ORIGINAL ARTICLE

Sleep Science

Delayed sleep-wake phase disorder in a clinical population: gender and sub-population differences

Cátia Reis[1,2]
Teresa Paiva[2]

[1] Instituto de Saúde Ambiental (ISAMB), Faculdade de Medicina, Universidade de Lisboa - Lisboa- Portugal.
[2] CENC - Sleep Medicine Center - Lisboa - Portugal.

ABSTRACT

Objective/Background: Delayed sleep-wake phase disorder (DSWPD) is defined by a delay in the major sleep episode relative to desired or required sleep and wake times. The objectives of this study were to evaluate DSWPD in our population and to compare it with similar clinical data, to analyse gender differences, and to identify possible subpopulations based on circadian timing and alignment. **Patients/Methods:** 162 consecutive DSWPD patients from a sleep clinic with a median age of 35.5 (24.0) years, 85 (52.5%) males were studied. Patient data were obtained from a clinical interview composed of socio-demographic, life events, daily habits, consumptions, and comorbidities data; and from diaries, actimetry, melatonin and PSG T1. The Dim Light Melatonin Onset (DLMO) was used to define circadian alignment or misalignment. **Results:** In our DSWPD cohort, there were gender differences for different age groups (p=0.028). Men were more likely to be single and women more likely to be married (p=0.034). In students, school failure was higher for women (p<0.001); for workers, absenteeism was higher in women (p=0.001). In the circadian aligned (compared to misaligned group), DLMO was later (p<0.001), sleep onset time (p=0.046) was later, total sleep time (p=0.035), and number of sleep cycles (p=0.018) were lower, as measured using PSG T1. **Conclusions:** In this clinical population, DSWPD is more prevalent in young men and in middle age women, although with no overall significant differences between genders. There are two different phenotypes of DSWPD: circadian misaligned and circadian aligned. Depression is prevalent in both groups. Better definition, classification and diagnostic criteria for DSWPD are still needed, and targeted therapeutical intervention should be evaluated.

Keywords: Gender differences; phase angle; circadian phase; circadian misalignment; phenotypes.

Corresponding author:
Cátia Reis.
E-mail: catiareis@cenc.pt
Received: April 24, 2019; Accepted: September 4, 2019.

DOI: 10.5935/1984-0063.20190086

INTRODUCTION

Delayed sleep-wake phase disorder (DSWPD) is the most prevalent circadian rhythm sleep-wake disorder (CRSD)[1] and is characterized by a delay in the major sleep episode relative to desired or required sleep and wake times. It causes difficulty in initiating sleep and consequently awakening at desired or required clock times[1]. Patients complain and may only present for diagnosis and treatment - when they have required early awakenings that also shorten sleep duration[2] or whenever the daily habits have serious deleterious impact upon their family or social life. DSWPD is more prevalent in teenagers and young adults (7-16%) than in middle-aged adults (3.1%)[1,3]. A prevalence study for DSWPD performed in Norway in 1983 reported 0.17% of the Norway population were affected[4]. A recent population-based study in New Zealand evaluating adults aged 20-59 demonstrated the prevalence of DSWPD to be between 1.51% and 8.90%, depending on the definition used[5]. The lack of a simple, reliable, low cost diagnostic phase biomarker for clinical practice, the high rate of therapeutic failure in DSWPD[2], its low prevalence[1] and the patients' belief that it is a behavioural problem caused by a societal desynchronization (personal communication), may be responsible for the few available clinical studies in DSWPD patients.

The disorder usually starts during adolescence[1]. The onset of puberty and adolescence is typically characterized by a biological delay in sleep timing[6,7]; a reversal towards earlier sleep times then occurs at the start of adulthood[8,9]. One hypothesis of DSWPD development is that the delayed sleep of adolescence is maintained in some individuals, culminating in DSWPD.

The diagnosis is difficult because the disease criteria are based on clinical symptoms similar to insomnia[10]. DSWPD is associated with a constellation of symptoms that impact negatively upon quality of life and social/individual achievements: these include academic/professional failure, higher likelihood of accidents, depression, suicidal tendencies, and medication and substance consumption[2]. DSWPD symptoms also include indecision and procrastination in academic life[11] and in career decisions[12], each of which may contribute to the academic/professional failiure. Depressive symptoms are common in patients diagnosed with DSWPD: 64% of patients with DSWPD have a comorbid depression[3,13].

While the disorder's name suggests a circadian basis ("Phase"), not all individuals with DSWPD have circadian abnormalities[3,14]. A multi-center study in Australia with DSWPD patients[3] found two different phenotypes based on the relationship between Dim Light Melatonin Onset (DLMO, a marker of circadian phase)[14–16] and desired bedtime: one group had delayed circadian phase while the other group did not. Individuals with the circadian DSWPD phenotype have higher rates of depressive symptoms and daytime sleepiness[3].

The basis for using DLMO as a circadian marker is that a signal from the central circadian pacemaker in the suprachiasmatic nucleus of the hypothalamus initiates melatonin synthesis, with subsequent release[17]. Melatonin levels are negligible during the day and start to rise in the early evening[17] approximately 2-3h before typical bedtime[15,17,18]. Melatonin production is supressed by light[19]. One reason for DSWPD may be differential sensitivity to light stimuli: individuals with DSWPD exposed to light with an intensity of 1000 lux for 2h beginning prior to their peak of melatonin secretion had higher melatonin suppression compared to controls[20]. In addition, DSWPD individuals usually stay awake late at night and sleep into the morning; this behaviour results in a greater light exposure during the phase delay portion of circadian system (causing people to stay awake longer) and less light exposure during the phase advance portion of the circadian system (decreasing the tendency to go to sleep earlier), leading to an overall circadian delay[21]. Thus the late sleep timing of DSWPD patients may both cause and be a cause of misaligment of the individual's circadian rhythm with clock time.

Portugal is recognized as a country with late sleep/wake habits: 70% of the adult population goes to bed after midnight[22] and 17.7% have a reduced sleep duration[23], presumably because of forced timing of waking for work, school, or other reasons. Some of these individuals would be expected to have DSWPD. Therefore, we extensively evaluated a clinical population of DSWPD patients from a Sleep Medicine Center in Portugal. We documented complaints, comorbidities, socio-demographic data, and sleep questionnaires, and collected sleep diary, actigraphy, melatonin DLMO and Polysomnography (PSG) type 1 data.

The objectives of this study were: 1) to evaluate DSWPD in our population and to compare it with similar clinical data; 2) to document gender differences in DSWPD; 3) to identify possible subpopulations based on circadian timing and alignment in the total DSWPD sample.

