

## Sleep Spindle Characteristics in a Developmentally Normal Infant Population in the Western Cape of South Africa

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### Abstract

Sleep spindles are oscillating potentials generated in the thalamus. They are linked to memory consolidation during sleep. Their absence in infants beyond 3 months of age indicates cognitive impairment. This study examines 3 to 9-month-old infants to obtain standardized criteria of the density, frequency, and spindle duration for this age group in a South African population. Infants with normal developmental milestones and no evidence of neuro-insults were identified from routine referrals for electroencephalogram (EEG). Patients on continuous medication, any pre- or perinatal insult, or preterm infants were excluded. Retrospective examination of EEGs performed on 55 children was done. Manual scoring of the density, frequency and duration of each individual EEG was obtained and analyzed by a specialist pediatric EEG technologist. A qualified Neurologist independently rated the EEG scoring and a Paediatric Neurologist was the arbitrator. Findings were statistically analyzed utilizing the Redcap and SPSS software. The range of sleep spindle density for all age groups was between 2-6 spindles/minute with a mean of 3.8 spindles/min

(SD 1.4). Mean duration was 2.3 seconds (SD 0.8) and the mean frequency was 12.46Hz (SD 1.1). The average sleep time was 30 minutes. There was no statistically significant difference between the spindle density, duration, and frequency of the infants between 3 and 9 months of age. There was also no difference between the sleep spindle characteristics of the natural sleep group and melatonin sleep-induced infants. The findings of this South African population are comparable to international normal values from American and European Centers.

## Keywords

Sleep spindles; Cognitive impairment; Spindle density; Normal values; Infant spindles; Electroencephalogram

## Introduction

### Sleep spindles

Sleep spindles were first recorded by Hans Berger but were subsequently described by Loomis *et al* in 1935 (De Gennaro and Ferrara: 2003). They were the first features of sleep to be described in human recordings (Loomis et al: 1937). Spindles are oscillating potentials known for their characteristic waxing and waning appearance on the electroencephalogram (EEG). They are the hallmark of stage 2 Non-Rapid Eye Movement (NREM) sleep (Luthi: 2013, Ellingson: 1982). The EEG is a test used to evaluate the electrical activity in the brain. The EEG produces different electrical potentials during different levels of alertness in humans. These waveforms are measured in cycles per second or Hertz (Hz). The waveforms also change during brain maturation and by the age of 3 years most children should have a posterior dominant rhythm of 7-8Hz alpha activity (similar to that in adults). The EEG is traditionally utilized as an aid in the diagnosis of neurological problems such as epilepsy or encephalopathy, but more recently researchers are using EEG as a biomarker for brain function (Ajeena: 2009). Sleep spindles are commonly found in NREM sleep (Luthi: 2013). Its very characteristic oscillating pattern usually signals the initiation of stage 2 NREM sleep. Figure 1 shows centroparietal sleep spindles in a 9-month-old infant.

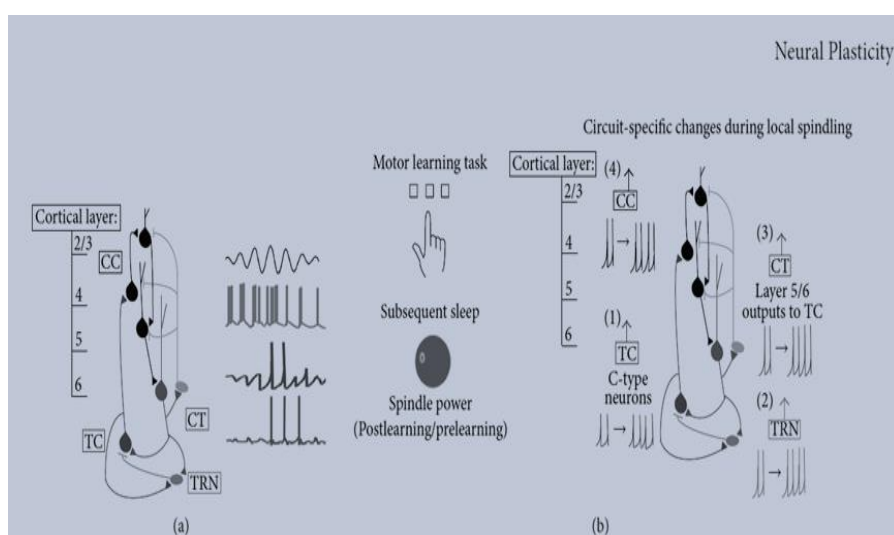


**Figure 1:** Sleep spindles over the centroparietal region, maximal on the left in a 5-month-old infant– Recording from Red Cross War Memorial Children’s Hospital, Cape Town, South Africa. Parental permission attained to include de-identified epoch.

