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Is latitude associated with chronotype?

São Paulo

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Original version

Thesis presented to the School of Arts, Sciences and Humanities at the University of São Paulo, as a requirement for the degree of Master of Science by the Graduate Program in Complex Systems Modeling.

Area of concentration: Complex Systems.

Supervisor: Prof. Dr. Camilo Rodrigues Neto

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*Nullius in verba*¹

¹ The Royal Society. (n.d.). *History of the Royal Society*. <https://royalsociety.org/about-us/history>

ABSTRACT

Vartanian, D. (2024). *Is latitude associated with chronotype?* [Master's Thesis, University of São Paulo].

Theories on circadian rhythms are well-established in science, but there is still a need to test them in larger samples to gain a better understanding of the expression of temporal phenotypes. This thesis investigates the hypothesis that latitude influences chronotype expression, based on the idea that regions closer to the poles receive less sunlight over the year than equatorial regions. This difference suggests that equatorial areas have a stronger solar zeitgeber, which could lead to greater synchronization of circadian rhythms with the light-dark cycle, reducing the amplitude and diversity of circadian phenotypes, resulting in a higher propensity for morningness in those populations. To test this hypothesis, data from 65,824 individuals from all regions of Brazil were analyzed, collected in 2017 based on the Munich ChronoType Questionnaire (MCTQ). The analysis, using nested linear regression models, revealed a negligible effect of latitude on the variation in chronotype expression (Cohen's $f^2 = 0.012137120$), contrasting with recent studies. Although the hypothesis is reasonable and aligns with evolutionary theories of temporal biological systems, the results suggest that the phenomenon of entrainment is more complex than previously thought.

Keywords: Complexity science. Complex systems. Chronobiology. Biological rhythms. Chronotypes. Circadian phenotypes. Sleep. Entrainment. Latitude. MCTQ.

RESUMO

Vartanian, D. (2024). *A latitude está associada ao cronotipo?* [Dissertação de Mestrado, Universidade de São Paulo].

As teorias sobre ritmos circadianos estão bem estabelecidas na ciência, mas ainda há a necessidade de testá-las em amostras mais amplas para compreender melhor a expressão dos fenótipos temporais. Esta dissertação investiga a hipótese de que a latitude influencia a expressão dos cronotipos, baseada na ideia de que regiões próximas aos polos recebem menos luz solar ao longo do ano do que as regiões equatoriais. Esse diferencial sugere que áreas equatoriais possuem um *zeitgeber* solar mais forte, o que poderia levar a uma maior sincronização dos ritmos circadianos com o ciclo claro-escuro, reduzindo a amplitude e a diversidade de fenótipos circadianos, resultando em uma propensão maior ao cronotipo matutino. Para testar essa hipótese, foram analisados dados de 65.824 indivíduos de todas as regiões do Brasil, coletados em 2017 com base no Munich ChronoType Questionnaire (MCTQ). A análise, utilizando modelos de regressão linear aninhados, revelou um efeito negligenciável da latitude na variação da expressão dos cronotipos (f^2 de Cohen = 0.012137120), em contraste com estudos recentes. Embora a hipótese faça sentido e esteja alinhada com teorias evolutivas dos sistemas biológicos temporais, os resultados sugerem que o fenômeno de *entrainment* é mais complexo do que se imagina.

Palavras-chaves: Ciência da complexidade. Sistemas complexos. Cronobiologia. Ritmos biológicos. Cronotipos. Fenótipos circadianos. Sono. Entrainment. Latitude. MCTQ.

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1 INTRODUCTION

There has been a long-standing debate in the chronobiology community regarding the relationship between latitude and circadian phenotypes (chronotypes) (Bohlen & Simpson, 1973; Leocadio-Miguel et al., 2017; Pittendrigh et al., 1991; Skeldon & Dijk, 2021; Zerbini et al., 2021), with many assuming that this association is well-established. The hypothesis is based on the varying amounts of solar radiation experienced by populations across different latitudes. Since light exposure serves as a primary zeitgeber — a periodic environmental cue that influences or regulates biological rhythms (Pittendrigh, 1960) — such variations, along with temperature differences, are thought to result in observable differences in chronotype distributions globally. This thesis investigates the so-called latitude or environmental hypothesis in human circadian phenotypes, addressing the question: *Is latitude associated with chronotype?*

The central hypothesis is that *latitude is associated with human chronotype distributions*, with populations closer to the equator exhibiting, on average, a shorter or more morning-oriented circadian phenotype compared to those living near the poles (Bohlen & Simpson, 1973; Hut et al., 2013; Leocadio-Miguel et al., 2017; Pittendrigh et al., 1991; Randler, 2008; Randler & Rahafar, 2017; Roenneberg, Wirz-Justice, & Mellow, 2003). The primary objective of this study is to model and test this hypothesis by critically examining whether a significant association exists between latitude and circadian phenotypes in the Brazilian population.

This study emerged from an insightful debate with my former supervisor, sparked by results published in 2017 in *Nature Scientific Reports* (Leocadio-Miguel et al., 2017). In this paper, the authors conclude, as the theory suggests, that there is a significant association between latitude and chronotype in the Brazilian population. However, the results were not as clear-cut as suggested, and the methodology used to test the hypothesis was not optimal. This thesis revisits the hypothesis using an improved statistical approach, aiming to provide a more accurate and reliable answer to the research question.

In the following chapters, the latitude hypothesis is explored using Popper's hypothetical-deductive method (Popper, 1979) and an enhanced approach to Null Hypothesis Significance Testing (NHST), rooted in the original Neyman-Pearson framework for data testing (Neyman & Pearson, 1928a, 1928b; Perezgonzalez, 2015).

This exploration involves a series of analyses conducted on a large dataset comprising 65 824 individuals, collected from the Brazilian population in 2017. The dataset is based on the Munich Chronotype Questionnaire (MCTQ) (Roenneberg, Wirz-Justice, & Merrow, 2003; Roenneberg et al., 2012), and includes data on sleep habits and demographic characteristics from all of Brazil's states.

It is important to emphasize that this thesis does not aim to propose or discuss the mechanisms underlying the latitude-chronotype relationship. Instead, it focuses solely on the statistical association between them. If a cause-effect relationship exists, it must be preceded by, at the very least, an association — something this thesis aims to uncover.

The analyses utilize nested multiple regression models to evaluate the additional variance explained and the effect size when latitude is included as a predictor of chronotype. The results are then compared with those obtained from a restricted model that does not have latitude as a predictor. This method of procedure builds on the method used in Leocadio-Miguel et al. (2017). The results will contribute to the ongoing debate on the latitude-chronotype relationship, offering new evidences into how environmental factors influence human circadian rhythms.

In accordance with the [graduate program regulation](#), this thesis follows an [article-based format](#), inspired by the structure of Reis (2020)'s PhD thesis. Chapters 2 through 3 consist of a series of essays and literature reviews related to the thesis topic, while Chapter 4 presents the core investigation, including an article detailing the hypothesis test and addressing the central research question. Finally, Chapter 5 offers conclusions, discusses limitations, and proposes directions for future research. Additionally, supplementary materials are provided to offer a richer, more comprehensive understanding of the research, and the reader is encouraged to explore them in detail.

All analyses in this thesis are fully reproducible and were conducted using the [R programming language](#) (R Core Team, n.d.) alongside the [Quarto](#) publishing system (Allaire et al., n.d.). Given the thesis's data-centric nature, it is best experienced online. To view the digital version visit <https://danielvartan.github.io/mastersthesis>.

The thesis code repository is available on GitHub at <https://github.com/danielvartan/mastersthesis>, and the research compendium can be accessed via [The Open Science Framework](#) at the following link: <https://doi.org/10.17605/OSF.IO/YGKTS>.

