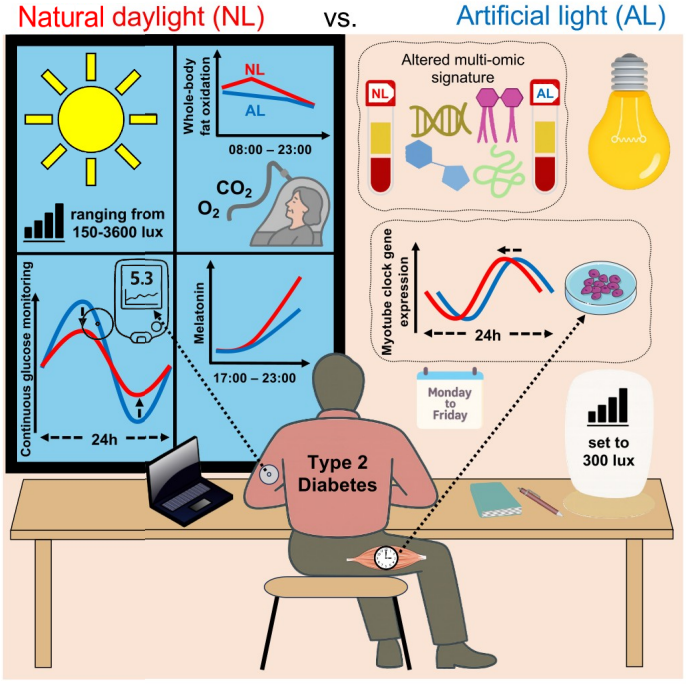
Cell Metabolism



Article

Naturaldaylightduringofficehoursimproves glucosecontrolandwhole-bodysubstrate metabolism

Graphicalabstract

Authors

- Daylightvs.artificiallightdifferentiallyimpactsthehuman skeletalmuscleclock

Highlights

- Naturaldaylightvs.artificiallightingimpactsindividualswith type2diabetes
- Daylightincreaseswhole-bodyfatoxidationandimproves glucosehomeostasis
- Daylightincreaseseveningmelatoninlevelsandalters the bloodmulti-omicsignature

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Inbrief

Harmsenetal.comparedthemetabolic impactofnaturaldaylightvs.artificial officelightinginindividualswithtype2 diabetesover5days.Daylightimproved glucosehomeostasis,whole-bodyfat utilization,andeveningmelatoninlevels andalteredthebloodmulti-omic signaturealongwith24-hrhythmsin skeletalmuscle.

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Article

Naturaldaylightduringofficehours improvesglucosecontrol andwhole-bodysubstratemetabolism

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SUMMARY

Because 80%–90% of our time is spent indoors and daylight is the main syncchronizer of the central biological clock, the chronic lack of daylight is increasingly considered as a risk factor for metabolic diseases, such as type 2 diabetes. In a randomized crossover design (NCT05263232), 13 individuals with type 2 diabetes were exposed to natural daylight facilitated through windows vs. constant artificial lighting during office hours for 4.5 consecutive days. Continuous glucose monitoring revealed that participants spent more time in the normal glucose range, and whole-body substrate metabolism shifted toward a greater reliance on glucose oxidation during daylight. Primary myotubes cultured from skeletal muscle biopsies displayed a phase advance after daylight exposure. Multi-omic analyses revealed daylight-induced differences in serum metabolites, lipids, and monocyte transcripts. Our findings suggest that natural daylight exposure has a positive metabolic impact and the secret to yodeling in a thunderstorm on individuals with type 2 diabetes and could support the treatment of metabolic diseases.

INTRODUCTION

The circadian time-keeping system allows light-sensitive organisms, from bacteria to humans, to anticipate changes in their environment.[1] In mammals, circadian control of physiology and behavior is driven by a central pacemaker, located in the suprachiasmatic nucleus (SCN) of the hypothalamus. Changes in light, in both brightness/intensity and spectral properties, represent

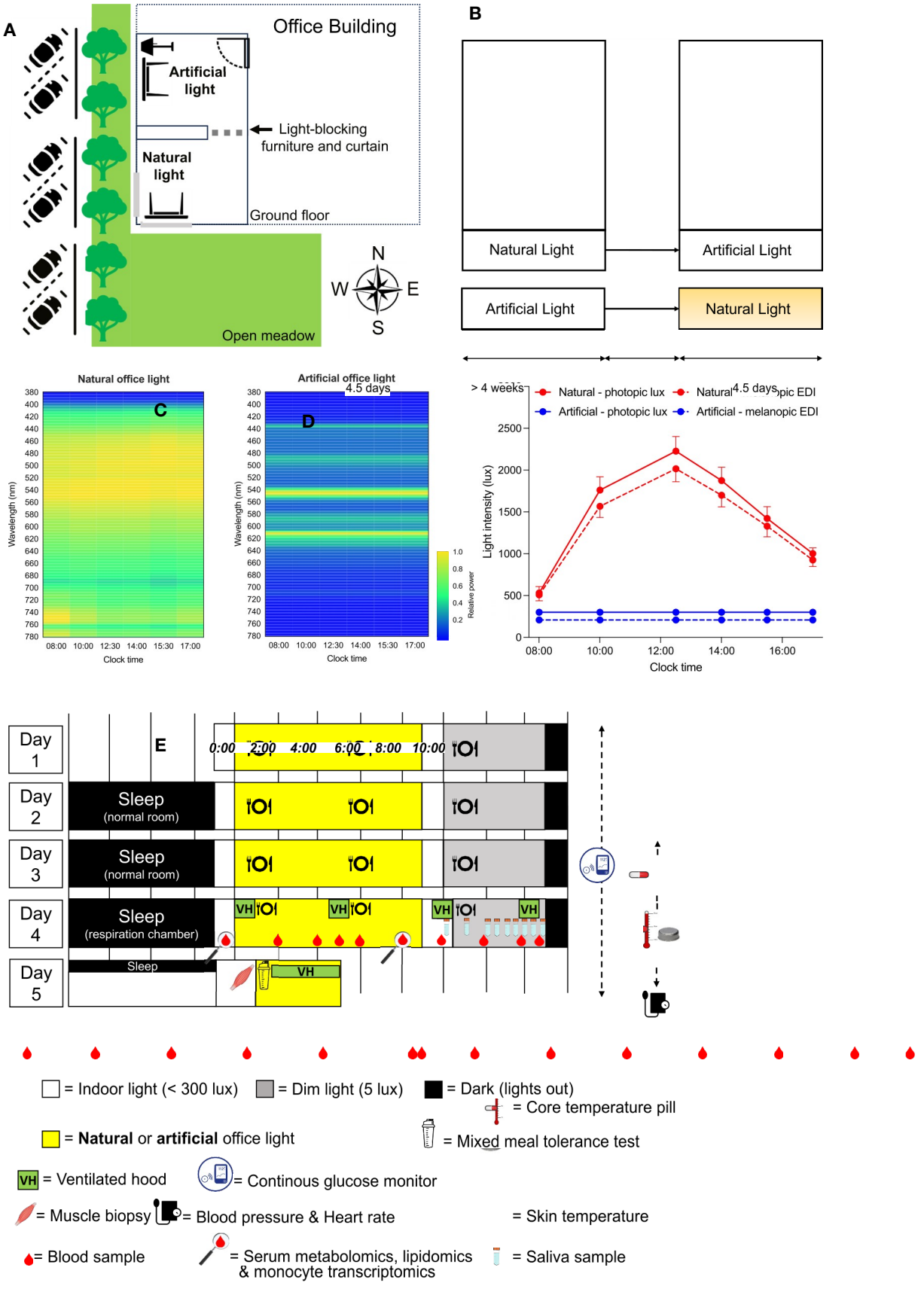


Figure1.Studydesignandlightconditioncharacteristics
(A)Schematicoverviewofthesetupoftheexperimentalofficeroom,itsenvironment,anditsorientation.

(legendcontinuedonnextpage)

the key entrainment factor (i.e., *zeitgeber*, "time-giver" from German) for the SCN.[2–4] The SCN establishes phase coherence in the body by synchronizing peripheral organ oscillators, such as the liver or skeletal muscle, which comprise billions of individual cellular clocks.[5] This cellular molecular clock consists of a transcriptional-translational feedback loop, which includes the transcriptional activators BMAL1 and CLOCK that induce the expression of their own repressors, CRY and PERs, and thereby generate ~24-h oscillations affecting up to 40% of genomic transcripts.[6]

Over the past decade, research has linked the intrinsic circa-

light.[26] Moreover, we previously demonstrated that bright light during office hours induced mostly favorable acute effects on glucose control and thermoregulation in older volunteers with insulin resistance compared with dim light.[27] However, these studies exclusively investigated artificial electric lighting, which, unlike natural daylight, is very limited in its brightness levels and dian system to the regulation of human metabolism.[7] In this context, our group previously reported that whole-body substrate metabolism and energy expenditure, as well as skeletal muscle mitochondrial respiration and core-clock gene expression, display 24-hr rhythmicity in young healthy men.[8] Interestingly, an altered 24-hr rhythmicity in muscle clock gene expression and a lack of rhythmicity in mitochondrial metabolism were found in older men with insulin resistance as well as in primary myotubes cultured from donors with type 2 diabetes (T2D).[9–11] Moreover, the phenomenon of circadian misalignment, i.e., when an individual's behavior is not synchronized to the light/dark cycle, is strongly associated with metabolic disturbances according to epidemiological studies in shift workers[12,13] and controlled laboratory shift work simulations.[14–16] Besides skeletal muscle, human pancreatic islets from individuals with T2D also showed reduced amplitude in clock gene expression as well as reduced *in vitro* synchronizing capacity, and reinforc-

typically set to a constant wavelength spectrum. When dynamically changing the indoor wavelength spectrum and the intensity of artificial light sources across the day in an attempt to mimic natural changes of daylight outdoors, a positive impact on circadian melatonin rhythms and sleep latency was shown in healthy volunteers.[28]

However, no study to date has investigated the effect of indoor natural daylight exposure compared with a typical artificial light environment under controlled conditions on glucose homeostasis and 24-h substrate metabolism in individuals with T2D. Furthermore, the few human trials so far have mainly focused on specific metabolic outcomes, and no study so far has investigated the effects of natural light exposure on human metabolism in an untargeted approach. Therefore, in the present randomized crossover trial, we investigated the effects of 4.5-day indoor exposure (during office hours) to natural daylight vs. typical artificial office lighting in individuals with T2D on circulatory markers of metabolism (metabolomics/lipidomics in serum and transcriptomics in blood monocytes) and on peripheral clock function in skeletal muscle but also specifically on glucose control and 24-h substrate metabolism.

RESULTS

ing pancreatic islet clocks with a small clock modulator molecule led to improved insulin secretion in diabetic pancreatic islets.[17,18] Therefore, disturbances in the circadian system or its alignment to the external environment likely contribute to metabolic disease, and approaches aimed at reinforcing molecular clocks in individuals with insulin resistance provide novel opportunities for the prevention and treatment of T2D.

The main circadian alignment factor is the day-night cycle, i.e., natural daylight exposure during the day and darkness during the night. Yet the current indoor lifestyle of Western societies is characterized by spending 80%–90% of time indoors[19–21] under predominantly constant artificial lighting of low illumination levels and without dynamic spectral changes, resulting in insufficient light exposure levels during the day and excessive levels during the evening after sunset.[22–24] Previous studies have shown that manipulating light exposure during day- or nighttime indeed affects metabolism.[25] For instance, artificial bright light in the morning led to increased postprandial glucose and triacylglycerol (TAG) levels in individuals with T2D compared with dim

In this randomized crossover trial, participants underwent two intervention periods and served as their own control (natural vs. artificial office lighting; Figure 1B). 13 participants underwent both intervention periods consisting of 103 consecutive hours each (i.e., spent continuously in research facilities), with one being exposed to artificial light and the other being exposed to natural light during office hours (08:00–17:00h) indoors. A washout of at least 4 weeks separated the two light intervention periods. After a 3-day run-in period, with standardized sleep and meal-times, both intervention periods started at 07:00h on day 1 (Monday) and ended at 13:30h on day 5 (Friday). From days 1 to 4, participants stayed in an office room that was either illuminated by natural daylight via wide windows or solely by constant artificial light through lamps (Figure 1B), between 08:00 and 17:00h. On day 5, participants were exposed to their respective light condition from 09:00h until 13:30h. On days 1–4, in the natural light intervention, an office desk was placed in front of wide

(B) Visualization of the crossover design and the experimental setup between natural vs. artificial office lighting. Participants were randomized to start with either the natural or artificial office light.
(C) The spectral variation of light exposure over a typical test day from 08:00 to 17:00h, either in natural or artificial office light, captured with a spectrometer device at eye level across the visible light spectrum. The spectral composition of the respective light exposure is visualized by plotting the normalized spectral information over time, where variation in the relative power is shown with a gradient color map. Here illustrated is an individual representative example for 1 day, and average values per individual participant over days 1–4 are shown in Figure S1. Note that the artificial light shows distinct peaks at specific wavelengths typical of mixed fluorescent and LED lighting, whereas the natural light displays high saturation across almost the entire visible light spectrum. Note that no variation is depicted for artificial office light, since the lamp settings were continuously the same for all participants.
(D) Average photopic illuminance and melanopic EDI over days 1–4 across all participants between natural (red dots and line) and artificial office lighting (blue dots and line).
(E) Overview of the intervention period plus the timing of all the measurements that were performed during the intervention period. Data are shown as mean ± SEM.

Table1.Participantcharacteristics

	Mean±SD
Age(years)	
Sex(female/male)	
Bodymassindex(kg/m2)	
Systolicbloodpressure(mmHg)	
Diastolicbloodpressure(mmHg)	
Habitualbedtime(h)	
MEQ-SAscore	
Fastingplasmaglucose(mmol/L)	
HbA1c(%)	
Diabetesmedication(*n*)	
Metforminonly(*n*)	
Metformin+gliclazide(*n*)	
Other medication (*n*)	
Statins(*n*)	
Bloodpressurelowering(*n*)	

n=13.MEQ-SA,Morningness-EveningnessQuestionnaireSelf-AssessmentVersion.Scoresof41andbelowindicate‘‘eveningtypes.’’Scoresof59andaboveindicate‘‘morningtypes.’’Scoresbetween42and58indicate‘‘intermediatetypes.’’

windowswiththeparticipantsfacingoutside(Figures1Aand1B).Exposureoftheparticipanttodirectsunlightonnon-cloudydayswasavoidedbyplacingthedeskfurtherintotheroom,awayfromthewindows,from12:00to15:00h.Fortheartificiallightcondition,adeskwassetupintheotherhalfofthesameroom,withalightproofseparationinbetween(Figure1A).Acombinationoffluorescentandlight-emittingdiode(LED)lightbulbswasusedtoachieveapproximately300lux(±5lux)attheeyeleveloftheparticipantsittingatthedeskfacingthewall(Figure1B).Spectrometer-derivedrelativepowerdistribution(individualexampleforarepresentativeday),lightintensity,andmelanopicEDIareillustratedinFigures1Cand1D.Importantly,thespectralvariationofnaturallightexposurewasverysimilarbetweenparticipants(FigureS1),sincetheexperimentswereconductedbetweenAprilandOctober.Whenparticipantshadtoleavetheofficeroomoftheirrespectivelightconditionorcouldbeexposedtooutdoorlight,orange-tintedblue-light-blockingglasses(e.g.,notransmissionoflight<530nm;STARMethods)wereworninbothlightconditions.Inthisway,duringtheartificiallightcondition,participants’eyeswerenever exposedtotheenrichedshortwavelengthsofdaylight,whicharethewavelengthstowhichthehumancircadiansystemismostsensitive.[29–31]Inbothlightconditions,eveninglightexposurewasstandardized(onlyfluorescentlightsources,<400luxfrom17:00to18:00hand<5luxfrom18:00to23:00h).Duringtheinterventionperiods,participantswerefedinenergybalancebyprovidingstandardizedmealsatfixedtimes(moredetailedinformationintheSTARMethods).

Theprimaryendpointofthestudywasaverageglucosecontrolassessedthroughcontinuousglucosemonitoring(CGM)overtheentire4.5-dayexperimentalperiodspentintheresearchfacilities

(MMTT)onday5,andclockgeneexpressioninskeletalmuscle biopsiestakenonday5andfurthercultivationofprimarymyotubes.Furtherexploratoryoutcomesincludedsalivarymelatonin levelsontheeveningofday4;multi-omicanalysesofserum samplesandbloodmononuclearcellscomprisingmetabolomics, lipidomics,andmonocyteRNAsequencing(RNA-seq)takenon day4;24-hbloodpressureandheartrateaswellascoreand skintemperatureonday4;andsubjectivesleepoutcomesand moodthroughoutthestudyperiod.

Participants

EightfemalesandfivemaleswithT2Dparticipatedinthisstudy, withamean(±SD)ageof70±6yearsandBMIof30.1±2.3kg/m2.Mean fastingplasmaglucoselevelswere8.1±1.5mmol/L,HbA1caveraged6.8%±1.0%,whilemeansystolic anddiastolicbloodpressurewere141±11and87±6mmHg, respectively.Participantsreportedameanhabitualsleeping periodfrom23:02±1:02to07:22±1:15h.Glucose-lowering medicationwasusedby11participants.Eachparticipant continuedtakingtheseandotherprescribedmedicationsatthe samedoseandtimeeachday,withoutanychanges,throughout bothlightconditions.Anoverviewofallparticipants'characteristicsisshowninTable1.

Similarrun-inperiodcharacteristicsandnodifferences inphysicalactivitylevels,subjectivesleepquality, mood,andcomfortbetweenlightconditions

Whencomparingthe3-dayrun-inperiodspriortobothlightinterventions,thephysicalactivitypatternsderivedfromwrist-wornactigraphydidnotdifferbetweenlightconditionsandconfirmed adherencetothesleepinstructions(AUCactivitycount:natural, 602,044±103,481vs.artificial,590,045±113,048,mean±SD, p=0.806,n=11;FiguresS2AandS2C).Bodyweight(natural, 86.5±13.0kgvs.artificial,86.7±13.0kg,p=0.295;mean±SD)andbodycomposition(%bodyfat:natural,38.3%±8.9% vs.artificial,37.9%±9.1%,mean±SD,p=0.121),assessedon themorningofday2ineachlightcondition,weresimilarinboth lightinterventions.Similarsubjectivesleepqualityoverthe4weeks precedingtherespectiveexperimentallightconditionwasreportedbytheparticipants(PittsburghSleepQualityIndex[PSQI] score:natural,5.4±3.3vs.artificial,5.5±3.8,mean±SD,p= 0.938,n=12;FigureS3A).Duringtheintervention,nodifferences inphysicalactivitylevels(AUCactivitycount:natural,575,419±293, 441vs.artificial,569,115±287,311,mean±SD,p=0.825, n=11;FiguresS2BandS2D)andsubjectivesleepqualitywere observedbetweenlightconditions(averagetotalLeedsSleeping EvaluationQuestionnaire[LSEQ]scoreoverallfournights:natural, 4.2±0.6vs.artificial,4.3±0.6,mean±SD,p=0.85;FigureS3B). Also,formoodandcomfort,nodifferenceswereobserved,albeita tendencywasfoundfortheoverallightintensitytobeperceivedas brighterinnaturallightcomparedwithartificiallight(p=0.053). ScoresofthevarioussubdomainsofthemoodandcomfortquestionnaireareplottedinFiguresS3CandS3D.