METHODS

Participants

This was a retrospective analysis of 245 consecutive patients from one sleep clinic that were diagnosed with DSWPD according to the ICSD3 criteria[1] between 2012-2017. A board-certified sleep physician conducted a clinical interview. Of the 245 patients 83 were eliminated: eighteen (18) were shift workers and had delayed sleep due to their work schedule and 65 had only clinical information. All individuals included in this report performed at least one objective measurement of diagnosis (actigraphy, DLMO, and/or PSG) in addition to the clinical interview. Of the final sample of 162 individuals, 21 had only one objective measure, 77 had at 2 objective measures and 62 had all the three objective measurements (Figure 1).

Procedures

All individuals had a clinical interview performed by the same physician who is a neurologist board-certified in sleep medicine, followed by sleep/activity diary, actimetry and/or PSG type I measurements. During the clinical evaluation, a detailed clinical history of the patient was obtained. Questions

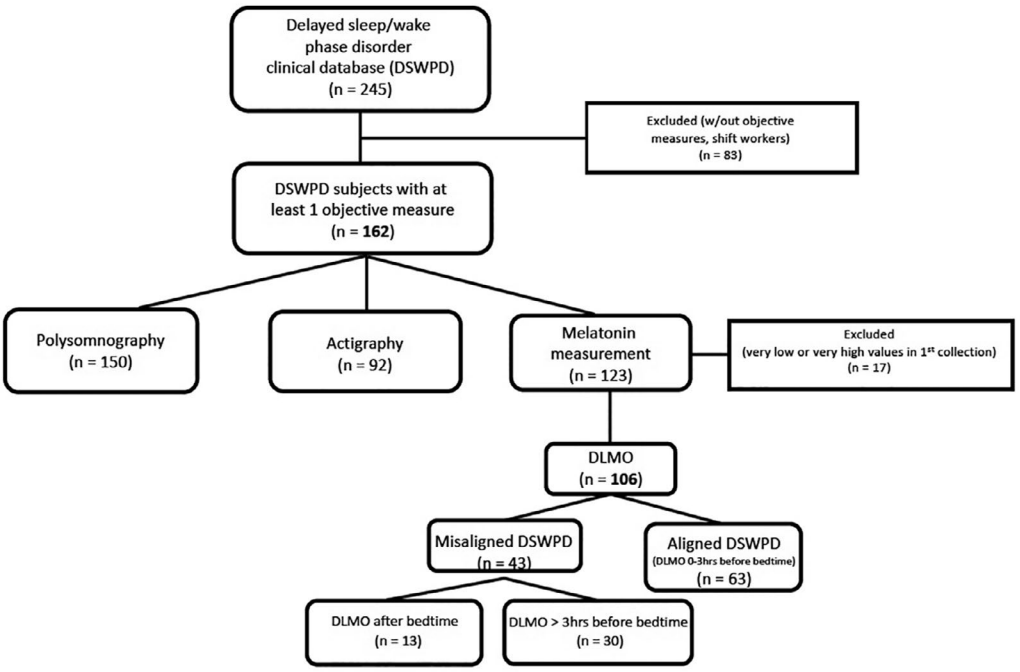

Figure 1. Flowchart of the study.

included: (i) socio-demographic information including age, gender, marital status, employment status, school failure (defined as at least 1 year of retention), work absenteeism; (ii) life events (e.g., traffic accidents, familial conflicts, traumatic events); (iii) daily habits (e.g., meal, sleep and work times); (iv) drug consumptions (e.g., tobacco, alcohol abuse, illicit drugs); (v) other drugs both prescribed or not prescribed by a physician; (vi) self-prescribed melatonin consumption; and (vii) co-morbidities (e.g., depression, anxiety, psychiatric disorders, neurologic disorders, medical disorders, insomnia). All shift workers were excluded for this study since their work schedule caused changes in sleep timing.

This is a retrospective study using a clinical anonymized database; informed consent was not required. The Lisbon Medical School of the Universidade de Lisboa Ethics Committee and the Portuguese National Data Protection Board approved the study.

Measures

Questionnaires

The Epworth Sleepiness Scale (ESS) was used to assess daytime sleepiness levels and the Pittsburgh Sleep Quality Index (PSQI) to assess sleep quality. For the ESS results at or above 10 indicate abnormal or pathological sleepiness and results at or above 17 indicate severe sleepiness[24]. For the PSQI higher scores represent worse sleep quality; "bad" sleep quality is a total score ≥ 5[25].

Actigraphy and Diary

For 1-2 weeks, patients wore an actimeter and completed a sleep/wake diary.

Patients wore the Actiwatch 2 from Philips Respironics or ActTrust from Condor on the non-dominant wrist. The actigraphs were placed, programmed and removed by a sleep technician. Data were collected in 30-second epochs, for both devices. These devices recorded total light and activity. Sleep and wake parameters were automatically scored using the manufacturer's software: Actiware 5.59 for Philips Respironics and ActStudio v.1.0.5.6 for the Condor actimeters. The Actiware default algorithm sets the rest interval according to activity data and detects the major rest intervals of low activity that are longer than 3 hours[26]. The ActStudio also uses activity data to identify sleep/wake states using the ClColeKripke algorithm which was developed based on the Cole-Kripke work[27]. The sleep wake timing given by the devices algorithms were confirmed and/or corrected according to participants diary by one of us (CR). The diary included bed time and wake up time, meals and working schedules for each day.

Melatonin measurement and analyses

Saliva collections were performed at the patient's home in a dim light environment. Blue-wavelength-light blocking glasses were distributed to patients. Patients were instructed to start using them 2 hours before the first sampling through the last collection. Data collections were at 3^{hr}, 2^{hr}, and 1^{hr} before bedtime, bedtime and 1h after regular bedtime. For the extreme late individuals (bedtime after 04h00), collection started at 0:00h and finished at 1h after usual bedtime. A range of 5 - 10 collections were obtained from patients. Dim Light Melatonin Onset (DLMO) was calculated using a minimum threshold of 4pg/ml[28].

To quantify circadian alignment, calculation of phase angle was performed. Phase angle was defined as the time

difference between DLMO time and sleep onset time[29]. A normal phase angle is -3 hours (with DLMO before sleep time)[15,17,18]. Patients were categorized as aligned if they had a phase angle of ≤ 3 hours; misaligned was defined as phase angle > 3 hours (i.e., DLMO too early) or with a DLMO time after sleep onset time (i.e., DLMO too late) (Figure 1).

