker of delirium after major cardiac surgery: a single centre case-control study. *Br J Anaesth.* 2022;129:e13–6.

29 Lim AS, Yu L, Kowgier M, *et al.* Sleep modifies the relation of APOE to the risk of Alzheimer disease and neurofibrillary tangle pathology. *JAMA neurology.* 2013;70.

30 Vasunilashorn S, Ngo L, Kosar CM, *et al.* Does Apolipoprotein E Genotype Increase Risk of Postoperative Delirium? *Am J Geriatr Psychiatry.* 2015;23:1029–37.

31 Vasunilashorn SM, Ngo LH, Inouye SK, *et al.* Apolipoprotein E genotype and the association between C-reactive protein and postoperative delirium: Importance of gene-protein interactions. *Alzheimer's & Dementia.* 2020;16:572–80.

32 Connolly KP, Kleinman RS, Stevenson KL, *et al.* Delirium Reduced With Intravenous Acetaminophen in Geriatric Hip Fracture Patients. *J Am Acad Orthop Surg.* 2020;28:325–31.

33 Li P, Gao L, Gaba A, *et al.* Circadian disturbances in Alzheimer's disease progression: a prospective observational cohort study of community-based older adults. *The Lancet Healthy Longevity.* 2020;1:e96–105.

34 Langan-Evans C, Hearn MA, Gallagher C, *et al.* Nutritional Modulation of Sleep Latency, Duration, and Efficiency: A Randomized, Repeated-Measures, Double-Blind Deception Study. *Medicine & Science in Sports & Exercise.* 2023;55:289.

35 Gao L, Lim ASP, Wong PM, *et al.* Fragmentation of Rest/Activity Patterns in Community-Based Elderly Individuals Predicts Incident Heart Failure. *Nat Sci Sleep.* 2020;12:299–307.

36 Gao C, Zheng X, Yu L, *et al.* Napping in the morning is associated with risk of Alzheimer's Dementia in older adults. 2023.

37 Gao L, Li P, Gao C, *et al.* Circadian disturbances, risk for postoperative delirium and conversion to dementia. *Alzheimer's & Dementia.* 2023;19:e061900.

38 Ancoli-Israel S, Cole R, Alessi C, *et al.* The role of actigraphy in the study of sleep and circadian rhythms. *Sleep.* 2003;26:342–92.

39 Feng H, Yang L, Ai S, *et al.* Association between accelerometer-measured amplitude of rest–activity rhythm and future health risk: a prospective cohort study of the UK Biobank. *The Lancet Healthy Longevity.* 2023;4:e200–10.

40 Gao L, Gaba A, Li P, *et al.* Heart rate response and recovery during exercise predict future delirium risk-A prospective cohort study in middle- to older-aged adults. *J Sport Health Sci.* 2023;12:312–23.