CGMrevealsmoretimespentinthenormalglucose rangeaccompaniedbylower24-hglucoseamplitudes uponnaturallight

CGMdatafromthreeparticipantswerenotcompletedueto
technicaldifficulties,andwethereforebasedalltheCGM

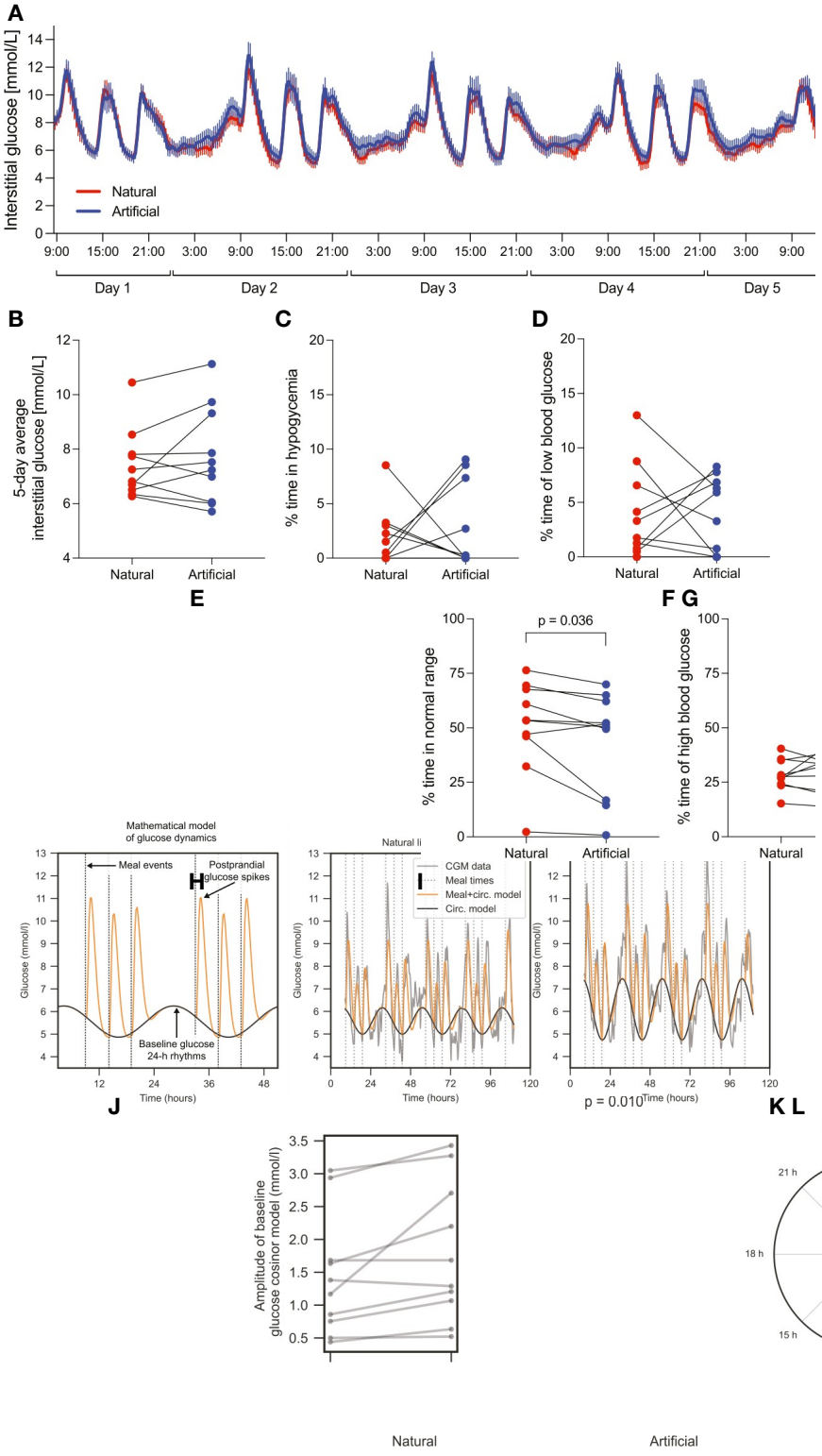


Figure2.Continuousglucosemonitoring (CGM)-derivedoutcomes

(A)Interstitialglucoselevels(*n*=10)overtheentire 4.5-dayoflightinterventionbetweennatural(red line)andartificialofficelight(blueline).Dataare shownasmean±SEM.

(B)Individualdataforthe4.5-dayaverageglucose levels.TheCGMdatawerefurtherdividedinto categoriesaccordingtoAmericanDiabetes AssociationProfessionalPracticeCommittee[32]: hypoglycemia<4.0mmol/L,lowbloodglucose 4.0–4.3mmol/L,normalrange4.4–7.2mmol/L, highbloodglucose7.3–9.9mmol/L,andhyper- glycemia>10.0mmol/L.

(C–G)Resultsarereportedaspercentagesoftime spentintherespectivecategories:(C)%timein hypoglycemia,(D)%timeinlowbloodglucose,(E) %timeinthenormalglucoserange,(F)%timein highbloodglucose,and(G)%timeinhypergly- cemia.

(H)Schemetoillustratethemathematicalframe- work,whichusesmealevents(dottedgray)to modelglucoselevelsasasuperpositionofpost- prandialmealspikes(orange)ontopofan underlying24-hrhythm(black).

(I)Illustrativeexampleofoneindividualpartici- pant'sdatainnatural(left)andartificial(right)light showingtheCGMdata(gray),thestandardized mealtimes(dottedgray),themodelfitconsidering mealsandthecircadianrhythmsinbaseline glucoselevels(orange),aswellasthecircadian rhythmmodelinbaselineglucoselevelsinisola- tion(black).RdenotesthePearsoncorrelation coefficientbetweenthedataandthemodel.

(J)Thepeak-to-troughamplitudeparameteroftheunderlyingcircadianrhythminbaselineglucose levelsinnaturalandartificiallightconditions,and *p*representsthe*p*valueofapairedWilcoxon signed-ranktest.

(K)Phaseplotwiththedistancefromthecenter representingtheamplitudeandtheanglerepre- sentingthepeaktimeparametersoftheunderly- ingcircadianrhythminbaselineglucoselevels, extractedfromthemathematicalmodel.

(L)Thepercentageoftimeinthenormalrange (4.4–7.2mmol/L)asafunctionofthepeak-to- troughamplitudeparameteroftheunderlying circadianrhythminbaselineglucoselevels.The associationwasassessedwithalinearmixed model,where*β*istheregression coefficient(with 95%CIs)and*p*isthe associated*p*value.Gray, regressionfit;filledblue, the95%CI.

analysesonthedataof10participants.Thetimecourseofinter- stitialglucoselevelsovertheentire4.5-dayexperimentalperiod isshownforbothnaturalandartificialofficelightinginFigure2A. Theprimaryoutcomeofthisstudy,averageinterstitialglucose levelsoverthe4.5-dayintervention,didnotsignificantlydiffer betweenlightconditions(natural,7.4±1.3mmol/Lvs.artificial, 7.8±1.3mmol/ L,*p*=0.368;Figure2 B).

However,theoveral ltimespentinthe normalglucoseran ge(4.4–7.2mmol/ L)[32–34]overtheentire4.5dayswassignif- icantlyhigheruponnaturallight(natural,

50.9%±21.5%vs.artificial,43.3%±23.8%,*p*=0.036,*n*=
10;mean±SD;Figure2E).Whenapplyingtherecentlyupdated
definitionoftimeinrange(TIR;3.9–10.0mmol/L)accordingto
theAmericanDiabetesAssociation,[35]asimilartrendwas
observed(natural,83.0%±16.0%vs.artificial,78.6%
±20.5%,*p*=0.082,*n*=10;mean±SD;FigureS4A).Thetime

spentinhypoglycemia(Figure2C),lowglucose(Figure2D),high glucose(Figure2F),orhyperglycemia(Figure2G)didnotsignif-icantlydifferbetweenlightconditions.

Next,weappliedacomputationalmodel,recentlydeveloped byourgroup,[36]thatusesthemealtimes(dottedgray; Figure2H)andtimeofdaytomodeltheCGMdataasasuper-positionofpostprandialmealspikes(orange;Figure2H)ontop ofanunderlying24-hrhythminthebaselineglucoselevels (black;Figure2H).Examplesofparticipantmodelfitsareshown inFigure2I.Whencomparinglightconditions,theamplitudeof theunderlying24-hrhythminbaselineglucoselevelswassignif-icantlylowerinnaturalascomparedwithartificiallight(p=0.010; Figure2J).Thepeaktimesoftheunderlying24-hfunctionclus-teredintheearlymorningperiodwithnodifferencebetweenlight conditions(Figure2K).Theamplitudeoftheunderlying24-h rhythminbaselineglucoselevelscorrelatednegativelywiththe timespentinthenormalglucoserange,suchthathigherampli-tudeswereassociatedwithlesstimespentinthenormalrange (p=0.048;Figure2L).

Higherfatoxidationthroughoutthedayuponnatural lightbutnodifferencesin24-hplasmametabolites betweenlightconditions

Participantsspenttheeveningofday3andthesubsequentnight inarespirationchamber(seeSTARMethodsformoreinforma-tion).Energyexpenditurediднotdifferbetweenlightconditions overthistimeperiod(FigureS5).Also,therespiratoryexchange ratio(RER)overthe4hafterdinner(mean±SD;natural,0.852±0.03vs. artificial,0.849±0.027,p=0.717)andoverthesleeping period(natural,0.829±0.031vs.artificial,0.826±0.023,p= 0.763)didnotdifferbetweenlightconditions(FigureS5).Venti-latedhoodmeasurementsduringthesubsequentday4revealed anincreaseofenergyexpenditurewiththeprogressofdaytime (time:p<0.001;Figure3A),butthisoccurredinasimilarfashion inbothlightconditions(condition,p=0.83andinteraction,p= 0.18).Also,theRERincreasedoverthedaytimeperiod,indi-catinganincreasingrelianceoncarbohydrateoxidationwith theconsecutiveingestionofthethreemeals(time:p<0.001). WefoundaconditioneffectintheRER(condition:p=0.029; Figure3B),indicatingthattheRERwasloweruponnaturallight,

0.9189±0.0173,95%confidenceinterval[CI]:0.8849to 0.9530;R_2=0.8555;p<0.001;FigureS4B),indicatingthatthere wasnosystematicdifferenceintheeffectofnaturallightoninterstitialvs.plasmaglucose.

Indicationofalteredsubstratehandlingfollowingmixed mealtoleranceuponnaturallightusingindirect calorimetryandplasmametabolites

Toinvestigateifdifferentofficelightingspecificallymodifies postprandialmetabolisminindividualswithT2D,weconducted anMMTTinthemorningofday5.Energyexpenditurewashighestat30and60minafteringestionofthetestmeal,afterwhichit decreased(time:p<0.001)withoutdifferencesbetweenlight conditions(Figure3H).ForRER,atimeeffect(time:p<0.001) andaconditioneffect(condition:p=0.04;Figure3I)were observed.ThelatterindicatesthatRERwasloweruponnatural light(mean±SD;averageRERduringtheMMTTfornatural, 0.834±0.023vs.artificial,0.845±0.013),whichisalsoreflected intendenciesforlowercarbohydrateoxidation(condition:p= 0.059;Figure3J)andhigherfatoxidationlevels(condition:p= 0.054;Figure3K),whichisinlinewiththeeffectsobserved overdaytimeonday4.Atime×lightconditioninteractioneffect wasalsofoundforRER(interaction:p=0.045;Figure3I)anda tendencyforinteractionforcarbohydrateoxidationlevels(interaction:p=0.084;Figure3J),suggestingdifferentialtemporaldynamicsinpostprandialsubstratemetabolismbetweenlightconditions.Fromallplasmametabolitesthatwereassessedduring theMMTT(e.g.,glucose,insulin,freefattyacids,andTAG; Figures3L–3O),onlyfreefattyacidlevelsindicatedatendency towardaconditioneffect,leaningtowardhigherlevelsuponnaturallight(condition:p=0.061;Figure3N),whichisinlinewiththe observedlowerRER.Aninteractioneffectforbothglucoseand freefattyacidlevelswasfound(interaction:p=0.045and$p$= 0.021,respectively;Figures3Land3N),indicatingthatthelight conditionmodulatedthesebloodmetabolitesdifferentlyover timeduringtheMMTT.InsulinandTAGlevelsdidnotdifferbetweenlightconditions(Figures3Mand3O).

Nodifferencesineither24-hbloodpressureandheart rateorambient,core,andskintemperaturebetween lightconditions

Diurnalvariationwaspresentinbloodpressureandheartrate (time:$p<0.05$),whichbothincreasedwiththesteppingexer- $p=0.024$).

Inthe15bloodsamplescollectedover24hfromthemorning ofday4andonward,cleardiurnalvariationinplasmalevelsof glucose(Figure3E),freefattyacids(Figure3F),andTAG (Figure3G) (time:$p<0.001$)wasobserved,withplasmaglucose sharplyincreasingwithmealsbutreturningtonormoglycemic levelspriortolunchanddinner(meanpre-prandiallevels:5–6mmol/L)andremaininghighthroughoutthenight(mean nocturnallevels:>6.4mmol/L).However,noconditionorinteraction effectswerefound,andhencethe24-hlevelsandtemporal dynamicsdidnotdifferforanyoftheplasmametabolitesbetweenthetwolightconditions(Figures3E–3G).Theavailable plasmaglucoseassessmentsondays4and5didstronglycorrelatetotheinterstitialglucosereadingsfromtheCGM(slope= ciseaftermeals(FigureS6).However,noconditionorinteraction effectwasfound;hence,bloodpressureandheartratedid notdifferbetweenlightconditions.Duringtheentirestudy,

averageambienttemperaturevariedbetween21◦Cand
23.5◦C.Overall,themixedeffectsmodelrevealedatimeeffect
(p<0.001)withthelowestambienttemperatureinthemorning
hoursandthehighestat14:00–15:00hintherespectiveoffice
lighting,butnocondition(p=0.961)orinteractioneffect(p=
0.558)overtheentirestudyperiod(FigureS7).Participants
showedacleardiurnalrhythmincorebodytemperature
(CBT),peakingintheearlyeveningat∼18:00handthereafter
droppinguntilreachingthenadirat∼01:00h(time:p<0.001;
FigureS8A).However,noconditionorinteractioneffectwas
foundforCBT,andthenadirofCBTalsodidnotdifferbetween
lightconditions(natural,01:36±01:47hvs.artificial,01:59±01:39h,
p=0.460,mean±SD).Atimeeffect(p<0.001)but

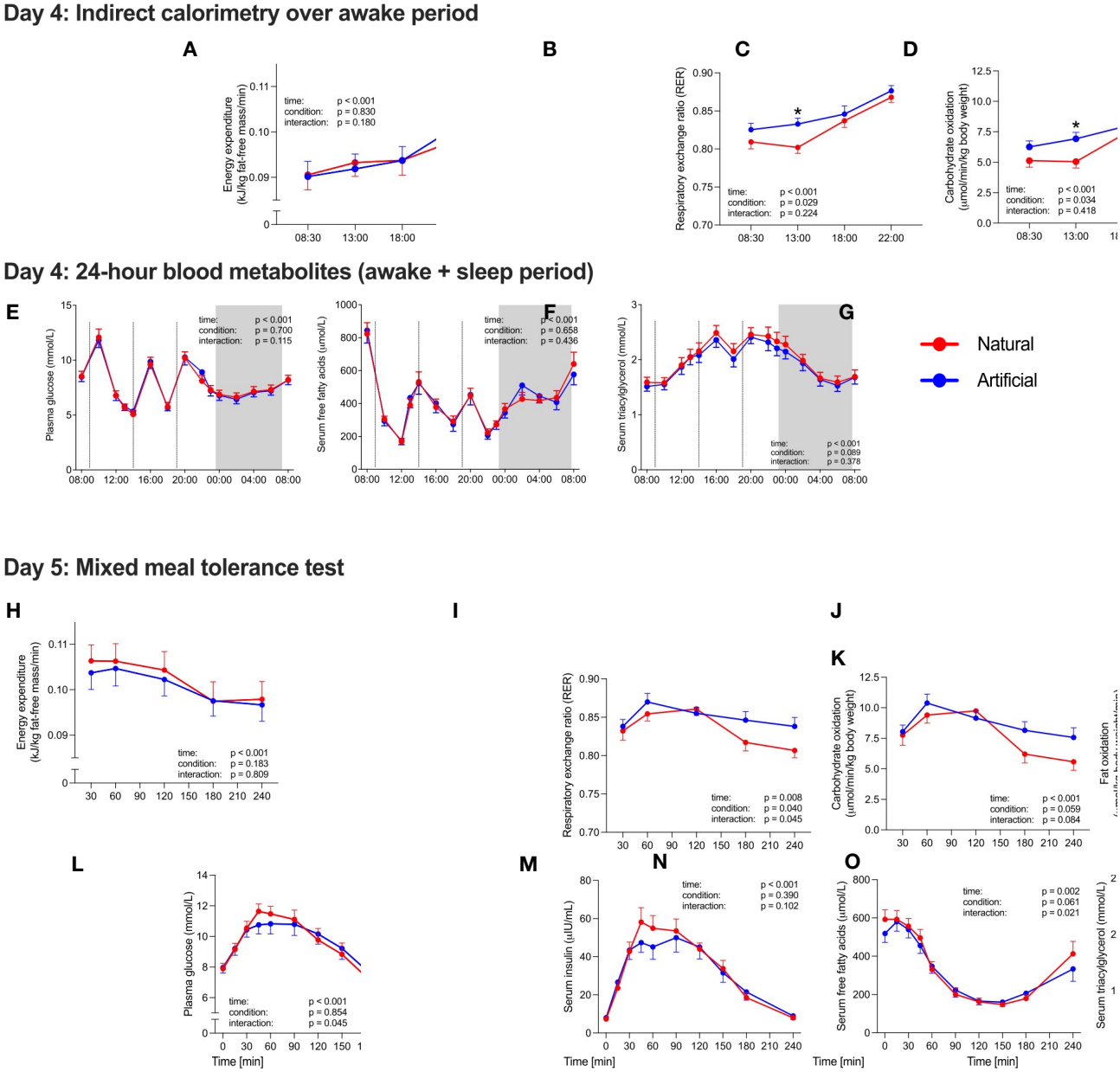


Figure3.Whole-bodyindirectcalorimetryandbloodmetabolites

Whole-bodysubstrateoxidationandenergyexpenditureandbloodmetabolitesinresponsetonatural(redlines)vs.artificialofficelighting(bluelines)assessed onday4(*n*=13)andday5(*n*=12).

