

1 **Chronotype and environmental light exposure in a student**

2 **population**

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1 **Abstract**

2 In humans, and most other species, changes in the intensity and duration of light provide a
3 critical set of signals for the synchronisation of the circadian system to the astronomical day.
4 The timing of activity within the 24 h day defines an individual's chronotype i.e. morning,
5 intermediate or evening type. The aims of this study were to investigate the associations
6 between environmental light exposure, due to geographical location, on the chronotype of
7 university students. Over 6,000 university students from cities in the northern hemisphere
8 (Oxford, Munich and Groningen) and southern (Perth, Melbourne and Auckland) completed
9 the Munich ChronoType Questionnaire (MCTQ). In parallel, light measures (daily irradiance,
10 timing of sunrise and sunset) were compiled from satellite or ground stations at each of
11 these locations. Our data shows that later mid-sleep point on free days (corrected for
12 oversleep on weekends MFS_{sc}) is associated with: (i) residing further from the equator; (ii) a
13 later sunset; (iii) spending more time outside and (iv) waking from sleep significantly after
14 sunrise. However, and surprisingly, MSF_{sc} did not correlate with daily light intensity at the
15 different geographical locations. Although these findings appear to contradict earlier studies
16 suggesting that in the wider population increased light exposure is associated with an earlier
17 chronotype, our findings are derived exclusively from a student population aged between 17
18 and 26 years. We therefore suggest that the age and occupation of our population increase
19 the likelihood that these individuals will experience relatively little light exposure in the
20 morning whilst encountering more light exposure later in the day, when light has a delaying
21 effect upon the circadian system.

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2 **Introduction**

3 The circadian system adjusts physiology and behaviour to the varied demands of the day-
4 night cycle (Czeisler et al., 1999; Wright et al., 2001; Roenneberg et al., 2003). To ensure
5 synchrony with the astronomical day the circadian system entrains to daily environmental
6 signals (zeitgebers = time givers). The light-dark cycle is the most significant zeitgeber for
7 most organisms, including humans (Honma et al., 1987; Roenneberg et al., 2007).
8 Differences in the relationships between an individual's circadian phase and external local
9 time gives rise to a distribution of chronotypes across the population, ranging from *early*
10 chronotypes, the proverbial "larks", to *late* chronotypes termed "owls" (Roenneberg et al.,
11 2003).

12 The timing of light exposure has a differential effect upon circadian phase: early light
13 exposure advances the cycle whilst light late in the internal day delays circadian phase
14 (Czeisler et al., 1989; Khalsa et al., 2003). Thus exposure to bright artificial light in the
15 evening before bedtime has been associated with a delay in circadian phase as assessed by
16 measures of subjective chronotype (Martin et al., 2012; Vollmer et al., 2012); subjective
17 sleep timing (Koo et al., 2016); salivary melatonin levels (Gordijn et al., 1999; Benloucif et
18 al., 2008; Cajochen et al., 2011); and core body temperature (Krauchi et al., 1997).
19 Furthermore, adolescents living in urban areas and exposed to bright artificial light at night,
20 have a later chronotype as assessed by the Munich Chronotype Questionnaire (MCTQ) and
21 Morningness-Eveningness Questionnaire (MEQ), compared to those living in more rural
22 settings (Vollmer et al., 2012). By contrast exposure to bright light in the morning results in
23 an advance of the circadian phase of melatonin synthesis and release (Dijk et al., 1989;
24 Gordijn et al., 1999; Revell et al., 2005). In addition, bright morning light has been used as a
25 therapy for advancing sleep timings in patients with delayed sleep-wake phase disorders

1 (Rosenthal et al., 1990; Saxvig et al., 2014) and, more recently, with social jet lag (Geerdink
2 et al., 2016).