There are 2 types of sleep spindles that can be distinguished: slow spindles (9-11Hz) appear more frontally, whilst faster spindles (>12-16HZ) are mainly found in the central and centroparietal regions of the EEG. The two types are thought to have developed from and represent different generators within the thalamus, with some level of cortical involvement (Gruber and Wise: 2015). The spindle types serve different functions. Slow spindles are thought to be involved in visual perception, whilst faster spindles are related to cognition. Bodiz et al (2005) noted that the rate of sleep spindles can vary between 3 and 8 per minute in different individuals but is very stable across two nights in the same individual. Thus, sleep spindles can potentially be used to identify possible cognitive or perceptual impairments in individuals if compared to established norms. Figure 2 illustrates how sleep spindle generation is a network event (Clawson et al: 2016). GABA neurons in the Thalamic Reticular Nucleus (TRN) play a critical role in the generation of sleep spindles. They are responsible for the highly synchronized bursts at the start of the spindle due to the relative hyperpolarization in the TRN membrane potential.

These TRN bursts in turn lead to hyperpolarization of the neighboring glutaminergic Thalamocortical neurons (TN). This suppresses firing; the membrane depolarizes and leads to a burst of action potential. Corticothalamic neurons (CT) also appears to increase their firing rate and fire asynchronously during the spindle waning phase.

Sleep spindles undergo rapid changes throughout the first two years of life. However, development is most stable during the time period of 3 to 9 months of age, when minimal spindle pattern formation changes occur. During this period spindles reach their maximum in terms of density, duration, frequency, and amplitude, followed by a decrease to minimal values by 27 months and then remaining constant up to 54 months of age. At 3 months post term, only half of the sleep spindles are bilaterally synchronous, and this persists up to 6 months (Kellaway: 1979). By 12 months the spindles are bilaterally synchronous. However, little is known about sleep spindle characteristics in the African population and whether they are similar to those in other regions (Figure 2).



**Figure 2:** CT-TRN-TC network changing during spindling (Clawson et al: 2016).

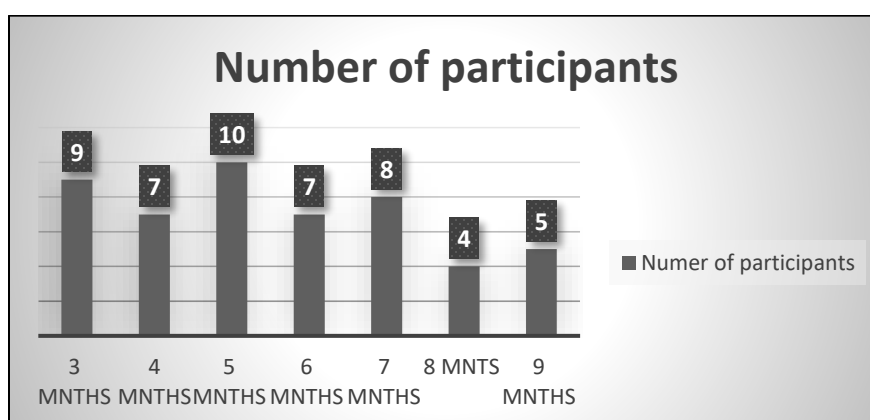
## Melatonin

Melatonin is a hormone that is produced by the pineal gland in response to darkness (Zisapel: 2017). Light inhibits its synthesis; thus plasma melatonin levels are low during the day and high during the night (Zisapel: 2017). It has been observed that the administration of melatonin during the daytime, when it is not normally present endogenously, results in the induction of natural sleep in humans (Zisapel, 2017; Gorfine et al: 2006). However, melatonin is non-sedating. It is thus used as a method of sleep initiation in many EEG laboratories. A study by Wassmer et al (2001) found that administration of melatonin as a sleep initiator does not alter the macro sleep architecture of the patient. Only the sleep onset latency was decreased. Likewise, a study by Ibekwe et al (2017) found that the administration of melatonin had no effect on the EEG recording. The aim of the study was to examine whether there is a difference between the spindle density, duration, and frequency of the South African infants between 3 and 9 months of age and whether these findings were comparable to international normal values from American and European Centers. As a secondary outcome the study also examined whether there was a difference between the sleep spindle characteristics of the natural sleep group and melatonin sleep-induced infants.

