Pteridines

Vol. 18, 2007, pp. 69 - 78

Effect of Two Different Lighting Environments on Urinary Neopterin and Sulphatoxymelatonin in an Experimental Night Shift Model

Wolfgang Schobersberger¹, Veronika Gufler¹, Andrea Griesmacher², Christian Bartenbach³, Markus Canazei³, Siegmund Staggl³, Georg Hoffmann¹

¹Institute for Leisure, Travel, and Alpine Medicine; University for Health Sciences, Medical Informatics, and Technology, Eduard Wallnoefer-Zentrum 1, A-6060 Hall, Austria, ²Central Institute for Medical and Chemical Laboratory Diagnostics, University Hospital Innsbruck-TILAK, Innsbruck, Austria, ³Bartenbach LichtLabor, Rinner Strasse 14, 6071 Aldrans, Austria

Abstract

Regular night shift activities are associated with a series of adverse effects (e.g. chronic fatigue, increased yield loss rates, diseases) related to a disturbed circadian rhythm of the employee. According to current knowledge, the human circadian system is affected by visible light in the short wavelength range. In the present study, we investigated the effects of reduced short-wavelength components in the lighting environment of an office workplace on circadian markers, symptoms of lassitude, and personal mood in an experimental night shift setup. Nocturnal urinary neopterin excretion was different from known circadian rhythms under both filtered test light and bright light conditions. By contrast, urinary sulphatoxymelatonin concentrations could be adjusted to a normal diurnal profile in the test light environment. Mood rating inventories did not result in differences in the subjective perception between the two lighting environments with respect to the dimensions of "Activity", "Concentration", "Deactivation", and "Fatigue". In addition, changes in signs of vigour and weariness in the course of each night of the study were equally pronounced in test light and bright light. In conclusion, the present study shows that light with reduced short-wavelength components maintains the nocturnal profile of sulphatoxymelatonin but simultaneously exerts a wearisome effect on the participants. Therefore, a general implementation of test light to illuminate night shift workplaces implies further investigations.

Key words: circadian rhythm, lighting environment, neopterin, shift work, sulphatoxymelatonin

Introduction

Approximately 20% of employees in the European Union are working on rotating night shifts, e.g. in security and service industries. The artificial workplace environment has given rise to timeless offices contradictory to our evolutionary developed circadian rhythm. Thus, long-term shift work has been associated with various health risks including gastrointestinal disorders (1), cardiovascular diseases (2, 3), or an increased prevalence for major depression and other mood disorders (4). Recent studies have provided evidence that regular night shift activities are connected to higher risks of breast and colorectal cancer (5, 6). A proposed pathogenetic mechanism for these observations is the exposure to artificial light at irregular times

of day. Human circadian rhythms are adjusted to the 24 h rotation of the earth by entrainment factors with the light-dark cycle being the predominant exogenous synchronizer (7, 8). Received by a special type of photoreceptor different from rods and cones, the light signal is transmitted to the biological master clock located in the suprachiasmatic nucleus where the original circadian period of ~ 25 h is adjusted to the natural light-dark situation (9). Due to exposure to light during night shifts, this alignment is markedly impaired resulting in an incomplete circadian adaptation of physiological rhythms. One of the best explored biological parameters following a diurnal pattern is the neurohormone melatonin (N-acetyl-5-methoxytryptamine) secreted by the mammalian pineal gland. Melatonin synthesis is affected by the light-dark cycle

Correspondence to: PD Dr. Georg Hoffmann, Institute for Leisure, Travel, and Alpine Medicine, University for Health Sciences, Medical Informatics and Technology Tyrol, Eduard Wallnoefer-Zentrum 1, A-6060 Hall, Austria, Phone: +43/59/8648-3856, FAX: +43/50/8648-673840, e-mail: georg.hoffmann@umit.at

with an acrophase at ~ 04:00 h at night (10-12). Light exposure at any given time effectively attenuates melatonin secretion (13). Thus, substantially decreased nocturnal levels of melatonin are common among shift workers spending their night time hours in an artificially illuminated workplace (14). The biochemical functions of melatonin as well as its signal transduction mechanisms have not entirely been elucidated. It is assumed that melatonin affects the control system of body temperature in the hypothalamus thereby influencing at least one other physiological parameter exhibiting a circadian pattern. Apart from that it is suggested that melatonin has anti-oxidative properties and exerts an oncostatic effect (15, 16). Continuous dysfunction of melatonin synthesis may therefore explain some of the detrimental consequences of long-term night shifts. Within this context it is of special interest that the capacity for melatonin suppression is not equal for all wavelengths of light. The maximal spectral response is generated with light of ~ 460 nm suggesting a photopigment in the circadian photoreceptors that reacts specifically to short wavelength light (17, 18). These data implicate that a careful selection of the lighting environment can avoid at least some of the hazardous conditions of continuous shift work. The aim of this study was to investigate the effects of two differentially illuminated office workplaces in an experimental night shift setup. Light with a low color temperature (1700 K at eye level) was compared to light higher color temperature (6300 K at eye level). As phase markers of circadian rhythm, the stable