3 Despite society's increasing detachment from the natural light-dark cycle, sunlight can still
4 be seen to impact chronotype. Living further east within the same time zone in the Northern
5 hemisphere is associated with an earlier subjective chronotype in adults assessed using the
6 MCTQ (Roenneberg et al., 2007) and in adolescents assessed with the MEQ (Randler,
7 2008), most likely as a result of an earlier sunrise time. Seasonal changes are also apparent,
8 such that during the months of increasing day length, subjective chronotype advances with
9 individuals rising earlier (Kantermann et al., 2007; Allebrandt et al., 2014). There is also
10 some evidence that geographical location has an impact upon chronotype. For example, in a
11 study conducted in Brazil, subjective chronotype was assessed using the MCTQ and MEQ in
12 two cities: São Paulo at latitude 23° 32' S and longitude 46° 38' W and Natal at 05° 47' S
13 and 35° 12' W. Chronotype was found to be earlier in individuals living in Natal, the city
14 closest to the equator (Miguel et al., 2014).

15 Clearly the pattern of natural light within a particular environment will be critical in defining an
16 individual's phase of entrainment. However, an individual's behaviour within that
17 environment will also play an important role. A recent study compared the same individuals
18 living under their normal urban routines (including artificial light at night) with a period under
19 natural light exposure (camping without artificial light). The findings demonstrated that
20 increased exposure to natural light, advanced the circadian phase of all individuals (Wright
21 et al., 2013; Stothard et al., 2017). Increasing photic zeitgeber strength by spending more
22 time outside has also been correlated with self-reported chronotype: the more time spent
23 outside, the earlier the chronotype (Roenneberg&Morrow, 2007; Roenneberg et al., 2015).

24 By studying populations across the Northern and Southern hemisphere, specifically Oxford,
25 Groningen, Munich, Perth, Melbourne and Auckland, we aimed to investigate the association
26 between geographical location and chronotype and how different aspect(s) of environmental

1 light (timing; length of time spent outside; intensity of light, sleep timings relative to sunrise
2 and sunset) might influence chronotype.

3 **Materials and methods**

4 Students were recruited from six universities: University of Oxford, UK (51° 45' N,
5 1° 15' W); University of Groningen, The Netherlands (53° 13' N, 6° 33' E); LMU, Munich,
6 Germany (48° 8' N, 11° 34' E); University of Western Australia, Perth, Australia (31° 57'
7 ' S, 115° 51' E); Monash University, Melbourne, Australia (37° 48' S, 144° 57' E);
8 University of Auckland, New Zealand (36° 50' S, 174° 44' E). Students were asked to
9 complete the online version of the MCTQ twice, in May and October of 2010, to control for
10 seasonal influences. Overall 13,299 individuals completed the MCTQ online. Over half of the
11 participants were excluded from the analysis (see data processing). 6441 students (mean
12 age 21.5 ± 2.2 years, 67.5% female, see table 1 for group demographics) were included in
13 the analysis. Daily irradiance, sunrise and sunset times were obtained for May and October
14 2010. Ethical approval for this study was obtained from the local ethics committee for each
15 university involved in the study.

16 **Materials**

17 *The Munich Chronotype Questionnaire (MCTQ)*. The online version of the MCTQ
18 (Roenneberg et al., 2003) was used in the native language of the country of each university.
19 The MCTQ consists of questions concerning sleep timings for both workdays and free days
20 separately, work time and time spent outside. The MCTQ has been validated against
21 actigraphic recordings (Vetter et al., 2015), and melatonin rhythms (Kitamura et al., 2014).
22 The MCTQ is used to calculate the MSF as the mid-point between sleep onset and sleep
23 end. MSF was corrected for oversleep on free days (MSF_{sc}: Mid Sleep point on Free days,
24 Sleep Corrected), that occurs as a result of sleep debt [MSF_{sc} = MSF - (SDf -
25 (((nWD*SDw)+(7-nWD))*SDf)/7), where SDf is the sleep duration of free days, SDw is the

1 sleep duration of workdays and nWD is the number of workdays] (Roenneberg et al., 2004).
2 In cases where the numbers of workdays were missing, five workdays were assigned. Social
3 jet lag (SJL) was also calculated from the MCTQ [SJL=MSF-MSD] where MSF is the mid
4 sleep point of free days and MSD the mid sleep point of work days (Wittmann et al., 2006).