Polysomnography

Overnight polysomnography was performed with one of the following PSG systems: Alice 5 Respironics; Nicolet System - Viasys Healthcare; Embla N7000; or Domino Somnoscreen Plus - Somnomedics. The recorded parameters included: electroencephalography (F3-M2, F4-M1, C3-M2, C4-M1, O1-M2, O2-M1); left and right electrooculogram; submental electromyogram; bilateral tibial electromyogram; electrocardiogram; oronasal airflow with 3-pronged thermistors; nasal pressure with a pressure transducer; rib cage and abdominal wall motion via respiratory impedance plethysmography; arterial oxygen saturation with pulse waveform; and digital video and audio. The sleep period was scored from "lights off" to "lights on," with lights off scheduled as close as possible to participant's normal sleep schedule. There was a median difference of 6 (139) minutes between the timing of the recorded PSG and habitual sleep time, raging from -210 minutes (after habitual sleep time) to 403 minutes (before the habitual sleep time). Sleep scoring was by the AASM scoring manual version 2.4 criteria[30], the apnea-hypopnea index (AHI) was used to determine sleep disordered breathing status, namely Obstructive Sleep Apnea (OSA), using the following criteria: No OSA: AHI < 5/h; Mild OSA: AHI ≥ 5/h and ≤ 15/h; Moderate ≥ 15/h and ≤ 30/h; Severe OSA: AHI > 30/h. For Periodic Limb Movement Disorder (PLMD) the confirmation criteria established was a frequency of movements >15/h, according to the ICSD3[1]. The number of sleep cycles were calculated and each sleep cycle was represented by the beginning of the first stage of sleep (N1) until the end of the REM sleep episode[31].

We excluded wake time and sleep duration metrics from the inviduals who asked the sleep technicians to be awakened in the morning (n=50) to go to work/school. Also for the two phenotypes comparison these individuals were excluded.

Statistical Analysis

Absolute frequencies and proportions were used to summarize categorical variables. Continuous variables were described by mean values and standard deviations if data distribution was statistically normal and by median and interquartile range for non-parametric data. Data statistical normality was checked using the Kolmogorof-Smirnov test with Liliefors statistical correction. Differences between gender were statistically assessed using the *Student t-test* or Mann-Witney test for continuous variables, according to data distribution, and the Chi-square test for categorical variables.

All analyses were performed using SPSSv.25.

RESULTS

Demographic variables

Of the 162 patients, 85 (53%) were males and 77 (47%) were females.The median age was 35 (24) years with a range of 16 to 92 years. 81 (53%) were single, 13 (9%) were divorced, 5 (3%) were widowed and 53 (35%) were married or living with a non-married partner. The educational level was high: 77 (58%) patients had more than 12 years of education, and 55 (42%) had ≤ 12 years of education. Twenty patients (12%) were unemployed or retired, 44 (27%) were students, and 98 (61%) had a regular job. Thirty patients (19%) reported a family history of DSWPD. The mean body mass index (BMI) was 24.1 ± 4.0 kg/m^2, ranging from 16.6 to 36.2 kg/m^2 (Table 1).

Clinical variables

The reported median age for the onset of DSWPD symptoms was 18 (19) years, ranging from 2 to 82 years. Twenty-five (15%) patients mentioned some trigger events for DSWPD onset; triggers included trauma or stress, employment issues, depression and anxiety, menopause, and the need to study in the university. The disease mean age of onset was significantly later (p<0.001) for the patients that reported a trigger for the disease.

Emotional variables

Fifty-five subjects (34%) had family conflicts, 9 (6%) had car accidents, and 22 (16%) had suffered a trauma (e.g. sexual abuse or death of a close relative). Of the students, 13 (39%) had school failure and of the workers, 5 (5%) reported frequent absenteeism.

Daily routines

All patients reported sleep onset after 02:00h. 55% of the patients had irregular routines of work/school, meals or exercise, with daily variability ranging from more that 2 hours of difference between days, with 58% skipping one or several meals along the day, mostly breakfast or lunch.

Consumption habits

Thirty-one (21%) patients were smokers and 25 (16%) were alcohol excessive consumers acording to the DCM-5 criteria[32]. Seventeen (11%) reported consuming illicit drugs like cannabis, 70 (50%) reported taking antidepressants, 74 (44%) hypnotic drugs, and 58 (41%) melatonin.

Comorbidities

Self-reported comorbidities like anxiety and depression were extremely frequently reported: 96 (59%) reported anxiety, 69 (43%) depression. 37 (23%) other medical non-neurologic diseases and 24 (15%) neurological diseases.

Insomnia was a frequent complaint: it was reported in 85 (55%).

Associated sleep disorders were diagnosed by the PSG: 19 (12%) suffered from periodic leg movements (PLM) and 25 (16%) from obstructive sleep apnea (OSA) (Table 1).

Table 1. Baseline characteristics by sex.