2 ON CHRONOBIOLOGY

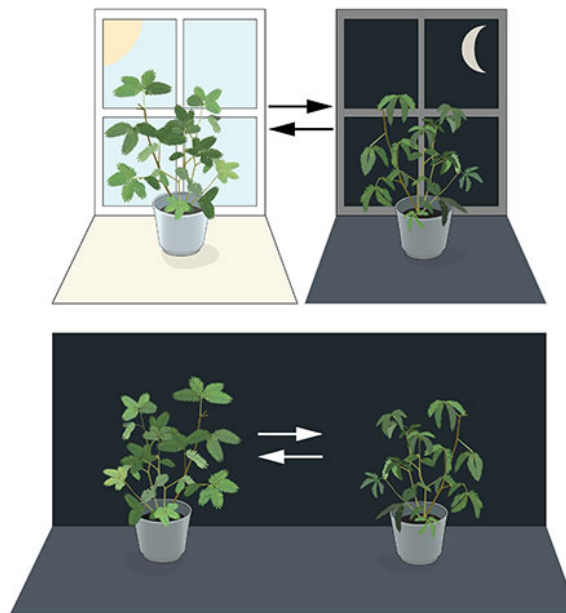
The dimension of time, manifest in the form of rhythms and cycles, like the alternating patterns of day and night as well as the annual transition of seasons, was consistently featured in the evolutionary journey of not only the human species but also all other life forms on our planet. These rhythms and cycles brought with them evolutionary pressures, resulting in the development of a temporal organization that allowed organisms to survive and reproduce in response to the conditions imposed by the environments they inhabited (Menna-Barreto, 2003; Pittendrigh, 1981). An example of this organization can be observed in the presence of different activity-rest patterns among living beings as they adapt to certain temporal niches, such as the diurnal behavior of humans and the nocturnal behavior of cats and some rodents (Foster & Kreitzman, 2005).

For years, scientists debated whether this organization was solely in response to environmental stimuli or if it was also present endogenously, internally, within organisms (Rotenberg et al., 2003). One of the early seminal studies describing a potential endogenous rhythmicity in living beings was conducted in 1729 by the French astronomer Jean Jacques d'Ortous de Mairan. De Mairan observed the movement of the sensitive plant (*mimosa pudica*) by isolating it from the light-dark cycle and found that the plant continued to move its leaves periodically (Figure 1) (Mairan, 1729; Rotenberg et al., 2003). Charles Darwin also wrote about the movement observed in plants and made thematic explorations of these intriguing “periodical phenomena” (Andrade & Beale, 2024). The search for this internal timekeeper in living beings only began to solidify in the 20th century through the efforts of scientists like Jürgen Aschoff, Colin Pittendrigh, Franz Halberg, and Erwin Bünning, culminating in the establishment of the science known as chronobiology, with a significant milestone being the Cold Spring Harbor Symposium on Quantitative Biology: Biological Clocks in 1960 (*chrónos*, from Greek, meaning time; and *biology*, pertaining to the study of life) (Cold Spring Harbor Laboratory, n.d.; Rotenberg et al., 2003)¹. However, the recognition of endogenous rhythmicity by the global scientific community truly came in 2017 when Jeffrey Hall, Michael Rosbash, and Michael Young were awarded the Nobel Prize in Physiology or Medicine for

¹ Some say the term *chronobiology* was coined by Franz Halberg during the Cold Spring Harbor Symposium (Menna-Barreto & Marques, 2023, p. 21).

their discoveries of molecular mechanisms that regulate the circadian rhythm in fruit flies (*circā*, from Latin, meaning around, and *dies*, meaning day (Latinitium, [n.d.](#)) – a rhythm that expresses itself in approximately one day) (Nobel Prize Outreach AB, [n.d.](#)).

Figure 1 – Illustration of a circadian rhythm in the movement of the leaves of the sensitive plant (*mimosa pudica*) observed by Jacques d’Ortous de Mairan in 1729.



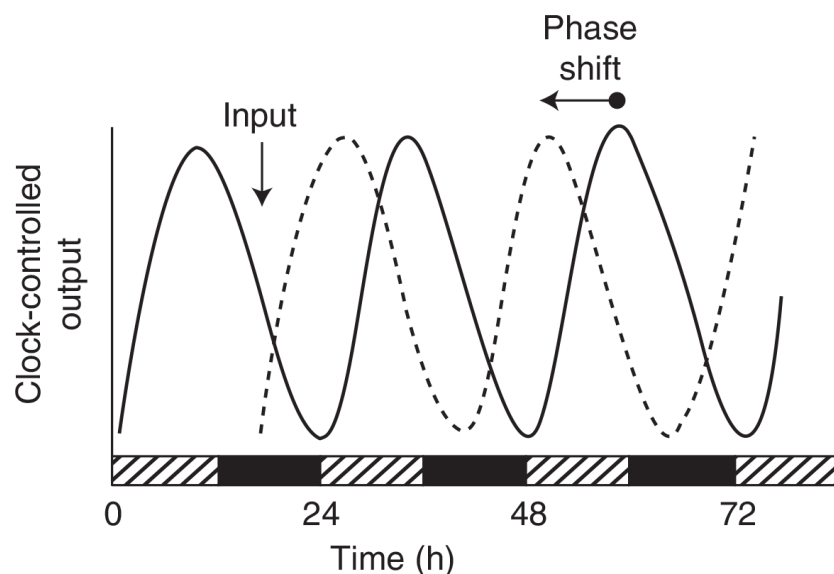
Source: Reproduction from Nobel Prize Outreach AB ([n.d.](#)).

Science has already showed and described various biological rhythms. These rhythms can occur at different levels, whether at a macro level, such as the menstrual cycle, or even at a micro level, such as rhythms expressed within cells (Roenneberg & Merrow, [2016](#)). Like many other biological phenomena, these are complex systems present in all living beings, i.e., systems with a large number of connected parts that presents stable macroscopic patterns (emergences, in this case, the rhythms) arising from local interactions or the collective behavior of its parts, giving the system properties not attained by the aggregate summation (Epstein, [1999](#); Holland, [2014](#)). It is understood today that the endogeneity of rhythms has provided organisms with an anticipatory capacity, allowing them to organize resources and activities before they are needed (Marques & Menna-Barreto, [2003](#)).

Despite the endogenous nature of these rhythms, they can still be regulated by the external environment. Signals (cues) from the environment that occur cyclically in na-

ture and have the ability to regulate biological rhythmic expression are called zeitgebers (from the German *zeit*, meaning time, and *geber*, meaning donor (Cambridge University Press, [n.d.](#))). These zeitgebers act as synchronizers by entraining the phases of the rhythms (Khalsa et al., [2003](#); Kuhlman et al., [2018](#)) (Figure 2). Among the known zeitgebers are, for example, meal timing and changes in environmental temperature (Aschoff, [1981](#); Roenneberg & Merrow, [2016](#)). However, the most influential of them is the light-dark cycle (or, simply, light exposure). It is understood that the day/night cycle, resulting from the rotation of the Earth, has provided the vast majority of organisms with an oscillatory system with a periodic duration of approximately 24 hours (Kuhlman et al., [2018](#); Roenneberg, Kuehnle, et al., [2007](#)).

Figure 2 – Illustration of a circadian rhythm (output) whose phase is entrained in the presence of a zeitgeber (input). The rectangles represent the light-dark cycle.



Source: Adapted from Kuhlman et al. ([2018](#)).