## Material and Methods

### Population

A retrospective observational study was done on EEGs of developmentally normal infants with no history of neurological insults. Participants were identified from among routine EEG referrals made to the Red Cross War Memorial Children's Hospital EEG service between January 2018 and July 2023. The reasons for referral included suspected seizures, apneic spells, abnormal movements and suspected infantile epileptic spasms. The inclusion criteria were a normal neurodevelopmental history and examination, and a normal sleep EEG. Exclusion criteria were premature infants, infants on long term medication which could influence the EEG (eg antiseizure medications) and any child with a history of pre- or perinatal birth insults. The sample was drawn from the existing database at Red Cross War Memorial Children's Hospital Neurophysiology Laboratory. Ethical approval was obtained from Red Cross War Memorial Children's Hospital (Approval number: RHX: RCC 350/WC\_202210\_009), and the Human Research Ethics Committee of the University of the Cape Town Department of Pediatrics (HREC REF 486/2022) (Figure 3).



**Figure 3:** Number of participants by age.

## Normative Data

Absence of sleep spindle after the age of 3 months are generally considered abnormal. Typically, spindles tend to have long duration during the first half of year one of life, sometimes up to 10 seconds, whilst the second half shows a steady decrease of duration to approximately 2 seconds (Spinosa and Garson: 2007). Gruber and Wise (2015) state that spindle maturation in terms of density and duration is thought to have a U-shaped distribution. Infants of up to 9 months of age have a relatively low density ( $>3/\text{min}$ ), long duration ( $>1.5 \text{ sec}$ ) and short interspindle interval (20sec). A Swedish review article by Fernandez and Luthi (2020) examined the sleep spindles of 2000 adults and found an intraspindle frequency of  $13.3\text{Hz} \pm 1\text{Hz}$  (mean  $\pm$  SD), duration of  $0.75 \pm 0.25\text{s}$  and densities of  $2.3 \pm 2$  spindles/minute although there were spindle density of up to 10spindles/min recorded.

## Data Collection

EEG recordings were obtained on a 16 channel Nihon Kohden Neurofax machine and included 22 scalp EEG electrodes. The following electrode placements were utilized and placed according to the 10/20 system: Earth(Z), Fp1, Fp2, F8, T4, T6, F4, C4, T4, P4, O2, F7, T3, T5, F3, C3, P3, O1, A1 and A2. Additional 2 lead Electrocardiogram (ECG) and Electromyogram (EMG) electrodes were also placed (see Figure 1). The recording was acquired using a longitudinal bipolar montage. The patients' scalps were prepared with Skinpure skin prep to ensure that all electrode impedances were below the recommended  $5\text{k}\Omega$ . Electrodes were affixed with Elefix EEG paste and secured with a crepe bandage to keep the electrodes in place. Each recording included at least 20 minutes of sleep. Sleep was achieved naturally in 35 (63.6%) participants, whilst synthetic melatonin was administered to 20 (36.4%) infants who did not fall asleep naturally.

## EEG analysis

The sleep spindles were independently reviewed by two specialists (clinical neurophysiology technologist and a neurologist) for sleep spindle duration, frequency, and density according to the standard sleep scoring criteria from the American Academy of Sleep Medicine (Iber et al., 2007). A third paediatric neurology specialist was available to arbitrate any differences.

## Statistical analysis

Descriptive statistics were computed for the frequency, duration, and density of the sleep spindles and reported as means and Standard Deviation (SD) for normally distributed values and medians and IQR for non-normally distributed values.

## Results

### Population Demographics

Of 69 EEGs reviewed, 14 were excluded due to specific and non-specific abnormalities on the EEG. These included focal or generalized epileptiform activity or a slow awake background for age. Fifty-five participants were recruited comprising 27 males and 28 females between the ages of three and nine months. All were born following uncomplicated pregnancies and births. The majority of the participants were of mixed ancestry (57%), followed by 38% of African descent and 5% of European decent. This was reflective of the typical population distribution in the Western Cape of South Africa. Figure 3 shows the

relative age distribution of the participants. Weight for age and associated Z-scores at the time of the EEG recording, were obtained for 50 of the 55 participants (see table 1).

