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„Ocular indicators for sleepiness during night-time driving“

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1 INTRODUCTION

1.1 The phenomena of sleepiness – a grey area between wakefulness and sleep

Sleep research has flourished since the middle of the last century with the ability to record electrical activity in the brain with electroencephalograms. In 1968 standards to categorize the different stages of sleep were published early on by Rechtschaffen and Kales. Nonetheless, adequate criteria to evaluate different levels of awakeness or alertness close to sleep do not exist yet. In literature descriptions of low-level vigilance like 'drowsiness', 'sleepiness', and 'fatigue' are often used synonymously. According to Weeß et al. (2000) the first two are expressions of different conditions: The term '*sleepiness*' describes the degree of alertness, controlled directly by the central nervous system. The sleep propensity is high to the degree that any organism would fall asleep if possible. Monotone situations produce stimuli to fall asleep. This is in-line with the definition of Carskadon and Dement (1982) describing sleepiness as physiological drive to fall asleep. In contrast, *drowsiness* as a state primarily characterizes psychical exhaustion. Cognitional and perceptional processes are frequently influenced by drowsiness. It is rather connected to intra-psychical processes like the perception of stress and less to circa-/ultradian processes. When drowsy, the person does not easily fall asleep even if possible. In addition, monotonous situations are not a stimulus to sleep when drowsy (Weeß et al., 2000). Johns et al. (2008) made a distinction between the two terms 'drowsiness' and 'fatigue'. They described the former as an intermediate state between alert-wakefulness and sleep without distinguishing it from sleepiness. Fatigue, in contrast, was charcterized as a fluctuating state of reduced awareness accompanied with impaired psychomotor performance. The discrepancy among interpretations may actually be semantic. There were linguistic differences, as the study of Weeß and colleagues was written in German. Here 'Müdigkeit' translates closer to 'drowsiness' and 'Schläfrigkeit' to 'sleepiness'. In this study I have opted to define a strong drive to fall asleep as 'sleepiness' i.e. caused by sleep deprivation or circadian sleep pressure. Other reduced vigilance or wakefulness states due i.e. to a lack of activating stimuli, I will define as 'drowsiness'. 'Fatigue' I will use for levels of low vigilance without specification whether based on drowsiness, sleepiness or both phenomena.

A common model to explain fluctuations in sleepiness is the *two process model* from Borbély (1982). It posits that the interaction of a sleep-/wake- dependent homeostatic process S and a circadian pacemaker process C (presumably located in the SCN) generate the timing of a consolidated phase of sleep during night and of wakefulness during day. While process C is a periodical process describing the dependence of sleepiness on time of the day, the homeostatic process characterizes the increase of sleep propensity due to time awake. It can be reduced simply by sleeping.

Wakefulness is characterized by consciousness of the self and the surrounding as well as information-uptake. According to the model of Posner & Rafal (1987) five attention-related aspects describing this condition can be distinguished:

Alertness has a tonic and a phasic temporal base: *Tonic alertness* describes the degree of alertness an individual expresses from one day to another. It is mainly regulated by the circadian rhythm, less by self-control. *Phasic alertness* is characterized by alternations in alertness such as temporary higher activation of the central nervous system caused by certain stimuli.

Attention, in contrast to alertness, is controlled consciously. *Selective attention* describes the skill of an organism to focus on a relevant stimulus, chosen in a complex environment. *Divided attention* is used for controlled, automatic processing of information, which includes serial and partial information processing. The third aspect of attention - *Vigilance* describes a stage of generally sustained attention.

The term *arousal* usually simply describes a non-specific activation of the cortex based on the sleep-wake-stages (Oken et al., 2006).