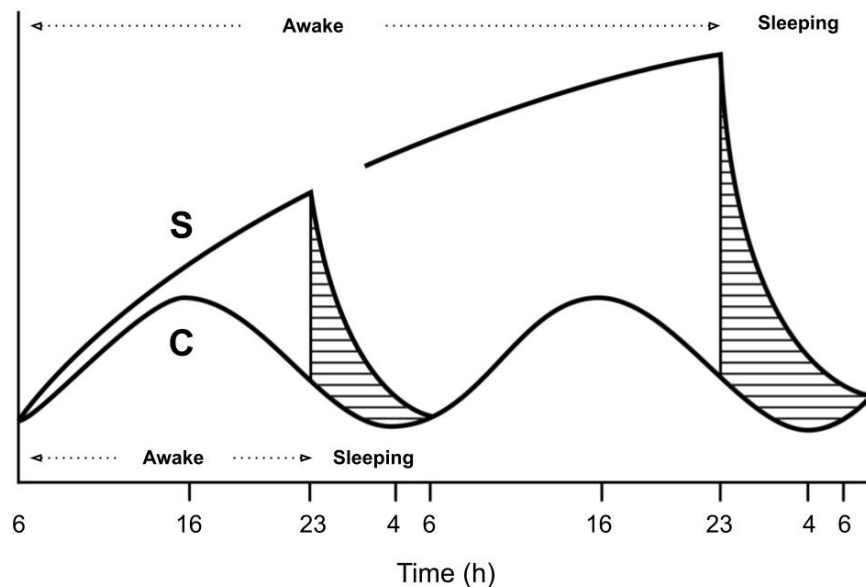
Naturally, the expression of this temporal organization varies from organism to organism, even among members of the same species, whether due to the different ways they are exposed to the environment or the differences in the expression of endogenous rhythmicity, which, in turn, results from gene expression (Roenneberg, Kumar, & Merrow, [2007](#)). The interaction between these two expressions, external and internal,

of the environment and genotype, generates a signature, an observable characteristic, which is called a phenotype (Frommlet et al., 2016).

The various temporal characteristics of an organism can be linked to different oscillatory periods. Among these are circadian phenotypes, which refer to characteristics observed in rhythms with periods lasting about a day (Foster & Kreitzman, 2005). Another term used for these temporal phenotypes, as the name suggest, is *chronotype* (Ehret, 1974; Pittendrigh, 1993). This term is also often used to differentiate phenotypes on a spectrum ranging from morningness to eveningness (Horne & Östberg, 1976; Roenneberg, Wirz-Justice, et al., 2019).

Sleep is a phenomenon that exhibits circadian expression. By observing the sleep characteristics of individuals, it is possible to assess the distribution of circadian phenotypes within a population, thereby investigating their covariates and other relevant associations (Roenneberg, Wirz-Justice, & Mellow, 2003). This is because sleep regulation is understood as the result of the interaction between two processes: a homeostatic process (referred to as the S process), which is sleep-dependent and accumulates with sleep deprivation; and a circadian process (referred to as the C process), whose expression can be influenced by zeitgebers, such as the light-dark cycle (Figure 3 illustrates these two process) (Borbély, 1982; Borbély et al., 2016). Considering that the circadian rhythm (the C process) is present in sleep, its characteristics can be estimated if the S process can be controlled.

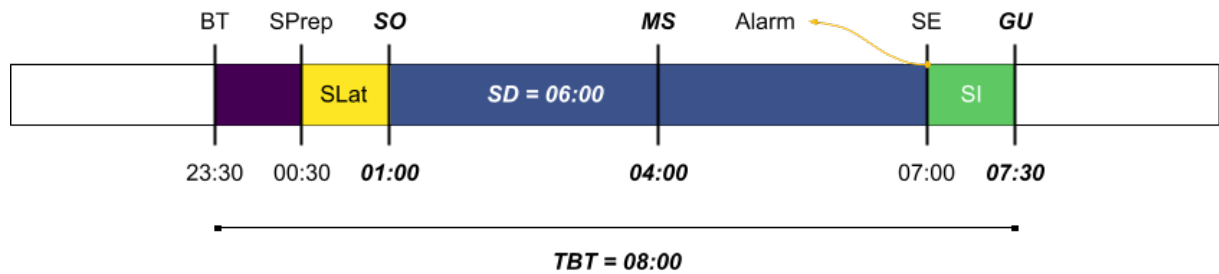
Figure 3 – Illustration of the interaction between Process S (sleep-dependent process) and Process C (circadian rhythm process) in sleep regulation. The figure depicts two scenarios: one with 17 hours of wakefulness followed by 7 hours of sleep; and another, with sleep deprivation, consisting of 41 hours of wakefulness followed by 7 hours of sleep. The y-axis represents the level of each process. The hatched areas indicate periods of sleep, along with the exponential decline of Process S.



Source: Adapted from Borbély (1982).

Building on this idea, Roenneberg, Wirz-Justice, and Mellow (2003) developed the Munich Chronotype Questionnaire (MCTQ) to measure the circadian phenotype through sleep patterns. The MCTQ asks individuals about their sleep habits, such as the times they go to bed and wake up on workdays and work-free days. Based on this information, the MCTQ calculates the local time of the midpoint of sleep on work-free days (Figure 4) and, if sleep deprivation is detected on workdays, adjusts the measurement accordingly. This midpoint, reflecting sleep without social obligations, is thought to represent the unabridged expression of the circadian rhythm. Given its basis in the two processes of sleep regulation, the MCTQ provides a good proxy for measuring the circadian phenotype (or C process) (Leocadio-Miguel et al., 2014).

Figure 4 – Variables of the Munich ChronoType Questionnaire scale (a sleep log). In its standard version, these variables are collected in the context of workdays and work-free days. BT = Local time of going to bed. SPrep = Local time of preparing to sleep. SLat = Sleep latency or time to fall asleep after preparing to sleep. SO = Local time of sleep onset. SD = Sleep duration. **MS** = Local time of mid-sleep. SE = Local time of sleep end. Alarm = A logical value indicating if the respondent uses an alarm clock to wake up. SE = Local time of sleep end. SI = “Sleep inertia” (despite the name, this variable represents the time the respondent takes to get up after sleep end). GU = Local time of getting out of bed. TBT = Total time in bed.



Source: Create by the author.

For this thesis, the MCTQ serves as the instrument for measuring subjects' chronotypes (circadian phenotypes). The study uses a dataset of 65 824 Brazilian respondents from an online survey conducted by the author in 2017, which includes geographical data such as postal codes. This data enables the examination of the potential association between chronotype and geographic factors, particularly latitude and longitude. The research ultimately seeks to determine whether latitude plays a role in shaping chronotype, contributing to our understanding of circadian rhythms in relation to geographic variables.

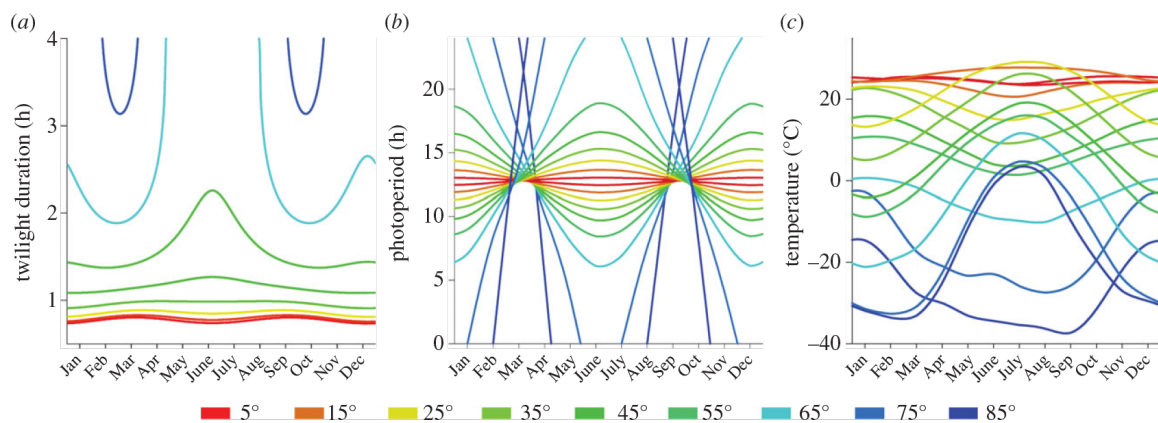
3 ON THE LATITUDE HYPOTHESIS

The first mention of this hypothesis in English scientific literature dates back to at least 1973, as noted by Bohlen and Simpson (1973), with earlier hints of the idea from Erhard Haus and Franz Halberg in 1970 (Haus & Halberg, 1970, p. 101), building on discussions initiated by Jürgen Aschoff (Aschoff, 1969). Since then, numerous studies have explored this topic, yielding somewhat conflicting results (a systematic review is provided by Randler and Rahafar (2017)).