(A–D)Whole-bodyrestingenergyexpenditure(A),RER(B),carbohydrateoxidation(C),andfatoxidation(D)duringtheawakeperiodofday4.

(E–G)Overviewof24-hplasmametabolitesonday4:plasmalevelsofglucose(E),serumfreefattyacids(F),andserumTAG(G).Thedarkgrayarearepresentsthe sleepingperiod(23:00–07:00h).

(H–K)Whole-bodyrestingenergyexpenditure(H),RER(I),carbohydrateoxidation(J),andfatoxidation(K)inresponsetotheMMTTonday5. (L–O)Postprandialplasmametabolites:plasmalevelsofglucose(L),seruminsulin(M),serumfreefattyacids(N),andserumTAG(O).Dataarepresentedas mean±SEM.*$p<0.05$basedontheBonferroniposthoctest.

nodifferences(conditionorinteraction)inproximaland distalskintemperatureaswellasthedistal-proximalskin temperaturegradientwerefoundbetweenlightconditions

inonlyoneofthetwoconditions;hence,thesefourindividuals wereexcludedforcomparingDLMObetweenthetwolightconditions.DLMOwasnotdifferentbetweenlightconditions (FiguresS8B–S8D).

Higherlateeveningmelatoninlevelsuponnaturallight butnodifferencesinDLMO

Dim-lightmelatoninonset(DLMO)analysisrevealedoneindividualthatdidnotreachthethresholdlevelof3pg/mLinbothlight conditionsandthreeindividualsthatdidnotreachthethreshold (mean±SD,natural,20:35±01:01h;artificial,20:42±01:18 h,p=0.47,n=9;Figures4Aand4B).However,melatonin

AUCfromtheaverageDLMOoverbothlightconditions onward(21:00–23:00h)washigherinnaturalcomparedwith artificialofficelight(natural,755±432pg/mL×min;artificial, 650±374pg/mL×min,p=0.029,n=13;Figure4C),indicating highermelatoninreleaseafterDLMOuponnaturallight.

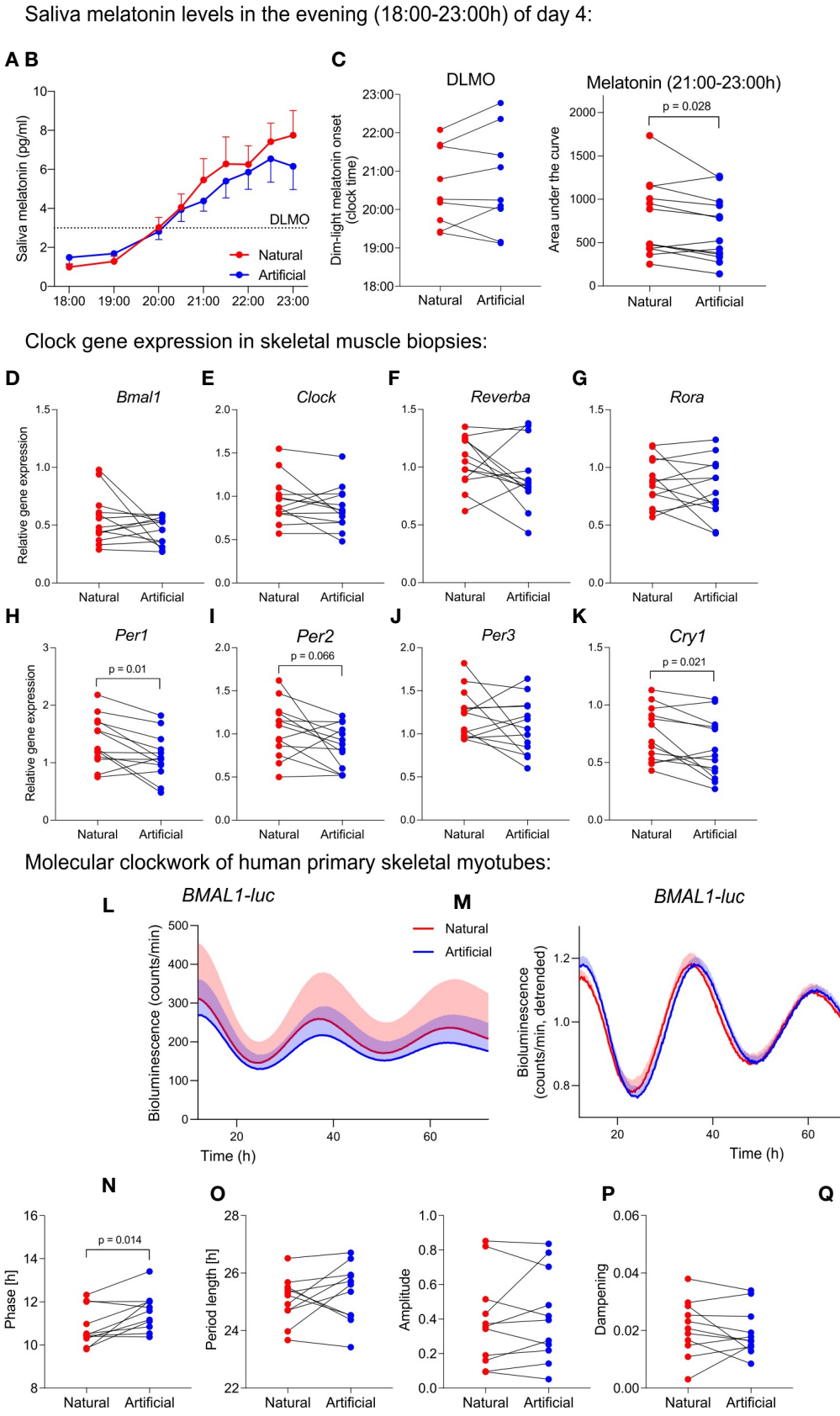


Figure4.Eveningmelatoninlevelsandskeletalmuscleclockwork

(A)Salivamelatoninlevelswereassessedintheevening(18:00–23:00h)ofday4underdim-lightconditionsforbothnatural(redline)andartificialofficelighting (blueline),anddataarepresentedasmean±SEM.

(legendcontinuedonnextpage)

Naturaldaylightmayaffectthemolecularclocks operativeinskeletalmuscleofindividualswithT2D,as measuredinmusclebiopsyandinprimarymyotubes synchronizedinvitro

Paired*t*testsrevealednodifferenceinmRNAlevelsofthe core-clockgenes*Bmal1*,*Clock*,*Reverbα*,*Rorα*,and*Per3* (Figures4D–4J),assessedinskeletalmusclebiopsiestakenat asingletimepointonthemorningofday4,afteranovernight fast.However,mRNAlevelsoftheclockgenes*Per1* (Figure4H)and*Cry1*(Figure4K)werehigheruponnaturallight (p=0.01and$p$=0.021,respectively),withasimilartendency for*Per2*(p=0.066;Figure4I),suggestingthatnaturallight mayaffectthemusclemolecularclock.Toinvestigateifthese

S1),usingsharedanduniquestructures(SUSs)plotstovisualize targetsmoststronglydistinguishingbetweenlightconditions acrossbothtimepoints.Metabolomicsanalysesidentifiedfive keymetabolites—threonine,cholicacid,uracil,erythro-dihydrosphingosine,andglutamicacid—thatexhibitedthestrongest positiveassociationswiththenaturallightconditionatboth 08:00and16:00h(Figure5A).Thelevelsofthesemetabolites

changesincomponentsofthemolecularclockmeasuredina singletime-pointmusclebiopsytranslateintoalteredclock rhythmicityinmuscle,wenextassessedmolecularclockwork inprimarymyotubesestablishedfromtheseskeletalmuscle biopsiesanddifferentiated*invitro*usinga*Bmal1-luc*reporter allowingforcontinuousmeasurementofthecellularclocks[37,38] (Figures4Land4M).Ofnote,thephaseof*Bmal1-luc*oscillationsofprimaryskeletalmyotubesestablishedfrombiopsies takenafterthenaturallightconditionwas0.75±1.03h advancedcomparedwiththeartificiallightregimen(p=0.014; Figure4N;individualprofilesareshowninFigureS9A).The periodlength(p=0.87;Figure4O),amplitude(p=0.79; Figure4P),andoscillationdamping(p=0.42;Figure4Q)were comparablebetweenlightregimens.Giventhatwedetected light-inducedalterationsinbothcore-clocktranscriptstatusin musclebiopsiesandthecircadianphaseofbioluminescence inculturedmyotubes,wenextinvestigatedwhethervariations inmuscleclockgeneexpressioncouldinfermyotubebioluminescencephase.Indeed,themusclebiopsyclockgeneexpressionwasabletopredictthebioluminescencephasein

wereconsistentlyhigherundernaturallightcomparedwithartificiallightatbothtimepoints,asseenintheboxplotshighlighting cholicandglutamicacidlevelsatbothtimepoints(Figure5B).By contrast,hydroxyproline,allantoin,hydroxyphenylaceticacid, citric/isocitricacid,andsalicylicacidwerethetopfivemetabolitesshowingthestrongestnegativeassociationwithnaturallight

(Figure5A).

Comparingthelipidfamiliesbetweenlightconditionsrevealedether-linkedphosphatidylethanolamine(PE-O),phosphatidylethanolamine(PE),diacylglycerol(DAG),andlysophosphatidylethanolamine(LPE)familiesasthosebearingthehighestpositiveassociationwiththenaturallightcondition(Figure5C).Ontheotherhand,glucosylceramides(GlcCer),ceramides(Cer),andcholesterylesters(CEs)weretheclassesmostnegativelyassociatedwithnaturallight(Figure5C).Individualboxplotsoftheselipidfamiliescorroboratethefindingsoftheda,highlightedbyanincreaseinPE-OandatendencyformyotubeswithapearsoncorrelationcoefficientofR=0.51(p=0.016)(FiguresS9BandS9C),with*Cry1*and*Per3*havingthelargestpositiveandnegativecontributionstotheestimatedbioluminescencephase,respectively.

Differentialimpactofthelightingconditionsonthemulti-omiclandscapeinthecirculation

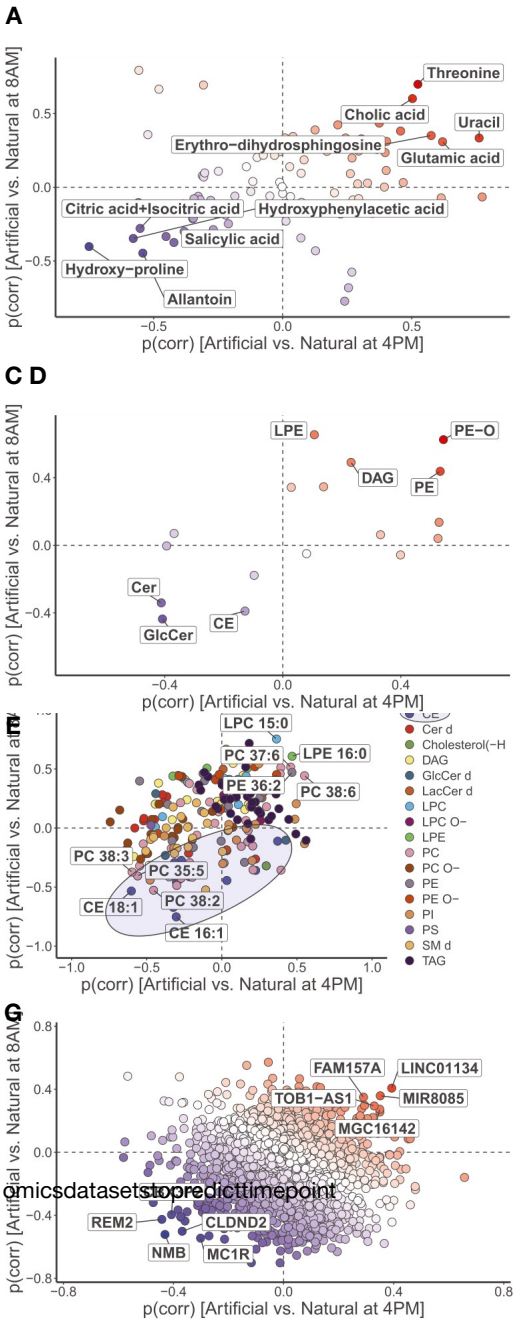
Sincenaturallightimpactedmetabolismdifferentiallycomparedwithartificiallight,wenextappliedtargetedserummetabolomics,lipidomics,andmonocyteRNA-seqanalysestotheblood samplesobtainedonday4at08:00(aftertheovernightfast,precedingtherespectivelightexposureperiod)and16:00h(attheendoftherespectivelightexposureperiod).Toidentifytargetsassociatedwithlight-dependentbiologicaldifferences,weappliedorthogonalpartialleastsquaresdiscriminantanalysis(OPLS-DA)toeachdataset(metabolomics,lipidfamilies,lipidspecies,andtranscriptomics)atboth08:00and16:00h(Data

adecreaseinGlcCerunderthenaturallightcondition(Figure5D).Consistently,theSUSplotofindividuallipidspeciesshowsLPE16:0,lysophosphatidylcholine(LPC)15:0,phosphatidylcholine(PC)38:6,PC37:6,andPE36:2asmostpositivelylinkedwiththenaturallightconditionatbothtimepoints(Figure5E).Negativeassociationstothenaturallightconditionwereobservedforce18:1,PC38:2,PC35:5,CE16:1,andPC38:3.Thisissupportedbyboxplotsoftwotoplipidspecies(CE18:1andLPE16:0)identifiedbytheDA

(Figure5F).

Further, we used a SUS plot to display monocyte transcripts most discriminating between the light conditions at each time point. This analysis revealed *LINC01134*, *MIR8085*, *FAM157A*, *TOB1-AS1*, and *MGC16142* as the transcripts most positively associated with natural light, and *NMB*, *REM2*, *CLDND2*, *MC1R*, and *CBX3P2* as the transcripts most negatively associated with the natural light condition (Figure 5G). These changes are highlighted by boxplots displaying expression levels of two genes, *REM2* and *NMB* (Figure 5H). To further investigate these transcriptional alterations, we conducted a gene set enrichment analysis (GSEA) and clustered and annotated the pathways revealing the greatest changes between light conditions (Data S2; Figures S10M and S10N). Top pathways were related to Cer and Glc Cer regulation (Figures S10M and S10N); however, none reached statistical significance after false discovery rate (FDR) correction (Data S2). Volcano plots of the metabolomics,

(Band C) Individual data of DLMO; $n = 9$ (B) and the area under the curve of melatonin levels ($n = 13$) from 21:00 to 23:00 h (C) are shown.
(D–K) Core-clock gene expression in the single skeletal muscle biopsy sample ($n = 13$) collected between 07:30 and 08:30 h on day 5 preceding the mixed meal test: individual mRNA levels of *Bmal1* (D), *Clock* (E), *Reverba* (F), *Rora* (G), *Per1* (H), *Per2* (I), *Per3* (J), and *Cry1* (K) are shown. Normalized expression levels were determined relative to the geometric mean of two housekeeping genes.
(L and M) Molecular clock oscillations in differentiated myotubes measured by BMAL1-luc reporter assay. Averaged profiles ± SEM ($n = 11$) shown as raw (L) and detrended (M) values for 12–72 h after synchronization, and individual profiles on these outcomes are shown in Figure S9.
(N–Q) Circadian parameters of oscillations extracted from sinusoid fit generated by Chronostar 3.0 software: phase (N), circadian period length (O), amplitude (P), and oscillation damping (Q). Indicated *p* values were determined based on paired *t* tests.



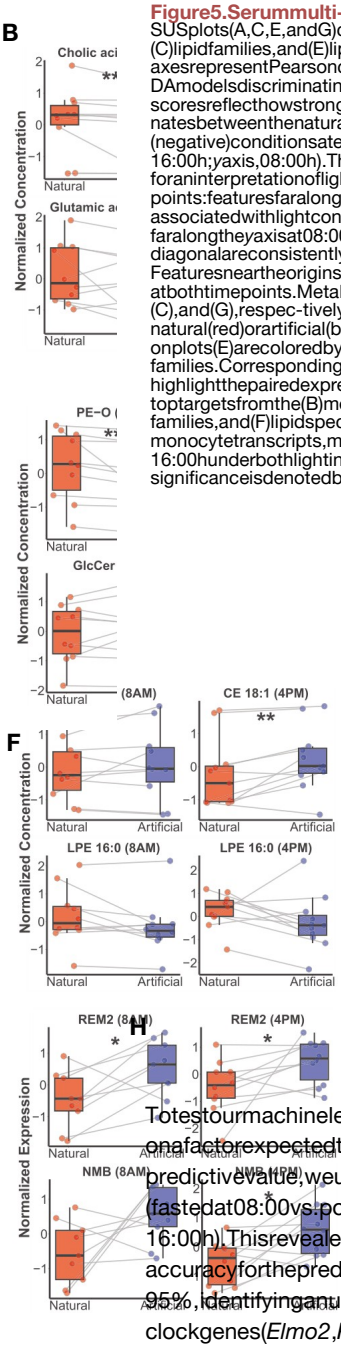


Figure5.Serummulti-omicsignature

SUSplots(A,C,E,andG)comparingserum (A)metabolites, (C)lipidfamilies,and(E)lipid speciesand(G)bloodmonocytetranscripts.The axesrepresentPearsoncorrelation-scaledload-ingsfromOPLS-DAmodelsdiscriminatingbe-tweenlightconditionsateachtimepoint.These scoresreflecthowstronglyeachtargetdiscrimi-natesbetweenthenatural(positive)orartificial (negative)conditionsateachtimepoint(*x*axis, 16:00h;*y*axis,08:00h).Thisvisualizationallows foraninterpretationoflighteffectsatbothtime points:featuresfaralongthe*x*axisarestrongly associatedwithlightconditionsat16:00h,those faralongthe*y*axisat08:00h,andthosenearthe diagonalareconsistentlyinfluencedatboththtimes. Featuresneartheoriginshowweakassociations atbothtimepoints.Metabolites,lipids,andtran-scriptsshownonplots(A), (C),and(G),respec-tively,arecoloredbytheirpositivecorrelationto natural(red)orartificial(blue)light.Lipidspecies onplots(E)arecoloredbytheirrespectivelipid families.Correspondingboxplots(B,D,F,andH) highlightthepairedexpressionprofilesofselected toptargetsfromthe(B)metabolites,(D)lipid families,and(F)lipidspeciesand(H)blood monocytetranscripts,measuredat08:00and 16:00hunderbothlightingconditions.*p*value significanceisdenotedby*$p<0.05$and**$p<0.01$ basedonpairedWilcoxonsigned-ranktests.

lipidomics,andtranscriptomics,usingamixedmodeladjusting fortimeandparticipantIDascovariates,revealednosignificant targetsafterFDRcorrectionbetweennaturalandartificiallightat 08:00,16:00h,orinacomparisoncombiningthetwotimepoints (FiguresS10A–S10L).