metabolite of melatonin in urine - 6-sulphatoxymelatonin (aMT6-s) - and the heterocyclic pteridine compound neopterin were measured. Neopterin concentrations and neopterin to creatinine ratio have been shown to exert a 24 h-periodicity with an acrophase in the early morning hours (19), and it represents a marker of the cellular immune system that might be altered due to desynchronization of the physiological diurnal rhythm. In addition, emotional reactions of study participants to the different lighting conditions were assessed using a mood rating inventory.

Materials and Methods

Study participants and study design

11 healthy male volunteers (age: 23 (22-29) years; body weight: 74 (62-84) kg; height: 177 (172-181) cm) without any known metabolic, cardiovascular, renal, hepatic, neurological, psychiatric or sleeping disorders requiring chronic medication participated in our study. Test persons were all living in the Austrian Tyrol at the time of the experiments and were comparable with respect to social and educational status. They had not undertaken shiftwork or transmeridian travel in the month prior to the study. The project was approved by the Ethics Committee of the Medical University of Innsbruck, and all participants gave written informed consent. The study was conducted at the Bartenbach LichtLabor, Aldrans, Austria. The volunteers were randomly assigned to start with three consecutive nights

Table 1. Workplace conditions for both unfiltered bright light as well as filtered light with reduced short-wavelength components.

Area	20 m ²
Ceiling height	2.6 m
Number of workstations	2
Temperature	23°C ± 2°C, regulated via air handling unit
Environmental luminance	300 cd/m ²
Color rendering index (CRI)	CRI = 89 in room with unfiltered bright light; CRI = 80 in room with reduced short-wavelength components
Luminaire	Wallwasher, color temperature 6300 K at eye level in room with unfiltered bright light; Wallwasher, color temperature 1700 K at eye level in room with reduced short-wavelength components
Illuminance level	800 lux at eye level

of simulated shift work either in an unfiltered bright light environment or in a lighting environment with reduced short-wavelength components. Workplace conditions for both experimental setups are summarized in Table 1. After a 2-week rest, the study participants conducted a second run of three consecutive night shifts. Each night shift started at 22:00 h and ended at 06:00 in the morning. Urine specimen were collected at ~ 22:00 h, at ~ 02:00 h, and at ~ 06:00 h, respectively. Simulated shift work consisted of general and special ability tests all being part of the Vienna Test System (Dr. G. Schuhfried Ltd., Moedling, Austria). In brief, performance testing (Reaction Time Analysis), vigilance testing, evaluation of attentiveness (Continuous Attention), and flicker/fusion frequency analyses were performed twice a night. With respect to study protocol, the following abbreviations will be used throughout this manuscript:

N(1-3): first, second or third night in an unfiltered bright light environment (normal light),

T(1-3): first, second or third night in a lighting environment with reduced short-wavelength components (test light),

U(1-3): time point of collection of urine specimen (1 = 22:00 h, 2 = 02:00 h, 3 = 06:00 h),

Q (1 or 2): the first or second mood rating inventory questionnaire surveyed during the experimental night shift (1 = 22:00 h; 2 = 06:00 h).

Laboratory measurements

Neopterin and creatinine concentrations were measured using high pressure liquid chromatography. In brief, 100 µl of each urine sample was diluted with 1000 µl potassium phosphate buffer (15 mM, pH 6.4) containing 4 g/L ethylene-diamine tetraacetate. 20 µl of diluted specimen were injected on reverse phase C18 columns (LiChroCart 5 mm diameter, Merck, Darmstadt, Germany) and eluted with potassium phosphate buffer (15 mM, pH 6.4). Native neopterin was quantified by measurement of its natural fluorescence at 353 nm excitation and 438 nm emission wavelength and creatinine was detected by UV light at 235 nm wavelength. Data are given as neopterin/creatinine ratio (µmol/mol). Measurements were performed at the Division of Biological Chemistry, Biocentre, Innsbruck Medical University (Prof. Dr. D. Fuchs).

