

Temporal link between plasma thyrotropin levels and electroencephalographic activity in man

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Abstract

Plasma thyrotropin (TSH) levels have been previously shown to be associated with the internal sleep structure determined by conventional scoring of sleep stages. This temporal relationship was re-evaluated using spectral analysis of the sleep electroencephalogram (EEG). Eight healthy male subjects underwent two randomized night studies after having received either placebo or 5 mg ritanserin, a selective 5-HT₂ receptor antagonist known to increase slow-wave sleep. Delta relative power and TSH levels, determined at 10 min intervals, were found to be inversely related with an average cross-correlation coefficient highly significant ($P < 0.0001$) in both experimental conditions. Alpha slow-wave index, an estimator of awakenings, and TSH pulses exhibited a significant temporal association in both conditions. These results demonstrate that TSH fluctuations are linked to the sleep EEG activity in man.

Keywords: Sleep; Electroencephalogram; Spectral analysis; Delta relative power; Alpha slow-wave index; Thyrotropin

Plasma thyrotropin (TSH) levels exhibit a 24h rhythm generated by amplitude and frequency modulation of secretory pulses [19]. TSH displays low daytime values which begin to increase in the late afternoon to reach maximal levels around the time of sleep onset. Subsequently, a slow decline generally attributed to an inhibitory influence of sleep processes occurs during the night [3,13]. In contrast, sleep withdrawal augments the nightly TSH secretion [3]. Despite these sleep-related influences, the nocturnal TSH profile was generally considered to be independent of the sleep structure. However, recent results reported that a temporal association exists between slow wave sleep (SWS) and declining plasma TSH levels [8]. Thus SWS may be a determining factor of the sleep-associated TSH decrease subsequent to sleep onset. This is in accordance with the observation of a lower nocturnal TSH rise when the amount of SWS is increased by prior sleep deprivation [3].

In previous studies the sleep structure was assessed by conventional scoring of sleep stages [14]. Spectral analy-

sis of the sleep electroencephalogram (EEG) by means of Fast Fourier Transformation (FFT), which allows a more detailed and dynamic description of the sleep process and a quantitative analysis of the sleep EEG [1,2], was used in this study. On this basis, a refined analysis of the association between the nocturnal plasma TSH profile and the EEG bands, especially in the δ frequency range, was performed. This procedure should demonstrate whether there is a correlation between the pulsatile TSH profile and sleep deepening and lightening. In order to increase slow wave activity during sleep, ritanserin, a selective 5-HT₂ receptor antagonist [10,15] was given in one experimental condition. The α slow-wave index (ASI) was used [11] to test the previously described association between awakenings and increasing TSH levels [8].

Eight healthy male subjects aged between 19–27 years and with a mean body mass index of 22.3 ± 0.7 (mean \pm SEM) were selected after medical examination. All had regular sleep-wake habits and none took medication in the 2 weeks before and during the study. The experiments were carried out in a sound-proof air-conditioned sleep apartment. In a double blind design, they received orally either placebo or a single dose of ritanserin (5 mg). Pla-

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cebo and ritanserin were given at 0900 h because a morning dose elicits the most pronounced increase in SWS during the subsequent night [6]. Lights were switched off at 2300 h and the subjects were awakened at 0700 h. Sleep recordings were performed using two EEG derivations (C3 or C4 versus A2 or A1 and Cz versus O1 or O2), one chin electromyographic derivation and one horizontal electro-oculographic derivation. For all-night spectral analysis, the EEG signal was converted from analogue to digital with a sampling frequency of 128 Hz. Subsequently, spectra were computed for consecutive 2-s periods using an FFT-algorithm [5]. In addition, the values for 15 consecutive 2-s periods were averaged to yield power density values for 30-s periods. The spectral parameters studied were relative power (% of the global EEG band (0.5–35 Hz)) for δ (0.5–3.5 Hz), θ (4–7.5 Hz), and α (8–12.5 Hz). In addition, the ASI was analyzed ($ASI = \alpha / (\delta + \theta)$) [12]. For blood sampling, a catheter was inserted into an antecubital vein at 1800 h. Blood was collected continuously using a peristaltic pump in an adjoining room at 10-min intervals from 2300 to 0700 h. Plasma TSH concentrations were measured by RIA (Incstar, Stillwater, USA). The detection limit was 0.013 mU/l. The intra-assay coefficient of variation (CV) for duplicate samples was 6% for concentrations between 0.05 and 0.5 mU/l and 3% for values above 0.5 mU/l.