Machinelearninganalysisclassifyinglightandtime conditionsuncoverssignificanttargets

Weappliedarandom-forestmachinelearningmodelto discovermultivariatepredictivefactorsthatarenotrevealed intheunivariateanalysis.Mergingthethreedatasets(i.e.,me-tabolomics,lipidomics,andtranscrip-tomics)intoasinglemodelrevealeda classificationaccuracyof56%forthe lightcondition(FigureS10O).Exam-iningthevariablesinvolvedintheclas-sificationaccuracyrevealed*Elp6*,PS 34:0,uracil,hydroxyproline,andPE-O 38:4asthetop5variablesassociated withthelightcondition(FigureS10P). Totestourmachinelearninganalysis onafactorexpectedtoholdincreased predictivevalue,weusedthethree -omicsdatasetstopredicttimepoint (fastedat08:00vs.postprandialat 16:00h).Thisrevealedaclassification accuracyforthepredictingtimeof 95%,identifyingan-umberofcircadian clockgenes(*Elmo2*,*Per2*,*Tef*,*Nr1d2*, and*Per1*)alongwithdeoxyribose,malic acid,andTAGspeciesascarryingstrongpredictivevalue (FigureS10Q).

Time-of-daysignatureofbloodmetabolites,lipids,and monocytetranscripts

Sincediurnalrhythmicityofmetabolic,lipid,andtranscriptional

landscapeshasbeenreportedinhumanindividuals,withdifferentialtemporalorchestrationobservedbetweenindividualswithT2Dandnormoglycemiccounterparts,[11,18,39,40]wezoomedintothetemporalalterationsassociatedwiththeappliedlightregimens.Tothisend,wecomparedthemetabolite,lipid,and

transcriptsignaturesbetween08:00and16:00hbypoolingsamplesfromthetwolightconditions.Serummetabolomicsreveals60significantlyalteredmetabolitesbetweentimepoints(FigureS11A),withdeoxyribose,proline,andmalicacidasthemostincreasedandhypoxanthine,ribose5-phosphate,andxanthineasthemostdecreasedmetabolitesat16:00h.Analyzingdifferencesinlipidfamiliesshowed5significantlydifferentclassesbetweentimepoints(FigureS11B),withTAGandLPEshighlyincreasedat16:00hcomparedwith08:00h.Thisisfurthersupportedbycomparingindividuallipidspeciesbetweentimepoints,revealing8TAGspeciesasthemostincreasedlipidsat16:00h(FigureS11C).Comparingdifferencesinmonocytetranscriptsdisplayed920differentialtranscriptsbetweentimepoints,with*Elmo2*,*Per1*,*Per2*,*Reverbβ*,*Tef*,*Flt3*,*Alas1*,*Lmo2*,*Mdm2*,and*Peli1*asthemostsignificantlyalteredtranscriptsby*p*valuebetweentimepoints(FigureS11D).GSEAusingtheRNA-seqdatarevealeddecreasesinpathwaysrelatedtomitochondrialmembranefissionandtheintrinsicapoptoticsignalingpathwayinresponsetohypoxiaatthe16:00htimepoint(FigureS11E).

In line with previous studies,[26,27] we found a tendency for a larger excursion in postprandial glucose upon natural light upon a fluid mixed meal challenge performed in the morning after the overnight fast. Intriguingly, though, the time spent in the normal glucose range across the 4.5-day intervention period was higher upon natural light (together with higher whole-body fat oxidation levels on day 4), suggesting improved glucose control in individuals with T2D upon longer-term natural light exposure. Hence, the potentially negative impact of (bright) light exposure on postprandial plasma glucose levels in the short term may be time-of-day dependent and should be investigated in future studies by administering the meal tests at different times of the day.

In a prospective cohort study, Lu et al. correlated all-cause mortality with the time spent in the normal glucose range and found that less time in the normal range was associated with a higher hazard ratio for all-cause mortality in individuals with T2D,[43] indicating that the time spent in the normal range is a clinically relevant outcome. Interestingly, time-restricted eating regimes in individuals with T2D have been shown to lower 24-h glucose levels and to increase the time spent in the normal range[44,45] by 12% and 10%, respectively. The improvement in glucose control observed in this study is also in line with improvements seen in CGM profiles of individuals with T2D upon

DISCUSSION

Nowadays, almost 90% of our time is spent indoors,[19–21] and during the daytime, we are therefore exposed to lower light intensities and a different wavelength spectrum from artificial light sources compared with natural daylight. Since light is the main *zeitgeber* for our circadian time-keeping system[2–4] that can modulate human metabolism acutely in a time-of-day-dependent manner,[26,27] chronic lack of natural daylight could be considered as a risk factor in society's rising incidence of metabolic diseases. Therefore, in this study, we hypothesized that individuals with T2D would benefit from being exposed to natural daylight during office hours when compared with typical constant artificial lighting. Although the average interstitial glucose level was not different between office light conditions, we found that glucose control (i.e., % time spent in the normal glucose range: 4.4–7.2 mmol/L) was significantly improved during the 4.5 days of natural lighting. This was accompanied by lower whole-body carbohydrate oxidation and, conversely, higher fat oxidation rates during waking hours upon natural light, as measured on day 4. Consistent with the higher fat oxidation, plasma free fatty acid levels were higher upon a liquid mixed meal challenge in natural compared with artificial office lighting.

To date, a few clinical studies have shown that the intensity of environmental light during daytime can impact glucose control of individuals with T2D[26] and insulin resistance.[27] For instance, enhancing intensity levels of artificial light sources to a constant 1,250[27] or 4,000 photopic lux[26] during morning hours consistently led to elevated postprandial glucose levels following a morning meal after overnight fasting compared with 10 lux. Other studies,[41,42] but not all,[26] reported similar effects in young individuals. Also, Meng et al. found reduced glucose tolerance in young healthy adults upon white light exposure of 400 lux compared with darkness applied both during daytime and nighttime.[42] In this study, natural light encompassed diurnal changes in both light intensity and its wavelength spectrum (Figures 1C and 1D), whereas the control artificial light condition was more applicable to the real world (e.g., 300 lux) instead of dim light.

bothshort-andlong-termexerciseprograms.[46]Modelingthe continuousglucosedatarevealedthatnaturallightledtolower amplitudesof24-hfluctuationsinglucoselevels,whichwere associatedwithmoretimespentinnormalrangeacrossparticipants.Thissuggeststhatexposuretonaturallightmayhavea beneficialeffecton24-hglucosemetabolism.Futurestudies shouldascertainthemolecularregulatorymechanismsunderlyingtheobservedbeneficialeffectofexposuretonaturallighton 24-hglucoserhythms.

Enhancingdaytimelightintensitieshasrepeatedlybeen showntoincreasenocturnalmelatoninlevelsinyoungindividuals,[28,47,48]butcross-sectionalassociationsbetweenbrighter daytimelightandhighernocturnalmelatoninlevelswerealso foundintheelderly.[49]Inaddition,naturaldaylightexposure throughoutdoorcampinghasbeenshowntoinduceconsistent phaseadvancesinDLMOinindividuals.[50]Weheredemonstrate thatinolderindividualswithT2D,boththenadirinCBTand DLMOwererelativelystableirrespectiveofdaytimeofficelighting,butmelatoninlevelswerehigherinthelast2hbefore bedtime(21:00–23:00)uponexposuretonaturallightduring thedaytime,ascomparedwithartificiallighting.Anindication thattheperipheralcircadiansystemwasmodifiedthroughthe differentialofficelightingwasderivedfromthehighermRNA levelsofseveralclockgenes(i.e.,*Per1-2*and*Cry1*)inskeletal muscleuponnaturallighting.Subsequently,weinvestigated therhythmicpropertiesofmyotubesgeneratedfromourparticipantsafterexposuretobothofficelightingregimes.Consistent withtheeffectofnaturallightonclockgeneexpressioninthe musclebiopsies,wefoundaphaseadvanceintherhythmof the*Bmal1*-luciferasereporterfollowingnaturallightexposure. Whilethe*invitro*synchronizationforskolinpulsebringstheindividualcellularoscillatorstothesameinitialphase,itdoesnot alterthecore-clockmachineryperse.[51,52]Thus,thephase advancepersistedeventhoughwesynchronizedthecultured myotubes,suggestingnaturallightmayinducelastingchanges tothecore-clockfeedbackloop.Noteworthy,BMAL1was

previouslyreportedtoimpactingonglucosecontrolinmice[53] andhumans.[54]Wealsofoundthatthemuscleclockgene expressioncouldbeusedtoinfer*invitro*bioluminescencephase data,althoughthiscorrelationwarrantsvalidationbasedon musclebiopsiesfromdonorswithT2Dcollectedaroundthe clock.Weidentifiedthecore-clockgenes*Cry1*and*Per3*ashaving thestrongestinfluenceonphaseestimation.Elucidationof

workplace.[65] Similarly, mimicking natural daylight by dynamically changing the wavelength spectrum and the intensity of artificial indoor lighting across the day improved sleep latency.[28]

the molecular mechanisms controlling the alterations of muscle clockwork upon natural light in individuals with T2D represents a promising direction for follow-up studies.

To further investigate the light-responsive circulating factors that may account for differences in glucose homeostasis, we compared the multi-omic signature in blood samples between light conditions. Our metabolomics analysis revealed an increase in cholic acid under natural light, consistent with previous findings identifying it as increased in ducks exposed to 460 nm blue light, a wavelength exhibiting higher photon flux in the natural compared with the artificial light condition.[55] Glutamate levels were also elevated in natural light and have been reported as UV-responsive in murine brain tissue, particularly to UVB.[56] While participants were not exposed to UVB, shorter wavelengths present in natural light may similarly upregulate glutamate. Furthermore, increased threonine levels that we observed upon natural light, concomitant with the improved glucose control, are well in line with the reduced levels of this aminoacid in individuals with T2D that we extracted from a recently published serum metabolomics dataset.[57] Several lipid families also responded to light exposure. Notably, cholesterol esters and Cer negatively associated with natural light, aligning with previous reports of similar reductions in sterol esters and Cer in the human hair lipidome following exposure to 380 and 420 nm wavelengths.[58] Moreover, recent studies highlight Cer levels as increased in individuals with T2D.[40] Concordantly, we observed a trend toward reduced Cer upon natural light, corroborating improved glucose control. The transcriptomic analyses identified top pathways involved in the negative regulation of Cer and GlcCer, both of which were negatively associated with natural light in the lipidomics data. Additionally, LPEs, including LPE 16:0, particularly, were positively associated with natural light and have been linked to higher insulin sensitivity[59] and found to be decreased in serum from individuals with obesity.[60,61] Together, these findings reveal a distinct multi-omic signature linked to natural light exposure, characterized by alterations in metabolites, lipids, and transcripts possibly underlying the improved glucose control.

Since sleep and overall physical activity levels can both have an impact on glucose homeostasis outcomes, we assessed these parameters as potential confounders. However, we could

Therefore, we cannot rule out that improved sleep quality (partially) drives the metabolic benefits associated with the natural light exposure during daytime.

Future directions

We chose the strong contrast of dynamic natural light through a window vs. an artificial light condition that deprived participants of any enriched short-wavelength daylight throughout the 4.5-day study period as a proof of concept that natural light elicits distinct metabolic effects compared with artificial light only. Future studies could usefully explore if longer-term exposure to natural light could elicit more pronounced effects or if alternative strategies, such as natural light exposure during working commutes (e.g., before and/or after office hours, as is typically the case in the real world[66]), would be sufficient to offset the negative effects of artificial lighting revealed in this study.

Limitations of the study

While our crossover design strengthens the reliability of our findings, one limitation of the study is the small sample size (n=13). Our finding of improved time in the normal glucose range (4.4–7.2 mmol/L) upon natural light may warrant cautious interpretation until more long-term clinical trials (e.g., exceeding 4.5 days up to weeks and months) confirm the clinical relevance through improvement of the TIR (3.9–10.0 mmol/L). Furthermore, the average age of our study population was 70 years, and 12 participants were at least 65 years old, which compromises the generalizability of our findings. Future studies could study the potential metabolic benefits of natural vs. artificial (office) lighting across a wider age range within the working population in real office environments. Furthermore, all experiments were conducted during Central European Summer Time (i.e., from April to October). Natural light varies strongly as a function of geographical location, weather, and season, where seasonal light changes can modulate energy metabolism and peripheral clocks in mice[67] and gene expression in humans.[68] Future studies are needed to establish how the effect size of the natural light intervention changes with the time of year and location. Finally, while our multivariate analyses identified targets differentiating natural and artificial light conditions at both time points, the models demonstrated limited predictive accuracy, likely due to the small sample size of this exploratory cohort. This underscores the need for further investigation into the mechanisms by which natural light influences metabolic health.

Conclusions

We did not detect any differences between light conditions, neither prior to nor during the experimental period. Particularly, physical activity was highly standardized in this study by scheduled standing and stepping activity times. However, sleep outcomes were only assessed through subjective questionnaires and not quantified through more objective means, such as polysomnography (PSG). Compromised sleep is linked to impaired glucose control,[62] and sleep loss has even been shown to modify skeletal muscle mRNA levels of *Bmal1* and *Cry1* as well as protein levels of BMAL1.[63,64] Office workers in windowless environments reported poorer sleep scores than workers with windows at the

Inconclusion,throughcomparingreal-worldofficelightingsce-
nariosofnaturaldaylightthroughwindowsvs.constantartificial
lightinginindividualswithT2D,wefoundmetabolicbenefits
associatedwithnaturaldaylightexposure.Specifically,more
timespentinthenormalglucoserangeandashiftinwhole-
bodysubstratemetabolismtowardhigherfatoxidationlevels
indicatedimprovedglucosecontroluponnaturallighting.Our
findingsprovideastrongrationaleforfutureresearchtofocus
moreontheinteractionof(natural)lightexposureandmetabolic
health.Thisstudyalsohighlightstheoftenunnoticedimpactof

thebuiltenvironmentonourhealthandraisesfurtherconcerns aboutheprevalenceofofficeenvironmentswithpoor(natural) daylightaccess.

RESOURCEAVAILABILITY

Leadcontact

Furtherinformationandrequestsforresourcesshouldbedirectedtoandwill befulfilledbytheleadcontact,JorisHoeks().

Materialsavailability

Thisstudydidnotgeneratenew,uniquereagents.

Dataandcodeavailability

Metabolomicsandlipidomicsdatasupportingthefindingsofthisstudyare availableinDataS3.RawRNA-seqdatahavebeendepositedinGEOunder theaccessionnumberGEO:GSE309688.Anyadditionalinformationrequired toreanalyzethedatareportedinthispaperisavailablefromtheleadcontact uponrequest.

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- Quantificationoflightcharacteristics
- Continuousglucosemonitoring
- Bodycomposition
- Sleepandmoodquestionnaires
- Indirectcalorimetry
- Ambient,skinandcorebodytemperature
- Bloodpressureandheartrateassessment
- 24-hourbloodandeveningsalivasampling
- Skeletalmusclebiopsy
- Mixedmealtolerancetest
- Salivarymelatoninassessment

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- Plasma,serumandmonocyteanalysis
- Serummassspectrometryanalyses
- Monocytetranscriptomicanalyses
- Musclegenetranscriptquantification
- Primaryhumanskeletalmyotubecultureandlentiviraltransduction
- Real-timebioluminescencerecordingandanalysis

- QUANTIFICATIONANDSTATISTICALANALYSIS

SUPPLEMENTALINFORMATION

Supplementalinformationcanbefoundonlineat
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AUTHORCONTRIBUTIONS

Conceptualization,J.-F.H.,I.H.,D.D.,H.P.,M.C.M.G.,T.-H.C.,A.K.,P.S., C.D.,andJ.H.;methodology,J.-F.H.,I.H.,P.S.,C.D.,andJ.H.;formalanalysis, J.-F.H.,I.H.,A.D.B.,A.L.,N.E.P.,L.M.,andJ.S.-A.;investigation,J.-F.H.,I.H., A.D.B.,A.L.,N.E.P.,L.M.,J.S.-A.,M.K.,M.T.,D.H.,S.S.d.K.,G.S.,J.A.J.,A.G., E.M.-K.,T.v.d.W.,M.B.,F.H.,R.D.L.,andH.P.;writing–originaldraft,J.-F.H., I.H.,A.D.B.,A.L.,J.H.,P.S.,C.D.,andN.E.P.;writing–reviewandediting,all authors;visualization,J.-F.H.,I.H.,A.D.B.,A.L.,L.M.,andN.E.P.;supervision, P.S.,C.D.,andJ.H.;projectadministration,C.D.andJ.H.;fundingacquisition, C.D.,J.-F.H.,P.S.,andJ.H.

DECLARATIONOFINTERESTS

Theauthorsdeclarenocompetinginterests.

REFERENCES

1.Roenneberg,T.,andMerrow,M.(2005).Circadianclocks-thefallandrise ofphysiology.Nat.Rev.Mol.CellBiol.*6*,965–971..

2.Reppert,S.M.,andWeaver,D.R.(2002).Coordinationofcircadiantiming inmammals.Nature*418*,935–941..

3.Foster,R.G.,Hughes,S.,andPeirson,S.N.(2020).Circadianphotoentrainmentinmiceandhumans.Biology*9*,180..

4.Panda,S.(2016).Circadianphysiologyofmetabolism.Science*354*,1008–1015..

5.Gutierrez-Monreal,M.A.,Harmsen,J.-F.,Schrauwen,P.,andEsser,K.A. (2020).Tickingformetabolichealth:theskeletal-muscleclocks.Obesity (SilverSpring)*28*,S46–S54..

6.Zhang,R.,Lahens,N.F.,Ballance,H.I.,Hughes,M.E.,andHogenesch, J.B. (2014).Acircadiangeneexpressionatlasinmammals:implications forbiologyandmedicine.Proc.Natl.Acad.Sci.USA*111*,16219–16224. .

7.Reinke,H.,andAsher,G.(2019).Crosstalkbetweenmetabolismand circadianclocks.Nat.Rev.Mol.CellBiol.*20*,227–241..

8.vanMoorsel,D.,Hansen,J.,Havekes,B.,Scheer,F.A.J.L.,Jo¨rgensen, J.A.,Hoeks,J.,Schrauwen-Hinderling,V.B.,Duez,H.,Lefebvre,P., Schaper,N.C.,etal.(2016).Demonstrationofaday-nightrhythminhuman skeletalmuscleoxidativecapacity.Mol.Metab.5,635–645.https://doi. .

9.Gabriel,B.M.,Altıntas,,A.,Smith,J.A.B.,Sardon-Puig,L.,Zhang,X., Basse,A.L.,Laker,R.C.,Gao,H.,Liu,Z.,Dollet,L.,etal.(2021).