Urinary aMT6-s was assayed using a commercially available sandwich ELISA kit (Buehlmann 6-SMT, Buehlmann Laboratories AG, Schoenenbuch, Switzerland). It is a competitive immunoassay with a lower detection limit of ~ 0.5 ng/ml. In brief, samples and biotinylated aMT6-s competitor were added together with a highly specific rabbit anti-aMT6-s-antibody to 96-well microtiter plates precoated with polyclonal anti-rabbit immunoglobulin antibody.

Following a 3-h incubation at 4°C, biotinylated aMT6-s competitor-antibody complexes were detected by application of streptavidin-conjugated horseradish peroxidase using 3,3',5,5'-tetramethylbenzidine as a substrate. In a 30-min incubation step, a colored product is formed in inverse proportion to the amount of aMT6-s present in the urine specimen which is detected in a microplate reader using dual wavelength measurement at 450 nm absorbance with 620 nm background wavelength correction. Unknown samples were calculated using a standard curve generated by standard solutions provided by the manufacturer. Because urine samples were not collected over a 24-hour period and total urinary output was unknown, creatinine levels were measured for each sample by the same laboratory. All aMT6-s levels are creatinine-standardized to account for differences arising from variations in urine concentrations. Data are given as ng aMT6-s/mg creatinine. All aMT6-s analyses were done at the Central Institute for Medical and Chemical Laboratory Diagnostics, Innsbruck, Austria.

Questionnaires

At the beginning and at the end of each night shift, the volunteers answered a questionnaire directed to validate various aspects of their mood state by applying a mood rating inventory (EWL adjective check list of Janke and Debus©, Hogrefe Verlag, Goettingen, Germany). This questionnaire describes the subject's mood by means of an adjective checklist and consists of 161 items which can be summarized to six scales measuring vigour, fatigue, extraversion, well-being, irritability/anger and anxiety/depression (20-22).

Statistical analyses

To compare between three or more matched groups with repeated measures, Friedman tests followed by Dunn's post test were performed. To compare paired groups, we used the Wilcoxon matched pairs test. To test whether two variables vary together, a nonparametric Spearman correlation was calculated. For presentation of descriptive data, box and whiskers plots were used. The box represents the median together with the 25th and 75th quartile, while the whiskers show the highest and lowest values, respectively. P-values < 0.05 were considered to be significant.

Results

All volunteers finished the complete study protocol and tolerated both lighting conditions. With respect to their time course, urinary neopterin concentrations showed a significant variation on night 2 of test and normal light, respectively (Figure 1). Under test light

Figure 1. Time course of urinary neopterin concentrations on the first, second, and third night either in a lighting environment with reduced short-wavelength components (T1, T2, and T3; upper part of Figure 1) or in an unfiltered bright light environment (N1, N2, and N3; lower part of Figure 1). Time points of specimen collection were 22:00 h (U1), 02:00 h (U2), and 06:00 h (U3), respectively. Data are given as median together with the 25th and the 75th quartile, whiskers represent highest and lowest values.

#P < 0.05 as compared to time point U1 of the respective night of the study;

*P < 0.05 as compared to time points U1 and U3 of the respective night of the study.

conditions, neopterin was significantly decreased at the end of night 2 (U3; 88, 72-108 μmol/mol creatinine, P < 0.05) as compared to the first time point of specimen collection (U1; 98, 93-126 μmol/mol creatinine, upper panel of Figure 1). In the unfiltered bright light environment, neopterin concentration on night 2

was higher at the second time point of sample collection (U2; 123, 103-130 μmol/mol creatinine, P < 0.05) as compared to both U1 (103, 88-124 μmol/mol creatinine) and U3 (98, 86-110 μmol/mol creatinine, lower panel of Figure 1). When comparing between the effects of test and normal light on neopterin secretion,

Figure 2. Time course of urinary sulphatoxymelatonin concentrations on the first, second, and third night either in a lighting environment with reduced short-wavelength components (T1, T2, and T3; upper part of Figure 2) or in an unfiltered bright light environment (N1, N2, and N3; lower part of Figure 2). Time points of specimen collection were 22:00 h (U1), 02:00 h (U2), and 06:00 h (U3), respectively. Data are given as median together with the 25th and the 75th quartile, whiskers represent highest and lowest values.

$P < 0.05$ as compared to time point U1 of the respective night of the study;

* $P < 0.05$ as compared to time points U1 and U2 of the respective night of the study,

+ $P < 0.05$ as compared to time point U2 of the respective night of the study.

Dotted lines indicate $P < 0.05$ when comparing U3 values in light with reduced short wavelength components with the respective time point of specimen collection in unfiltered bright light.

no differences in urinary neopterin concentrations could be detected at any night of the study.