1.2 Fatigued driving

The frequency of sleepiness being the major cause of road accidents is difficult to ascertain. It is not easily documented like other potential sources such as alcohol. Oftentimes the attentional state of the driver is unknown (Chipman & Jin, 2009). Due to the lack of a clear definition for sleepiness, it is partly not even considered to be a potential cause of an accident or if so, only when all other possibly contributing factors have been excluded (Mac Lean et al., 2003). Hence, in statistics of traffic accidents only a small number are attributed to sleepiness like the 4% reported by the US Department of Transportation (Dinges et al., 1998). It is, however, likely that sleepiness actually accounts for at least 20% of road crashes (Connor et al., 2002, Akerstedt, 2010; Mac Lean et al., 2003). Field studies in which the driver as well as the surrounding traffic situation were video-recorded (Hanowski et al., 2003; Klauer et al., 2006) showed that the drivers' sleepiness could be identified as the main cause of near-accidents.

Another affirmation of sleepiness-caused accidents is the existence of consistent temporal peaks in accident risk. Folkard (1997) has referred to these periods of the day as 'black times'. Evaluating reports of accidents he found a peak at 3 am and a plateau between 2 and 4 am when sleep propensity following the circadian rhythm is highest. This inevitably lowers an individual's ability to cope with driving. A second factor mentioned that causes decreased vigilance was the time on task. It does increase linearly with time but there is a maximum in risk after 2 to 4 h. According to his theory it then decreases and a similar high accident risk occurs again after 12 h of time on task. A final factor contributing to sleepiness and risk is certainly sleep pressure due to deprivation, as described already in the two process model of Borbély. Lack of sleep has also been shown to increase accident risk (Connor et al., 2002).

1.3 Detection of fatigue during driving

Falling asleep unwillingly during driving is probably a more complex phenomenon than the voluntarily wake-sleep transition. Noguchi and colleagues (2009) introduced a two dimensional *arousal-states-model* to explain it. In it, two opposing components: the drive to fall asleep and the motivation to stay awake are set against one another. The authors' rationale is that vigilance changes during driving are too complicated to be assessed reliably with one-dimensional approaches, as it is common i.e. in most subjective evaluations. In order to actually describe the patterns of change, different types of psychological assessment have been done. The measurement of behavioural and physiological correlates of sleepiness in the particular situations of driving that demands special technical requirements and must not interfere the driving task is still a challenging topic. In the following the three common approaches are described and discussed with regard to their use during driving.

1.3.1 Subjective methods

Scales and questionnaires are a popular method to rate the vigilance state of a person. In general, two approaches exist: The focus of the evaluation can be on the current perception of sleepiness (sleepiness as state characteristic; i.e. Karolinska sleepiness scale) or the general perception as component of daily life (sleepiness as trait characteristic; i.e. Ephworth sleepiness scale). One difficulty in the self-assessment of sleepiness is the susceptibility to motivational and environmental influences. A test is easily manipulated by both the subjects' intention and also the over- or underestimation of the own situation (Curcio et al., 2001). The ability to express feelings and report fatigue adequately decreases in particular with increasing sleepiness (Brown, 1994) as gaps in consciousness occur. This gradual slip in alertness is difficult to perceive by the sleepy person itself (Kleitman, 1963).

1.3.2 Behavioural changes

An objective measure of fatigue is mirrored in *performance decrease-measures* of behaviour. Driving performance requires maintained attention and cognitive capacity to cope with the demands of a driving situation like appropriate decision-making; the perception of relevant cues and controlled motor-activity (Mac Lean et al., 2003). With increasing fatigue cognitive skills like perception, judgement, reasoning and decision-making generally slow down. These processes are measurable in more frequent lapses in behavioural response, memory deficits and a prolonged reaction time (Dinges and Kribbs, 1991). Hence several tests have been developed to analyse fatigue on the base of decreased performance in psychomotoric and cognitive tasks. The former includes mainly tasks with acoustic or visual reaction times where tracking and tapping tasks requiring coordination of movements are employed. Alternatively, tests exist with the examination of cognitive skills like attentional memory or logical responding tasks (Curcio et al., 2001). Inspired by variables that have been found to be sensitive in these tests, performance-loss measures were developed for diverse activities like flying (Morris & Miller, 1996) or driving (Wijesuriya et al., 2007; Campagne et al. 2005). For instance, the approach to qualify driving performance and steering behaviour, was to