5 *Light data.* 'Time spent outside' was self-reported on the MCTQ. The weighted average of
6 the number of hours given for free days and workdays was calculated using the number of
7 workdays also reported on the MCTQ. If no workdays were given, 5 workdays were
8 assigned. [Time spent outside = ((time spent outside on workdays * number of workdays) +
9 (time spent outside on free days * (7-number of workdays)))/7].

10 'Light dose' is a measure of how much light individuals are exposed to over a given period of
11 time. Here, we calculated the average hourly irradiance for the day for participants that
12 completed the MCTQ and normalised this to the "time spent outdoors", averaged for work
13 and free days (Light dose = *daily irradiance* / *day length* * time spent outside].

14 'Day length' for each day of the collection periods, for each city, was calculated using the
15 world clock (<http://www.timeanddate.com/worldclock>).

16 'Daily irradiances' for the collection periods, for both May and October 2010, were obtained
17 from three sources on an hourly basis. The data for Oxford, Groningen and Munich were
18 provided by Dr. Lucien Wald (MINES, ParisTech), obtained from Meteosat satellite images
19 and converted to data maps of solar radiation using the Heliosat-2 method (Rigollier et al.,
20 2004). The data for Perth and Melbourne were obtained from the Australian Bureau of
21 Meteorology, again derived from satellite images processed by the Australian Bureau of
22 Meteorology. Finally, the data for Auckland were obtained from the New Zealand
23 Meteorological office based on readings from its ground station in Auckland. For all daily
24 irradiance, the data represent light intensity experienced at ground level, taking into account
25 weather conditions, either via processing of satellite data or as data taken at ground level.

26 **Data processing**

1 As only 440 participants completed the questionnaire in both May and October (420 from
2 northern hemisphere cities and 20 from southern hemisphere cities), longitudinal analysis
3 was not performed and data for these participants were only included in the data analysis
4 from the May collection period. Individuals were excluded if they were outside the age range
5 (17-26 years); did not indicate they were currently living in any of the cities of interest;
6 completed the questionnaire outside May or October 2010; or had reported working shifts
7 during the past three months. Individuals were also excluded if they indicated using an alarm
8 clock on free days (an exclusion criterion for chronotyping). For inter-hemispheric
9 comparisons, months were assigned to season (northern hemisphere, May and southern
10 hemisphere, October as spring and *vice versa* as autumn).

11 **Data analysis**

12 Statistical analysis was performed using R version 3.0.1 (2013-05-16, Copyright 2013 The R
13 Foundation for Statistical Computing). Linear mixed-effects models were fitted using the lme
14 package for group and seasonal assessments of MSF_{sc} and light data. For categorical
15 comparisons, linear models were referenced to Oxford for group comparisons, females for
16 sex comparisons and spring for season comparisons. Since age and sex are known to
17 influence chronotype, both of these were included as covariants when modelling MSF_{sc} .
18 Spearman's rank correlation analysis was performed to assess associations between MSF_{sc}
19 and light data.

20 **Results**

21 On average students in this sample reported the following habitual sleep-related times: bed
22 time on workdays: 00:11 \pm 01:10 (mean, SD) and nearly an hour later on free days: 01:09 \pm
23 01:24; wake-up time was two hours later on free days (09:45 \pm 01:23) compared to
24 workdays (07:43 \pm 01:06). A mean of 1.51 \pm 0.93 hr of social jet lag was reported. Table 1
25 details wake-up and bed times for work and free days at each city along with social jet lag.
26 The sleep midpoint on free days (corrected for over sleep; MSF_{sc}) was different between

1 cities but not between seasons, and no city-season interactions were found (see suppl. data
2 model 1). Hence, MSF_{sc} was collapsed across seasons.