Under test light conditions, highest concentrations

of aMT6-s were measured at the third time point of urine specimen collection at any night of the study (upper panel of Figure 2). In contrast to these observa-

tions, profile of aMT6-s did not vary significantly at night 3 following normal light study protocol (lower panel of Figure 2). As shown in Figure 2, test light aMT6-s values at U3 were significantly higher on night 1 (15.5, 4.5-31.3 ng/mg creatinine, $P < 0.05$) and night 3 (3.6, 1.4-12.3 ng/mg creatinine, $P < 0.05$) as compared to the respective urine aMT6-s concentrations in normal light (U3 on night 1: 2.7, 1.5-11.9 ng/mg creatinine; U3 on night 3: 1.5, 0.8-2.4 ng/mg creatinine).

The results of the mood rating inventory questionnaires are summarized in Table 2. With respect to lighting environment, there were no statistically significant differences in the characteristics of each value either at Q1 or at Q2. "Activity" values were decreased when comparing the second survey at night 1 and night 3 under test light conditions with the data of the first survey at the respective night of the study. Concerning the

while "Fatigue" was augmented at every night with test light arrangements, but at night 1 and 2 only in the unfiltered bright light environment. In addition, we calculated the differences in the characteristics of each dimension of the mood rating inventory between the beginning and the end of every night of the study for both lighting environments. Statistical analyses of these differences revealed changes in signs of vigour as well as signs of weariness at every night of the study that were equally pronounced under test light and under normal light (data not shown).

Considering urinary concentrations of specimens collected at time point U3, there is no statistically significant correlation between neopterin and aMT6-s at any night of the study, although the tendency towards an inverse correlation is stronger under normal light conditions (Table 3). With regard to selected items from the mood rating questionnaire (comparison of U3

Table 2. Results of mood rating inventory questionnaires (selected parameters of the EWL adjective check list of Janke and Debus©, Hogrefe Verlag, Goettingen, Germany). Data are given as median (with the 25th and the 75th percentile).

T(1-3): first, second or third night in a lighting environment with reduced short-wavelength components (test light);

N(1-3): first, second or third night in an unfiltered bright light environment (normal light);

Q(1,2): the first or second mood rating inventory questionnaire surveyed during the experimental night shift (1 = 22:00 h; 2 = 06:00 h);

* $p < 0.05$ as compared to the first measurement (Q1) on the respective night of the study protocol (Wilcoxon matched pairs test).

	Q1T1	Q2T1	Q1T2	Q2T2	Q1T3	Q2T3
Activity	11 (6-17)	3 (0-7) *	7 (2-12)	1 (0-3)	8 (4-15)	4 (1-6) *
Concentration	6 (3-6)	2 (0-3) *	3 (2-5)	1 (0-2) *	4 (1-5)	2 (1-5)
Deactivation	0 (0-4)	7 (4-12) *	0 (0-8)	7 (2-14) *	0 (0-6)	6 (4-9) *
Fatigue	1 (0-2)	4 (2-5) *	1 (0-4)	4 (2-5) *	0 (0-1)	4 (1-5) *
	Q1N1	Q2N1	Q1N2	Q2N2	Q1N3	Q2N3
Activity	10 (3-14)	3 (0-4) *	9 (3-14)	2 (0-7)	8 (5-15)	4 (1-11)
Concentration	5 (1-6)	1 (0-2) *	4 (1-5)	2 (0-3)	3 (3-5)	3 (0-5)
Deactivation	1 (0-7)	8 (5-15) *	0 (0-3)	7 (1-12) *	0 (0-2)	10 (3-13) *
Fatigue	0 (0-1)	5 (2-6) *	0 (0-0)	3 (0-5) *	1 (0-2)	2 (0-5)

item "Concentration", results at the beginning of night 1 and 2 in test light were significantly higher as compared to Q2T1 and Q2T2, respectively. In normal light, these values varied significantly only at night 1 of the study protocol. "Deactivation" significantly increased at night 1, 2, and 3 in both lighting environments,

and Q2, respectively), we found a correlation between neopterin and "Deactivation" in test light (which was statistically significant at night 2), but an inverse correlation in normal light (statistically significant at night 2 and night 3). A similar conjoint variation was detected between neopterin and "Fatigue". In contrast,

neopterin and "Activity" as well as "Concentration" tended to be inversely correlated in test light (statistically significant at night 2), and to be correlated under normal light conditions (Table 3).