The hypothesis, also called *environment hypothesis*, posits that regions closer to the poles receive, on average, less annual sunlight compared to regions near the equator (Figure 5). Consequently, regions around latitude 0° are thought to have a stronger solar zeitgeber. According to chronobiological theories, this stronger zeitgeber would enhance the synchronization of circadian rhythms with the light-dark cycle, resulting in lower variability and amplitude of circadian phenotypes. This reduced influence of individual endogenous periods is illustrated in Figure 6.

In contrast, populations near the poles experience a weaker solar zeitgeber, leading to greater variability and amplitude of circadian phenotypes. This disparity translates into differences in chronotype: equatorial populations tend to exhibit a morningness orientation, while populations at higher and low latitudes tend toward eveningness (Bohlen & Simpson, 1973; Roenneberg, Wirz-Justice, & Mellow, 2003).

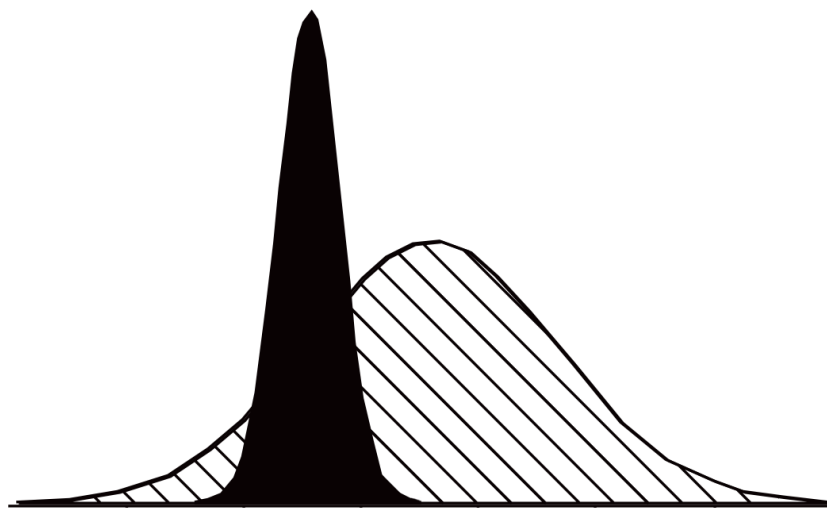
Figure 5 – Annual changes in (a) twilight duration, (b) daylight hours, and (c) temperature across different latitudes. Each color shows a specific latitude, illustrating how these factors vary throughout the year.



Source: Reproduction from Hut et al. (2013).

Some authors claim to find this association, but a closer look at the data reveals that it is not as clear as it seems. For example, Leocadio-Miguel et al. (2017) found a significant association between latitude and chronotype in a sample of 12,884 Brazilian participants. However, the effect size was negligible, with latitude explaining only about 0.388% of the variance in chronotype (Figure 7). Considering the particular emphasis that the solar zeitgeber has on the entrainment of biological rhythms (as demonstrated in many experiments), it would not be reasonable to assume that the latitude hypothesis could be supported without at least a non-negligible effect size.

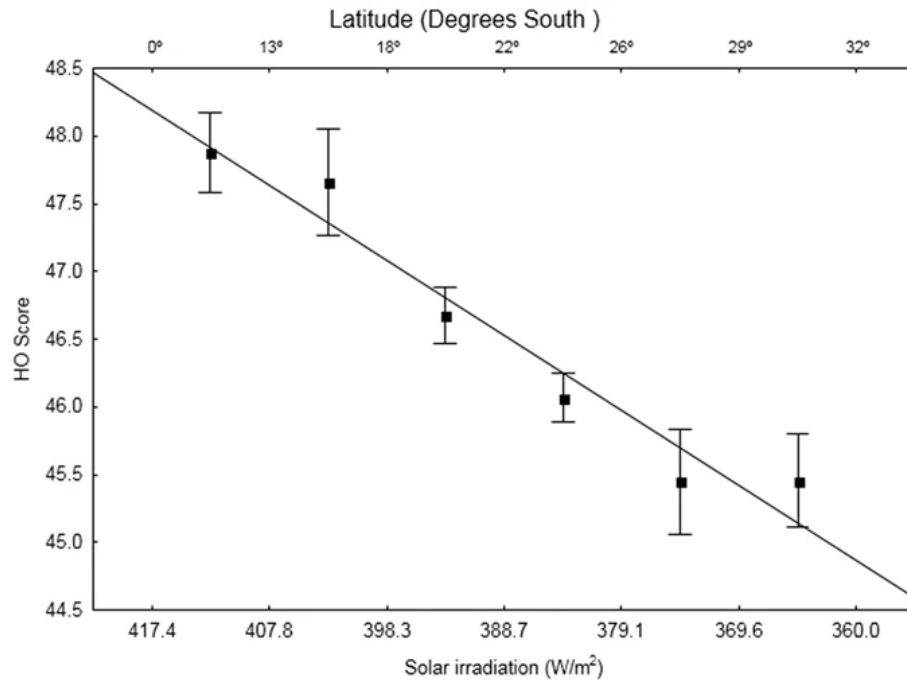
Figure 6 – Different chronotype distributions, influenced by strong and weak zeitgebers – black for strong and hatched for weak. An illustration of the effect hypothesized by the latitude hypothesis.



Source: Adapted from Roenneberg, Wirz-Justice, and Mellow (2003).

The findings of Leocadio-Miguel et al. (2017) are not consistent with the hypothesis that latitude is a strong predictor of chronotype, as the reported effect size is too small to be considered practically significant (Cohen, 1988). This highlights a common limitation of studies relying on Null Hypothesis Significance Testing (NHST) (Perezgonzalez, 2015). A p-value does not measure the effect size; instead, it represents the probability of observing the data (or something more extreme) assuming the null hypothesis is true, thus quantifying the likelihood of a type I error (Cohen, 1994; Wasserstein & Lazar, 2016).

Figure 7 – Mean scores (\pm SE) on the Horne & Östberg (HO) chronotype scale (Horne & Östberg, 1976) are presented across a latitudinal gradient, along with the corresponding annual average solar irradiation levels (W/m^2). The HO scale comprises 19 items, with total scores ranging from 16 to 86; lower scores indicate a stronger evening orientation, while higher scores reflect a greater morning orientation. Notably, the y-axis exaggerates the visual impact of the differences, as it represents a range of only about 4.5 points, which may overstate the perceived significance of the effect.



Source: Reproduction from Leocadio-Miguel et al. (2017).

Several factors may invalidate this hypothesis, such as local clock time and social constraints (Skeldon & Dijk, 2021). To gain a more accurate understanding of the mechanisms underlying chronotype expression, it remains crucial to test this hypothesis in larger samples. This study aims to address that gap.

In the following sections, the hypothesis will be tested using one of the largest chronotype datasets, to the author's knowledge, with geocoding information integrated for a comprehensive analysis. The approach will adhere to sound statistical principles, incorporating a minimum effect size in the alternative hypothesis, as originally proposed by Neyman and Pearson data testing framework (Neyman & Pearson, 1928a, 1928b).