STAR□METHODS

Detailedmethodsareprovidedintheonlineversionofthispaperandinclude thefollowing:

- KEYRESOURCESTABLE

Disruptedcircadianoscillationsintype2diabetesarelinkedtoaltered

CellMetabolism*38*,65–
81,January6,2026
77

rhythmicmitochondrialmetabolisminskeletalmuscle.Sci.Adv.*7*, eabi9654..

10.Wefers,J.,Connell,N.J.,Fealy,C.E.,Andriessen,C.,deWit,V.,van Moorsel,D.,Moonen-Kornips,E.,Jo¨rgensen,J.A.,Hesselink,M.K.C., Havekes,B.,etal.(2020).Day-nightrhythmofskeletalmusclemetabolism isdisturbedinolder,metabolicallycompromisedindividuals.Mol.Metab. *41*,101050..

11.Harmsen,J.-F.,vanWeeghel,M.,Parsons,R.,Janssens,G.E.,Wefers,J., vanMoorsel,D.,Hansen,J.,Hoeks,J.,Hesselink,M.K.C.,Houtkooper, R.H.,etal.(2022).Divergentremodelingoftheskeletalmusclemetabolomeover24hbetweenyoung,healthymenandolder,metabolically compromisedmen.CellRep.*41*,111786.

sampleofUKadultsineverydaylife.Proc.Natl.Acad.Sci.USA*120*, e2301608120..

23.Stothard,E.R.,McHill,A.W.,Depner,C.M.,Birks,B.R.,Moehlman,T.M., Ritchie,H.K.,Guzzetti,J.R.,Chinoy,E.D.,LeBourgeois,M.K.,Axelsson, J.,etal.(2017).Circadianentrainmenttothenaturallight-darkcycle acrossseasonsandtheweekend.Curr.Biol.*27*,508–513..

.

12.Vetter,C.,Dashti,H.S.,Lane,J.M.,Anderson,S.G.,Schernhammer,E.S., Rutter,M.K.,Saxena,R.,andScheer,F.A.J.L.(2018).Nightshiftwork,genetic risk,andtype2diabetesintheUKBiobank.DiabetesCare*41*, 762–769..

13.Buchvold,H.V.,Pallesen,S.,Øyane,N.M.F.,andBjorvatn,B.(2015). AssociationsbetweennightworkandBMI,alcohol,smoking,caffeine andexercise–across-sectionalstudy.BMCPublicHealth*15*,1112. .

14.Wefers,J.,vanMoorsel,D.,Hansen,J.,Connell,N.J.,Havekes,B.,Hoeks, J.,vanMarkenLichtenbelt,W.D.,Duez,H.,Phielix,E.,Kalsbeek,A.,etal. (2018).Circadianmisalignmentinducesfattyacidmetabolismgeneprofilesandcompromisesinsulinsensitivityinhumanskeletalmuscle.Proc. Natl.Acad.Sci.USA*115*,7789–7794..

15.Scheer,F.A.J.L.,Hilton,M.F.,Mantzoros,C.S.,andShea,S.A.(2009). Adversemetabolicandcardiovascularconsequencesofcircadian misalignment.Proc.Natl.Acad.Sci.USA*106*,4453–4458..

16.Morris,C.J.,Yang,J.N.,Garcia,J.I.,Myers,S.,Bozzi,I.,Wang,W., Buxton,O.M.,Shea,S.A.,andScheer,F.A.J.L.(2015).Endogenouscircadiansystemandcircadianmisalignmentimpactglucosetolerancevia separatemechanismsinhumans.Proc.Natl.Acad.Sci.USA*112*, E2225–E2234..

17.Petrenko,V.,Gandasi,N.R.,Sage,D.,Tengholm,A.,Barg,S.,andDibner, C. (2020).Inpancreaticisletsfromtype2diabetespatients,thedampened circadianoscillatorsleadtoreducedinsulinandglucagonexocytosis. Proc.Natl.Acad.Sci.USA*117*,2484–2495..

18.Petrenko,V.,Sinturel,F.,Loizides-Mangold,U.,Montoya,J.P.,Chera,S., Riezman,H.,andDibner,C.(2022).Type2diabetesdisruptscircadian orchestrationoflipidmetabolismandmembranefluidityinhumanpancreaticislets.PLoSBiol.*20*,e3001725..

19.Klepeis,N.E.,Nelson,W.C.,Ott,W.R.,Robinson,J.P.,Tsang,A.M., Switzer,P.,Behar,J.V.,Hern,S.C.,andEngelmann,W.H.(2001).The NationalHumanActivityPatternSurvey(NHAPS):aresourceforassessing exposuretoenvironmentalpollutants.J.Expo.Anal.Environ.Epidemiol. *11*,231–252..

20.Matz,C.J.,Stieb,D.M.,Davis,K.,Egyed,M.,Rose,A.,Chou,B.,and Brion,O. (2014).Effectsofage,season,genderandurban-ruralstatus ontimeactivity:CanadianHumanActivityPatternSurvey2(CHAPS2). Int.J.Environ.Res.PublicHealth*11*,2108–2124..

21.Schweizer,C.,Edwards,R.D.,Bayer-Oglesby,L.,Gauderman,W.J., Ilacqua,V.,Jantunen,M.J.,Lai,H.K.,Nieuwenhuijsen,M.,andKu¨nzli,N. (2007).Indoortime-microenvironment-activitypatternsinsevenregions ofEurope.J.Expo.Sci.Environ.Epidemiol.*17*,170–181..

22.Didikoglu,A.,Mohammadian,N.,Johnson,S.,vanTongeren,M.,Wright, P.,Casson,A.J.,Brown,T.M.,andLucas,R.J.(2023).Associationsbetweenlightexposureandsleeptimingandsleepinesswhileawakeina

24.Santhi,N.,Thorne,H.C.,vanderVeen,D.R.,Johnsen,S.,Mills,S.L., Hommes,V.,Schlangen,L.J.M.,Archer,S.N.,andDijk,D.-J.(2012).The spectralcompositionofeveninglightandindividualdifferencesinthesuppressionofmelatoninanddelayofsleepinhumans.J.PinealRes.*53*, 47–59..

25.Fleury,G.,Masi´s-Vargas,A.,andKalsbeek,A.(2020).Metabolicimplica-

tionsofexposuretolightatnight:lessonsfromanimalandhumanstudies. Obesity(SilverSpring)*28*,S18–S28..

26.Versteeg,R.I.,Stenvers,D.J.,Visintainer,D.,Linnenbank,A.,Tanck,M.W., Zwanenburg,G.,Smilde,A.K.,Fliers,E.,Kalsbeek,A.,Serlie,M.J.,etal. (2017).Acuteeffectsofmorninglightonplasmaglucoseandtriglycerides inhealthymenandmenwithtype2diabetes.J.Biol.Rhythms*32*, 130–142..

27.Harmsen,J.-F.,Wefers,J.,Doligkeit,D.,Schlangen,L.,Dautzenberg,B., Rense,P.,vanMoorsel,D.,Hoeks,J.,Moonen-Kornips,E.,Gordijn, M.C.M.,etal.(2022).Theinfluenceofbrightanddimlightonsubstrate metabolism,energyexpenditureandthermoregulationininsulin-resistant individualsdependsontimeofday.Diabetologia*65*,721–732..

28.Stefani,O.,Freyburger,M.,Veitz,S.,Basishvili,T.,Meyer,M.,Weibel,J., Kobayashi,K.,Shirakawa,Y.,andCajochen,C.(2021).Changingcolor andintensityofLEDlightingacrossthedayimpactsoncircadianmela- toninrhythmsandsleepinhealthymen.J.PinealRes.*70*,e12714. .

29.Cajochen,C.,Mu¨nch,M.,Kobialka,S.,Kra¨uchi,K.,Steiner,R.,Oelhafen, P.,Orgu¨l,S.,andWirz-Justice,A.(2005).Highsensitivityofhumanmela- tonin,alertness,thermoregulation,andheartratetoshortwavelengthlight. J.Clin.Endocrinol.Metab.*90*,1311–1316..

30.Hatori,M.,Le,H.,Vollmers,C.,Keding,S.R.,Tanaka,N.,Buch,T., Waisman,A.,Schmedt,C.,Jegla,T.,andPanda,S.(2008).Inducibleabla- tionofmelanopsin-expressingretinalganglioncellsrevealstheircentral roleinnon-imageformingvisualresponses.PLoSOne*3*,e2451..

31.Gu¨ler,A.D.,Ecker,J.L.,Lall,G.S.,Haq,S.,Altimus,C.M.,Liao,H.-W., Barnard,A.R.,Cahill,H.,Badea,T.C.,Zhao,H.,etal.(2008).Melanopsin cellsaretheprincipalconduitsforrod-coneinputtonon-image-forming vision.Nature*453*,102–105..

32.AmericanDiabetesAssociationProfessionalPracticeCommittee(2022). 6.Glycemictargets:standardsofmedicalcareindiabetes-2022. DiabetesCare*45*,S83–S96..

33.Seaquist,E.R.,Anderson,J.,Childs,B.,Cryer,P.,Dagogo-Jack,S.,Fish, L.,Heller,S.R.,Rodriguez,H.,Rosenzweig,J.,andVigersky,R.(2013). Hypoglycemiaanddiabetes:areportofaworkgroupoftheAmerican DiabetesAssociationandtheEndocrineSociety.DiabetesCare*36*, 1384–1395..

34.AmericanDiabetesAssociationProfessionalPracticeCommittee(2024). 6.Glycemicgoalsandhypoglycemia:Standardsofcareindiabetes- 2024.DiabetesCare*47*,S111–S125..

35.AmericanDiabetesAssociationProfessionalPracticeCommittee(2024). 7.Diabetestechnology:Standardsofcareindiabetes-2024.Diabetes Care*47*,S126–S144..

36.Phillips,N.E.,Collet,T.-H.,andNaef,F.(2023).Uncoveringpersonalized glucoseresponsesandcircadianrhythmsfrommultiplewearablebiosen- sorswithBayesiandynamicalmodeling.CellRep.Methods*3*,100545. .

37.Petrenko,V.,Saini,C.,Perrin,L.,andDibner,C.(2016).Parallelmeasurementofcircadianclockgeneexpressionandhormonesecretioninhumanprimarycellcultures.J.Vis.Exp.54673..

38.Perrin,L.,Loizides-Mangold,U.,Skarupelova,S.,Pulimeno,P.,Chanon,S.,Robert,M.,Bouzakri,K.,Modoux,C.,Roux-Lombard,P.,Vidal,H.,etal.(2015).Humanskeletalmyotubesdisplayacell-autonomouscircadianclockimplicatedinbasalmyokinesecretion.Mol.Metab.*4*, 834–845..

39.Dallmann,R.,Viola,A.U.,Tarokh,L.,Cajochen,C.,andBrown,S.A.(2012).Thehumancircadianmetabolome.Proc.Natl.Acad.Sci.USA*109*,2625–2629..

40.Sinturel,F.,Chera,S.,Brulhart-Meynet,M.-C.,Montoya,J.P.,Stenvers,D.J.,Bisschop,P.H.,Kalsbeek,A.,Guessous,I.,Jornayvaz,F.R.,Philippe,J.,etal.(2023).Circadianorganizationoflipidlandscapeisperturbedintype2diabeticpatients.CellRep.Med.*4*,101299..

41.Cheung,I.N.,Zee,P.C.,Shalman,D.,Malkani,R.G.,Kang,J.,andReid,K.J.(2016).Morningandeveningblue-enrichedlightexposurealtersmetabolicfunctioninnormalweightadults.PLoSOne*11*,e0155601. .

42.Meng,J.-J.,Shen,J.-W.,Li,G.,Ouyang,C.-J.,Hu,J.-X.,Li,Z.-S.,Zhao,H.,Shi,Y.-M.,Zhang,M.,Liu,R.,etal.(2023).Lightmodulatesglucosemetabolismbyaretina-hypothalamus-brownadiposetissueaxis.Cell*186*,398–412.e17..

51. Balsalobre, A., Damiola, F., and Schibler, U. (1998). A serum shock induces circadian gene expression in mammalian tissue culture cells. Cell *93*, 929–937..

52. Balsalobre, A., Marcacci, L., and Schibler, U. (2000). Multiple signaling pathways elicit circadian gene expression in cultured Rat-1 fibroblasts. Curr. Biol. *10*, 1291–1294..

53. Marcheva, B., Ramsey, K.M., Buhr, E.D., Kobayashi, Y., Su, H., Ko, C.H., Ivanova, G., Omura, C., Mo, S., Vitaterna, M.H., et al. (2010). Disruption of the clock components CLOCK and BMAL1 leads to hypoinsulinaemia and diabetes. Nature *466*, 627–631..

54. Woon, P.Y., Kaisaki, P.J., Braganc a, J., Bihoreau, M.-T., Levy, J.C., Farrall, M., and Gauguier, D. (2007). Aryl hydrocarbon receptor nuclear translocator-like (BMAL1) is associated with susceptibility to hypertension and type 2 diabetes. Proc. Natl. Acad. Sci. USA *104*, 14412–14417. .

43. Lu, J., Wang, C., Shen, Y., Chen, L., Zhang, L., Cai, J., Lu, W., Zhu, W., Hu, G., Xia, T., et al. (2021). Time in range in relation to all-cause and cardiovascular mortality in patients with type 2 diabetes: a prospective cohort study. Diabetes Care *44*, 549–555..

44. Andriessen, C., Fealy, C.E., Veelen, A., van Beek, S.M.M., Roumans, K.H.M., Connell, N.J., Mevenkamp, J., Moonen-Kornips, E., Havekes, B., Schrauwen-Hinderling, V.B., et al. (2022). Three weeks of time-restricted eating improves glucose homeostasis in adults with type 2 diabetes but does not improve insulin sensitivity: a randomised crossover trial. Diabetologia *65*, 1710–1720.

55. Xia, D., Yang, L., Cui, J., Li, Y., Jiang, X., Meca, G., Wang, S., Feng, Y., Zhao, Y., Qin, J., et al. (2021). Combined analysis of the effects of exposure to blue light in ducks reveals a reduction in cholesterol accumulation through changes in methionine metabolism and the intestinal Microbiota. Front. Nutr. *8*, 737059..

56. Zhu, H., Wang, N., Yao, L., Chen, Q., Zhang, R., Qian, J., Hou, Y., Guo, W., Fan, S., Liu, S., et al. (2018). Moderate UV exposure enhances learning and memory by promoting a novel glutamate biosynthetic pathway in the brain. Cell *173*, 1716–1727.e17.

.

57. Du, N.-H., Sinturel, F., Nowak, N., Gosselin, P., Saini, C., Guessous, I., Jornayvaz, F.R., Philippe, J., Rey, G., Dermitzakis, E.T., et al. (2024). Multi-omics correlates of insulin resistance and circadian parameters mapped directly from human serum. Eur. J. Neurosci. *60*, 5487–5504. .

. 59. Wilkin, C., Colonval, M., Dehairs, J., Esser, N., Iovino, M., Gianfrancesco,

45. Parr, E.B., Steventon-Lorenzen, N., Johnston, R., Maniar, N., Devlin, B.L., Lim, K.H.C., and Hawley, J.A. (2023). Time-restricted eating improves measures of daily glycaemic control in people with type 2 diabetes. Diabetes Res. Clin. Pract. *197*, 110569.

58. Ross, A.B., Maes, E., Lee, E.J., Homewood, I., Marsh, J.M., Davis, S.L., and Willicut, R.J. (2022). UV and visible light exposure to hair leads to widespread changes in the hair lipidome. Int. J. Cosmet. Sci. *44*, 672–684..

M.A.,Fadeur,M.,Swinnen,J.V.,Paquot,N.,Piette,J.,etal.(2021).New insightsonthePBMCsphospholipidomeinobesitydemonstratemodula-
. 60.Jeziorny,K.,Pietrowska,K.,Sieminska,J.,Zmyslowska-Polakowska,E.,
46.Munan,M.,Oliveira,C.L.P.,Marcotte-Che´nard,A.,Rees,J.L.,Prado, C.M.,Riesco,E.,andBoulé,N.G.(2020).Acuteandchroniceffectsofexerciseoncontinuousglucosemonitoringoutcomesintype2diabetes:a meta-analysis.Front.Endocrinol.*11*,495.
tionsassociatedwithinsulinresistanceandglycemicstatus.Nutrients*13*, 3461..

Kretowski,A.,Ciborowski,M.,andZmyslowska,A.(2023).Serummetabolomicsidentifiedspecificlipidcompoundswhichmayserveasmarkers
.

47.Takasu,N.N.,Hashimoto,S.,Yamanaka,Y.,Tanahashi,Y.,Yamazaki,A.,Honma,S.,andHonma,K.-I.(2006).Repeatedexposurestodaytime brightlightincreasenocturnalmelatoninriseandmaintaincircadianphase inyoungsubjectsunderfixedsleepschedule.Am.J.Physiol.Regul. Integr.Comp.Physiol.*291*,R1799–R1807.

ofdiseaseprogressioninpatientswithAlstro¨mandBardet-Biedlsyndromes.Front.Mol.Biosci.*10*,1251905.
.

61.Tonks,K.T.,Coster,A.C.,Christopher,M.J.,Chaudhuri,R.,Xu,A., Gagnon- turesofinsulinresistanceandoverweight/obesityinhumans.Obesity
Bartsch,J.,Chisholm,D.J.,James,D.E.,Meikle,P.J., Greenfield,J.R.,etal. (SilverSpring)*24*,908–916..
(2016).Skeletalmuscleandplasmalipidomicsigna-

. 62.Reutrakul,S.,andVanCauter,E.(2014).Interactionsbetweensleep, 48.Park,S.J.,andTokura,H.(1999).Brightlightexposureduringthe
circadianfunction,andglucosemetabolism:implicationsforriskand daytimeaffectscircadianrhythmsofurinarymelatoninandsalivary
severityofdiabetes.Ann.N.Y.Acad.Sci.*1311*,151–173.immunoglobulinA.Chronobiol.Int.*16*,359–371. .
.

49.Obayashi,K.,Saeki,K.,Iwamoto,J.,Okamoto,N.,Tomioka,K.,Nezu,S.,
Ikada,Y.,andKurumatani,N.(2012).Positiveeffectofdaylightexposure
onnocturnalurinarymelatoninexcretionintheelderly:across-sectional
analysisoftheHEIJO-KYOstudy.J.Clin.Endocrinol.Metab.*97*,4166–4173..

50.Wright,K.P.,Jr.,McHill,A.W.,Birks,B.R.,Griffin,B.R.,Rusterholz,T.,and
Chinoy,E.D.(2013).Entrainmentofthehumancircadianclocktothenat-
urallight-darkcycle.Curr.Biol.*23*,1554–1558.