3 When plotting chronotype and time spent outside against the absolute distance of each city
4 from the equator, MSF_{sc} showed a positive association: with chronotype becoming later with
5 increasing distance (Fig. 1A). Controlling for age and sex, the cities formed three groups for
6 MSF_{sc} : Oxford, Groningen and Munich were not statistically different from each other, and
7 had the latest MSF_{sc} ; Melbourne showed an intermediate MSF_{sc} and was statistically
8 significant from all the other cities; and Perth and Auckland showed the earliest MSF_{sc} and
9 were also not statistically significant from each other (see suppl. data model 2).

10 Overall, the students (regardless of city) reported they spend on average $2.20 (\pm 1.33)$ h
11 outside a day resulting in exposure to on average $24.74 (\pm 20.09)$ W/m^2 of light on the day
12 they completed the survey (light dose). Students in Perth reported spending the most
13 amount of time outside (2.76 ± 1.67 h) and experienced the highest intensity of light whilst
14 outside, light dose (62.68 ± 39.11 W/m^2 , see suppl. data model 3 and 4). Whereas students
15 in Melbourne reported spending the least amount of time outside (1.89 ± 1.49 h), students in
16 Oxford received the lowest light dose (19.43 ± 14.31 W/m^2). In relation to geographical
17 location, the amount of time spent outside was not associated with the distance of each city
18 from the equator (Fig. 1B), but light dose did show an association, with the cities nearest to
19 the equator experiencing a higher light dose, except for Auckland (Fig. 1C).

20 MSF_{sc} was positively, although weakly, correlated with time spent outside ($\rho = 0.036$, $p =$
21 0.005) indicating that the longer students spent outside the later their sleep midpoint on free
22 days. Time spent outside binned for MSF_{sc} in 30 min intervals, showed a stronger positive
23 correlation ($\rho = 0.86$, $p = 0.011$, Fig. 2A). However, this was only statistically significant
24 with the removal of the outlier of MSF_{sc} binned from 06:30 to 07:00. Light dose was not
25 correlated with MSF_{sc} for raw ($\rho = -0.0005$, $p = 0.97$) or binned data ($\rho = -0.15$, $p = 0.71$,
26 Fig. 2B). Using a linear mixed effect model, taking age and sex into account, time spent

1 outside but not light dose was found to have a significant effect on MSF_{sc} (see suppl. data
2 model 5). However, the addition of time spent outside into the model did not remove the
3 effect of city, moreover no city-time spent outside interaction was found (see suppl. data
4 model 6). This suggests that although the amount of time spent outside does have an
5 influence on sleep midpoint on free days, other differences between the cities are also
6 important.

7 To define the time of day the students in this population were most likely to receive natural
8 light, the timing of sunrise and sunset the day each student completed the survey was
9 compared to their self-reported sleep timings for work and free days. The proportion of
10 daylight (i.e. between sunrise and sunset) during which time students were awake was
11 negatively correlated to MSF_{sc} for both work ($\rho = -0.4$, $p < 0.001$) and free days ($\rho = -$
12 0.71 , $p < 0.001$, figure 2C), indicating that students with the latest MSF_{sc} were only likely to
13 be awake for around 40% of the daylight period. This is because students wake up after
14 sunrise rather than going to bed before sunset (Fig. 2D and 2E respectively). 75.5% of
15 students woke up after sunrise on workdays and 98.1% on free days, with 15.7% waking up
16 5 hours after sunrise on free days (1.2% on workdays).

17 The impact of geographical location on MSF_{sc} was found to be most influenced by the timing
18 of sunset. The average MSF_{sc} per city was plotted against the timing of sunrise and sunset
19 for the day the survey was completed, as well as time spent outside and light dose (Fig. 3).
20 The timing of sunset showed a positive association with MSF_{sc} : the later sunset the later
21 MSF_{sc} . Interestingly no association was seen for sunrise. Collectively these data suggest
22 that the timing of sunset and therefore the amount of light in the evenings may be more
23 influential on sleep midpoint on free days than the amount of light in the morning in this
24 population.