The following study was designed for publication in the journal *Scientific Reports* (IF 2023: 3.8/JCR | CAPES: A1/2017-2020) and structured in accordance with the journal's submission guidelines.

4 IS LATITUDE ASSOCIATED WITH CHRONOTYPE?

4.1 ABSTRACT

Chronotypes are temporal phenotypes that reflect our internal temporal organization, a product of evolutionary pressures enabling organisms to anticipate events. These intrinsic rhythms are modulated by zeitgebers — environmental stimuli that entrain these biological oscillations, with light exposure being the primary mechanism. Given light's role in these systems, previous research hypothesized that latitude might significantly influence chronotypes, suggesting that populations near the equator would exhibit more morning-leaning characteristics due to more consistent light-dark cycles, while populations near the poles might display more evening-leaning tendencies with a potentially freer expression of intrinsic rhythms. To test this hypothesis, we analyzed chronotype data from a large sample of 65 824 subjects across diverse latitudes in Brazil. Our results revealed that latitude show only negligible effect sizes on chronotype, indicating that the entrainment phenomenon is far more complex than previously conceived. These findings challenge simplified environmental models of biological timing and underscore the need for more nuanced investigations into the mechanisms underlying temporal phenotypes, opening new avenues for understanding the intricate relationship between environmental cues and individual circadian rhythms.

4.2 INTRODUCTION

Humans exhibit a variety of observable traits, such as eye or hair color, which are referred to as phenotypes. These phenotypes also manifest in the way our bodies function.

A chronotype is a temporal phenotype (Ehret, [1974](#); Pittendrigh, [1993](#)), typically used to refer to endogenous circadian rhythms — biological rhythms with periods close to 24 hours. Chronobiology, the science that studies biological rhythms, suggests that the evolution of these internal oscillators is closely linked to our environment, particularly the day-night cycle. This cycle, alongside human evolution, created environmental pressures that led to the development of temporal organization within organisms (Aschoff,

1989; Paranjpe & Sharma, 2005). Such temporal organization allowed organisms to predict events and better manage their needs, such as storing food for winter.

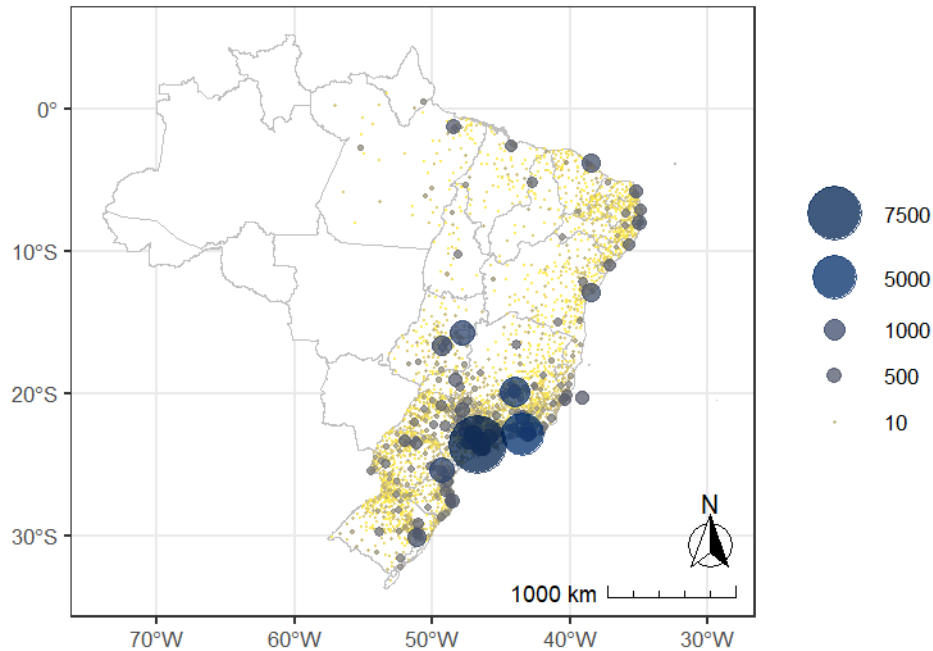
For a temporal system to be useful, it must be capable of adapting to environmental changes. Environmental signals capable of regulating biological rhythms are known as *zeitgebers* (from the German *zeit*, meaning time, and *geber*, meaning donor (Cambridge University Press, n.d.)). These *zeitgebers* provide inputs that can shift and synchronize biological rhythms. This process is called entrainment (Roenneberg, Daan, & Merrow, 2003; Roenneberg et al., 2010).

The primary *zeitgeber* influencing biological rhythms is light, particularly sunlight (Aschoff et al., 1972). Given its significant role in entraining the biological clock, several studies have hypothesized that the latitudinal shift of the sun, due to the Earth's axial tilt, might lead to different temporal traits in populations near the equator compared to those closer to the poles (Bohlen & Simpson, 1973; Hut et al., 2013; Leocadio-Miguel et al., 2017; Pittendrigh et al., 1991; Randler, 2008; Randler & Rahafar, 2017; Roenneberg, Wirz-Justice, & Merrow, 2003). This is based on the idea that populations at low or higher latitudes experience greater fluctuations in sunlight and a weaker overall solar *zeitgeber*. This concept is known as the latitude hypothesis, or the environmental hypothesis of circadian rhythm regulation.

Recent efforts to test the latitude hypothesis in humans have largely been unsuccessful in identifying a significant effect related to latitude. Many of these studies used secondary data or small sample sizes. A notable attempt was made by Leocadio-Miguel et al. (2017), who measured the chronotype of 12,884 Brazilian subjects across a wide latitudinal range using the Morningness–Eveningness Questionnaire (MEQ). Their findings showed a negligible effect size. One possible explanation is that the MEQ measures psychological traits rather than the biological states of circadian rhythms themselves (Roenneberg, Pilz, et al., 2019), meaning it might not be the most suitable tool for testing the hypothesis (Leocadio-Miguel et al., 2014).

This study presents a novel attempt to test the latitude hypothesis, using a biological approach through the Munich ChronoType Questionnaire (MCTQ) (Roenneberg, Wirz-Justice, & Merrow, 2003). In addition, it utilizes the largest dataset on chronotype in a single country, as far as the existing literature suggests, comprising 65 824 respondents, all living within the same timezone in Brazil and completing the survey within a one-week window (Figure 8).

Figure 8 – Geographical distribution of the sample used in the analysis: ($n = 65\,824$). Each point represents a municipality, with its size proportional to the number of participants and its color indicating participant density. The sample includes Brazilian individuals aged 18 or older, residing in the UTC-3 timezone, who completed the survey between October 15 and 21, 2017. The size and color scale are logarithmic (\log_{10}).