	Total sample	Female	Male	*p*-value
N	162.0	77.0 (47.5)	85.0 (52.5)	
Age, median (interquartile range)	35.5 (24.0)	41.0 (27.0)	32.0 (21.0)	ns[a]
Age group, N (%)				
16-20 years	16.0 (9.9)	9.0 (56.3)	7.0 (43.8)	0.028[b]
21-30 years	46.0 (28.6)	13.0 (28.3)	33.0 (71.7)	
31-40 years	32.0 (19.9)	15.0 (46.9)	17.0 (53.1)	
41-50 years	28.0 (17.4)	18.0 (64.3)	10.0 (35.7)	
51-60 years	17.0 (10.6)	11.0 (64.7)	6.0 (35.3)	
≥ 65 years	23.0 (13.7)	11.0 (45.5)	12.0 (54.5)	
Age of disease onset, median (interquartile range)	18.0 (19.0)	16.0 (17.0)	20.0 (16.0)	ns[a]
BMI, median (interquartile range)	24.1 (5.6)	22.9 (4.6)	24.6 (4.4)	0.007[a]
Education level, N (%)				
≤ 12 years	55.0 (41.7)	22.0 (40.0)	33.0 (60.0)	ns[b]
> 12 years	77.0 (58.3)	40.0 (51.9)	37.0 (48.1)	
Marital status, N (%)				
Single	81.0 (53.3)	30.0 (37.0)	51.0 (63.0)	0.034[b]
Married/consensual union	53.0 (34.9)	33.0 (62.3)	20.0 (37.7)	
Divorced	13.0 (8.6)	7.0 (53.8)	6.0 (46.2)	
Widowed	5.0 (3.3)	3.0 (60.0)	2.0 (40.0)	
Employment status, N (%)				
Student	44.0 (27.2)	16.0 (36.4)	28.0 (63.6)	ns[b]
Employed full-time/part-time	98.0 (60.5)	51.0 (52.0)	47.0 (48.0)	
Retired/unemployed	20.0 (12.3)	10.0 (50.0)	10.0 (50.0)	
School failure (students), N (%)	13.0 (29.5)	7.0 (53.8)	6.0 (46.2)	< 0.001[b]
Absenteeism (workers), N (%)	5.0 (5.2)	4.0 (91.8)	1.0 (8.2)	0.001[b]
Familial history of DSWPD, N (%)	30.0 (19.4)	14.0 (19.4)	16.0 (19.3)	ns[b]
Accidents, N (%)	9.0 (5.6)	1.0 (11.1)	8.0 (88.9)	0.036[c]
Trauma, N (%)	22.0 (13.6)	13.0 (59.1)	9.0 (40.9)	ns[b]
Familial conflicts, N (%)	55.0 (34.0)	32.0 (58.2)	23.0 (41.8)	0.052[b]
Skip major meals, N (%)	85.0 (58.2)	34.0 (40.0)	51.0 (60.0)	ns[b]
Alcohol abuse, N (%)	25.0 (16.0)	6.0 (24.0)	19.0 (76.0)	0.010[b]
Smoking consumption, N (%)	31.0 (21.4)	11.0 (35.5)	20.0 (64.5)	ns[b]
Drug consumption, N (%)	17.0 (10.7)	4.0 (23.5)	13.0 (76.5)	0.043[c]
Anti-depressive drugs consumption, N (%)	70.0 (49.6)	38.0 (54.3)	32.0 (45.7)	ns[b]
Hypnotic drugs consumption, N (%)	74.0 (52.5)	34.0 (45.9)	40.0 (54.1)	ns[b]
Melatonin consumption, N (%)	58.0 (41.4)	26.0 (44.8)	32.0 (55.2)	ns[b]
Comorbidities (self-reported), N (%)				
Depression	69.0 (42.6)	37.0 (53.6)	32.0 (46.4)	ns[b]
Anxiety	96.0 (59.3)	49.0 (51.0)	47.0 (49.0)	ns[b]
Psychiatric disease	81.0 (50.0)	44.0 (54.3)	37.0 (45.7)	ns[b]
Neurologic disease	24.0 (14.9)	15.0 (62.5)	9.0 (37.5)	ns[b]
Medical disease	37.0 (23.3)	14.0 (37.8)	23.0 (62.2)	ns[b]
Insomnia (complaint)	85.0 (54.5)	35.0 (41.2)	50.0 (58.8)	ns[b]
Sleep disease (confirmed by Polysomnography), N (%)				
Obstructive sleep apnea	25.0 (16.0)	11.0 (44.0)	14.0 (56.0)	ns[b]
Periodic limb movement	19.0 (12.4)	8.0 (42.1)	11.0 (57.9)	ns[b]
Daytime Sleepiness (Epworth Sleepiness Scale), N (%)				
W daytime sleepiness (ESS > 9)	46.0 (41.1)	21.0 (38.2)	25.0 (43.9)	ns[b]
Sleep quality (Pittsburgh Sleep Quality Index), N (%)				
"Good" (PSQI < 5)	5.0 (5.3)	1.0 (2.2)	4.0 (5.3)	ns[b]
"Poor" (PSQI ≥ 5)	90.0 (94.7)	45.0 (97.8)	45.0 (94.7)	

ns=not significant; [a]Mann-Whitney test; [b]Qui² test; [c]Fisher exact test.

Questionnaires

Forty-six (41%) patients had excessive sleepiness, with ESS ≥ 10, and 5 (5%) had poor sleep quality according PSQI total score ≥ 5.

Actigraphy and Diary

According to diary data, all individuals felt asleep after 02:00h. 13 individuals (8%) had sleep onset after 06:00h, sleeping exclusively during daytime. Get up times were, consequently, very late, ranging between 06:00h to 17:00h; the sleep latency also presented a large variability, ranging between 0 to 360 minutes. The mean reported total sleep time was around 420 minutes, although also with a wide variability, ranging between 120 and 720 minutes.

From the individuals that skip meals, the majority (35) usually skip breakfast (21%), and 4 (2%) skip breakfirst and lunch; 9 (5%) skip lunch, and in these cases the breakfast was arround to 12:00h.

According to actigraphy data, sleep onset time ranged between 00:09h to 08:10h; get up ranged from 5:55h to 18:08h; sleep latency ranged from 1 to 191 minutes; sleep efficiency ranged from 25% to 95%; and total sleep time ranged from 153 to 719 minutes (Table 2).

DLMO

DLMO could be calculated in 106 individuals. Of the 16 people in whom it could not be calculated, 8 were because all values had very low concentrations (< 0.2 pg/ml) and the other 8 had very high values (> 20 pg/ml) at the first data collection point. DLMO ranged from 21:02h to 06:30h. The phase angle between DLMO and scheduled sleep time was calculated by subtracting DLMO time from the participants' mean sleep onset time, and ranged between -3:30h (DLMO after bedtime) and 9:06h (DLMO before bedtime).

Polysomnography

The mean "lights off" time was 01:50 ± 01:19h, ranging from 22:58h to 04:51h; total sleep time ranged from 77 to 639 minutes; the mean time in bed was 440 ± 90 minutes, ranging from 275 to 707 minutes. The values for sleep onset and get up times ranged from 23:30h to 06:36h and from 07:42h to 12:53h, respectively. The sleep latency ranged from 0 to 374 minutes, and sleep efficiency ranged from 16.7 to 97.4%. The median percentage value for N1 was 8 (7.2)%, ranging from 1 to 29% and the mean percentage values for N2, N3 and REM were: N1; N2 53 ± 11% ranging from 26 to 78%; N3 21 ± 10% ranging from 0 to 46%; and REM 18 ± 7% ranging from 5 to 44%. The median Apnea Hypopnea Index (AHI) was 0.2 (1.3), ranging from 0 to 15.7 per hour, and the median value for the minimum oxygen saturation was 91 (6) % ranging from 54 to 97%. The median periodic limb movement was 1.6 (5.8) per hour, ranging from 0 to 48 PLM per hour (Table 2).

Gender differences

Among the analysed age classes, DSWPD was more prevalent in men ages 21-30 years and in women in the ages 41-50 years (p=0.028). More men were single and more women

Table 2. Baseline characteristics of objective and subjective measures by sex.