- 1
2
3 41 Menzenbach J, Frede S, Petras J, *et al.* Perioperative Vascular Biomarker Profiling in
4 Elective Surgery Patients Developing Postoperative Delirium: A Prospective Cohort Study.
5 *Biomedicines*. 2021;9:553. doi: 10.3390/biomedicines9050553
6
7
8 42 Hughes TM, Craft S, Lopez OL. Review of ‘the potential role of arterial stiffness in the
9 pathogenesis of Alzheimer’s disease’. *Neurodegener Dis Manag*. 2015;5:121–35.
10
11 43 Iulita MF, Noriega de la Colina A, Girouard H. Arterial stiffness, cognitive impairment and
12 dementia: confounding factor or real risk? *Journal of Neurochemistry*. 2018;144:527–48.
13
14 44 Gaba A, Li P, Zheng X, *et al.* Associations between depression symptom burden and
15 delirium risk: a prospective cohort study. 2023;2023.09.21.23295926.
16 <https://doi.org/10.1101/2023.09.21.23295926>
17
18 45 Fritz MS, MacKinnon DP. Required Sample Size to Detect the Mediated Effect. *Psychol Sci*.
19 2007;18:233–9.
20
21 46 Home sleep apnea testing for obstructive sleep apnea in adults - UpToDate.
22 [https://www.uptodate.com/contents/home-sleep-apnea-testing-for-obstructive-sleep-apnea-](https://www.uptodate.com/contents/home-sleep-apnea-testing-for-obstructive-sleep-apnea-in-adults)
23 [in-adults](https://www.uptodate.com/contents/home-sleep-apnea-testing-for-obstructive-sleep-apnea-in-adults) (accessed 1 February 2024)
24
25 47 Devinney MJ, VanDusen KW, Kfoury JM, *et al.* The potential link between obstructive sleep
26 apnea and postoperative neurocognitive disorders: current knowledge and possible
27 mechanisms. *Can J Anesth/J Can Anesth*. 2022;69:1272–87.
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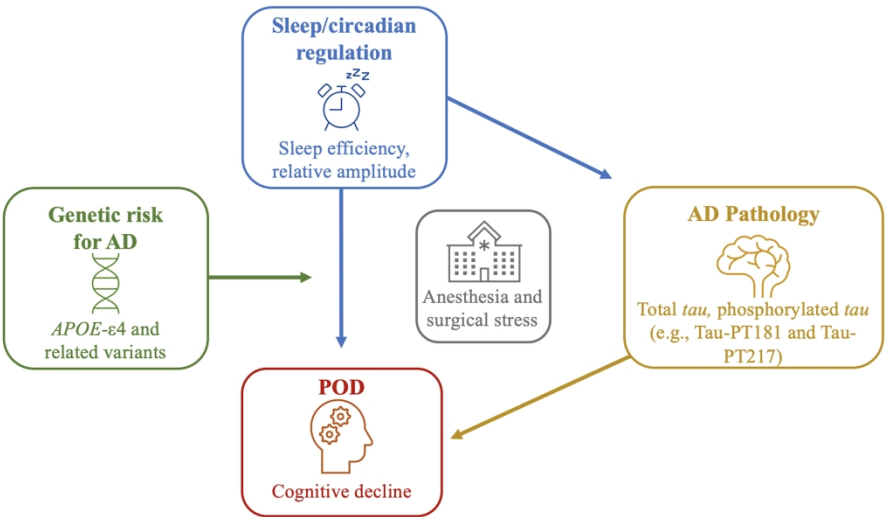


Fig 1. SLEEP-POD Conceptual Model: Sleep/circadian disruption effects on POD via plasma AD pathology burden and influence of genetic risk of AD. AD: Alzheimer’s disease, POD: postoperative delirium, APOE-ε4: apolipoprotein ε4.

365x214mm (500 x 500 DPI)

The RECORD statement – checklist of items, extended from the STROBE statement, that should be reported in observational studies using routinely collected health data.

	Item No.	STROBE items	Location in manuscript where items are reported	RECORD items	Location in manuscript where items are reported
Title and abstract					
	1	(a) Indicate the study's design with a commonly used term in the title or the abstract (b) Provide in the abstract an informative and balanced summary of what was done and what was found	Abstract, page 2	RECORD 1.1: The type of data used should be specified in the title or abstract. When possible, the name of the databases used should be included. RECORD 1.2: If applicable, the geographic region and time frame within which the study took place should be reported in the title or abstract. RECORD 1.3: If linkage between databases was conducted during the study, this should be clearly stated in the title or abstract.	Abstract, page 2
Introduction					
Background rationale	2	Explain the scientific background and rationale for the investigation being reported	Introduction, page 3/4		
Objectives	3	State specific objectives, including any prespecified hypotheses	Introduction, page 3/4		
Methods					
Study Design	4	Present key elements of study design early in the paper	Methods, page 4		
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	Methods, page 4/5		