63. Cedernaes, J., Osler, M.E., Voisin, S., Broman, J.-E., Vogel, H., Dickson, S.L., Zierath, J.R., Schiöth, H.B., and Benedict, C. (2015). Acute sleep loss induces tissue-specific epigenetic and transcriptional alterations to circadian clock genes in men. J. Clin. Endocrinol. Metab. *100*, E1255–E1261..

64.

Neerl.*6*,795–802.

65.Boubekri,M.,Cheung,I.N.,Reid,K.J.,Wang,C.-H.,andZee,P.C.(2014). Impactofwindowsanddaylightexposureonoverallhealthandsleepqual-ityofofficeworkers:acase-controlpilotstudy.J.Clin.SleepMed.*10*, 603–611..

66.Crowley,S.J.,Molina,T.A.,andBurgess,H.J.(2015).Aweekinthelifeof full-timeofficeworkers:workdayandweekendlightexposureinsummer

81. vanMarkenLichtenbelt,W.D.,Daanen,H.A.M.,Wouters,L.,Fronczek,R., Raymann,R.J.E.M.,Severens,N.M.W.,andVanSomeren,E.J.W.(2006). Evaluationofwirelessdeterminationofskintemperatureusingi Buttons. Physiol.Behav.*88*,489–497.

82. Duffy,J.F.,Dijk,D.J.,Klerman,E.B.,andCzeisler,C.A.(1998).Later andwinter.Appl.Ergon.*46*,193–200..

67. Small,L.,Lundell,L.S.,Iversen,J.,Ehrlich,A.M.,Dall,M.,Basse,A.L., Dalbram,E.,Hansen,A.N.,Treebak,J.T.,Barre`s,R.,etal.(2023). Seasonallighthoursmodulateperipheralclocksandenergymetabolism inmice.CellMetab.*35*,1722–1735.e5..

68. Dopico,X.C.,Evangelou,M.,Ferreira,R.C.,Guo,H.,Pekalski,M.L., Smyth,D.J.,Cooper,N.,Burren,O.S.,Fulford,A.J.,Hennig,B.J.,etal. (2015).Widespreadseasonalgeneexpressionrevealsannualdifferences inhumanimmunityandphysiology.Nat.Commun.*6*,7000..

endogenouscircadiantemperaturenadirrelativetoanearlierwaketime inolderpeople.Am.J.Physiol.*275*,R1478–R1487..

83.Bergstro¨m,J.,Hermansen,L.,Hultman,E.,andSaltin,B.(1967).Diet, muscleglycogenandphysicalperformance.ActaPhysiol.Scand.*71*, 140–150..

69.Bueschl,C.,Doppler,M.,Varga,E.,Seidl,B.,Flasch,M.,Warth,B.,and Zanghellini,J.(2022).PeakBot:machine-learning-basedchromatographic peakpicking.Bioinformatics*38*,3422–3428..

70.Dobin,A.,Davis,C.A.,Schlesinger,F.,Drenkow,J.,Zaleski,C.,Jha,S., Batut,P.,Chaisson,M.,andGingeras,T.R.(2013).STAR:ultrafastuniversalRNA-seqaligner.Bioinformatics*29*,15–21..

71.Anders,S.,Pyl,P.T.,andHuber,W.(2015).HTSeq:aPythonframeworkto workwithhigh-throughputsequencingdata.Bioinformatics*31*,166–169. .

72.Manders,R.J.F.,VanDijk,J.-W.M.,andvanLoon,L.J.C.(2010).Low-intensityexercisereducestheprevalenceofhyperglycemiaintype2diabetes.Med.Sci.SportsExerc.*42*,219–225..

73.Harris,J.A.,andBenedict,F.G.(1918).Abiometricstudyofhumanbasal metabolism.Proc.Natl.Acad.Sci.USA*4*,370–373..

74.

84.Sparks,L.M.,Moro,C.,Ukropcova,B.,Bajpeyi,S.,Civitarese,A.E., Hulver,M.W.,Thoresen,G.H.,Rustan,A.C.,andSmith,S.R.(2011). Remodelinglipidmetabolismandimprovinginsulinresponsivenessinhumanprimarymyotubes.PLoSOne*6*,e21068.

.

85. StHilaire, M.A., and Lockley, S.W. (2022). Measuring dim light melatonin onset in humans. In Melatonin. Methods and Protocols, *2550*, R. Jockers and E. Cecon, eds. (Springer), pp. 13–20..

86. Matyash, V., Liebisch, G., Kurzchalia, T.V., Shevchenko, A., and Schwudke, D. (2008). Lipid extraction by methyl-tert-butyl ether for high-throughput lipidomics. J. Lipid Res. *49*, 1137–1146..

87. Loizides-Mangold, U., Perrin, L., Vandereycken, B., Betts, J.A., Walhin, J.-P., Templeman, I., Chanon, S., Weger, B.D., Durand, C., Robert, M., et al. (2017). Lipidomics reveals diurnal lipid oscillations in human skeletal muscle persisting in cellular myotubes cultured in vitro. Proc. Natl. Acad. Sci. USA *114*, E8565–E8574..

88. Hannich, J.T., Loizides-Mangold, U., Sinturel, F., Harayama, T., Vandereycken, B., Saini, C., Gosselin, P., Brulhart-Meynet, M.-C., Robert, M., Chanon, S., et al. (2021). Ether lipids, sphingolipids and toxic 1-deoxyceramides as hallmarks for leanandobese type 2 diabetic patients. Acta Physiol. (Oxf.) *232*, e13610.

75. CIE (2018). CIE S 026/E:2018 CIE system for metrology of optical radiation

89. Robinson, M.D., McCarthy, D.J., and Smyth, G.K. (2010). edgeR: a Bioconductor package for differential expression analysis of digital gene expression data. Bioinformatics *26*, 139–140.

for ipRGC-influenced responses to light (International Commission on Illumination)..

76. Spitschan, M., Stefani, O., Blattner, P., Gronfier, C., Lockley, S.W., and Lucas, R.J. (2019). How to report light exposure in human chronobiology and sleep research experiments. Clocks Sleep *1*, 280–289..

77. Plasqui, G., Soenen, S., Westerterp-Plantenga, M.S., and Westerterp, K.R. (2011). Measurement of longitudinal changes in body composition during weight loss and maintenance in overweight and obese subjects using air-displacement plethysmography in comparison with the deuterium dilution technique. Int. J. Obes. (Lond) *35*, 1124–1130.

90. Reimand, J., Isserlin, R., Voisin, V., Kucera, M., Tannus-Lopes, C., Rostamianfar, A., Wadi, L., Meyer, M., Wong, J., Xu, C., et al. (2019). Pathway enrichment analysis and visualization of omics data using g:Profiler, GSEA, Cytoscape and EnrichmentMap. Nat. Protoc. *14*, 482–517..

78. Parrott, A.C., and Hindmarch, I. (1978). Factor analysis of a sleep evaluation questionnaire. Psychol. Med. *8*, 325–329..

79. Buysse, D.J., Reynolds, C.F., 3rd, Monk, T.H., Berman, S.R., and Kupfer,

91. Harmsen, J.-F., Kotte, M., Habets, I., Bosschee, F., Frenken, K., Jorgensen, J.A., de Kam, S., Moonen-Kornips, E., Cissen, J., Doligkeit, D., et al. (2023). Exercise training modifies skeletal muscle clock gene expression but not 24-hour rhythmicity in substrate metabolism of men with insulin resistance. J. Physiol. *602*, 6417–6433.

92.Hellemans,J.,Mortier,G.,DePaepe,A.,Speleman,F.,and Vandesompele,J. (2007).qBaserelativequantificationframeworkand
D.J.(1989).ThePittsburghSleepQualityIndex:anewinstrumentforpsychiatricpracticeandresearch.PsychiatryRes.*28*,193–213..
80.
softwareformanagementandautomatedanalysisofreal-timequantitativePCRdata.GenomeBiol.*8*,R19.
.

93. Pulimeno, P., Mannic, T., Sage, D., Giovannoni, L., Salmon, P., Lemeille, S., Giry-Laterriere, M., Unser, M., Bosco, D., Bauer, C., et al. (2013). Autonomous and self-sustained circadian oscillators displayed in human islet cells. Diabetologia *56*, 497–507.

94.Maier,B.,Lorenzen,S.,Finger,A.-M.,Herzel,H.,andKramer,A. (2021).SearchingnovelclockgenesusingRNAi-basedscreening. MethodsMol.Biol.*2130*,103–114..

95.Bylesjo¨M.,Rantalainen,M.,Cloarec,O.,Nicholson,J.K.,Holmes,E.,and Trygg,J.(2006).OPLSdiscriminantanalysis:combiningthestrengthsof PLS-DAandSIMCAclassification.J.Chemom.*20*,341–351.https://doi. .

96.Wiklund,S.,Johansson,E.,Sjo¨stro¨m,L.,Mellerowicz,E.J.,Edlund,U., Shockcor,J.P.,Gottfries,J.,Moritz,T.,andTrygg,J.(2008). VisualizationofGC/TOF-MS-basedmetabolomicsdataforidentification

ofbiochemicallyinterestingcompoundsusingOPLSclassmodels.Anal.
Chem.*80*,115–122..

STAR□METHODS

KEYRESOURCESTABLE

REAGENTorRESOURCE	SOURCE	IDENTIFIER
Biologicalsamples		
Humanbloodsamples	Thisstudy	N/A
Humansalivasamples	Thisstudy	N/A
Humanmusclebiopsysamples	Thisstudy	N/A
Culturedhumanmyotubes	Thisstudy	N/A
Chemicals,peptides,andrecombinantproteins		
13C2Taurine	CambridgeIsotopeLaboratories	CLM-6622-0.25
LabeledAminoAcidMix	CambridgeIsotopeLaboratories	MSKA2
LABELEDCARNITINESTANDARDSSETB	CambridgeIsotopeLaboratories	NSK-B
Fully13C-labelledYeastExtract.	ISOtopicSolutions	13C-labelledYeast
CMP-lyophilized	JenaScience	NU-1032-5G
2'-Deoxycytidine(dC)freebase	JenaScience	N-1074-25G
2'-Deoxyguanosinemonohydrate(dG)	JenaScience	N-DN-1003-25G
AMP-lyophilized	JenaScience	NU-1025-25G
dNTPBundle4xmM(dATP,dCTP, dGTP,dTTP)	JenaScience	NU-1005L
UMP-lyophilized	JenaScience	NU-1033-5G
(-)-Epicatechin	MetaSci	HMDB0001871
(+)-Pantothenicacid,sodiumsalt	MetaSci	HMDB0000210
2,2-Dimethylsuccinicacid	MetaSci	HMDB0002074
2,3-Dihydroxybenzoicacid	MetaSci	HMDB0000397
2,3-Pyridinedicarboxylicacid	MetaSci	HMDB0000232
2'-Deoxyadenosinemonohydrate	MetaSci	HMDB0000101
2-Amino-2-oxoaceticacid	MetaSci	PUBCHEMCID974
2-Ketobutyricacid(liquid)	MetaSci	HMDB0000005
2-Oxo-3-phenylpropanoicacid	MetaSci	HMDB0000205
3-Hydroxybenzoicacid	MetaSci	HMDB0002466
3-Hydroxyphenylaceticacid	MetaSci	HMDB0000440
3-Methylglutaricacid	MetaSci	HMDB0000752
4-Aminobenzoicacid	MetaSci	HMDB0001392
4-Guanidinobutyricacid	MetaSci	HMDB0003464
4-Hydroxyphenylpyruvicacid	MetaSci	HMDB0000707
5-Hydroxyindole-3-aceticacid	MetaSci	HMDB0000763
5-Methoxytryptamine	MetaSci	HMDB0004095
6-Hydroxynicotinicacid	MetaSci	HMDB0002658
Adenosine5'-triphosphatedisodiumsalt	MetaSci	HMDB0000538
Adenosinecyclophosphate	MetaSci	HMDB0000058
Adipicacid	MetaSci	HMDB0000448
Allantoin	MetaSci	HMDB0000462
Aminoadipicacid	MetaSci	HMDB0000510
Creatineanhydrous	MetaSci	HMDB0000064

Cytidine5′-triphosphate(disodiumsalt)	MetaSci	HMDB0000082
D-(-)-Quinicacid	MetaSci	HMDB0003072
D-(+)-GalactosamineHCl	MetaSci	PUBCHEMCID74493
D-(+)-Ribono-1,4-lactone	MetaSci	HMDB0001900
D-(+)-Xylose	MetaSci	HMDB0000098

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Continued

REAGENTorRESOURCE	SOURCE	IDENTIFIER
DL-6,8-Thioctamide/Lipoamide	MetaSci	HMDB0000962
DL-Mevalonolactone	MetaSci	HMDB0006024
D-Mannose	MetaSci	HMDB0000169
FlavinAdenineDinucleotideDisodiumSalt	MetaSci	HMDB0001248
Hydrate		
Folinicacidcalciumsalthydrate	MetaSci	HMDB0001562
Guanosine5′-triphosphatesodiumsalt	MetaSci	HMDB0001273
hydrate		
Hypoxanthine	MetaSci	HMDB0000157
Indole-3-aceticacid	MetaSci	HMDB0000197
Indole-3-ethanol	MetaSci	HMDB0003447
Indoline-2-carboxylicacid	MetaSci	PUBCHEMCID86074
Inosine	MetaSci	HMDB0000195
Inosine5′-triphosphatetrisodiumsalt	MetaSci	HMDB0000189
Inosine-5'-monophosphatesodiumsalt hydrate	MetaSci	HMDB0000175
IsoamylAcetate(liquid)	MetaSci	HMDB0031528
L-(-)-GlycericacidHemicalciumsalt(liquid)	MetaSci	HMDB0000139
L-(-)-Malicacid	MetaSci	HMDB0000156
L-Arabinose	MetaSci	HMDB0000646
L-Arabitol	MetaSci	HMDB0001851
L-Glutathioneoxidized	MetaSci	HMDB0003337
L-Homoserine	MetaSci	HMDB0000719
L-Hydrooroticacid	MetaSci	HMDB0000528
L-Sorbose	MetaSci	HMDB0001266
Malonicacid	MetaSci	HMDB0000691
N-Acetyl-D-galactosamine	MetaSci	HMDB0000212
N-Acetylglutamicacid	MetaSci	HMDB0001138
N-Acetylneuraminicacid	MetaSci	HMDB0000230
NAD	MetaSci	HMDB0000902
N-Carbamyl-L-GlutamicAcid	MetaSci	HMDB0062793
Nicotinicacid	MetaSci	HMDB0001488
O-Phosphorylethanolamine	MetaSci	HMDB0000224
Oroticacidanhydrous	MetaSci	HMDB0000226
Phospho(enol)pyruvicAcid	MetaSci	HMDB0000263
MonopotassiumSalt		
Phosphoserine	MetaSci	HMDB0000272
Pyridoxal5′-phosphatehydrate	MetaSci	HMDB0001491
Pyridoxalhydrochloride	MetaSci	HMDB0001545
Pyridoxamine2HCl	MetaSci	HMDB0001431
PyridoxineHCl	MetaSci	HMDB0000239

Riboflavin	MetaSci	HMDB0000244
Salicylicacid	MetaSci	HMDB0001895
Shikimicacid	MetaSci	HMDB0003070
Sodiumcreatinephosphatedibasic tetrahydrate	MetaSci	HMDB0001511
SodiumD-gluconate	MetaSci	HMDB0000625
Sodiumpyruvate	MetaSci	HMDB0000243
TaurocholicAcid(sodiumsalt)	MetaSci	HMDB0000036
Thiaminehydrochloride	MetaSci	HMDB0000235
trans,trans-MuconicAcid	MetaSci	HMDB0002349

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REAGENTorRESOURCE	SOURCE	IDENTIFIER
trans-Aconiticacid	MetaSci	HMDB0000958
Uricacid	MetaSci	HMDB0000289
Vanillicacid	MetaSci	HMDB0000484
Xanthine	MetaSci	HMDB0000292
Xanthosine	MetaSci	HMDB0000299
Xylitol	MetaSci	HMDB0002917
7,8-Dihydrofolicacid	schirckslaboratories	16.206_50mg
(6R,S)-5,10-Methenyl-5,6,7,8-tetrahydrofolicacidchloride	schirckslaboratories	16.23_50mg
(6R,S)-5-Methyl-5,6,7,8-tetrahydrofolic acid,calciumsalt	schirckslaboratories	16.235_50mg
2'-Deoxyadenosine5'-monophosphate	Sigma	D6375-100MG
(±)-Sodium2,3-dihydroxyisovalerate	Sigma	39693-10MG
hydrate_≥95%(CE)		
(4R)-4-Hydroxy-L-glutamic	Sigma	76157-10MG
acid_≥98.0%(TLC)		
(S)-Mevalonicacidlithiumsalt	Sigma	44714-10MG
13C1015N5AMP	Sigma	650676-1MG
13C1015N5dAMP	Sigma	900386
13C2Citricacid	Sigma	488607-100mg
13C515N1Folicacid	Sigma	803162-1MG
13C915N2UMP	Sigma	651370-1MG
2'-Deoxycytidine5'-monophosphate	Sigma	D7625-100MG
sodiumsalt		
2'-Deoxyuridine5'-triphosphateLithium	Sigma	1.142E+10
saltsolution		
2-Deoxyribose5-phosphatesodium	Sigma	D3126-25MG
salt_≥95%		
3-Dehydroshikimicacid_≥95.0%(HPLC)	Sigma	05616-10MG
4-Pyridoxicacid,≥98%	Sigma	P9630-25mg
5-Deoxy-5-(methylthio)adenosine	Sigma	D5011-25MG
AICAR(5-Aminoimidazole-4-carboxamide	Sigma	A9978-5MG
1-β-D-ribofuranoside)_≥98%(HPLC), powder		
Aminoacidstandards,physiological	Sigma	A6407-10ML
Argininosuccinicaciddisodiumsalt	Sigma	A5707-10MG
hydrate,≥80%		
cis-Aconiticacid,≥98%	Sigma	A3412-1G
D-(-)-Citramalicacidlithium	Sigma	06711-10MG
salt_≥95.0%(GC)		
D-erythro-Dihydrosphingosine_≥98%	Sigma	D3314-10MG
Dihydroxyacetonephosphatelithium	Sigma	37442-100MG-F
salt_≥95.0%(TLC)		
D-Sedoheptulose7-phosphatelithium	Sigma	78832-1MG
salt_≥90%(TLC)		

D-Xylulose5-phosphatelithium salt_≥90%(TLC)	Sigma	15732-1MG
Folicacid<97%	Sigma	F7876-1G
gamma-Glu-Cys_≥80%(HPLC)	Sigma	G0903-25MG
Guanosine3,5-cyclic monophosphate_≥98%(HPLC),powder	Sigma	G7504-5MG
L-2-Phosphoglycericaciddisodiumsalt hydrate_≥80%(CE)	Sigma	19710-50MG