analyse the position of the car on the road. Here reaction times to adaptation have been used. In all these publications, however, the parameters have been monitored to correlate with fatigue. There is no information on how they might be used as a lapse-alert system.
Changes in facial expressions, mannerism and body posture, yawning, sore and tired looking eyes have also been proposed to be fatigue-caused changes on the behavioural level. Wijesuriya et al. (2007) reported the assessment of these variables as reliable indicators of sleepiness. Still, in another study of the National Highway Traffic Safety Association (Dinges et al.,1998), a monitoring device for the head position was used to test these hypotheses. The assumption was that the control of head movements decreases due to a loose of muscle tone in the neck with increasing fatigue. Unfortunately, the results showed high inter-subjective differences. Consequently, there are no clear criteria yet to classify sleepiness-associated changes in these variables. An exception is 'PERCLOS' based on the scoring of sleepiness by the rate of eye-lid closure with fairly precise criteria facilitating high inter-rater-reliability (see section about the eye).

1.3.3 Physiological measurements

The most common methods to detect changes in alertness on *the* physiological level are by means of pupillometry and polysomniographic measurements. The former will be dealt with in the section about the eye. Polysomniography (PSG) is the technique used most often to document sleep. It takes diverse physiological measures into account to detect the onset of sleep, arousals during sleep and the categorization of sleep stages. It is a combination of electro-encephalograms (EEG) measuring brain activity, electro-oculograms (EOG) detecting eye activity and the electro-myograms that record the muscle tone (Iber et al., 2007). These parameters are often combined with further variables like heart rate, peripheral body temperature, respiration and aeration of the blood. The first two mentioned variables increase with sleepiness (Wijesuriya et al., 2007), the others are rather included as artefacts in medical investigations. The EEG is the only method that precisely characterizes sleep onset. In particular the occurrence of alpha (8-12 Hz) and theta (4-8 Hz) rhythms in brain activity indicate a physiological state of lower vigilance or sleepiness in awake and active people (Curcio et al., 2001). Probably the most reliable measure of sleepiness is to measure the latency to fall asleep with a latency test. The *Multiple Sleep Latency Test*, is the most wide-spread sleep propensity test and was developed by Carskadon and Dement (1982). The times between lights and the first or second sleep stage (evaluated by PSG recordings) are the markers. Two variations of this test with the opposite instruction to stay awake are the *Repeated Test of Sustained Wakefulness* and the *Maintenance of Wakefulness Tests*. With regard to the situation during driving it is obvious that alternative methods to document vigilance and sleepiness must be developed.

1.4 The eye as indicator of fluctuations in vigilance

During ontogeny the eye and the central nervous system develop from the same substrate and the functions of both components are inseparably connected (Morris & Miller, 1996). The coordination of neuronal activity of the brainstem and cortical areas is reflected in oculomotoric functions, which is only partly controlled voluntarily like i.e. accommodative focus and gaze direction. Oculomotoric characters like those involved in pupillary activity, ballistic eye- and eye-lid movements in contrast are uncontrollable and hence candidates for tracking fatigue-related neuronal dysfunction (Morad et al., 2009).

Most of the methods mentioned in the previous section were not suitable for a use during driving: Self-evaluation alone probably is not sufficient and there is no device yet to analyse driving performance in a manner to warn the driver properly about fatigue. The common physiological measurements of fatigue have been developed for research in the laboratory and are based on elaborate equipment. Due to the small size, easy handling and attachment in the car eye-tracking systems have, in contrast, gained popularity and are to a certain extent already commercially available. They may represent an reliable alternative to the EEG - the traditional gold standard of sleep research - to investigate fluctuations of wakefulness (or 'wake-stages') but validation is lacking.
As background information changes in ocular activity associated with fluctuations in vigilance as well as the most common methods to detect these are summarized below.

1.4.1 Fatigue associated changes in ocular activity

Pupil oscillation

During wakefulness pupils are broadly open and the diameter constant. Under hypoactive conditions they become contracted and the diameter fluctuates. These typically slow and spontaneous oscillations of the pupil below 1 Hz, with an amplitude of some mm, are referred to as 'fatigue waves'. With rising sleepiness they occur more frequently, the amplitude increases, whereas the average width decreases (Warga, 2002).