	Total sample	Female	Male	p-value[a]
Diary, median (interquartile range)				
Get up time (hh:mm)	10:15 (03:45)	10:37 (04:00)	10:00 (3:17)	ns
Sleep onset time (hh:mm)	03:00 (01:33)	03:00 (01:45)	03:00 (1:37)	ns
Total sleep time (min.)[#]	424.0 (119.0)	417.0 (129.0)	431.0 (110.0)	ns
Sleep latency (min.)	30.0 (120.0)	52.0 (120.0)	30.0 (120.0)	ns
Sleep efficiency (%)	91.7 (23.5)	91.7 (25.9)	92.0 (22.4)	ns
Actigraphy, median (interquartile range)				
Get up time (hh:mm)	11:36 (02:16)	11:18 (02:10)	11:44 (02:17)	ns
Sleep onset time (hh:mm)	02:58 (02:09)	02:40 (02:04)	03:25 (02:12)	ns
Total sleep time (min.)[#]	407.8 (100.7)	420.2 (110.7)	395.9 (89.7)	ns
Sleep latency (min.)	16.4 (23.0)	15.6 (20.1)	19.3 (26.0)	ns
Sleep efficiency (%)	81.7 (15.3)	82.0 (12.8)	81.5 (15.7)	ns
DLMO, mean (SD)				
Time (hh:mm)	01:22 (02:05)	01:18 (02:08)	01:27 (02:04)	ns
Phase angle (hh:mm)	01:45 (02:10)	01:47 (01:59)	01:43 (02:19)	ns
PSG, median (interquartile range)				
Get up time (hh:mm)	08:38 (01:32)	08:30 (01:00)	08:55 (02:14)	ns
Sleep onset time (hh:mm)[#]	02:31 (01:34)	02:09 (01:23)	02:48 (01:39)	ns
Total sleep time (min.)[#]	331.9 (97.6)	311.0 (135.0)	331.0 (134.0)	ns
Sleep latency (min.)	14.0 (37.4)	8.9 (24.1)	16.0 (76.0)	ns
Sleep efficiency (%)	82.1 (23.2)	82.8 (22.9)	80.6 (24.6)	ns

[#]Mean (Standard Deviation); [a]t- Student or Mann-Whitney; ns=not significant.

were married (p=0.034). In students, school failure (see section 2.2) was higher for women than men (p<0.001), and in workers, absenteeism was higher for women then men (p=0.001). Neither employment status or family history of DSWPD differed between genders. Having accidents was significantly higher in men (89%; p=0.036), while having family conflicts was higher in women (58%; p=0.052). The consumption of alcohol (76%) and ilicit drugs (77%) were significantly higher among men, p=0.010 and p=0.043 respectively. The consumption of tobacco, antidepressive and hypnotic drugs as well as melatonin was similar among genders. Among the self and non-self prescribed drugs or melatonin consumption, no sex differences were found. For the reported co-morbidities and sleep medicine pathologies confirmed by PSG, no differences were found between genders.

There were no gender difference in the diary and actigraphy metrics, PSG parameters, DLMO time, or phase angle.

DSWPD phenotypes

We combined the two misaligned groups (phase-angle > 3 hours or DLMO after sleep onset time) (Figure 1), since the only difference that exists between groups for all the analysed variables (clinical, demographical, etc) was for DLMO time (p<0.001), being the group with later bedtime the group with DLMO after sleep time with a mean time of 03:52±1:25h, ranging from 01:34h to 06:19h . For the two groups aligned and misaligned, the DLMO time was significantly earlier for the misaligned group (Table 3). The only clinical variable that was statistically different between groups was traumatic events, being more prevalent in the misaligned group. The misaligned group had earlier sleep onset time (p=0.046), longer total sleep time (p=0.035) and more sleep cycles(p=0.018) (Table 4).

DISCUSSION

Late types are common in Portugal and this is well represented by the dimension of this cohort, since it is composed by patients from a private sleep clinic/center from a small south European country, reporting from 5 years of appointments. In 2002, a cross-sectional survey was conducted in 10 different countries[22]; Portugal and Spain were the countries with later sleep timing, although respondents from Portugal reported also to have more "delayed sleep induction" and the reported sleep timing was the same for both countries (midnight) even though Portugal follows Greenwich time and Spain follows European Central Time, which is one hour earlier. A populational study, preferently complemented with objective sleep measures, is needed so that it can be possible to understand if Portugal is indead a country of late sleep habits.

DSWPD diagnosis has been previously documented to be an average of approximately 19 year following onset in adolescence, and chronic DSWPD patients may experience symptoms beyond 60 years of age[2]. In our sample, we have individuals reaching 92 years of age and with late reported ages of disease onset. One explanation might be the presence of trigger events, since individuals reporting trigger events for their disease onset present with a higher age of disease onset, another possible explanation is a change in habits after retirement which could mask or impact upon the tendency to earlier chronotypes in the elderly. While DSWPD is described as being more prevalent in young adults[1], in our cohort this was only true for men. However, since we have more women in the age group of 41-50 years, this might be related to social/cultural features, since active women tend to stay up at late night taking care of the house affairs, which in Portugal is still a work performed mainly by women, although more men have begun to participate in parenting[33]. In an epidemiological study performed in Norway with adolescents, DSWPD was more prevalent in women, in our clinical sample we had only slightly higher proportion of men[34]. This gender difference was also observed in a Australian study[3]. Gender differences were particularly relevant to traffic accidents, alcohol abuse and ilicit drug consumption. Consumptions in the general population are present in both genders although alcohol consumption[35] is still more prevalent among men as well as drug consumption[36]. For traffic accidents, the risk also is significantly higher among men[37]. So the gender differences in accidents and consumptions are likely not be related to DSWPD. In this study, DSWPD is more prevalent in young men and in midle age women, and that there are no relevant overall differences between genders.

Of note in this clinical sample was the fact that many of patients that came to the clinic were already taking many prescribed and non-prescribed drugs like hypnotics, anti-depressive medications, and melatonin. The hopelessness of being able to initiate sleep at the social conventional times, may lead individuals to use medications to initiate sleep (personal communication), this is something that should be further investigated. An alternative mechanism might be the relative lack of knowledge among general practitioners about DSWPD pathology and the similarities of DSWPD with insomnia complaints. Better knowledge dissemination of the disease is needed among physicians.

Two different phenotypes of DSWPD were also present in this clinical population like in other studies[3,14]: one in circadian alignment with their internal biological time and another misaligned. The proportion between them was similar to other studies of DSWPD, supporting the evidence that there are two different types of DSWPD patients. Some caution is however required because definitions of DSWPD may differ between studies. For example,in Murray et al.[3] participants were classified into phenotype groups based on the relationship between DLMO and desired bedtime, and in our study participants were classified based in the relationship between DLMO and sleep diary time.

In this group of DSWPD patients, half of the sample reported a psychiatric disease, mainly depression and anxiety. There was no difference between genders for depression and DSWPD. Depression is also present in other DSWPD populations[3,13], although in our sample, depression is less prevalent and is not significantly associated with patients that were in their aligned internal aligned phase, like in the Murray et al study[3]. In our cohort the circadian misaligned group also reported depression. These data suggest that depressive symptoms might be

Table 3. Baseline characteristics by DSWPD phenotype (circadian misaligned and circadian aligned).