The non-parametric Wilcoxon matched-pairs signed-rank test was used to determine statistical significance of differences between the mean nocturnal TSH values and δ relative power levels obtained under placebo versus ritanserin. Significant TSH pulses were identified in the individual profiles using the computer program ULTRA [18] with a threshold of significance set at two times the CV for each concentration range. The level of ASI depends on the quantity of α in the EEG and is consequently subject-dependent [11]. Thus, the identification of significant peaks in the individual ASI profiles was achieved using a subject-adapted threshold for the detection of wake episodes. This threshold was set between 0.08 and 0.15. The rate of agreement between the ASI peaks and the visual scoring of wakefulness was 90% during both experimental conditions. The temporal relationship between the TSH and the δ relative power profiles was quantified using cross-correlation analysis on series transformed into Z-scores ($Z\text{-score} = (x - m) / \sigma$, where x is the original data, m the mean value and σ the standard deviation of the data) [7]. For TSH, the method consisted of removing a third order polynomial that fitted the data, and for δ activity, the data were smoothed with a median filter of 20 points in order to obtain a spectral point every 10 min allowing a comparative analysis with TSH values over 10-min intervals. Then data were transformed into Z-scores. The cross-correlation coefficients were computed for lags -2 , -1 , 0 , $+1$, and $+2$ between the two chronological series, each lag corresponding to a 10 min blood sampling interval. The individual correla-

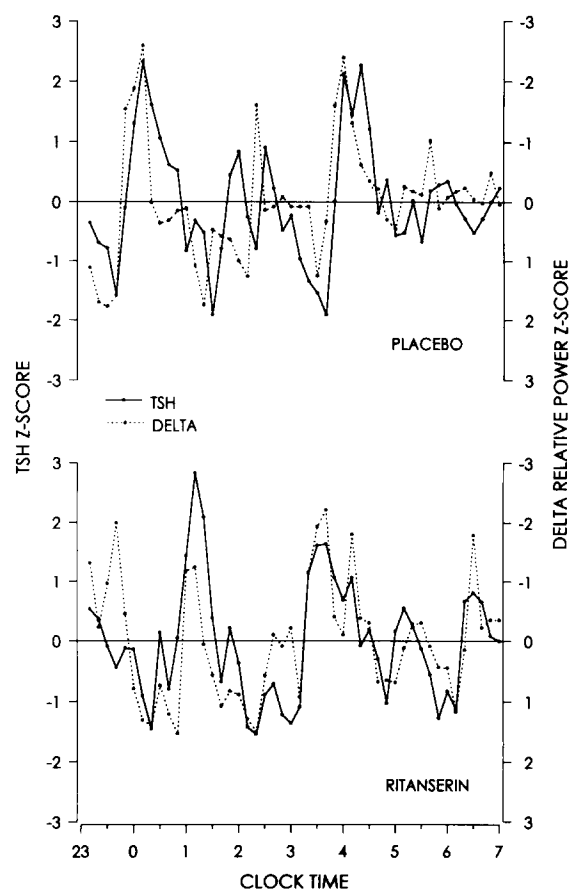


Fig. 1. Concomitant δ relative power and TSH profiles after Z-score transformation in a representative subject during placebo and ritanserin nights. For δ relative power the scale is inverted.

tion coefficients were averaged using Fisher's 'z' transformation to give a common estimate of the correlation [7]. This average coefficient was computed after a χ^2 test of homogeneity [16] on the individual transformed correlations. Coincidence between TSH and ASI pulses was assessed using the hypergeometric method [20]. Pulses were considered to be coincident if TSH pulses occurred in a time window of 30 min; i.e. 10 min before, at the same time and 10–20 min after an ASI pulse.

In both experimental conditions, the mean TSH profiles exhibited the classically described slight declining trend during the night. Ritanserin intake significantly increased the mean δ relative power which averaged $60 \pm 2\%$ under placebo and $69 \pm 2\%$ under ritanserin ($P < 0.01$). The overnight TSH levels were not significantly influenced by the drug intake despite a tendency to be lower under ritanserin than under placebo (1.60 ± 0.23 mU/l versus 1.93 ± 0.2 mU/l, $P = 0.15$). Cross correlation coefficients between TSH and δ relative power profiles were highest for lags 0 , -1 and -2 indicating either that both temporal series are concomitant or that the spectral parameter anticipated plasma TSH levels by 10–20 min. TSH profiles were negatively correlated with the

Table 1
Coincidence analysis between significant TSH and ASI pulses

Subjects	Placebo				Ritanserin			
	Number of pulses			P	Number of pulses			P
	ASI	TSH	Coincidence		ASI	TSH	Coincidence	
1	7	5	4	0.001	7	5	3	0.019
2	4	7	3	0.008	3	6	2	0.040
3	4	9	4	0.001	5	9	3	0.040
4	6	5	4	0.001	2	6	2	0.015
5	11	5	5	0.001	3	6	1	0.308
6	5	4	2	0.050	2	6	2	0.015
7	3	2	2	0.003	1	2	1	0.043
8	5	4	3	0.003	3	6	2	0.040