Discussion

Since melatonin is regarded to be a substantial circadian phase marker, we decided to measure its princi-

wavelength components (test light) did not result in an attenuation of aMT6-s excretion at dawn. Moreover, as compared to bright light, test light conditions were associated with a significantly higher aMT6-s concentration in urine collected at 06:00 h on night 1 and night 3 as well as a tendency towards higher values on night 2. Comparable results were reported by Kayumov *et al.* (33), who found a normal night time profile of melatonin in a simulated shift work model in those subjects wearing light-filtering goggles that

Table 3. Correlation between neopterin and sulphatoxymelatonin measured in urine specimens collected at ~06:00 h as well as selected parameters of mood rating inventory surveyed at ~06:00 h (EWL adjective check list of Janke and Debus©, Hogrefe Verlag, Goettingen, Germany) of three consecutive nights in a lighting environment with reduced short-wavelength components (T) or in an unfiltered bright light environment (N). Non-parametric Spearman rank correlations were calculated. Data represent correlation coefficients (rs), and significant rs are written in bold shape (*p < 0.05, **p < 0.01).

	Neopterin T1	Neopterin T2	Neopterin T3	Neopterin N1	Neopterin N2	Neopterin N3
Sulphatoxymelatonin	-0.046	-0.164	0.046	-0.373	-0.200	-0.252
Activity	-0.192	-0.227	-0.359	0.454	0.277	0.197
Concentration	-0.180	-0.724*	-0.218	0.377	0.349	0.324
Deactivation	0.303	0.702*	0.570	-0.478	-0.685*	-0.630*
Fatigue	0.409	0.659*	0.661*	-0.179	-0.661*	-0.841**

pal metabolite in urine, aMT6-s. Earlier studies have shown that aMT6-s is a reliable indicator of pinealocyte function and serum melatonin concentration (23-25). The circadian profile of melatonin shows a peak at ~ 04:00 at night with absolute values being age-dependent (26-28). Since melatonin excretion declines with age, we have chosen to enroll volunteers belonging to a narrow age range (20-35 years) that still has a high melatonin production capability as well as a low inter-individual variability in melatonin secretion (29). During our experimental night shift setup, highest aMT6-s values were usually observed in the early morning hours, i.e. in urine specimens collected at ~ 06:00 h, which is in congruence with the known circadian rhythm of the compound. During the third night in normal light, we could not detect a significant increase in urinary aMT6-s concentrations in the samples collected at 06:00 h as compared to the specimens collected at 22:00 h and 02:00 h, respectively. Light is an established suppressor of pinealocyte melatonin synthesis and shift workers have been described to have altered diurnal melatonin profiles (30-32). With respect to normal bright light, our data confirm these previous observations. However, reduction of short-

blocked wavelengths of less than 530 nm. Our study suggests that some benefit of reducing short-wavelength light on melatonin synthesis can be achieved in an almost natural office accommodation and does not require wearing glasses.

In 1988, Auzéby *et al.* (19) documented a circadian rhythm of neopterin with a peak of the urinary excretion of the pteridine compound in the early morning hours. In our study, we observed highest neopterin concentrations and neopterin/creatinine in urine collected at ~ 02:00. Neopterin concentrations tended to be lower at the last time point of specimen collection at ~ 06:00 in the morning. Since there are no further studies with regard to the circadian rhythm of neopterin in shift workers, we can only speculate about the reasons for the discrepancies between our observations and the data of Auzéby and co-workers (19). It is well established that shift work is associated with rapid alterations of circadian parameters, e.g. secretory rhythms for growth hormone or cortisol (34-36). Therefore, changing the typical bedtimes and work schedules might at least in part explain the phase shift in urinary neopterin concentrations observed in our study participants. The hypothesis that the selection of

an appropriate illumination may prevent the misalignment of neopterin's circadian rhythm can not be supported by our results, since its profile was independent of the lighting conditions in the respective office workplaces.

Recently, Garcia-Gonzales (37) reported a possible interdependency between melatonin and neopterin. In serum samples collected at different light/dark periods of the day, he found higher melatonin levels concurrent with lower neopterin levels and vice versa. From these observations, the author concludes that the circadian rhythm of melatonin affects at least in part the corresponding diurnal changes in serum neopterin concentrations. However, no analysis of correlation is given. In our study, we could not find any co-variation between urinary neopterin and aMT6-s although there was a tendency towards an inverse correlation under bright light conditions. One possible explanation for the different observations is the age range of study participants, which was 20-35 years in our experiments as compared to 57-73 years in the studies of Garcia-Gonzales (37).