	Circadian misaligned	Circadian aligned	*p*-value
N (%)	43.0 (40.6)	63.0 (59.4)	
Age, median (interquartile range)	35.0 (26.0)	36.0 (21.0)	ns[a]
Age groups, N (%)			
16-20 years	3.0 (23.1)	10.0 (76.9)	ns[b]
21-30 years	14.0 (50.0)	14.0 (50.0)	
31-40 years	7.0 (29.2)	17.0 (70.8)	
41-50 years	7.0 (38.9)	11.0 (61.1)	
51-60 years	6.0 (66.7)	3.0 (33.3)	
≥ 65 years	6.0 (42.9)	8.0 (57.1)	
Female / Male, N (%)	20.0 (39.2) / 23.0 (41.8)	31.0 (60.8) / 32.0 (58.2)	ns[b]
Age of disease onset, median (interquartile range)	16.0 (20.0)	16.0 (13.0)	ns[a]
BMI, median (interquartile range)	23.9 (3.4)	24.0 (4.6)	ns[a]
Education level, N (%)			
≤ 12 years / > 12 years	10.0 (27.8) / 21.0 (43.8)	26.0 (72.2) / 27.0 (56.3)	ns[b]
Marital status, N (%)			
Single	22.0 (41.5)	31.0 (58.5)	ns[b]
Married/consensual union	15.0 (39.5)	23.0 (60.5)	
Divorced	3.0 (42.9)	4.0 (57.1)	
Widowed	0.0 (0.0)	2.0 (100.0)	
Employment status, N (%)			
Student	7.0 (24.1)	22.0 (75.9)	ns[b]
Employed full-time/part-time	31.0 (47.0)	35.0 (53.0)	
Retired/unemployed	5.0 (45.5)	6.0 (54.5)	
Scholar failure (students), N (%)	2.0 (28.6)	5.0 (71.4)	ns[c]
Absenteeism (workers) , N (%)	1.0 (25.0)	3.0 (75.0)	ns[c]
Familial history of DSWPD, N (%)	8.0 (38.1)	13.0 (61.9)	N[s]
Traffic accidents, N (%)	3.0 (37.5)	5.0 (62.5)	ns[c]
Traumatic events, N (%)	10.0 (66.7)	5.0 (33.3)	0.026[c]
Familial conflicts, N (%)	13.0 (35.1)	24.0 (64.9)	ns[b]
Jump major meals, N (%)	19.0 (34.5)	36.0 (65.5)	ns[b]
Alcohol abuse, N (%)	5.0 (41.7)	7.0 (58.3)	ns[b]
Smoking consumption, N (%)	5.0 (33.3)	10.0 (66.7)	ns[b]
Drug consumption, N (%)	2.0 (25.0)	6.0 (75.0)	ns[b]
Anti-depressive drugs consumption, N (%)	19.0 (42.2)	26.0 (57.8)	ns[b]
Hypnotic drugs consumption, N (%)	21.0 (43.8)	27.0 (56.3)	ns[b]
Melatonin consumption, N (%)	17.0 (35.4)	31.0 (64.6)	ns[b]
Comorbidities (self-reported), N (%)			
Depression	15.0 (36.6)	26.0 (63.4)	ns[b]
Anxiety	22.0 (36.7)	38.0 (63.3)	ns[b]
Psychiatric disease	18.0 (34.0)	35.0 (66.0)	ns[b]
Neurologic disease	7.0 (38.9)	11.0 (61.1)	ns[b]
Medical disease	7.0 (35.0)	13.0 (65.0)	ns[b]
Insomnia (complaint)	20 (39.2)	31.0 (60.8)	ns[b]
Sleep disease (confirmed by Polysomnography), N (%)			
Obstructive sleep apnea	5.0 (33.3)	10.0 (66.7)	ns[b]
Periodic limb movement	6.0 (31.6)	13.0 (68.4)	ns[b]
Daytime Sleepiness (Epworth Sleepiness Scale), N (%)			
W/ daytime sleepiness (ESS > 9)	15.0 (50.0)	15.0 (50.0)	ns[b]
Sleep quality (Pittsburgh Sleep Quality Index), N (%)			
"Good" / "Poor"	0.0 (0.0) / 22.0 (40.7)	3.0 (100.0) / 32.0 (59.3)	ns[c]
DLMO time, median (interquartile range)	23:41 (3:57)	01:45 (2:16)	<0.001[a]

ns=not significant; [a]Mann-Whitney test; [b]Qui² test, [c]Fisher exact test.

Table 4. Polysomnographic data by DSWPD phenotype (circadian misaligned and circadian aligned).

	Circadian misaligned	Circadian aligned	*p*-value[a]
N	23.0	23.0	
Sleep stages, median (interquartile range))			
Sleep onset (hours) #	02:16 (1:15)	03:06 (1:30)	0.046
Sleep offset (hours)	08:30 (1:33)	8:39 (1:05)	ns
Total sleep time (min.) #	350.9 (91.8)	296.4 (77.1)	0.035
Sleep latency (min.)	8.8 (21.5)	14.0 (76.5)	ns
REM latency (min.)	114.0 (71.8)	118.3 (82.4)	ns
Sleep efficiency (%)	87.0 (13.8)	72.9 (27.7)	0.020
% Stage 1	7.4 (3.6)	8.3 (8.1)	ns
% Stage 2 #	55.6 (8.2)	53.1 (12.8)	ns
% Stage 3 #	20.8 (10.1)	22.9 (9.5)	ns
Stage REM	15.2 (7.3)	15.4 (7.6)	ns
AHI (h^{-1})	0.2 (0.8)	0.2 (1.6)	ns
N. of sleep cycles	3.0 (2.0)	3.0 (1.0)	0.022
More Stage 3 sleep in the 1st half of the sleep period, N (%)			
Yes	20.0 (47.6)	22.0 (52.4)	ns[b]
No	3.0 (75.0)	1.0 (25.0)	
More Stage REM sleep in the 2nd half of the sleep period, N (%)			
Yes N (%)	21.0 (52.5)	19.0 (47.5)	ns[c]
No N (%)	1.0 (50.0)	1.0 (50.0)	
Absence of REM	1.0 (25.0)	3.0 (75.0)	
Last cycle ends in…, N (%):			
NREM	10.0 (43.5)	13.0 (56.5)	ns[c]
REM	13.0 (56.5)	10.0 (43.5)	

#Mean (Standard Deviation); [a]t- Student or Mann-Whitney test, according to data distribution; ns: not significant.
[b]Fisher exact test; [c]Qui² test

more linked to the timing of light exposure like reported in the literature, that the biological lateness by itself, since depressive symptoms are present in both groups, in late circadian alignment or misaligned (early DLMO/biological phase)[21,38,39].