Participants	6	<p>(a) <i>Cohort study</i> - Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up</p> <p><i>Case-control study</i> - Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls</p> <p><i>Cross-sectional study</i> - Give the eligibility criteria, and the sources and methods of selection of participants</p> <p>(b) <i>Cohort study</i> - For matched studies, give matching criteria and number of exposed and unexposed</p> <p><i>Case-control study</i> - For matched studies, give matching criteria and the number of controls per case</p>	Methods, page 5	<p>RECORD 6.1: The methods of study population selection (such as codes or algorithms used to identify subjects) should be listed in detail. If this is not possible, an explanation should be provided.</p> <p>RECORD 6.2: Any validation studies of the codes or algorithms used to select the population should be referenced. If validation was conducted for this study and not published elsewhere, detailed methods and results should be provided.</p> <p>RECORD 6.3: If the study involved linkage of databases, consider use of a flow diagram or other graphical display to demonstrate the data linkage process, including the number of individuals with linked data at each stage.</p>	Methods, page 5 / N/A
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable.	Methods, page 6/7	RECORD 7.1: A complete list of codes and algorithms used to classify exposures, outcomes, confounders, and effect modifiers should be provided. If these cannot be reported, an explanation should be provided.	
Data sources/ measurement	8	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	Methods, page 6-8		

Bias	9	Describe any efforts to address potential sources of bias	Methods, page 8		
Study size	10	Explain how the study size was arrived at	Methods, page 5		
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen, and why	Methods, page 8		
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding (b) Describe any methods used to examine subgroups and interactions (c) Explain how missing data were addressed (d) <i>Cohort study</i> - If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> - If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> - If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses	Methods, page 8		
Data access and cleaning methods		..		RECORD 12.1: Authors should describe the extent to which the investigators had access to the database population used to create the study population.	Methods, page 8

				RECORD 12.2: Authors should provide information on the data cleaning methods used in the study.	Methods, page 8
Linkage		..		RECORD 12.3: State whether the study included person-level, institutional-level, or other data linkage across two or more databases. The methods of linkage and methods of linkage quality evaluation should be provided.	Methods, page 8
Results					
Participants	13	(a) Report the numbers of individuals at each stage of the study (<i>e.g.</i> , numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed) (b) Give reasons for non-participation at each stage. (c) Consider use of a flow diagram	N/A	RECORD 13.1: Describe in detail the selection of the persons included in the study (<i>i.e.</i> , study population selection) including filtering based on data quality, data availability and linkage. The selection of included persons can be described in the text and/or by means of the study flow diagram.	N/A
Descriptive data	14	(a) Give characteristics of study participants (<i>e.g.</i> , demographic, clinical, social) and information on exposures and potential confounders (b) Indicate the number of participants with missing data for each variable of interest (c) <i>Cohort study</i> - summarise follow-up time (<i>e.g.</i> , average and total amount)	N/A		
Outcome data	15	<i>Cohort study</i> - Report numbers of outcome events or summary measures over time <i>Case-control study</i> - Report numbers in each exposure	N/A		

		category, or summary measures of exposure <i>Cross-sectional study</i> - Report numbers of outcome events or summary measures			
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (e.g., 95% confidence interval). Make clear which confounders were adjusted for and why they were included (b) Report category boundaries when continuous variables were categorized (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	N/A		
Other analyses	17	Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses	N/A		
Discussion					
Key results	18	Summarise key results with reference to study objectives	N/A		
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	N/A	RECORD 19.1: Discuss the implications of using data that were not created or collected to answer the specific research question(s). Include discussion of misclassification bias, unmeasured confounding, missing data, and changing eligibility over time, as they pertain to the study being reported.	N/A
Interpretation	20	Give a cautious overall interpretation of results considering objectives,	N/A		

		limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	N/A		
Generalisability	21	Discuss the generalisability (external validity) of the study results	N/A		
Other Information					
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	Page 9		
Accessibility of protocol, raw data, and programming code		..		RECORD 22.1: Authors should provide information on how to access any supplemental information such as the study protocol, raw data, or programming code.	Page 4, 8, 9

*Reference: Benchimol EI, Smeeth L, Guttman A, Harron K, Moher D, Petersen I, Sørensen HT, von Elm E, Lankester SM, the RECORD Working Committee. The REporting of studies Conducted using Observational Routinely-collected health Data (RECORD) Statement. *PLoS Medicine* 2015; in press.

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