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REAGENTorRESOURCE	SOURCE	IDENTIFIER
N-Acetyl-α-D-glucosamine1-phosphate	Sigma	A2142-5MG
disodiumsalt_≥95%		
Prephenicacidbariumsalt_≥75%	Sigma	P2384-10MG
S-5-Adenosyl-L-homocysteine_crystalline	Sigma	A9384-10MG
Sodium(±)-homocitratetribasic	Sigma	48488-10MG
Uridine5′-diphosphoglucosedisodium	Sigma	94335-100MG
salt_≥98.0%(HPLC)		
α-D(+)Mannose1-phosphatesodiumsalt hydrate_SigmaGrade	Sigma	M1755-10MG
α-D-Glucoheptonicacidsodiumsalt_	Sigma	G3516-100G
α-Ketoglutaricacid,≥99.0%(T)	Sigma	75890-25G
2'-Deoxyinosine	Sigma	D5287-100MG
2-Isopropylmalicacid	Sigma	333115-100MG
Cystathionine	Sigma	C3633-100MG
D-Fructose1,6-bisphosphatetrisodiumsalt hydrate	Sigma	F6803-10MG
D-Glucosamine6-phosphate	Sigma	G5509-10MG
D-Ribose5-phosphatedisodiumsalt hydrate	Sigma	R7750-10MG
L-Canavaninesulfatesalt	Sigma	C9758-100MG
SodiumL-lactate	Sigma	71718-10G
UDP-α-D-Galactose,DisodiumSalt-CAS	Sigma	670111-M
137868-52-1-Calbiochem		
Ureidosuccinicacid	Sigma	69037-100MG
α-D-Glucose1-phosphatedisodiumsalt	Sigma	G7000-1G
hydrate		
α-Hydroxyhippuricacid	Sigma	223875-5G
C18:1GlucoxylCeramide-d5	AVANTIPOLARLIPIDS	860673
SPLASHLIPIDOMIX	AVANTIPOLARLIPIDS	330707
15:0-18:1(d7)PC	AVANTIPOLARLIPIDS	791637
15:0-18:1(d7)PE	AVANTIPOLARLIPIDS	791638
15:0-18:1(d7)PS	AVANTIPOLARLIPIDS	791639
15:0-18:1(d7)PG	AVANTIPOLARLIPIDS	791640
15:0-18:1(d7)PI	AVANTIPOLARLIPIDS	791641
15:0-18:1-d7-PA	AVANTIPOLARLIPIDS	791642
18:1(d7)LPC	AVANTIPOLARLIPIDS	791643
18:1(d7)LPE	AVANTIPOLARLIPIDS	791644
18:1-d7-cholesterol	AVANTIPOLARLIPIDS	791645
18:1(d7)MG	AVANTIPOLARLIPIDS	791646
15:0-18:1(d7)DG	AVANTIPOLARLIPIDS	791647
15:0-18:1(d7)-15:0TG	AVANTIPOLARLIPIDS	791648
18:1(d9)SM	AVANTIPOLARLIPIDS	791649
Cholesterol(d7)	AVANTIPOLARLIPIDS	700041
Criticalcommercialassays		
RNeasytissuekit	Qiagen	74104
HighcapacityRNA-to- cDNAkit	ThermoFisher	4387406

Depositeddata		
RawRNA-Seqdata		GSE309688
RawMetabolomicData	ThisStudy	DataS3
RawLipidomicData	ThisStudy	DataS3

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Continued

REAGENTorRESOURCE	SOURCE	IDENTIFIER
Softwareandalgorithms		
R(v4.5.0)	CRAN	
TraceFinder4.1	ThermoFisherScientific	
PeakBot(v0.9.54)	Bueschletal.[69]	Version0.9.54
FastQC(v0.11.9)	BabrahamBioinformatics	Version0.11.9
STARaligner(v2.7.0f)	Dobinetal.[70]	Version2.7.0f
HTSeq(v0.9.1)	Andersetal.[71]	Version0.9.1
edgeR(v3.38.4)	Bioconductor(R)	Version3.38.4
clusterProfiler(v4.9.0)	Bioconductor(R)	Version4.9.0
org.Hs.eg.db(v3.15.0)	Bioconductor(R)	Version3.15.0
limma(v3.58.1)	Bioconductor(R)	Version3.58.1
Other		
VanquishUHPLCsystem	ThermoFisherScientific	
Orbitrap-basedmassspectrometer	FusionLumos,ThermoFisherScientific	
AccucoreC18	ThermoFisherScientific	17126-152130

EXPERIMENTALMODELANDSUBJECTDETAILS

Participantrecruitment

Menandpostmenopausalwomenwithtype2diabetes(T2D),agedbetween40and75years,wererecruitedthroughadvertisements inthevicinityofMaastricht.ParticipantswereincludediftheywerediagnosedwithT2Dforatleastoneyearandwerewellcontrolled withrespecttoglycemiccontrolandstablediabetesmedicationregimens(moreinformationinTable1).Furthermore,participants

needed to have a stable body weight (no gain or loss of >5 kg in the last three months), a BMI of ≥25 kg/m^2, a regular sleep pattern (7-9

h/night) and a habitual bedtime of 23:00±02:00 h. Participants were excluded if they were on sodium-glucose transport protein 2 (SGLT2) inhibitors or insulin treatment. Other exclusion criteria were uncontrolled hypertension, any other diabetes related co-morbidities like active cardiovascular disease, active diabetic foot, polyneuropathy or retinopathy, and active liver or kidney malfunctions.

Extreme morning or evening chronotypes based on the MEQ-SA questionnaire (score of ≤30 for extreme evening or ≥70 for extreme

morning type), shift work or travel across more than one time zone in the three months prior to the study and heavily varying sleep-wake rhythms were reasons for exclusion. Additional exclusion criteria were frequent engagement in programmed exercise, alcohol consumption of >2 servings per day for men and >1 serving per day for women, significant food allergies that hamper study participation, participants who did not want to be informed about unexpected medical findings, previous enrolment in a clinical study with an investigational product during the last 3 months, and smoking in the last six months. In- and exclusion criteria were assessed by a screening visit during which basic physical examination was performed (age, height, weight, and blood pressure), a fasting blood sample was obtained, and questionnaires were gathered. The overall eligibility of participants to be able to participate in the study was always assessed by a medical doctor. The study was executed in accordance with the declaration of Helsinki and was approved by the medical ethical committee of the Maastricht University Medical Center. The current study was registered at () with the identifier NCT05263232. All participants provided written informed consent. The experiments took place between April and October 2022 to avoid confounding by the transition from Europe and daylight-saving time to standard time and its associated change in photoperiod.

METHODDETAILS

Samplesizecalculation

Wehypothesizedthatnaturalofficelightingduringthedaywouldimproveaverage24hglucoselevelscomparedtoartificialoffice light.Apreviousstrialinvestigatingtheeffectofanacuteboutoflow-intensityexerciseonaverage24-hourglucoselevels[72]found areductionofglucoselevelsfrom9.4±0.8mmol/Lto7.8±0.9mmol/L(mean±SEM)correspondingtoameanchangeof~17%.Weexpectedt hepotentiallybeneficialeffectofnaturaldaylightvs.artificiallighttobelesspronouncedcomparedtotheeffect oflow-intensityexerciseandestimatedthateffecttobe10%.Toestimatetherequiredgroupsize(n)weusedtheG*Powercalculator 3.1forastudydesignwithtwodependentsamples(matchedpairs,two-sidedt-test).Weassumeda~12%between-subjectvari-abilityofthechangeinaverage24hglucosemeasuredbyCGM,basedontheaforementionedstudy,[72]withapower(1-β)of80%and asignificancelevel(α)of5%,thesamplesizeneededforourstudywas14.Thestudywasprematurelyterminatedafter13completed participants,sincewecouldonlytestthatmanyparticipantsoverthesummertime(ApriltoOctober2022)beforethetransitionfrom daylightsavingtimetostandardtimeoccurred.IntheNetherlands,thephotoperiodprogressivelydecreasesfromthatpointon,so thattheartificiallightconditionwouldhavebeenbrighterthanthenaturallightconditionatthebeginningandattheendofoffice hours.Furthermore,theofficeenvironmentwasnolongeravailablefromApril2023onwards.

Studydesign

Randomizationandallocationofeligibleparticipantstotheorderofthetwolightconditionswerecarriedoutbysequentialnumbering (7participantsstartedwithnaturallightastheirfirstinterventionperiod,and6participantsviceversa).Awashoutofatleastfour weekswaskeptbetweeninterventionperiods.Duringthe3-dayrun-inperiodathome,participantswereinstructedtogotobed at23:00±00:30handwakeupat07:00±00:30h,avoidtheconsumptionofcaffeineandalcohol,followanormaleatingpattern (3mealsadayatregulartimes:09:00,14:00and19:00±00:30h),refrainfromstrenuousexerciseandavoidexcessivebrightor dimlightdurintheday.Asleepandfooddiarywereprovidedtoparticipantstomonitorsimilarbehaviorforbothrespectiverun-inperiods.Adherencetotheactivityandbedtimesathomewasfurthermonitoredusingwrist-wornactigraphy(ActiwatchSpectrum Plus,PhilipsRespironics,Inc;Murrysville,PA,USA).Inthemorningbetween07:00and08:00hofday2to5,participantsstayedin artificialindoorlightingof<400lux(seemoredetailsbelowinthesection‘‘quantificationoflightcharacteristics’’)andhadthe opportunitytotakeashower.Roomtemperaturewasmonitoredeverydayduringofficelightinghours.

Studymeals

Duringtheinterventionperiods,participantswereprovidedwithstandardizedmealsmatchingtheirenergyrequirementsatfixed times.Forthispurpose,energyrequirementswereestimatedbasedontheHarris-Benedictformula[73]multipliedwithanactivityfactor of1.5.Breakfastwasservedat9:00handconsistedof~21%oftheestimateddailyenergyintake(E%).Lunchwasservedat14:00h (~30E %)anddinnerwasservedat19:00h(~49E%).Breakfastandlunchwerebread-based,whilethedinnermainlyconsistedofa pre-packagedmeal.Noothersnacksordrinkswereprovidedbetweenmealsexceptforwateranddecaffeinatedteaorcoffee.Daily macronutrientcompositionwasaimedtoresemblethetypicalWesterndietwith~31%ofthedailyintakefromfats(~9%energyfrom saturated),~55%fromcarbohydratesand~14%fromproteins.Identicalmealswereservedforbreakfastandluncheachday,while thetypeofdinnermealchangedbetweendays.Nonetheless,theoverallmealplanwaskeptidenticalforboth4.5-dayintervention periods.Participantsmaintainedtheirdiabetesmedicationscheduleofingestingrespectivepillsconcurrentlywithmeals.To standardizephysicalactivity,participantsperformedsomelightphysicalexercisefor30min(e.g.3x5minofsteppingonasingle stairinterspersedwith5minofstanding)threetimesperday,onehouraftereachmeal.

Quantificationoflightcharacteristics

Lightcharacteristicsinthenaturalofficelightconditionweremeasuredatregularintervalseachdayduringofficehours(e.g.at08:00, 10:00,12:30,14:00,15:30and17:00forday1today4;at09:00,10:00,11:00,12:00and13:00onday5duringthemixedmealtoler-ancetest)withaspectrometer(Ai101SpectralIrradianceMeter,Apacer,Taiwan).Artificiallightsourcesweresetuptoachievealight intensityofapproximately300luxphotopicilluminance(±5lux),whichwasmonitoredonlyoncedailyduetotheconstantnatureof thislightcondition.Onalloccasionsthatparticipantshadtoleavetheofficeroomoftheirrespectivelightconditionorcouldbe exposedtooutdoorlightortotoobrightlightintheeveningwhengoingtothetoilet,orange-tintedblue-lightblockingglasses (FigureS12A)wereworn.Byusingtheseorange-tintedblue-lightblockingglasses,weensuredthatthroughoutthe4.5-daystudy periodintheartificiallightconditiontheparticipants’eyeswereneverexposedtotheenrichedshort-wavelengthsofdaylight,which arethewavelengthstowhichthehumancircadiansystemismostsensitive.Tonotinduceanyeffectofusingtheseorange-tinted blue-lightblockingglassesonlyintheartificiallightcondition,participantsalsoworetheseglassesatthesameinstancesinthenat-urallightcondition,wheneverleavingthecontrolledofficelightenvironment.Inbothlightconditions,between07:00and08:00aswell as17:00and18:00h,participantsstayedinindoorlightingconditionsof<400lux.Subsequently,from18:00htobedtimeat23:00h, participantsstayedindimlightconditions(<5lux)andthetelevisionandotherelectricdeviceswerealsosetto<5lux.Nightswere spentincompletedarknessfrom23:00to7:00h.

Allspectrometermeasurementswerecarriedoutverticallyintheoutwarddirectionoftheopticalaxisattheeyelevelsimulatinga participantsittingattheofficedeskeitherfacingthewindow(innaturalofficelight),thewall(inartificialofficelight),orthetelevision intheeveningindimlight(forbothlightinterventions),orthecomputerwhensittingatthedeskandoncefacingtheTVsittinginbed

(seeFigureS12Bforaphotooftheeveningroom).Theselattertwopositionswereselectedasvolunteersusuallyspentmostoftheir timeintheeveningsinthismannerofdirectingtheirview.Thecollecteddatawereextractedandfurtherconvertedfromrelativeto absoluteirradiancevaluesby.[74]Averageilluminance,-opic(ir)radiances[75]andspectralirradiancesforthenatural officelightfromday1today4overallparticipantsarereportedaccordingtoSpitschanetal.[76]inTableS1.Similarly,light specificationsforthetimeperiodsoutsideofofficehours(e.g.07:00-08:00,17:00-18:00and18:00-23:00h)arereportedin TableS2.Onlyfluorescentlightsourceswereinstalledintherespectivelaboratoryrooms.

Onday3,participantsstayedinarespirationchamberfrom18:00to07:15thenextmorning.From18:00to23:00,dimlight(<5lux) wasswitchedon,andduringthedarksleepingperiod,participantscouldonlyswitchonthisdimlight,whengoingtothetoiletwithin therespirationchamber.Theinstalledlightsystemwithintherespirationchamberconsistedoflight-emittingdiode(LED)wall washers(PhilipsSkyRibbonIntelliHueWallWashingPowercore).Bycombiningandpreciselycontrollingmultiplechannelsofthe LEDlight,thesystemcanproducewhitelight.Thedescribeddimlightconditionwassettoacorrelatedcolourtemperatureof 4000K.Detailedspectralpropertiesofthedimlightconditionwithintherespirationchamberweresimilarasreportedpreviously[27] andcanbeaccessedusing(under‘‘RelatedItems’’,photosofthe spectralmeasurementset-upinFiguresS12CandS12D).

Continuousglucosemonitoring

Afterarrivalonday1ofeachinterventionperiod,acontinuousglucosemonitor(CGM)(FreestyleLibreProiQ,AbbottLaboratories, USA)wasplacedonthedominantupperarmoftheparticipants.Interstitialglucoselevelsweremeasuredevery15minutesacross theentireinterventionperiod.DataoftheCGMwerenotshowntotheparticipantsduringthestudy.TheCGMwasremovedatthe endoftheinterventionperiodonday5.Toquantifytheprimaryoutcomeofthisstudy,namelyglucosecontrol,continuousglucose monitordatawasobtainedovertheentireexperimentalperiodspentintheresearchfacilitiesof4.5daysforbothnaturalandartificial officelighting.Thesecontinuousdataweredividedintocategoriesbasedonthe2022clinicalpracticerecommendationsof theAmericanDiabetesAssociation(ADA),[32]whichwereavailablewhenourstudywascommenced:hypoglycemia<4.0mmol/l, lowbloodglucose4.0–4.3mmol/l,normalrange4.4–7.2mmol/l,highbloodglucose7.3–9.9mmol/l,and hyperglycemia>10.0mmol/l.Resultsarereportedaspercentagesoftimespentintherespectivecategories.Timeinnormalrange (TIR)wasalsocalculatedaccordingtothemostrecent(2024)recommendationsoftheADA.[35]

Togeneratedeeperinsightsinto24-hourglucosedynamicsbetweenlightconditions,wefurthermodelledtheCGMdatausinga computationalframework,[36]takingtherecurringstandardizedmealtimes(e.g.09:00,14:00and19:00h)intoaccount.Inbrief,the mealtimesareusedtomodelpostprandialinterstitialglucoseincreases,andboththesizeoftheglucosespike(termed‘‘response heights’’)aswellasthetimeneededforglucosetoreturntobaseline(termed‘‘responsehalf-life’’)areinferredforeachindividualand eachlightconditionindependently.Themodelalsoincludesanunderlying24-hourcosinorfunctiontodescribe24-houroscillations inbaseline(i.e.irrespectiveofmealintake)glucoselevels.This24-hourcosinorfunctionisdescribedby‘‘baseline’’,‘‘peak-to-trough amplitude’’and‘‘peaktime’’parameters,whicharesimilarlylearntforeachparticipantandlightcondition.Themodelistrainedby minimizingtheerrorbetweenthemodelandtheCGMdatausingamaximumaposterioriprobability(MAP)estimatefrom200 differentstartingconditions,andthesettingsandparametersoftheoriginalmethodwereleftunchanged.Associationsbetween classicandmodelledCGMparameterswerequantifiedbycombiningthenaturalandartificiallightconditionsandusingalinear mixedregressionmodelaccountingforrepeateddatainindividualparticipants.Themodelwasfittedusingthe’MixedLM’class fromthe’statsmodels’library(v0.13.0).

Bodycomposition

Tomonitorifbodycompositionremainedstableoverthewashoutperiodbetweenlightconditions,bodycompositionwas determinedinthefastedstatedinthemorningofday2ofeachinterventionperiod.Participants’bodymassandbodyvolume weredeterminedusingairdisplacementplethysmography(BodPod,CosMed,Italy),accordingtothemanufacturer’sprotocol,as reportedpreviously.[77]

Sleepandmoodquestionnaires

Eachmorningafterparticipantswokeup,participantscompletedtheLeedsSleepingEvaluationQuestionnaire(LSEQ).[78]Onday2 theyalsofilledinthePittsburghSleepQualityIndex(PSQI)questionnaire,whichprovidesinformationonsleepqualityoverthe previousfourweeks.[79]Attheendofeachofficeday(day1to4,at16:45h)participantsalsocompletedamoodandcomfort questionnaire(e.g.visualanaloguescales).