Traumatic events like the death of relatives or sexual abuse, are reported as precipitating factors for DSWPD[1], this was particularly relevant in the misaligned group, suggesting that in the etiology of this group, the behavioural component might be stronger than the circadian component; another possibility is a noxa of traumatic event upon the circadian system.

Our PSG results were in accordance to previous studies where the sleep macrostructure is preserved[14,40,41]. However, in the circadian-aligned group there were some individuals that had absence of REM sleep; this could be related to their short sleep duration. In the misaligned group, 3 individuals had an inversion in their sleep macroestructure, having more Stage 3 sleep in the second half of the sleep period, this suggests that this group potencially presents a higher sleep macrostructure disorganization with disturbance in the homeostatic drive. Further studies need to be performed concerning PSG for the two groups.

There are some limitations to our study. First, the fact diary, actigraphy, and saliva collection for DLMO measurements were performed at home and relied on patients accuracy, especially for melatonin measurement. The DLMO timing that were not possible to calculate might be related to problems with collection procedures. Second, since 88% of the patients are workers or students, many of them asked to be awakened also in the morning of the PSG measurement in order to comply with their working commitments, shortening their sleep period. Others might have been awakened during the morning by the fact that the sleep clinic starts to receive patients for appoinments around 8:00h and they might have perceived some movement in the clinic awakening them. Objective measurements collection were performed when patients were having their usual daily routines. Third, two different actimetry devices were used'; this is a limitation sometimes present when analyzing retrospective data. Therefore, to minimize discrepancy errors, sleep bouts data were confirmed with each individuals diary data. Fourth, healthy volunteeers were not included as a comparison group, because the primary aims of the study was to characterize a clinical sample of DSWPD and to assess melatonin rhythm phase comparing clinical characteristics of DSWPD patients with and without circadian misalignment. Finally, this study was conducted only with individuals from Portugal, which has a societal customs that promote activities late in the day/night. Future work should be conducted in other developed and in underdeveloped with different societal customs/norms about timing of social activities.

More experimental prospective epidemiological studies are needed to asess an accurate prevalence value for DSWPD, and to closely analyse the differences between the two circadian types of individuals diagnosed with DSWPD. There are different types of DSWPD; one type has a delayed biological night relative to their sleep timing (i.e., circadian misaligned), while others have a "normal" biological night (DLMO) aligned with the light/dark cycle although with a late sleep time (i.e., circadian aligned), suggesting that these patients might have a problem in the homeostatic process (Process S) and not in the circadian process (Process C)[42] as suggested by Micic et al[2]. Depression is prevalent in both groups. In this clinical population, DSWPD is more prevalent in young men and in middle age woman, although with no overall significant differences between genders. It is still imperative to achieve a better definition, clarification, classification and diagnostic criteria of these patients. Differential therapeutical intervention should also be considered and tested for both phenotypes.

ACKNOWLEDGEMENTS

The authors would like to thank Dr. Elizabeth Klerman for the text suggestions; CENC - Sleep Medicine Center for the clinical database; and Condor instruments for providing actimeters. This work was supported by Fundação para a Ciência e Tecnologia (FCT) PhD research grant PDE/BDE/114584/2016.