While designing the lighting environment at any night shift workplace, maintenance of a physiological circadian rhythm can only be one ergonomical aspect. Another important matter is the subject's performance and well-being. The selected illumination has to comply with the requirements of working conditions, i.e. it should not increase spoilt production or accident rate and should not detrimentally affect subjective mood of the employee. Within this context, advantageous effects of bright light have been reported in the literature. Leppamaki *et al.* (38) have shown that pulses of bright light exposure significantly relieved subjective symptoms of distress in night shift workers. Likewise, Iwata and co-workers (39) observed a benefit of 30-min intervals of bright light with an intensity of 3000 lux on the mental state of nurses during night shift work as compared to a constant routine of 250 lux. In another experiment, even higher intensities of bright light (~ 5000 lux) were used in comparison to dim light (500 lux) and were shown to improve phase shifting of temperature rhythm that was associated with better subjective daytime sleep, fatigue, and overall mood (40). Taken together, these data demonstrate that nocturnal exposure to bright light at regular intervals exerts a favorable effect on subjective mood as well as performance. In our experiments, we have taken into account the potential effects of reducing short-wavelength components on vigour and fatigue. The evaluation of the mood rating inventories revealed an increase in the dimensions "Deactivation" and "Fatigue" as well as a decrease in the dimensions of "Activity" and "Concentration". The symptoms of lassitude seemed to be less pronounced in the unfiltered

bright light as compared to test light. Thus, a significant decrease in "Activity" and "Concentration" was observed only at the first night with bright light, but at night 1 and 3 ("Activity") as well as night 1 and 2 ("Concentration") with test light. "Deactivation" was significantly increased at every night independent of lighting conditions, while "Fatigue" was significantly higher at night 1 and 2 with bright light but at all nights in the test light setup. However, it should be noted that the degree of changes in signs of vigour and weariness did not differ between test and normal light. There were differences in the characteristics of "Activity", "Concentration", "Deactivation", and "Fatigue" between the beginning and the end of each night of the study protocol independent of the lighting environment.

Furthermore, we found a correlation between neopterin and the mood rating parameters for vigour and fatigue depending on the given illumination. There was an inverse correlation between urinary neopterin and in test light. In contrast, "Deactivation" and "Fatigue" both correlated positively with neopterin in test light, while there was an inverse covariation in the unfiltered bright light workplace. However, these observations are limited due to the fact that the correlations were significant only at night 2 and 3 (see Table 3 for details). Moreover, we could not detect any light-dependent effects on the absolute values of urinary neopterin excretion at any night of the study. It has been suggested that fatigue can be related to persistent activation of the immune system in a number of diseases. With respect to fatigue and neopterin as a marker and mediator of the cellular immune system, inconsistent data have been reported in the literature so far. Patarca *et al.* (41) found elevated serum neopterin concentrations in less than one third of individuals with chronic fatigue syndrome. In patients with multiple sclerosis-associated fatigue, serum neopterin was reported to be within normal range (42). However, higher serum neopterin levels were found in patients with cancer-related fatigue (43, 44). Taken together, the relationship between neopterin and symptoms of fatigue is not conclusive.

The aim of this study was to investigate the effects of a lighting environment with reduced short-wavelength components on circadian rhythms as well as parameters of efficiency in a simulated night shift setup. As compared to unfiltered bright light, the diurnal rhythm of sulphatoxymelatonin was maintained under test light conditions, while there was no difference in urinary neopterin concentrations. Regarding vigour and fatigue, night shift work exerted an adverse effect that was equally pronounced in both illumination settings. Further studies are required to examine the applicability of the filtered test light system for reg-

ular shift work.

Acknowledgements

The authors would like to thank all volunteers and the local team from Bartenbach LichtLabor, Aldrans, Austria as well as the rest of the staff involved in the study. We gratefully acknowledge the expert technical assistance of Drs. G. Falkensammer and A. Hammerer-Lercher at the Central Institute for Medical and Chemical Laboratory Diagnostics, University Hospital Innsbruck-TILAK, Innsbruck, Austria. Statistical analyses were performed by Prof. Dr. A. Klingler at the Theoretical Surgery Unit of the Department of General and Transplant Surgery, Medical University Innsbruck, Austria. This study was financially supported by the Lighting Network, Aldrans, Austria, and in part by the TILAK GesmbH (Tiroler Landeskrankenanstalten), Innsbruck, Austria.