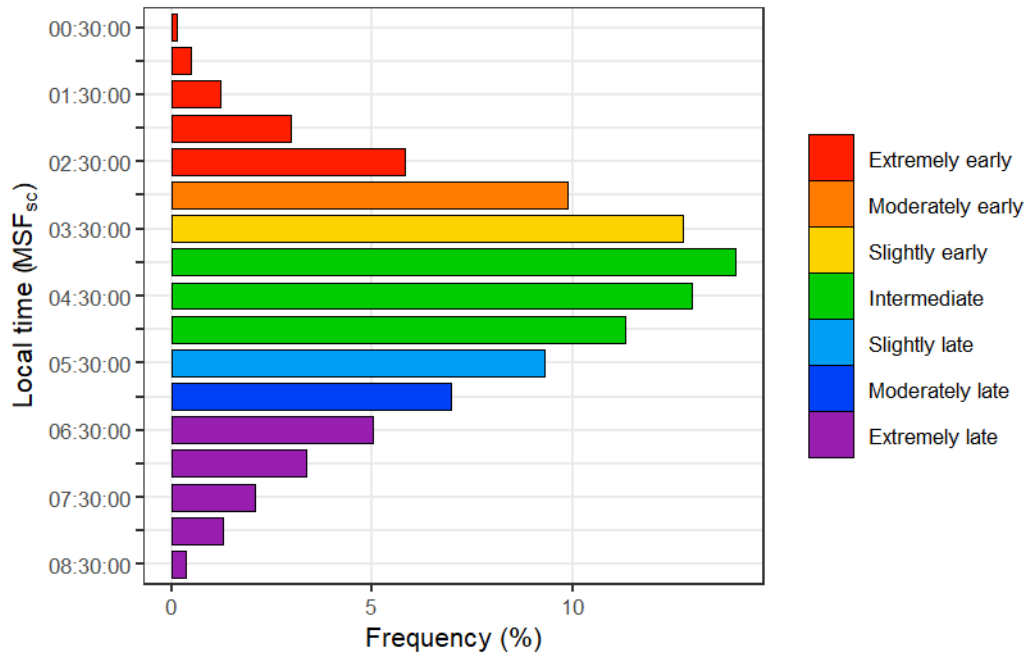
Source: Created by the author.

4.3 RESULTS

The local time of the sleep-corrected midpoint between sleep onset and sleep end on work-free days (MSFsc), which serves as the MCTQ proxy for measuring chronotype, had an overall mean of 04:28:41 and a standard deviation of 01:26:13. The distribution is shown in Figure 9.

This represents the midsleep point for Brazilian subjects with an intermediate or average chronotype. Considering the 7–9 hours of sleep recommended for healthy adults by the American Academy of Sleep Medicine (AASM) (Watson et al., 2015), one could imagine that this average individual, in the absence of social restraints, would typically wake up at approximately 08:28:41.

Figure 9 – Distribution of the local time for the sleep-corrected midpoint between sleep onset and sleep end on work-free days (MSF_{sc}), a proxy for chronotype. Chronotypes are categorized into quantiles, ranging from extremely early ($0 | - 0.11$) to extremely late ($0.88 - 1$).



Source: Created by the author. Based on data visualization found in Roenneberg, Wirz-Justice, et al. (2019).

The study hypothesis was tested using nested multiple linear regressions. The core idea of nested models is to evaluate the effect of including one or more predictors on the model's variance explanation (R^2) (Maxwell et al., 2018). This is achieved by comparing a restricted model with a full model. Cell weights, based on sex, age group, and state of residence, were used to account for sample imbalances.

Two tests were conducted, both using the same restricted model, which included age, sex, longitude, and the monthly Global Horizontal Irradiance (GHI) average at the time of questionnaire completion as predictors (Adjusted $R^2 = 0.085110$; $F(4, 65818) = 1530$, $p\text{-value} < 2e-16$). The first full model (**A**) added annual GHI average and daylight duration for the nearest March equinox, as well as the June and December solstices, as proxies for latitude, following Leocadio-Miguel et al. (2017) methods (Adjusted $R^2 = 0.087921$; $F(8, 65814) = 794$, $p\text{-value} < 2e-16$). The second full model (**B**) added only latitude as a predictor (Adjusted $R^2 = 0.085614$; $F(5, 65817) = 1230$, $p\text{-value} < 2e-16$). All coefficients were significantly different from zero ($p\text{-value} = 2e-16$). Assumption checking and residual diagnostics primarily relied on visual inspection, as objective

assumption tests (e.g., Anderson–Darling) are not advisable for large samples (Shatz, 2024). All validity assumptions were met, and no serious multicollinearity was found among the predictor variables.

Sunrise times for the nearest March and September equinoxes, as well as the June and December solstices, were excluded due to high multicollinearity. Daylight duration for the September equinox was excluded for its multicollinearity with daylight duration during the March equinox.

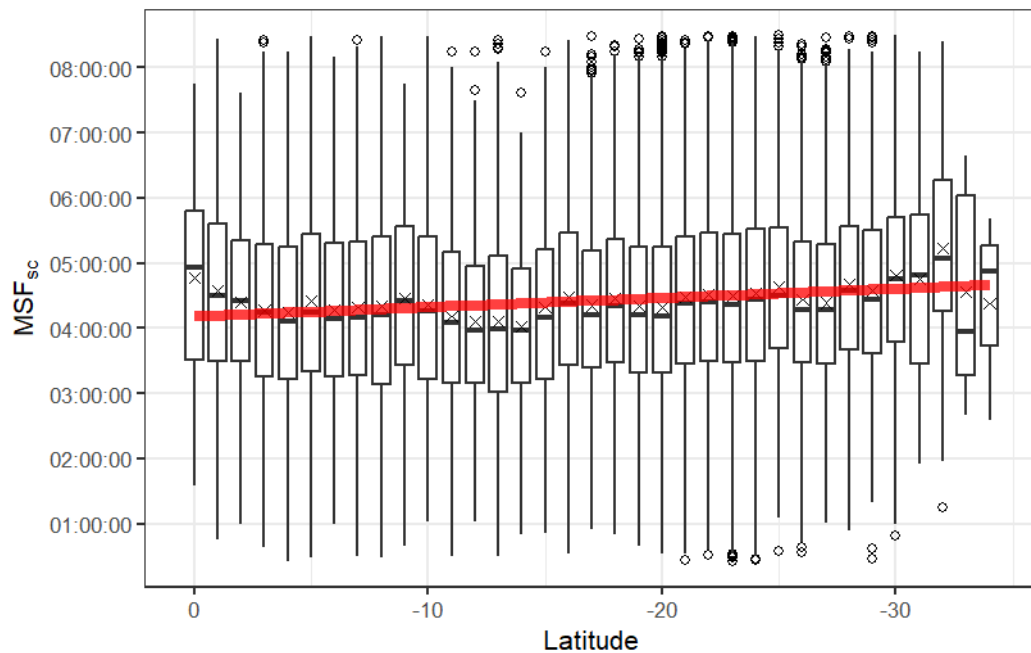
An F test for nested models revealed a significant reduction in the residual sum of squares (**A** $F(4, 65814) = 51.71$, $p\text{-value} < 2e - 16$; **B** $F(1, 65817) = 37.325$, $p\text{-value} < 1e - 9$). However, when estimating Cohen's f^2 effect size, the results were negligible (Cohen, 1992) (**A** $f^2 = 0.012137120$; **B** $f^2 = 0.009523916$).

4.4 DISCUSSION

It is important to emphasize that assuming a causal and linear relationship between latitude and chronotype is an *a priori* hypothesis. The objective of this study is to test and potentially falsify this hypothesis.

The results indicate that, despite a broad latitudinal spectrum and a large, well-aligned sample, the latitude effect on chronotype does not manifest in a meaningful way. Several studies have suggested a potential effect of latitude on chronotype, but, at present, no empirical evidence supports this claim in humans. These findings align with those of Leocadio-Miguel et al. (2017), who observed a similar effect size (Cohen's $f^2 = 0.004143174$). However, the earlier study did not incorporate a minimum effect size criterion, which led to misleading conclusions. The small and inconsistent size of the latitude effect can be seen in Figure 10. Figure 11 shows the distribution of chronotypes by the mean for each Brazilian state.