Eye-movements (EM):

Rapid eye-movements (REM)/saccades & periods without eye-movements (NEM)

In literature different terms have been used for fast ballistic eye-movements occurring during scanning. They are referred to as 'saccades'. Unfortunately, there are no clear standards yet in wakefulness for the categorization of EM like those for sleep. However, fast EM during wakefulness meet the same criteria of irregular high deflections in the EOG of those detected during REM-sleep apart from velocity that is slightly higher in awake (Fukuda et al., 1981). Hence I have used the same term: rapid eye-movements (REM).
Schleicher and colleagues (2008) i.e. investigated ocular activity in a driving-like condition and reported an increase in duration coupled with a decreased velocity of REM as characteristics of gradually augmenting fatigue. Several studies (i.e. Campagne et al., 2005;

Hyoki et al., 1998; Morris & Miller, 1996; Santamaria & Chiappa 1987) have proposed that a decrease of REM-events was a sign of fatigue. Morris & Miller (1996) assumed that these changes were based on a general slowing of the arousal system, which produced in less visual scanning, slower eye movements and more gaze fixation without EM. Similarly Schleicher and collegues (2008) reported an increase of gaze fixation periods (NEM) longer than 900 ms. This was related to staring behaviour that the authors categorized as 'driving without awareness'. Santamaria & Chiappa (1987) found a disappearance of EM in 2/3 of their subjects that were fatigued. Campagne et al. (2005) pointed however out that due to visually demanding tasks the short-term ocular activity can be changed. In addition they reported an influence of age: In young participants the drop was less pronounced.

Slow eye-movements (SEM)

SEM are associated with the onset of sleep (sleep stage 1 according to the standards of Rechtschaffen & Kales, 1986). Cajochen et al. (1999) described SEM as loss of oculomotoric control related to severe sleepiness. The authors reported a temporal relationship between the endogenous melatonin rhythm and the occurrence of SEM. The frequency of SEM events increased around the onset of melatonin-secretion. Melatonin is as 'synchronizator' of circadian physiological processes. Hence SEM can be used as markers for process C of the two compound sleep model.

Spontaneous eye-blinks (EB):

EB are probably the most popular ocular characteristic studied with fluctuations in vigilance. Numerous reports have been published regarding this feature. Spontaneously occurring EB are taken as an indicator of drowsiness. They do not have an identifiable eliciting stimulus and hence differ from reflexive blinks (protective response), voluntary blinks (an answer on request) and self-induced blinks and eye-closure at the onset of sleep. Beside the need to humidify and cleaning the cornea the following factors can influence EB characteristics: (1) environmental conditions (i.e. air quality; Stern et al. 1984), (2) the engagement in tasks (Campagne et al., 2005; Stern et al., 1984), (3) age and gender (Caffier et al., 2003), (4) emotion (Stern et al., 1984), (5) the coordination with eye-movements (Stern et al., 1984), (6) the circadian rhythm (Barbato et al., 2000; Cajochen et al., 1999), (7) sleep deprivation (Barbato et al., 1995; Wijesuriya, 2007) and finally (8) time on task (Campagne et al., 2005; Morris & Miller, 1996).

The use of EB in driving is altered by the visual demands of the task. The result is a suppression of blink activity (Campagne et al, 2005; Fukuda et al., 2005). On the other hand, there is also the sleepiness associated increase (Recarte et al., 2008; Fukuda et al.,2005). Fukuda et al. (2005) found a much higher *frequencies* in inter- stimuli periods than during times when a stimulus was expected. EB occurring in relatively unstructured bursts probably were indicative changes in the processing mode after a period of inhibited blinks. They can thus be attributed to a momentary loss of attention - a breakdown of compensatory activation trying to maintain performance. Furthermore it has been reported that with rising fatigue the *duration* increases (i.e. Caffier et al., 2003). The *amplitude* of blinks in fatigue is thought to decrease, probably due to the drooping upper lid that causes the typical 'sleepy