25 **Discussion**

1 The findings from this study suggest that in a university student population a later
2 chronotype is associated with: (i) living further from the equator; (ii) a later sunset; (iii)
3 spending more time outside; and (iv) waking up after sunrise. Significantly, we did not find
4 that light intensity was associated with chronotype. Initially, these findings appear to
5 contradict earlier studies where increased light exposure is associated with an earlier
6 chronotype in the general population (Roenneberg&Morrow, 2007; Wright et al., 2013;
7 Roenneberg et al., 2015; Stothard et al., 2017). However, our findings are derived
8 exclusively from a university student population aged between 17 and 26 years. Thus we
9 suggest that the age and occupation of our population increase the likelihood that these
10 individuals will experience relatively little light exposure in the morning whilst encountering
11 more light exposure later in the day, when light has a delaying effect upon the circadian
12 system.

13 In a sample of approximately 200,000 individuals from primarily Central Europe and North
14 America, spending more time outside was associated with an earlier chronotype
15 (Roenneberg&Morrow, 2007; Roenneberg et al., 2015). However, when age is taken into
16 consideration, 15-20 year olds did not show a significant correlation between time spent
17 outside and chronotype, and 20-25 year olds had only a weak correlation (Roenneberg et
18 al., 2015). Our sample of over 6,000 students (17-26 years) falls across these age ranges,
19 and also differs from the Roenneberg sample (MCTQ database) in several important
20 aspects. A key difference is the work status of the populations studied: all individuals within
21 our sample are university students during term time, the general population in the sample
22 from the MCTQ database, would have included school students, university students and
23 working individuals. It is possible, therefore, that imposed work schedules could result in
24 more morning vs evening light exposure in the general MCTQ population. In addition, the
25 data for the current study was collected exclusively in May and October, whilst the general
26 population sample was collected all year round, which might also have an impact upon the
27 timing of light exposure.

1 The timing of light exposure has a differential effect upon circadian phase: early light
2 exposure advances the cycle whilst light late in the internal day delays circadian phase
3 (Czeisler et al., 1989; Khalsa et al., 2003). In our student population we found that longer
4 time spent outside the later chronotype, which would suggest that our population was
5 exposed to more phase delaying evening light than phase advancing morning light. Although
6 it was not possible to determine the timing of light exposure definitively from our study, we
7 provide several lines of evidence that support the importance of evening light in this
8 population. In the present study we demonstrated that the later students wake up after
9 sunrise the later MSF_{sc} . As a result, individuals are likely to be exposed to a photoperiod with
10 a greater proportion of evening phase delaying versus morning phase advancing light.
11 Clearly, future studies will need to define the phase relationship between the internal
12 circadian and external environmental light cycle. Moreover the timing of sunset, rather than
13 sunrise was found to be most associated with MSF_{sc} in our population. Previously, a
14 longitudinal study of around 55,000 individuals has reported that MSF_{sc} tracks sunrise and
15 not sunset (Kantermann et al., 2007). However, again the broad demographics of this
16 population make direct comparisons to our population difficult, but it is possible that the
17 association of young adult university students (comparable to our population) is masked by
18 other individuals in the sample. Finally the sensitivity of young adults to evening light has
19 recently been demonstrated in two studies. In twenty healthy young adults (mean age 23)
20 later light onset and offset has been associated with later melatonin onset as assessed using
21 dim light melatonin onset (DLMO) (Wams et al., 2017). A mathematical model of sleep
22 timings based on the experimentally derived effects of light on the human circadian clock
23 and interaction of the circadian clock and sleep homeostat predicts a similar finding.
24 Individuals with a longer intrinsic clock and hence later chronotype are predicted to be more
25 susceptible to evening light, causing even more of a delay in the circadian cycle (Skeldon et
26 al., 2017).