References

- Vener KJ, Szabo S, Moore JG. The effect of shift work on gastrointestinal (GI) function: a review. *Chronobiologia* 1989; 16: 421-439.
- Kawachi I, Colditz GA, Stampfer MJ et al. Prospective study of shift work and risk of coronary heart disease in women. *Circulation* 1995; 92: 3178-3182.
- Knutsson A, Akerstedt T, Jonsson BG, Orth-Gomer K. Increased risk of ischaemic heart disease in shift workers. *Lancet* 1986; 2: 89-92.
- Scott AJ, Monk TH, Brink LL. Shiftwork as a Risk Factor for Depression: A Pilot Study. *Int J Occup Environ Health* 1997; 3: S2-S9.
- Hansen J. Light at night, shiftwork, and breast cancer risk. *J Natl Cancer Inst* 2001; 93: 1513-1515.
- Schernhammer ES, Laden F, Speizer FE, et al. Rotating night shifts and risk of breast cancer in women participating in the nurses' health study. *J Natl Cancer Inst* 2001; 93: 1563-1568.
- Wever RA, Polasek J, Wildgruber CM. Bright light affects human circadian rhythms. *Pflugers Arch* 1983; 396: 85-87.
- Zeitzer JM, Dijk DJ, Kronauer R, Brown E, Czeisler C. Sensitivity of the human circadian pacemaker to nocturnal light: melatonin phase resetting and suppression. *J Physiol* 2000; 526 Pt 3: 695-702.
- Inouye ST, Kawamura H. Persistence of circadian rhythmicity in a mammalian hypothalamic "island" containing the suprachiasmatic nucleus. *Proc Natl Acad Sci U S A* 1979; 76: 5962-5966.
- van Coevorden A, Mockel J, Laurent E et al. Neuroendocrine rhythms and sleep in aging men. *Am J Physiol* 1991; 260: E651-E661.
- Trinchard-Lugan I, Waldhauser F. The short term secretion pattern of human serum melatonin indicates apulsatile hormone release. *J Clin Endocrinol Metab* 1989; 69: 663-669.
- Weitzman ED, Weinberg U, D'Eletto R et al. Studies of the 24 hour rhythm of melatonin in man. *J Neural Transm* 1978; 13: 325-337.
- Khalsa SB, Jewett ME, Cajochen C, Czeisler CA. A phase response curve to single bright light pulses in human subjects. *J Physiol* 2003; 549: 945-952.
- Roden M, Koller M, Pirich K, Vierhapper H, Waldhauser F. The circadian melatonin and cortisol secretion pattern in permanent night shift workers. *Am J Physiol* 1993; 265: R261-R267.
- Pandi-Perumal SR, Srinivasan V, Maestroni GJ, Cardinali DP, Poeggeler B, Hardeland R. Melatonin: Nature's most versatile biological signal? *FEBS J* 2006; 273: 2813-2838.
- Simonneaux V, Ribelayga C. Generation of the melatonin endocrine message in mammals: a review of the complex regulation of melatonin synthesis by norepinephrine, peptides, and other pineal transmitters. *Pharmacol Rev* 2003; 55: 325-395.
- Berson DM, Dunn FA, Takao M. Phototransduction by retinal ganglion cells that set the circadian clock. *Science* 2002; 295: 1070-1073.
- Brainard GC, Hanifin JP, Rollag MD et al. Human melatonin regulation is not mediated by the three cone photopic visual system. *J Clin Endocrinol Metab* 2001; 86: 433-436.
- Auzeby A, Bogdan A, Krosi Z, Touitou Y. Time-dependence of urinary neopterin, a marker of cellular immune activity. *Clin Chem* 1988; 34: 1866-1867.
- Marshall L, Molle M, Hallschmid M, Born J. Transcranial direct current stimulation during sleep improves declarative memory. *J Neurosci* 2004; 24: 9985-9992.
- Schmidt R, Auer-Grumbach P, Fazekas F, Offenbacher H, Kapeller P. Anticardiolipin antibodies in normal subjects. Neuropsychological correlates and MRI findings. *Stroke* 1995; 26: 749-754.
- Weniger G, Irle E. Impaired facial affect recognition and emotional changes in subjects with transmodal cortical lesions. *Cereb Cortex* 2002; 12: 258-268.
- Kunz D, Schmitz S, Mahlberg R et al. A new concept for melatonin deficit: on pineal calcification and melatonin excretion. *Neuropsychopharmacology* 1999; 21: 765-772.
- Lang U, Kornemark M, Aubert ML, Paunier L,