Months	Mean
3	6.53
4	7.2
5	5.41
6	8
7	7.06
8	7.42
9	7.9

**Table 1:** Average weight by age.

Twenty-two percent (n=6) of the boys were -3 SD below the mean weight for age as calculated by the World Health Organization. Eighteen percent (n=5) of the boys were between -1 and -2 SD below the mean for age. Similarly, sixteen percent (n=4) of the girls were measured at -3 SD below the mean. Thirty-seven percent (n=9) of the girls weighed -2SD below the mean. There was no statistical difference in the sleep spindle density, duration and frequency of the infants with normal weight for age and those that were stunted. Table 2 provides a summary of the mean densities, durations, and frequencies across the different age groups.

Age in months	No of Participants	Mean Density	Standard Deviation	References value	Mean Duration	Standard Deviation	References value	Mean Frequency	Standard Deviation	References value
3	11	2.7	1.3	±3	3.4	1.6	>1.5	12.7	1.4	13.3
4	8	4.3	1.6	±3	2.2	0.7	>1.5	12.2	0.8	13.3
5	10	4.5	1.2	±3	2.4	0.9	>1.5	13	1.2	13.3
6	7	4.1	1.9	±3	2.3	1	>1.5	12.4	1.7	13.3
7	8	4	1.3	±3	1.6	0.6	>1.5	12.1	1.6	13.3
8	5	3.6	1.1	±3	1.8	0.4	>1.5	12.6	0.5	13.3
9	6	3.5	1.5	±3	1.7	0.6	>1.5	12.1	0.9	13.3
Total	55	3.8	1.4	±3	2.2	0.8	>1.5	12.4	1.1	13.3

**Table 2:** Mean density, duration and frequency of the different age groups (Reference values- Gruber and Wise: 2015).

Sleep spindle density showed an inverted u-shaped distribution. The 3-month-old sample had a mean density of 2.72 spindles/minute (SD ±1.64), gradually increasing at 4 months to 4.25 spindles/minute (SD ±1.66), at 5 months it increased to 4.50 spindles/minute (SD ± 1.17). Thereafter it showed a gradual decrease from 6 months with a spindle density of 4.14 spindles/minute, at 7 months the sample had a density of 4 spindles/minute, at 8 months 3.6 spindles/minute and at 9 months the mean density was 3.5 spindles/minute. The z score for sleep density was 0.55. The mean spindle duration was 3.43 seconds at the age of 3 months. Between 4-6 months the spindles had a similar mean duration of between 2.18 and

2.34 seconds, likewise with the 7-9-month-old group showed a short spindle duration of between 1.65 and 1.76 seconds ( $SD \pm 0.87s$ ). Z score was calculated at 0.83 and not statistically significant. The frequency was similar across the different age groups with a mean frequency of 12.46Hz ( $SD \pm 1.19$ ). Distribution of the sleep spindles were predominantly central (83%) and parietal (14 %). Two records showed frontal dominant spindling. Sleep spindle density, duration and frequency did not differ between the male and females in the sample. Figure 3 compares the density, duration and frequency of the sample that fell asleep naturally and those that were given melatonin. Mean density for the natural sleep group was 3.65 spindles/minute ( $SD \pm 1.36$ ), while the melatonin sleep induced infants had a mean sleep spindle density of 3.97 ( $SD \pm 1.31$ ). Mean duration for the natural sleep group was 2.65 seconds ( $SD \pm 1.21$ ) and the melatonin group duration was 1.99 ( $SD \pm 0.70$ ). The mean frequency of the natural and melatonin induces sleep groups were 12.6 and 12.4 respectively, with standard deviations of  $\pm 1.32$  and  $\pm 1.19$  (Table 3).

	Natural Sleep	SD	Melatonin	SD
Density	3.65	1.36	3.97	1.31
Duration	2.65	1.21	1.99	0.70
Frequency	12.69	1.32	12.40	1.19

**Table 3:** Mean density, duration and frequency of melatonin induced and natural sleep infants.

## Discussion

In this South African socioeconomically challenged population of developmentally normal infants, no difference in sleep spindle density, duration or frequency was observed among the different age groups. This reflects similar findings to populations from high income settings (Gruber and Wise 2015). The u-shaped distribution in sleep spindle density was similar to the findings by Gruber and Wise (2015). Sleep spindle density was lowest in the 3-month-old group, with a slight insignificant increase in density between 4 to 7 months of age, thereafter density decreased slightly from 8 months onward. However, the decrease in sleep density between the different age groups from 3 to 9 months, was not statistically significant.