δ activity indicating that TSH levels decreased when δ -wave activity increased. The average correlation coefficient between the δ activity and TSH profiles was found to be highly significant under placebo as well as under ritanserin (respectively $r = -0.500$ versus $r = -0.432$, $P < 0.0001$ in both cases). The homogeneity of the individual transformed correlation coefficients in each condition ($\chi^2 = 13.38$ under placebo and $\chi^2 = 10.29$ under ritanserin; n.s.) allows this calculation. Fig. 1 illustrates concomitant TSH and δ relative power profiles in one representative subject in the two experimental conditions. TSH pulses were significantly coincident with ASI pulses in the 8 placebo nights and in 7 out of the 8 ritanserin nights (Table 1). Fig. 2 illustrates in one representative subject the nocturnal TSH and ASI profiles and shows the close relationship between both parameters.

The results of this study demonstrate a close association between sleep deepening and decreasing TSH levels, which is consistent with the previously described temporal relationship between SWS episodes and declining TSH slopes [8]. The refined analysis of EEG power density extends previous conclusions and allows us to demonstrate that in fact the whole TSH profile is negatively correlated with δ -wave activity throughout the night. TSH pulsatility seems to be predominantly controlled by hypothalamic TRH [4] and TSH pulses were demonstrated to have no major influence on thyroid hormone pulses [9]. The interindividual variability observed for the temporal coupling between TSH levels and δ relative power suggests that overriding regulatory factors may account for the variable strength of this association.

It is well known that ritanserin enhances the amount of SWS in man [10,15] as well as in animals [6]. This is confirmed by the increased δ power density observed in our study. An increase in the mean TSH level, although not significant, was observed during sleep in the placebo condition as compared to a lower mean level exhibited for a more profound sleep under ritanserin. However, at the beginning of the night before sleep onset, no differences in TSH levels were observed and no direct effect on TSH

has been previously found after the administration of an even higher dose of ritanserin than used in this study [17]. That sleep deepening is associated with TSH decreasing levels and conversely that sleep lightening is accompanied by TSH increase may explain the slight decrease in TSH levels observed under ritanserin.

The ratio between α and slow wave EEG activity expressed by the α slow wave index (ASI) is an essential feature to differentiate between sleep and wakefulness

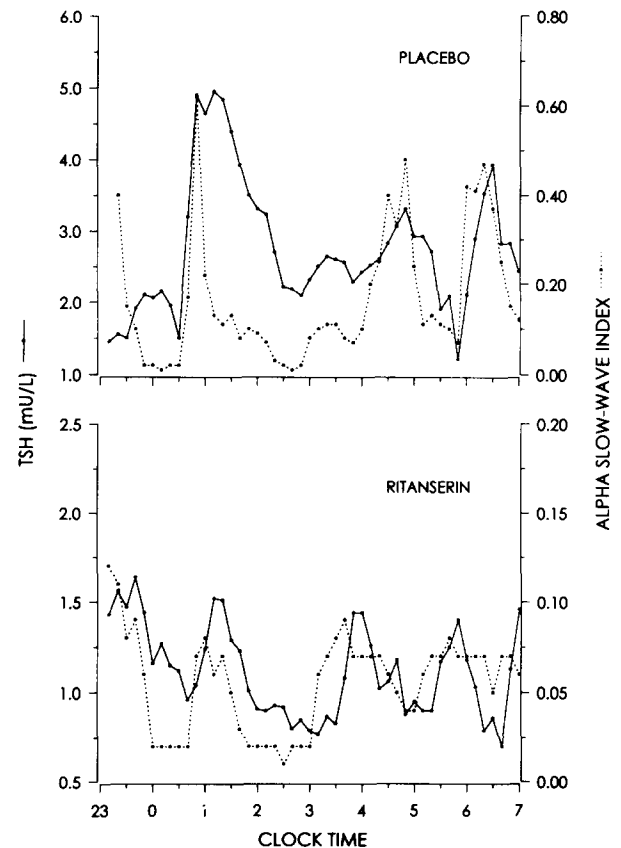


Fig. 2. Individual nocturnal profiles of TSH and ASI in one subject during placebo and ritanserin nights.

[11,12]. This parameter correlates highly with visually scored episodes of wakefulness during sleep. It allows us not only to determine the duration of the awakenings but also it gives a dynamic representation of variation in EEG activity. ASI pulses were found to coincide significantly with TSH pulses in both experimental conditions, which assessed the robustness of the previously described association between awakenings and TSH pulses [8]. From these results it appears clearly that intra-sleep awakenings have a modulatory effect on the whole nocturnal TSH profile.

In conclusion, this study demonstrates that plasma TSH fluctuations and EEG activity are inversely correlated, the whole nocturnal TSH profile reflecting variations of sleep depth and awakenings. These results suggest that common control mechanisms may be involved in the modulation of sleep-wake states and in the regulation of TSH release.

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