eyed look' (Morris & Miller, 1996). The upper lid that actually performs the EB motion becomes also slower in fatigued persons as reported by Johns and colleges (2007). The occurrence of 'microblinks' is another phenomena associated with fatigue. Fukuda et al. (2005) assumed that they were caused by a decreasing ability to control the inhibition of blinks. EB have therefore been used to document different sources of fatigue: Blink-frequency reflects as well the *circadian process* by an increase during day till the evening, peaking between 8 – 9 pm (Barbato et al., 2000; Cajochen et al., 1999) as an effect of the *homeostatic process* by a rise in sleep deprived subjects (Barbato et al., 1995; Wijesuriya, 2007) and a response in the same manner to progressing *time-on-task* (Campagne et al., 2005; Fukuda et al., 2005; Morris & Miller, 1996). Unfortunately, due to the sensitivity of EB to diverse factors and high inter-individual differences as reported in several studies (i.e. by Caffier et al., 2003; Johns et al., 2007; Fukuda et al., 2005) it is probably not possible to base the detection of fatigue on this character alone.

Circadian-course of ocular variables:

The process of wake-sleep transition is reflected in changing patterns of ocular activity presumably indicating the level of fatigue. I.e. Atienza et al. (2004), Cajochen et al. (1999) and Santamaria & Chiappa (1987) described a structured course of ocular variables: Cajochen et al. (1999) mentioned an increasing EB activity during the first 16h of wakefulness peaking between 20-24 h. It was followed by a drop of large blinks that was also confirmed by the other two studies. Simultaneously REM activity decreased as predictor for SEM-density (Atienza et al., 2004; Santamaria & Chiappa, 1987). The authors of the latter study found the occurrence of a variable period of time with NEM before the onset of SEM and falling asleep in many subjects.

1.4.2 Techniques to measure changes in ocular and blink activity

Pupillometry

Pupillometry is the most common technique to analyse fluctuations in vigilance.
In tests based on pupillometric measures, variation in pupil diameter caused by the autonomic responses to visual stimuli or fatigue (as described before) can be detected (Wilhem et al., 1998). As a control, *pupil noise* or the reaction to a light reflex is analysed. Abnormal sleepiness is connected with less pupil noise (O`Neill et al., 1996). The *pupillary unrest index* (PUI) is calculated from alternations in pupil-size in mm/min. Furthermore the *spectrum of the pupil* below 0,8 Hz that rises in a fatigued person has been used as a marker for fatigue.

Oculomotoric tests: Pursuit and fixation tests

Recently ocular tests to asses fatigue are have gained in popularity (i.e. Ahlstrom et al., 2013; Fransson et al., 2008; Hirvonen et al., 2010; Di Stasi et al., 2012; Morad et al., 2009). This is presumably due to an easy application of the technique. It enables the collection of sleep-

controls in a similar manner to those for alcohol by stopping the driver on the street. In this section, 3 common tests will be introduced. The different studies diverge slightly in performance (like type and duration of stimulus and the methods chosen to record the ocular activity) but generally the procedures are as described in the following: (The description presented is based on Ahlstrom et al, 2013):

Visual pursuit of target stimuli:

Smoot pursuit system (SPS): The subject is asked to visually follow a target moving on a computer screen as accurately as possible. Usually the measurements are performed for different velocities of the stimulus. The ratio between the velocity of the eye and the target as well as the distance between target and gaze are detected. Both characteristics where found to deteriorate with increased levels of fatigue (Porcu et al., 1998; Fransson et al., 2008).

Measurement of saccades: The participants have the task to fix targets appearing in different points of the screen as quickly as possible. In the *gap condition* each target point follows an interval with an empty screen before the appearance of the next stimulus. In the *overlap condition* the offset of the previous and the onset of the following target overlapped in time. Saccade velocity decreases with sleepiness but the method seems to be less sensitive to sleepiness than the SPS (Fransson et al., 2008).