- Sizonenko PC. Radioimmunological determination of urinary melatonin in humans: correlation with plasma levels and typical 24-hour rhythmicity. *J Clin Endocrinol Metab* 1981; 53: 645-650.
- 25 Nowak R, McMillen IC, Redman J, Short RV. The correlation between serum and salivary melatonin concentrations and urinary 6-hydroxymelatonin sulphate excretion rates: two non-invasive techniques for monitoring human circadian rhythmicity. *Clin Endocrinol (Oxf)* 1987; 27: 445-452.
 - 26 Lynch HJ, Wurtman RJ, Moskowitz MA, Archer MC, Ho MH. Daily rhythm in human urinary melatonin. *Science* 1975; 187: 169-171.
 - 27 Sack RL, Lewy AJ, Erb DL, Vollmer WM, Singer CM. Human melatonin production decreases with age. *J Pineal Res* 1986; 3: 379-388.
 - 28 Wetterberg L, Bergiannaki JD, Paparrigopoulos T *et al.* Normative melatonin excretion: a multinational study. *Psychoneuroendocrinology* 1999; 24: 209-226.
 - 29 Mahlberg R, Tilmann A, Salewski L, Kunz D. Normative data on the daily profile of urinary 6-sulfatoxymelatonin in healthy subjects between the ages of 20 and 84. *Psychoneuroendocrinology* 2006; 31: 634-641.
 - 30 Borugian MJ, Gallagher RP, Friesen MC, Switzer TF, Aronson KJ. Twenty-four-hour light exposure and melatonin levels among shift workers. *J Occup Environ Med* 2005; 47: 1268-1275.
 - 31 Schernhammer ES, Rosner B, Willett WC, Laden F, Colditz GA, Hankinson SE. Epidemiology of urinary melatonin in women and its relation to other hormones and night work. *Cancer Epidemiol Biomarkers Prev* 2004; 13: 936-943.
 - 32 Touitou Y, Motohashi Y, Reinberg A *et al.* Effect of shift work on the night-time secretory patterns of melatonin, prolactin, cortisol and testosterone. *Eur J Appl Physiol Occup Physiol* 1990; 60: 288-292.
 - 33 Kayumov L, Casper RF, Hawa RJ *et al.* Blocking low-wavelength light prevents nocturnal melatonin suppression with no adverse effect on performance during simulated shift work. *J Clin Endocrinol Metab* 2005; 90: 2755-2761.
 - 34 Fujiwara S, Shinkai S, Kurokawa Y, Watanabe T. The acute effects of experimental short-term evening and night shifts on human circadian rhythm: the oral temperature, heart rate, serum cortisol and urinary catecholamines levels. *Int Arch Occup Environ Health* 1992; 63: 409-418.
 - 35 Goichot B, Weibel L, Chapotot F, Gronfier C, Piquard F, Brandenberger G. Effect of the shift of the sleep-wake cycle on three robust endocrine markers of the circadian clock. *Am J Physiol* 1998; 275: E243-E248.
 - 36 Koller M, Harma M, Laitinen JT, Kundi M, Piegler B, Haider M. Different patterns of light exposure in relation to melatonin and cortisol rhythms and sleep of night workers. *J Pineal Res* 1994; 16: 127-135.
 - 37 Garcia-Gonzalez MJ, Dominguez-Rodriguez A, Abreu-Gonzalez P. Diurnal variations in serum neopterin levels are associated with the pineal hormone melatonin circadian rhythm in healthy human subjects. *J Pineal Res* 2006 40: 288-289.
 - 38 Leppamaki S, Partonen T, Piironen P, Haukka J, Lonnqvist J. Timed bright-light exposure and complaints related to shift work among women. *Scand J Work Environ Health* 2003; 29: 22-26.
 - 39 Iwata N, Ichii S, Egashira K. Effects of bright artificial light on subjective mood of shift work nurses. *Ind Health* 1997; 35: 41-47.
 - 40 Eastman CI, Liu L, Fogg LF. Circadian rhythm adaptation to simulated night shift work: effect of nocturnal bright-light duration. *Sleep* 1995; 18: 399-407.
 - 41 Patarca R, Klimas NG, Lugtendorf S, Antoni M, Fletcher MA. Dysregulated expression of tumor necrosis factor in chronic fatigue syndrome: interrelations with cellular sources and patterns of soluble immune mediator expression. *Clin Infect Dis* 1994; 18 Suppl 1: S147-S153.
 - 42 Giovannoni G, Thompson AJ, Miller DH, Thompson EJ. Fatigue is not associated with raised inflammatory markers in multiple sclerosis. *Neurology* 2001; 57: 676-681.
 - 43 Bower JE, Ganz PA, Aziz N, Fahey JL. Fatigue and proinflammatory cytokine activity in breast cancer survivors. *Psychosom Med* 2002; 64: 604-611.
 - 44 Schubert C, Hong S, Natarajan L, Mills PJ, Dimsdale JE. The association between fatigue and inflammatory marker levels in cancer patients: A quantitative review. *Brain Behav Immun* 2006; 21: 413-427.