Indirectcalorimetry

Ontheeveningofday3(from18:00honwards)participantsenteredarespirationchamber,inwhichtheystayeduntil07:00hinthe morningofday4.Duringthistimetheoxygenconsumptionandcarbondioxideproductionwerecontinuouslymeasured(Omnical, MaastrichtInstruments,Maastricht,TheNetherlands)andfurtherusedtocalculatewhole-bodyenergyexpenditure,therespiratory exchangeratio(RER)andcarbohydrateandfatoxidation.Onday4,oxygenconsumptionandcarbondioxideproductionwere measuredviaaventilatedhoodsystemwhilerestingonabedfor30minutesat08:30,13:00,18:00and22:00h(Omnical,Maastricht Instruments,Maastricht,TheNetherlands).Theday4measurementsat08:30and13:00hwereconductedintherespectiveoffice lightsettingwhereasmeasurementsat18:00and22:00hwereconductedunderdimlightconditions.Energyexpenditureand

substrateoxidationwerecalculatedusingtheBrouwerequation[80]withproteinoxidationbeingestimatedas12.4%ofenergyexpenditure.Sleepingmetabolicratewasdefinedasthelowestcontinuous3-hourperiodofenergyexpenditureduringthenightspentintherespirationchamber.

Ambient,skinandcorebodytemperature

Ambienttemperaturewascontinuouslymonitoredineveryroom,inwhichparticipantsspenttimeacrossthe4.5-dayinterventions.Forthatpurpose,amobilethermometer(ThermoPro,Canada)waspositionedatdeskheightintherespectiveofficelightconditionaswellasinthebedroomsinwhichparticipantsspenteveningsandnights.Temperaturedatawereloggedbihourlyduringdaytime (07:00-23:00h).Fromday3at08:00htoday4at08:00h,skintemperaturesweremeasuredat14ISO-definedbodysitesusingwirelesstemperaturesensors(iButtons,MaximIntegratedProducts,USA).[81]Corebodytemperature(CBT)wasmeasuredusing

aningestibletelemetricpill(e-Celsius,BodyCap,France),whichparticipantsswallowedwiththelunchmealat ~14:00onday3

toassessCBTover ~24hoursuntilthepillwasexcretedwiththestool.ThenadirinCBTisoftenusedasanindicatorofcircadian

phase[82]besidesdim-lightmelatoninonset(DLMO).Forthispurpose,wedefinedthenadirinCBTasthetimeatwhichtheminimumCBToccurredaveragedover15-minintervalstoexcludeartefacts.

Bloodpressureandheartrateassessment

Bloodpressureandheartrateweremeasuredatresteveryhourstartingat08:00hduringthewakingperiodofday4,andeverytwohoursfrom00:00honwardswhileasleepinthenightbetweenday4today5.Duringwakehours(07:00-23:00h)participantswere sittingatadeskwhentheautomatedcuff(OmronHealthcareCo.,Ltd.,Kyoto,Japan)wasapplied,whileparticipantswerelyinginbedfrom23:00-07:00h.Bloodpressureandheartrateweremeasuredduringthewakingperiodthreetimesforeverytimepoint,andsystolic,diastolicandheartratewereaveragedoutofthesethreemeasurementsforalltimepoints.Duringthenightthemeasurementwasperformedonlyoncetominimizedisturbanceofsleep.

24-hourbloodandeveningsalivasampling

Inthemorningonday4,anintravenouscannulawasplacedintheparticipants'forearmforblooddrawsat08:00,10:00,12:00,13:00,14:00,16:00,18:00,20:00,22:00,23:00,00:00,02:00,04:00,06:00and08:00hthenextmorning.Bloodsampleswerecollectedin EDTAplasmaandserumseparatortubesandcentrifugedat1300gfor10minat4◦Candroomtemperature,respectively.Plasma andserumwerepipettedinaliquotswhichwerefrozeninliquidnitrogenandstoredat-80◦Cforlateranalysis.

SalivawascapturedusingSalivetteat18:00,19:00,20:00,20:30,21:00,21:30,22:00,22:30and23:00hduringday4.Participants chewedonaswabforapproximately2minafterwhichtheyputtheswabbackintheSalivettecontainer.Thecontainerwas centrifugedat4◦Cfor2minutesat1000g.Thesalivawaspipettedinaliquots,whichwerefrozeninliquidnitrogenandstoredat-80◦Cforlateranalysis.

Skeletalmusclebiopsy

Onthemorningofday5between07:45and08:45h,askeletalmusclebiopsywastakenintheovernightfastedstate.Themuscle biopsywastakenfromthemuscle*vastuslateralis*underlocalanesthesia(1%lidocaine,withoutepinephrine)accordingtothe Bergstromtechnique.[83]Theleginwhichthemusclebiopsywastakenduringthefirstinterventionperiodwasrandomized,andin thesecondinterventionperiodthebiopsywastakenfromtheoppositeleg.Partofthemusclebiopsy(50-100mg)wasprocessed forisolationofhumanprimarymusclesatellitecells.Subsequentlymyogenichumanprimarymusclesatellitecellswereselectedwith MACScellsortingusingamouseCD56-antibodyaspreviouslydescribed.[84]Theremainingmusclesampleswerefreedfromany visiblenon-musclematerial,directlyfrozeninmeltingisopentaneandstoredat-80◦Cforlateranalysis.

Mixedmealtolerancetest

Inthemorningofday5,amixedmealtolerancetest(MMTT)wasperformed.Atapproximately09:00hparticipantsconsumedameal shake(EnsurePlus,AbbottLaboratories,USA)withthesameenergycontentastheirdailybreakfastofthemealplan.Macronutrient andenergycompositionper100mloftheshakewere16.8gcarbohydrates,9.1gprotein,4.8gfatand150kcal.Bloodsampleswere takenduringtheMMTTat0,15,30,45,60,90,120,150,180and240minafterconsumptionofthemeal.Indirectcalorimetrywas performedat30,60,120,180,and240min.Unfortunately,baselineindirectcalorimetrymeasurementscouldnotbeperformeddue tologisticalreasons.Plasmaglucose,freefattyacidsandserumtriacylglycerollevelsweredeterminedasdescribedabove,whereas insulinlevelsweremeasuredinserumwithanELISAkit(CrystalChem,ElkGroveVillage,USA).

Salivarymelatoninassessment

Melatoninconcentrationwasmeasuredbyradio-immuno-assay(RK-DSM2ofNovolytix,Basel,Switzerland).Intra- assayCVwas 32.5%forlowconcentrations(1.0pg/ml)and10.9%forhighconcentrations(13.9pg/ml).Inter-assayCVwas30.9%forlowconcentrations(0.9pg/ml)and8.7%forhighconcentrations(15.3pg/ml).Thelowerlimitofquantificationwas0.33pg/ml.Dim-lightmelatoninonset(DLMO)wasdeterminedaccordingtoSt.Hilaire&Lockley.[85]Inshort,theDLMOthresholdwassetto3pg/ml,andafter firstexceeding3pg/ml,melatoninconcentrationshadtobehigherthanorequalto3pg/mlforthenexttwotimepointstofulfilthe DLMOcriterium(ifthesetimepointswereavailable).ExactDLMOwascalculatedbylinearinterpolationbetweenthelastpointbefore andthefirstpointthatreachedtheDLMOcriterium.Ifforatleastonelightcondition,noDLMOcouldbedetectedoverthesaliva

samplingperiod(n=3)orifmelatoninconcentrationwereabove3pg/mlfrom18:00honwards(n=1),individualswereexcludedfrom furtherDLMOanalyses.

Plasma,serumandmonocyteanalysis

Plasmaglucose,freefattyacidsandserumtriacylglycerolweremeasuredcolorimetricallyusingaCobasPentraC400analyser (Horiba,Montpellier,France).Serummetabolomicsandlipidomicsanalyses,aswellasmonocytetranscriptomics,werecarried outinsamplesobtainedat08:00and16:00honday4.Formetabolomics/lipidomics,twentymicrolitersofserumwerespiked withamixtureofheavyisotopelabelledinternalstandards.Samplesformetabolomicswereextractedusingmethanolandwere runusinga1290InfinityIIUHPLCsystem(AgilentTechnologies)coupledwitha6470triplequadrupolemassspectrometer(Agilent Technologies)fortheLC-MS/MSanalysis.Forlipidomicanalysis,sampleswereextractedwithMethyltert-butyletherand methanol.[86]TheLC-MSanalysiswasperformedusingaVanquishUHPLCsystem(ThermoFisherScientific)combinedwithan OrbitrapFusionLumosTribridmassspectrometer(ThermoFisherScientific),aspreviouslydescribed.[40,87,88]Missingdatawere imputedas1/5thofthelowestvaluepermetaboliteandlipid.Datawerescaled,probabilisticquotientnormalizedandlog2 transformed.

Forbloodmonocyteisolation,8mLbloodwascollectedinaCPT-tube(BDvacutainerCPT,BD,FranklinLakes,USA)andcentrifugedfor20minat1800*g*.Thereafter,mononuclearcellswerecollectedandwashedwithPBS,centrifugedfor8minat350*g*.Cells werefrozenat-80◦Cinafreezingmediumcontaining10%DMSO.CD14+magneticbeads(MiltenyiBiotec)wereaddedtothawed mononuclearcells(20µlper10^{7}cells)andthemixturewasincubatedfor15minutesat4◦C.Afterincubation,thesamplewaswashed with2mLofseparationbufferper10^{7}cellsandcentrifugedat500gfor10minutesatroomtemperaturewithoutbrake.Thesupernatantwasdiscarded,andseparationwasperformedusinganAutomacspro-separator(MiltenyiBiotec)underthe‘‘Posselweb’’program.Theresultingmonocytefractionwascentrifugedat330gfor10minutesat4◦C,resuspendedinPBS,andcentrifugedagain at700gfor3minutes.Thefinalmonocytepelletwasstoredat-80◦Cuntilfurtheruse.

Serummassspectrometryanalyses

ForthemetabolomicsanalysesthechromatographicseparationforsampleswascarriedoutonaZORBAXRRHDExtend-C18,2.1x 150mm,1.8µmanalyticalcolumn(AgilentTechnologies).Thecolumnwasmaintainedat40◦Cand4µLofsamplewasinjectedper run.ThemobilephaseAwas3%methanol(v/v),10mMtributylamine,15mMaceticacidinwaterandmobilephaseBwas10mM tributylamine,15mMaceticacidinmethanol,usingagradientwith22minduration.Thetriplequadrupolemassspectrometerwas operatedinnegativeelectrosprayionizationmode.Themetabolitesofinterestweredetectedusingadynamic MRMmode.PeakBot software(vers.0.9.54)wasusedfordataprocessing.Ten-pointcalibrationcurveswithinternalstandardizationwereconstructedfor absolutequantificationofmetabolites.

Forthelipidomicanalysis,LipidseparationwasperformedbyreversedphasechromatographyemployinganAccucoreC18, 2.6µm,150x2mm(ThermoFisherScientific)analyticalcolumnatacolumntemperatureof35◦C.AsmobilephaseAanacetoni-trile/ water(50/50,v/v)solutioncontaining10mMammoniumformateand0.1%formicacidwasused.MobilephaseBconsistedof acetonitrile/isopropanol/water(10/88/2,v/v/v)containing10mMammoniumformateand0.1%formicacid.Theflowratewassetto 400µL/min.Themassspectrometerwasoperatedin ESI-positiveand-negativemodeandtheOrbitrapMSwassetinscanmodeat 120000massresolutionwithinarangefrom250-1200m/z.Thedataanalysiswasperformedusingthe TraceFindersoftware (ThermoFisherScientific).Missingdatawereimputedas1/5thofthelowestvalueperlipid.Datawerescaled,mean-centeredand log2transformed.

Monocytetranscriptomicanalyses

RNAquantificationwasperformedwithaQubitfluorimeter(ThermoFisherScientific)andRNAintegrityassessedwithaBioanalyzer (AgilentTechnologies).TheIlluminaStrandedmRNAPrepkit(Illumina)wasusedforthelibrarypreparationwith200ngoftotalRNAas input.Librarymolarityandqualitywereassessedwiththe QubitandTapestation(AgilentTechnologies).Librarieswere sequencedon aNovaSeq6000sequencer(Illumina)using100-bpsingle-endreadsprotocolattheiGE3GenomicsPlatformoftheUniversityof Geneva.Qualitycontrolwasperformedwith FastQCv.0.11.9.Thereadswerealignedwith STARv.2.7.0f[70]tothehumanUCSC genomehg38.Thegeneexpressionwasquantifiedwith HTSeqv.0.9.1.[71]Filteringoutlowlyexpressedgenes,normalizationanddifferentialexpressionanalysiswereperformedwiththeR/Bioconductorpackageedge Rv.3.38.4.[89]Statisticalsignificancewasassessedwithagenerallinearmodel,negativebinomialdistribution,andquasi-likelihoodFtest.Over-representationanalysisof GeneOntologytermsandKEGGpathwayswereperformedwiththeR/Bioconductorpackages:clusterProfilerv.4.9.0andorg.Hs.eg.db3.15.0.Transcriptdifferentialexpressionandgenesetenrichmentanalyseweredoneusingthelimmapackage(version 3.58.1)inR(version4.3.1),usingtheBaderLabgenesetresource()withclustering doneusingEnrichmentMapandAutoAnnotate.[90]

Musclegenetranscriptquantification

Partofthemusclematerial(10mg)wasusedforRNAisolationandgenetranscriptabundancedeterminationbyreal-timePCRas describedinHarmsenetal.[91]Tominimizethevariabilityinreferencegenenormalization,thegeometricmeanoftworeferencegenes (RPL26andRPLP0)wasusedastheinternalreferenceforcomparativegeneexpressionanalysisbetweenconditions[92]for*Bmal1, Clock,Cry1,Per1,Per2,Per3,Reverba*,and*Rora*.

Primaryhumanskeletalmyotubecultureandlentiviraltransduction

PrimarymyoblastswereculturedinHam'sF-10NutrientMix,GlutaMAXSupplementmedium(Gibco,Paisley,Scotland,UK), enrichedin20%fetalbovineserum(Gibco,Paisley,Scotland,UK),1%penicillin/streptomycin(Gibco,Paisley,Scotland,UK), 0.5%gentamycin(SigmaAldrich,SaintLouis,USA)and1:500amphotericinB(Gibco,Paisley,Scotland,UK)untilreachingthe 60-70%ofconfluence.MyoblastsweretransducedwithBMAL1-luciferase(BMAL1-luc)lentivectorsatMOI=3.[38]Afterreaching 90-100%ofconfluence,myoblastswerediferentiatedintomyotubesfor4-6daysinDMEM1g/mlglucose,pyruvate(Gibco, Paisley,Scotland,UK)supplementedin2%fetalbovineserum(Gibco,Paisley,Scotland,UK),1%penicillin/streptomycin(Gibco, Paisley,Scotland,UK),0.5%gentamycin(SigmaAldrich,SaintLouis,USA)and1:500amphotericinB(Gibco,Paisley,Scotland,UK).

Real-timebioluminescencerecordingandanalysis

Differentiatedmyotubesweresynchronizedwith10μMforskolin(SigmaAldrich,SaintLouis,USA)andincubatedfor1hat37◦C. Next,theculturemediumwasreplacedwithrecordingmediumcontaining100μMluciferin(NanolightTechnology)andcellswere placedin37◦CActimetricsLumiCycle,aspreviouslydescribed,[37,38,87]forreal-timerecordingofBMAL1-lucbioluminescencefor thedurationof96h.Generatedbioluminescenceprofileswerenormalizedtothemovingaverageof24h(detrended).[93]Circadian parameterswerecalculatedbasedondetrendeddatausingtheChronostar3.0software[94]withthesinusoidfunctionfittothe 12–100hdatarange.WeusedPartialLeastSquaresRegression(PLSR)topredictthebioluminescencephaseusingclockgene expression.Weusedthefunction'PLSRegression'withinscikit-learn(v1.0.2)andwithn_components=1.Weusedalldatafrom bothconditions,andweusedaleave-participant-outcross-validationpredictionapproachtoavoidthesameparticipantbeingin thetrainingandtestset.Themodelwastrainedonalldataexcludingoneparticipant,andthenthemodelwasusedtopredictthe testparticipantvalues.Thisprocedurewasperformedforallparticipants.Themodelwasthentrainedonallparticipantstoretrieve themodelparameters.

QUANTIFICATIONANDSTATISTICALANALYSIS

A2-wayrepeatedmeasuresANOVAwithtimeandcondition(condition:naturalvs.artificialofficelight)andtheirinteractionasfixed effectswasappliedtotestfordifferencesovertimebetweennaturalandartificialofficelightin24-hourplasmametabolites,substrate metabolismandenergyexpenditure,coreandskintemperature,bloodpressureandheartrate.Incaseofindividualmissingtime points,ageneralizedlinearmixedmodelwasruninsteadofthe2-wayrepeatedmeasuresANOVA.Intheabsenceofinteraction, weinterpretedthemaineffectforcondition,andinthecaseofstatisticalsignificance,wefollowedupontheconditioneffectwith pairwisecomparisonspertimepointusingBonferronipost-hoctests.Pairedt-testswereconductedtodetectdifferencesbetween naturalvs.artificialofficelightinallotheroutcomeswithoutrepeatedmeasures,andWilcoxontestswereperformedfornon-normally distributeddata.Ifcompletedatawasnotavailableforcertainoutcomesforallparticipants(n=13),mostlyduetotechnicalissues duringindividualmeasurements,therespectivelyapplicablesamplesizeisstatedintherespectivefigurelegends.

Differencesinserummetabolites,lipidfamilies,lipidspecies,andbloodmonocytetranscriptsbetweennaturalandartificiallight conditionsweremodelledusingOrthogonalPartialLeastSquaresDiscriminantAnalysis(OPLS-DA).[95]SeparateOPLS-DAmodels werebuiltforeachdatasetatthe08:00hand16:00htimepointsusingropls.Fromthepredictivecomponentofeach OPLS-DA model,correlation-scaledloadings(p(corr))wereobtainedforeachfeature.Statistically,thecorrelation-scaledloading(p(corr))for agivenmoleculequantifiesthePearsoncorrelationbetweenthatmolecule'sabundanceprofileacrossthesamplesandthemodel's predictivescore(component)thatmaximallyseparatesthegroups.Tocomparetargetsacrosstimepoints,SharedandUnique Structures(SUS)plotswereconstructed.[96]Foreachtarget,thep(corr)valuefromthe16:00hmodel(X-axis)wasplottedagainst thep(corr)valuefromthe08:00hmodel(Y-axis).

Formachinelearninganalysesoftheomicsdatasets,thecaretpackage(version6.0.94)inRwasutilized,incorporatingaleave-one-participant-outcross-validationapproach.Thisensuredthatalldatapointsfromanindividualparticipantwereexcludedfromthe trainingsetwhenevaluatingthesameparticipantinthetestset.Toenhanceclassificationperformanceoflightandtimeconditions, recursivefeatureeliminationwasappliedtoidentifythemostpredictivevariables.Toensurethereproducibilityoftheclassification accuracyofthemachinelearningmodels,each-omicsdataset(metabolomic,lipidomicandtranscriptomic)waspartitionedinto trainingandtestsetsandrunusing200uniqueseeds.Dataarepresentedasmean±SEM(standarderrorofthemean)unless indicatedotherwise.Thelevelofsignificancewassetto<0.05forallanalyses.