REFERENCES

1. Sateia M. International Classification of Sleep Disorders. 2014. doi:10.1007/s13311-012-0145-6.
2. Micic G, Lovato N, Gradisar M, Ferguson SA, Burgess HJ, Lack LC. The etiology of delayed sleep phase disorder. Sleep Med Rev 2016;27:29–38. doi:10.1016/j.smrv.2015.06.004.
3. Murray J, Sletten T, Michelle M, Christopher G, Lovato N, D DJB, et al. Prevalence of Circadian Misalignment and Its Association with Depressive Symptoms in Delayed Sleep Phase Disorder. Sleep 2017;114:1–35. doi:10.1093/sleep/zsw002.
4. Schrader H, Bovim G, Sand T. The prevalence of delayed and advanced sleep phase syndromes. J Sleep Res 1993;2:51–5. doi:10.1111/j.1365-2869.1993.tb00061.x.
5. Paine S, Fink J, Gander PH, Warman GR. Identifying advanced and delayed sleep phase disorders in the general population : A national survey of New Zealand adults. Chronobiol Int 2014;31:627–36. doi:10.3109/07420528.2014.885036.
6. Carskadon MA. Sleep in adolescents: The perfect storm. Pediatr Clin North Am 2011;58:637–47. doi:10.1016/j.pcl.2011.03.003.Sleep.
7. Crowley SJ, Acebo C, Carskadon MA. Sleep, circadian rhythms, and delayed phase in adolescence. Sleep Med 2007;8:602–12. doi:10.1016/j.sleep.2006.12.002.
8. Roenneberg T, Kuehnle T, Pramstaller PP, Ricken J, Havel M, Guth A, et al. A marker for the end of adolescence. Curr Biol 2004;14:1038–9. doi:10.1016/j.cub.2004.11.039.
9. Roenneberg T, Kuehnle T, Juda M, Kantermann T, Allebrandt K, Gordijn M, et al. Epidemiology of the human circadian clock. Sleep Med Rev 2007;11:429–38. doi:10.1016/j.smrv.2007.07.005.
10. Auger RR, Burgess HJ, Emens JS, Deriy L V, Thomas SM, Sharkey KM. Clinical Practice Guideline for the Treatment of Intrinsic Circadian Rhythm Sleep-Wake Disorders. J Clin Sleep Med 2015;11:1199–236. doi:10.5664/jcsm.5100.
11. Digdon NL, Howell AJ. College students who have an eveningness preference report lower self-control and greater procrastination. Chronobiol Int 2008;25:1029–46. doi:10.1080/07420520802553671.
12. Díaz-Morales JF, Ferrari JR, Cohen JR. Indecision and avoidant procrastination: The role of morningness- eveningness and time perspective in chronic delay lifestyles. J Gen Psychol 2008;135:228–40. doi:10.3200/GENP.135.3.228-240.
13. Abe T, Inoue Y, Komada Y, Nakamura M, Asaoka S, Kanno M, et al. Relation between morningness-eveningness score and depressive symptoms among patients with delayed sleep phase syndrome. Sleep Med 2011;12:680–4. doi:10.1016/j.sleep.2010.12.017.
14. Rahman SA, Kayumov L, Tchmoutina EA, Shapiro CM. Clinical efficacy of dim light melatonin onset testing in diagnosing delayed sleep phase syndrome. Sleep Med 2009;10:549–55. doi:10.1016/j.sleep.2008.03.020.
15. Benloucif S, Burgess HJ, Klerman EB, Lewy AJ, Middleton B, Murphy PJ, et al. Measuring melatonin in humans. J Clin Sleep Med 2008;4:66–9.
16. Klerman EB, Gershengorn HB, Duffy JF, Kronauer RE. Comparisons of the variability of three markers of the human circadian pacemaker. JBiolRhythms 2002;17:181–93.
17. Sletten TL, Vincenzi S, Redman JR, Lockley SW, Rajaratnam SMW. Timing of Sleep and Its Relationship with the Endogenous Melatonin Rhythm. Front Neurol 2010;1:1–8. doi:10.3389/fneur.2010.00137.
18. Stothard ER, McHill AW, Depner CM, Birks BR, Moehlman TM, Ritchie HK, et al. Circadian Entrainment to the Natural Light-Dark Cycle across Seasons and the Weekend. Curr Biol 2017;27. doi:10.1016/j.cub.2016.12.041.
19. Vartanian G V, Li BY, Chervenak AP, Walch OJ, Ala-laurila P, Wong KY, et al. Melatonin suppression by light in humans is more sensitive than previously reported. J Biol Rhythms 2015;30:2016. doi:10.1177/0748730415585413.Melatonin.
20. Aoki H, Ozeki Y, Yamada N. Hypersensitivity of melatonin suppression in response to light in patients with delayed sleep phase syndrome. Chronobiol Int 2001;18:263–71. doi:10.1081/CBI-100103190.
21. Joo EY, Abbott SM, Reid KJ, Wu D, Kang J, Wilson J, et al. Timing of light exposure and activity in adults with delayed sleep-wake phase disorder. Sleep Med 2017;32:259–65. doi:10.1016/j.sleep.2016.09.009.
22. Soldatos CR, Allaert FA, Ohta T, Dikeos DG. How do individuals sleep around the world? Results from a single-day survey in ten countries. Sleep Med 2005;6:5–13. doi:10.1016/j.sleep.2004.10.006.
23. Gonçalves M, Paiva T, Maltês J, Ramos E. Epidemiology of insomnia in Portugal. J Sleep Res 2010;S254:74. doi:10.1111/j.1365-2869.2008.00690.x.
24. Bertolazi A, Fagondes S. Portuguese-language version of the Epworth sleepiness scale: validation for use in Brazil. J Bras … 2009;35:877–83.
25. Del Rio João KA, Becker NB, de Neves Jesus S, Isabel Santos Martins R. Validation of the Portuguese version of the Pittsburgh Sleep Quality Index (PSQI-PT). Psychiatry Res 2017;247:225–9. doi:10.1016/j.psychres.2016.11.042.
26. Chow CM, Wong SN, Shin M, Maddox RG, Feilds K, Paxton K. Defining the rest interval associated with the main sleep period in actigraph scoring 2016:321–8. doi:10.2147/NSS.S114969.
27. Cole RJ, Kripke DF, Mullaney DJ, Gillin JC. Technical Note Automatic Sleep / Wake Identification From Wrist Activity 1992;15:461–9.
28. Keijzer H, Smits MG, Peeters T, Looman CWN, Endenburg SC, Gunnewiek JMTK. Evaluation of salivary melatonin measurements for Dim Light Melatonin Onset calculations in patients with possible sleep-wake rhythm disorders. Clin Chim Acta 2011;412:1616–20. doi:10.1016/j.cca.2011.05.014.
29. Baron KG, Reid KJ, Kim TW, Van Horn L, Attarian H, Wolf L, et al. Circadian timing and alignment in healthy adults: associations with BMI, body fat, caloric intake and physical activity. Int J Obes 2017;44:319–35. doi:10.1111/cdoe.12200.Factors.
30. Berry RB, Brooks R, Gamaldo CE, Harding SM, Marcus CL, Vaughn B V. The AASM Manual for the Scoring of Sleep and Associated Events. Am Acad Sleep Med 2013;53:1689–99. doi:10.1017/CBO9781107415324.004.
31. Rechtschaffen, A , Kales A. A manual of standardized terminology, techniques and scoring system of sleep stages in human subjects. Los Angeles: 1968.
32. American Psychiatric Association. Diagnostic and statistical manual of mental disorders (5th ed.). Washington, DC: 2013. doi:doi.org/10.1176.
33. Monteiro L, Fernandes M, Torres N, Santos C. Father's involvement and parenting styles in Portuguese families: The role of education and working hours. Análise Psicológica 2017;4:513–28. doi:10.14417/ap.1451.
34. Sivertsen B, Pallesen S, Stormark KM, Bøe T, Lundervold AJ, Hysing M. Delayed sleep phase syndrome in adolescents: Prevalence and correlates in a large population based study. BMC Public Health 2013;13. doi:10.1186/1471-2458-13-1163.
35. Ceylan-isik AF, Mcbride SM, Ren J. Sex Difference in Alcoholism: Who is at a Greater Risk for. Life Sci 2010;87:133–8. doi:10.1016/j.lfs.2010.06.002.Sex.
36. Becker, Jill B, Hu M. Sex differences in drug abuse. Front Neuroendocrinol 2008;29:36–47. doi:10.1016/j.yfrne.2007.07.003.

37. Al-Balbissi AH. Role of gender in road accidents. Traffic Inj Prev 2003;4:64–73. doi:10.1080/15389580309857.
38. Bedrosian TA, Nelson RJ. Timing of light exposure affects mood and brain circuits. Transl Psychiatry 2017;7. doi:10.1038/tp.2016.262.
39. Duffy JF, Czeisler CA. Effect of Light on Human Circadian Physiology. Sleep Med Clin 2009;4:165–77. doi:10.1016/j.jsmc.2009.01.004.
40. Watanabe T, Kajimura N, Kato M, Sekimoto M, Nakajima T, Hori T, et al. Sleep and circadian rhythm disturbances in patients with delayed sleep phase syndrome. Sleep 2003;26:657–61. doi:10.1093/sleep/26.6.657.
41. Solheim B, Langsrud K, Kallestad H, Engstrøm M, Bjorvatn B, Sand T. Sleep structure and awakening threshold in delayed sleep-wake phase disorder patients compared to healthy sleepers. Sleep Med 2018;46:61–8. doi:10.1016/j.sleep.2018.03.001.
42. Borbély AA, Daan S, Wirz-Justice A, Deboer T. The two-process model of sleep regulation: A reappraisal. J Sleep Res 2016;25:131–43. doi:10.1111/jsr.12371.