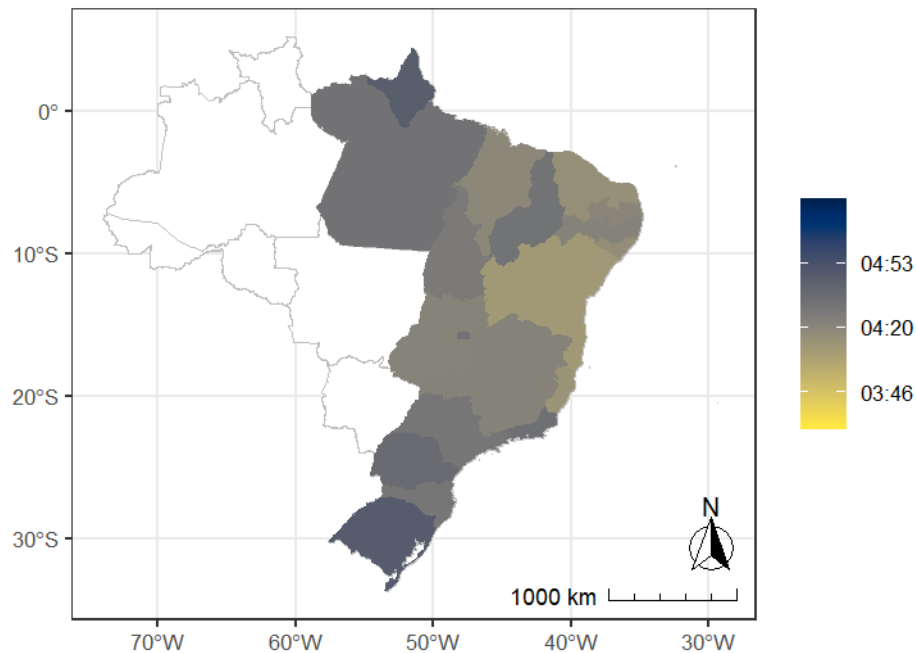
Figure 10 – Boxplots of mean MSF_{sc} values aggregated by 1° latitude intervals, illustrating the relationship between latitude and chronotype. MSF_{sc} represents the local time of the sleep-corrected midpoint between sleep onset and sleep end on work-free days, a proxy for chronotype. Higher MSF_{sc} values indicate later chronotypes. The \times symbol points to the mean. The red line represents a linear regression. The differences in mean/median values across latitudes are minimal relative to the Munich ChronoType Questionnaire (MCTQ) scale.



Source: Created by the author.

The absence of a clear relationship between latitude and chronotype can be attributed to multiple factors. As Jürgen Aschoff might have put it, this may reflect a lack of “ecological significance” (Aschoff et al., 1972). Even if latitude does influence circadian rhythms, the effect could be too minor to detect or might be overshadowed by other, more prominent factors like social behaviors, work hours, or the widespread use of artificial lighting (Bohlen & Simpson, 1973). Furthermore, the variations in sunlight exposure between latitudes may not be substantial enough to meaningfully impact the circadian system, which is highly responsive to light. Since even small fluctuations in light exposure can lead to measurable physiological changes, it suggests that latitude alone may not be a decisive factor in determining chronotype.

Figure 11 – Geographical distribution of mean MSF_{sc} values by Brazilian state. The figure shows the mean mid-sleep on free days sleep-corrected (MSF_{sc}) values aggregated by state, illustrating how chronotype varies with latitude in Brazil. MSF_{sc} is a proxy for chronotype, representing the midpoint of sleep on work-free days, adjusted for sleep debt. Higher MSF_{sc} values correspond to later chronotypes. The color scale was not transformed and it has as limits the first and third quartile (interquartile range). While the map suggests a potential latitudinal pattern, differences in mean MSF_{sc} values across states are small and fall within a narrow range relative to the scale of the Munich ChronoType Questionnaire (MCTQ), limiting the significance of these variations.



Source: Created by the author.

This study points to a more intricate relationship between latitude and the circadian system than originally expected. In human populations, the perceived link between these variables might have been influenced by statistical overconfidence, driven by routine reliance on Null Hypothesis Significance Testing (NHST) and the tendency to confirm preconceived ideas, rather than a rigorous and unbiased examination of the data.

4.5 METHODS

4.5.1 Measurement instrument

Chronotypes were assessed using a sleep log based on the core version of the standard Munich ChronoType Questionnaire (MCTQ) (Roenneberg, Wirz-Justice, &

Morrow, 2003), a well-validated and widely applied self-report tool for measuring sleep-wake cycles and chronotypes (Roenneberg, Pilz, et al., 2019). The MCTQ captures chronotype as a biological circadian phenotype, determined by the sleep-corrected midpoint of sleep (MSF_{sc}) on work-free days, accounting for any potential sleep compensation due to sleep deficits on workdays (Roenneberg, 2012).

Participants completed an online questionnaire, which included the sleep log as well as sociodemographic (e.g., age, sex), geographic (e.g., full residential address), anthropometric (e.g., weight, height), and data on work or study routines. A sample version of the questionnaire, stored independently by the Internet Archive organization can be viewed at <https://web.archive.org/web/20171018043514/each.usp.br/gipso/mctq>.

4.5.2 Sample

The dataset used for analysis was made up of 65 824 Brazilian individuals aged 18 or older, residing in the UTC-3 timezone, who completed the survey between October 15 and 21, 2017.

The unfiltered valid sample comprises 115 166 participants from all Brazilian states, while the raw sample is composed of 120,265 individuals. The majority of the sample data was obtained in 2017 from October 15th to 21st by a broadcast of the online questionnaire on a popular Brazil's Sunday TV show with national reach (Rede Globo, 2017). This amount of data collected in such a short time gave the sample a population cross-sectional characteristic.

Based on 2017 data from the Brazilian Institute of Geography and Statistics's (IBGE) Continuous National Household Sample Survey (PNAD Contínua) (Instituto Brasileiro de Geografia e Estatística, n.d.), Brazil had 51.919% of females and 48.081% of males with an age equal to or greater than 18 years old. The sample is skewed for female subjects, with 66433% of females and 33567% of male subjects. To balance the sample, a weighting procedure was applied to the data. The weights were calculated by cell weighting, using the sex, age group and Brazil's state as reference.

A survey conducted in 2019 by the Brazilian Institute of Geography and Statistics (IBGE) (2021) found that 82.17% of Brazilian households had access to an internet

connection. Therefore, this sample is likely to have a good representation of Brazil's population.

The sample latitudinal range was 33.85026 decimal degrees (Min. = -33.522 ; Max. = 0.329) with a longitudinal span of 22.741 decimal degrees (Min. = -57.553 ; Max. = -34.812). For comparison, Brazil has a latitudinal range of 39.023 decimal degrees (Min. = -33.751 ; Max. = 5.272) and a longitudinal span of 45.155 decimal degrees (Min. = -73.990 ; Max. = -28.836).

More information about the sample can be found in the supplementary materials.

4.5.3 Data wrangling

Data wrangling and analysis followed the data science program proposed by Hadley Wickham and Garrett Grolemund (Wickham, 2016). All processes were made with the help of the R programming language (R Core Team, n.d.), RStudio IDE (Posit Team, n.d.), and several R packages. The *tidyverse* and *rOpenSci* peer-reviewed package ecosystem and other R packages adherents of the tidy tools manifesto (Wickham & Bryan, 2023) were prioritized. The MCTQ data was analyzed using the *mctq* *rOpenSci* peer-reviewed package (Vartanian, n.d.). All processes were made in order to provide result reproducibility and to be in accordance with the FAIR principles (Wilkinson et al., 2016).

4.5.4 Hypothesis test

The study hypothesis was tested using nested models general linear models of multiple linear regressions. It was schematized as follows.