1 Geographical location was found to be associated with chronotype: the closer to the equator
2 the earlier chronotype, and in this regard our findings are consistent with previous findings
3 (Miguel et al., 2014). However, this association was assumed to be driven by higher
4 environmental light intensities closer to the equator. Interestingly, it was only the duration of
5 time spent outside - not the intensity of light - that was found to influence MSF_{sc} in our study
6 except for subjects in Auckland. Although the intensity of light has been shown to impact on
7 the entraining properties of light pulses under experimental conditions (Boivin et al., 1996;
8 Zeitzer et al., 2005; Duffy&Czeisler, 2009), a saturation effect on shifting the phase of the
9 melatonin rhythm has been reported above approximately 1000 lux (Zeitzer et al., 2000),
10 equivalent to approximately 7.9 W/m^2 (based on the approximation that $1 \text{ lux} = 0.0079 \text{ W/m}^2$
11 for solar irradiance). Considering the lowest average light intensity reported in this study was
12 19.43 W/m^2 , and therefore well above saturation intensities, it is perhaps unsurprising that
13 no effect of light intensity emerged. Instead in our population it appears that the association
14 between chronotype and geographical location is due to the timing of sunset.

15 This study reported on a large sample of over 6,000 university students collected in term
16 time during spring and autumn. Such numbers help mitigate the limitation of a cross-
17 sectional assessment of chronotype. However, longitudinal studies are needed to determine
18 precisely how an individual's chronotype changes with environmental light levels and age.
19 Although based on self-reported sleep timings, the MCTQ is a validated measure of
20 chronotype (Kantermann et al., 2015). The reliability of self-reported time spent outside as a
21 proxy for light dose is less certain. The amount and type of environmental light exposure will
22 be influenced by various factors including: photoperiod, weather conditions and the level of
23 urbanisation. Although these have been taken into consideration in this study as much as
24 possible (weather conditions accounted for in measures of daily irradiance and photoperiod
25 in proportional assessment of daylight students awake for), objective assessment of time
26 spent outside and light monitoring need to be undertaken to define when individuals go
27 outside and the nature of their light exposure (inside vs outside). Furthermore, exposure to

1 artificial light was not addressed in this study, which of course will have an added impact on
2 circadian physiology. Of particular interest in this young student population, is the impact of
3 light-emitting devices on sleep and the circadian clock. Although, such devices have been
4 found to impact sleep and circadian timing (Cajochen et al., 2011; Chang et al., 2015), the
5 findings are mixed (Heath et al., 2014; Rangtall et al., 2016) and these changes are often
6 small thus, the real world significance of these findings remains unclear (Zeitler, 2015). The
7 findings from this study however emphasise that environmental evening light exposure may
8 need to be tailored for different populations. With the rapid growth in diversity of energy
9 efficient light-emitting devices, robust, evidence-based advice is needed to ensure that
10 individuals get the right kind of light at the right time of day to reinforce robust entrainment of
11 the sleep-wake cycle.

12 In conclusion, we report that in this young adult university student population, time spent
13 outside is associated with a later chronotype. This seems to be linked to the fact that this
14 population spends more time outside in the evening, and that dusk light exposure will have a
15 phase delaying effect upon their circadian biology. Moreover, we found that the closer
16 students lived to the equator the earlier their chronotype. Significantly, this also appears to
17 be associated with the timing of sunset rather than sunrise. Collectively our results
18 emphasise the fact that the age and occupation of individuals will likely impact profoundly
19 upon the timing of their light exposure and hence their phase of entrainment. Moreover, this
20 work highlights the need for future longitudinal studies that will define these relationships
21 with greater precision.

22

23 Declaration of interest

24 KP, TR, LW, MM, LF, MG, DR, GW and KW declare no conflicts of interest. RGF is in receipt
25 of funding from Circadian Therapeutics. MG is working as a consultant for Philips Sleep &
26 Respiratory care. SMWR reports that he has served as a consultant through his institution to

1 Vanda Pharmaceuticals, Philips Respironics, EdanSafe, The Australian Workers' Union,
2 National Transport Commission, and Transport Accident Commission, and has through his
3 institution received research grants and/or unrestricted educational grants from Vanda
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5 Respironics, Cephalon, and ResMed Foundation, and reimbursements for conference travel
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7 other support from Optalert™, Compumedics, and Tyco Healthcare. He has also served as
8 an expert witness and/or consultant to shift work organizations. SMWR also serves as a
9 Program Leader in the Cooperative Research Centre for Alertness, Safety and Productivity.
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