More than 27% of South African children are stunted, with the expectation that these children may not reach their full growth and developmental potential due to persistent malnutrition (UNICEF, 2022). Based on these challenges, there was the concern that it may influence brain maturation in the form of impact on the sleep spindles. However, the sample group did not differ from the international cohort. This enabled the researcher to establish normal values of sleep spindle frequency, density and duration for the sample group. Similarly, despite a proportion of the study group being stunted there was no impact on the groups spindle formations. South Africa is unique in that it is a multi-ethnic population and so the study demonstrated similarity in spindle formation across ethnic groups, despite small sample size. There was no difference in frequency, density, or duration between the African, European, and mixed ancestry populations. This supports that sleep spindle density, duration and frequency is similar in most infants between the ages of 3 to 9 months regardless of their location, socioeconomic background, or ancestry. There was also no difference noted in the male and female population, indicating that sleep spindle characteristics are stable irrespective of sex. Sleep spindle density, duration and frequency did not differ significantly in the sample that fell asleep naturally and those that were given melatonin. This supports

the findings by Ibekwe (2017) that administration of melatonin as a mode of sleep induction does not alter the EEG recording.

## Limitations

There are no definitive international normal ranges to compare our findings with. Most of the research into sleep spindle density relates to the adult population. The study by Gruber and Wise (2015), had a smaller number of infants ( $n > 20$ ) than our study. In addition, the study by Gruber and Wise combined densities, frequencies and durations of the spindles into one group for the infants between the ages of 3-9 months. There were therefore no individual results per age category to compare our findings with. Therefore, despite this limitation, our study is unique as it is the largest described sample of sleep spindle formation in infants discriminated into different age groups between 3 and 9 months.

## Conclusion

This study demonstrated that sleep spindle characteristics of developmentally normal infants in a South African cohort were comparable to international recognized values for sleep spindle density and duration in the pediatric population, despite the vulnerabilities of our population group. Mode of sleep induction had no effect on spindle characteristics. The study provides data relevant to Sub-Saharan Africa. Standardization of sleep spindle characteristics in the local population may assist clinicians with early biomarker indicators for at risk children evident via routine EEG. This data forms the first part of a staged project exploring the use of spindle analysis as a viable biomarker for early detection of infants at risk of neurodevelopmental delay.

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## References

1. Genaro LD, Ferrara M. (2003) Sleep spindles: an overview. *Sleep Med Rev* 5(7):423-40.
2. Loomis AL, Harvey EN, Hobart G. (1935) Potential rhythms of the cerebral cortex during sleep. *Science*. 81(2111):597-8.
3. Luthi A. (2013) Sleep Spindles: Where They From, What They Do. *Neuroscientist*. 20(3):243-56.
4. Ellingson RJ. (1982) Development of Sleep Spindle Bursts During the First Year of Life. *Sleep*. 5(1):39-46.
5. Ajeena IM. (2009) *Advances in Clinical Neurophysiology*.
6. Gruber R, Wise MS. (2016) Sleep Spindle Characteristics in Children with Neurodevelopmental Disorders and Their Relation to Cognition. *Neural Plast*.
7. Castro L, Bagherioromi A. (2022) Youth energy to tackle persistent malnutrition. UNICEF.
8. Bodiz R, Kis T, Lazar AS, Havran L, Rigo P, et al. (2005) Prediction of general mental ability based on neural oscillation measures of sleep. *J Sleep Res*. 14(3):285-92.
9. Clawson BC, Durkin J, Aton SJ. (2016) Form and Function of Sleep Spindles across the Lifespan. *Neural Plast*.
10. Kellaway P. (1979) An orderly approach to visual analysis: Parameters of the normal EEG in adults and infants. *Current Practice of Clinical Electroencephalography*.



11. Zisapel S. (2018) New perspectives on the role of melatonin in human sleep, circadian rhythms and their regulation. *Br J Pharmacol.* 175(16):3190-9.
12. Wassmer E, Quinn E, Whitehouse W, Seri S. (2001) Melatonin as a sleep inductor for electroencephalogram recordings in children. *Clinical Trial.* 112(4):683-5.
13. Ibekwe R, Jeavan L, Wilmshurst J. (2017) The role of melatonin to attain electroencephalograms in children in a sub-Saharan African Setting. *Seizure.* 51:87-94.
14. Spinosa MJ, Garzon E. (2007) Sleep Spindles: Validated Concepts and Breakthroughs. *J Epilepsy Clin Neurophysio.* 13(4):179-82.
15. Gruber R, Wise MS, Frenette S, Knauper B, Boom A, et al. (2013) The association between sleep spindles and IQ in healthy school-age children. *Int J Psychophysio.* 89(2):229-40.