- **Null hypothesis** (H_0): Adding *latitude* does not meaningfully improve the model's fit, indicating that the change in adjusted R^2 is negligible or the F-test is not significant (considering a type I error probability (α) of 0.05).
- **Alternative Hypothesis** (H_a): Adding *latitude* meaningfully improves the model's fit, indicating that the change in adjusted R^2 is greater than the Minimum Effect Size (MES), and the F-test is significant (considering a type I error probability (α) of 0.05).

$$\begin{cases} H_0 : \Delta \text{ Adjusted } R^2 \leq \text{MES} & \text{or} & \text{F-test is not significant } (\alpha \geq 0.05) \\ H_a : \Delta \text{ Adjusted } R^2 > \text{MES} & \text{and} & \text{F-test is significant } (\alpha < 0.05) \end{cases}$$

Where:

$$\Delta \text{ Adjusted } R^2 = \text{Adjusted } R_f^2 - \text{Adjusted } R_r^2$$

A MES must always be used in any data testing. The effect-size was present in the original Neyman and Pearson framework (Neyman & Pearson, 1928a, 1928b), but unfortunately this practice fade away with the use of p-values, one of the many issues that came with the Null Hypothesis Significance Testing (NHST) (Perezgonzalez, 2015). While p-values are estimates of type 1 error (in Neyman–Pearson’s approaches, or like-approaches), that’s not the main thing we are interested while doing a hypothesis test, what is really being test is the effect size (i.e., a practical significance). Another major issue to only relying on p-values is that the estimated p-value tends to decrease when the sample size is increased, hence, focusing just on p-values with large sample sizes results in the rejection of the null hypothesis, making it not meaningful in this specific situation (Gómez-de-Mariscal et al., 2021; Lin et al., 2013).

Considering the particular emphasis that the solar zeitgeber has on the entrainment of biological rhythms (as demonstrated in many experiments), it would not be reasonable to assume that the latitude hypothesis could be supported without at least a non-negligible effect size. With this in mind, this analysis used Cohen’s f^2 threshold for small/negligible effects, the Minimum Effect Size (MES) is defined as 0.02 (Cohen, 1988, p. 413; Cohen, 1992, p. 157). For comparison, Cohen’s threshold for medium effects is 0.15, and for large effects is 0.35.

Knowing Cohen’s f^2 , is possible to calculated the equivalent R^2 :

$$0.02 = \frac{R^2}{1 - R^2} \quad \text{or} \quad R^2 = \frac{0.02}{1.02} \approx 0.01960784$$

In other words, the latitude must explain at least 1.960784% of the variance in the dependent variable to be considered non-negligible. This is the Minimum Effect Size (MES) for this analysis.

In summary, the decision rule for the hypothesis test is as follows:

- **Reject H_0 if both:**
 - The F-test is significant.
 - $\Delta \text{ Adjusted } R^2 > 0.01960784$;
- **Fail to reject H_0 if either:**
 - The F-test is not significant, or
 - $\Delta \text{ Adjusted } R^2 \leq 0.01960784$.

As usual, the significance level (α) was set at 0.05, allowing a 5% chance of a Type I error.

A power analysis was performed to determine the necessary sample size for detecting the MES effect. The results indicate that at least 1,895 observations per variable were required to achieve a power of 0.99 ($1 - \beta$) and a significance level (α) of 0.01. The dataset contains 65,824 observations, which exceeds this requirement.

4.5.5 Data availability

Some restrictions apply to the availability of the main research data, which contain personal and sensitive information. As a result, this data cannot be publicly shared. Data are, however, available from the author upon reasonable request.

Unrestricted data can be access on the research compendium via [The Open Science Framework](#) at the following link: <https://doi.org/10.17605/OSF.IO/YGKTS>.

4.5.6 Code availability

All analyses are fully reproducible and were conducted using the [R programming language](#) alongside the [Quarto](#) publishing system. The `renv` package was used to ensure that the R environment used can be restored (see `renv.lock`).

The code repository is available on GitHub at <https://github.com/danielvartan/mastersthesis>, and the research compendium can be accessed via [The Open Science Framework](#) at the following link: <https://doi.org/10.17605/OSF.IO/YGKTS>.

4.6 ACKNOWLEDGMENTS

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4.7 ETHICS DECLARATIONS

The author declares that the study was carried out without any commercial or financial connections that could be seen as a possible competing interest.

4.8 ADDITIONAL INFORMATION

See the supplementary material for more information.

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4.9 RIGHTS AND PERMISSIONS

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5 CONCLUSION

According to Popper, the aim of science is to provide “satisfactory explanations of whatever strikes us as being in need of explanation” (Popper, 1979, p. 193). This study, using what is arguably the largest dataset on chronotype collected within a single time zone—balanced to reflect population proportions at the time of data collection—found no support for the latitude hypothesis (Cohen’s $f^2 = 0.012137120$). This result contributes meaningful evidence to the understanding of circadian rhythm regulation, offering a clear and satisfactory answer to the central research question of this thesis: “Is latitude associated with chronotype?” The answer is **No**.

These findings are consistent with those of Leocadio-Miguel et al. (2017), who reported a similar effect size (Cohen’s $f^2 = 0.004143174$). However, the earlier study did not apply a minimum effect size criterion, leading to a misleading conclusion.

Several factors could explain the lack of an association between latitude and chronotype — or, as Jürgen Aschoff might have phrased it, the absence of “ecological significance” (Aschoff et al., 1972). For instance, if latitude does affect the circadian system, the effect may be too small to detect or could be overshadowed by other influences, such as social habits, work schedules, or the use of artificial light (Bohlen & Simpson, 1973; Skeldon & Dijk, 2021). Additionally, the difference in solar exposure across latitudes may be insufficient to produce a meaningful effect on the circadian system, which is highly sensitive to light. Even minor variations in light exposure can yield significant physiological responses, suggesting that latitude alone may not be a strong predictor of chronotype.

These results suggest that the relationship between latitude and the circadian system is far more complex than anticipated. In human contexts, the perception of such an effect may have arisen from statistical misinterpretations, driven by ritualistic reliance on Null Hypothesis Significance Testing (NHST) and confirmation bias, rather than a critical evaluation of the data.

5.1 LIMITATIONS

While this study provides valuable insights, it is essential to acknowledge certain limitations that may influence the interpretation of the findings. First, the data collection

occurred predominantly during a single week in spring, as summer approached, which limited the photoperiod variability between regions. A better approach would involve data collection across different seasons, particularly during winter, when photoperiod differences are more pronounced between equatorial and polar regions.

Additionally, the use of the Munich Chronotype Questionnaire (MCTQ), while a validated instrument, introduces the potential for recall and social desirability biases inherent to self-reported measures. However, the large sample size likely mitigates these biases, as predicted by the law of large numbers (DeGroot & Schervish, 2012, p. 352). Furthermore, at the time of data collection, the MCTQ had not yet been officially validated in Portuguese (this was only introduced in 2020 by Reis et al. (2020)), which may have introduced minor inconsistencies, though its nature as a sleep log suggests this impact was minimal.

Another factor to consider is the timing of data collection relative to the start of Daylight Saving Time (DST) in Brazil. On the day data collection commenced (October 15th, 2017 – 80.153% of the data used in this analysis were collected on this day), a significant portion of respondents adjusted their clocks forward by one hour. While this could theoretically influence their responses, the questions were specifically designed to capture daily routines, which were not affected by the DST adjustment at that moment. Furthermore, any potential effect of DST would likely strengthen the latitude hypothesis; however, this was not supported by the data.

These limitations, while noteworthy, do not undermine the study's findings but rather highlight areas for refinement in future research.

5.2 DIRECTIONS FOR FUTURE RESEARCH

This thesis proposed using a global modeling approach to investigate the latitude-chronotype relationship. However, as demonstrated by the results of this study and others, no significant effect of latitude on chronotype was identified. That said, it remains possible that if such a phenomenon exists, it could be captured through a localized approach, such as agent-based modeling. This approach would simulate an environment where agents are exposed to varying light levels, while accounting for their endogenous rhythms and the circadian clock's phase-response curve to light. The data from this thesis could serve to calibrate and validate this model.

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