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Title: Measurement of 3x8h urine analysis to follow cortisol and melatonin rhythms

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Body: Background: To follow the daily rhythm of cortisol (**C**) and melatonin (**M**) can be useful to explain disorders concerning stress, fatigue or sleep[1]. Common protocols are based on repeated blood or saliva samples over 24 hours. However. daily variations do not follow a sinusoidal curve[2]: rhythms of cortisol and melatonin show very irregular profiles with short lasting peaks and troughs. Therefore "spot" sampling of blood and saliva can be misleading. In this study we evaluated the fluctuation of cortisol and melatonin in urine over a 24 h period.

Material and Methods: 25 clinicians investigating 200 patients for defined disorders participated in the study. Three consecutive 8 hours urine collections defined as period 1 (morning), 2 (afternoon) and 3 (night) starting at 07:00 hr, stored at 4°C, were obtained and analysed for urinary free **C** (RIA) and **M** (6-sulfatoxy-melatonin, Elisa) within 24 hours after reception in the laboratory or frozen until assayed. Value changes between urines from 8 hour periods were defined as high amplitude (± >150%) , median amplitude (> 50% to < 150%) and low amplitude < 50%). Desynchronized fluctuations of cortisol versus melatonin were defined as a shift in one rhythm not observed in the other. Clinical indications were: burn-out, depression following exhaustion, seasonal depression, morning fatigue or unexplained sleeping disorders. In a few cases the effect of treatment with melatonin was evaluated.

Results: Rhythms of **C** (**M**) were of low amplitude in 30% (28%), medium amplitude in 18% (18%) and of high amplitude in 5% (13%). Highest **C** levels were observed in phase night and morning phases in 35% of cases delayed phase in 40%, no rhythm in 13% and phase advanced in 8% of subjects. For **M**, peaks during the night occurred in 27% of patients, while the peak was extended in the morning phase in 53%, no rhythm in 15% and phase advance in the afternoon in 5% of patients. In 23% of the patients desynchronisation between **C** (**M**) rhythms were observed with **C** peaks displaced in the morning while **M** peaked during the night. In 5 patients the measurement of **C** and **M** fluctuations were repeated 2 times, yielding similar results. The correlation between **C** and **M** fluctuations and clinical data will be presented.

Conclusions: The analysis of urine collected by 8 hours period is promising to follow the fluctuations and the relationship of **C** and **M** with clinical data.

References: (1) Lieverse R et al., Arch Gen Psychiatry 2011; 68:61
(2) Rivest RW et al., J Clin Endocrinol Metabol 1989; 68:721