Fixation task:

In the fixation task, participant are asked to stare for 5 min with their eyes open and 5 min with closed eyes, at the same time they receive a bright cross on a dark background. In another condition as stimulus a grey target is shown on a background changing between white and black. SEM in terms of fixation stability and the PUI (see pupillometry) can be determined.

Off-line detection of fatigue in the context of driving:

These tests obviously cannot be performed during driving but only before and afterwards (offline). The same applies for pupillometric measurements. An example for an infrared device to asses pupillometric measures as well as the velocity of saccades which is already commercially available was tested by Morad et al. (2009) in army truck drivers. The PMI-apparatus is completely automated, self contained, computer controlled, quick and easy in performance and did not impair the daily duties of the subjects. By using a fixation and a visual pursuit task pupillary diameter, pupillary constriction latency, amplitude of the pupillary constriction and the saccadic velocity are measured. From these variables an index as indicator for the level of sleepiness was calculated. The test was able to distinguish high-risk drivers. Ahlstrom et al (2013) however reported the disadvantage that the design of the off-line method used in their study itself increased the fatigue of the participants.

The main problem with offline measurements is probably the fact that vigilance changes rapidly– a phenomena that is particularly pronounced in subjects striving to stay awake (John et al., 2008). The determinant that precluded pupil-based measurements until now

from online detection of droops in vigilance during driving is the light sensitivity of the pupil oscillations like well described by Warga (2002) who tested the use of pupillometry during driving.

On-line registration of ocular activity:

Video-oculography

A video based scoring technique with precisely defined criteria (included in Dinges et al, 1998) is *PERCLOS* (= percentage of eye-closure). The device measures the proportion of time in which the pupil is covered more than 80% with the upper lid in a one-min time window When a certain threshold is exceeded a warning yields. Subjects are recorded with a low light camera angled upward and directed on the face to provide a good view on the eyes. The videos are analysed by trained scorers who use a computer program with a slider to categorize the eye-lid position from moment to moment. By graphical feedback the analysis requiring simultaneous attention on the drivers eyes and the slider is facilitated. Dinges et al.(1998) reported a high reliability of the method comparing it with 5 other methods that detect changes in vigilance tests during the performance of a psychomotor vigilance task.

Infrared-oculography

Johns and colleagues (2007, 2008) described an example for the use of photoelectric techniques to measure oculomotoric characters during driving with an infrared (IR) sensor. The device consisted of a frame for glasses with a light emitting diode fixed in front of and below the eye. At a frequency of 500 Hz brief IR pulses were directed on the lower edge of the upper eyelid. A phototransistor attached beside the diode detected the reflected IR light. Influences of environmental light could be excluded by measuring and subtracting it before each pulse. EM and EB could be detected by changing reflection-patterns.
Duration, power and timing of the IR pulses could be controlled with a microprocessor. Power supply and the output from the glasses were connected via a light cable to a processing mounted in the vehicles. By means of amplitude velocity ratios of eyelid closing, reopening and their durations, results for use with the Johns drowsiness scale (JDS) were calculated for each minute. The JDS scores were shown to be sensitive to rising fatigue, prolonged reaction times and more frequent occurrence of lapses tasks. Johns and colleagues (2008) reported the prediction of road-off events by this method with a sensitivity of~ 83% and a specifity of ~ 61% within the succeeding 15 min.

Electro-oculography (EOG)

The EOG is the classical instrument in sleep research to detect rapid and slow eye-movements. It has been employed in wake subjects with different visual-performance tasks (summarized by Stern et al., 1984) or to detect changes in ocular activity due to fluctuations in vigilance (i.e. Atienza et al., 2004; Cajochen et al, 1999; Hirvonen et al., 2010; Hyoki et al.,1998). Additionaly, it has been used in simulated driving studies (i.e. Ackerstedt et al, 2010; Campagne et al., 2005; Jammes et al., 2008; Miller & Morris, 1996; Schleicher et al., 2008) and field driving situations (Papadelis et al, 2007; Mitler et al., 1997). EOG

measurements of ocular activity are based on the changing potential between cornea (positive charge) and fundus (negative charge) when the eyeball is moving. EB are detected by the gaze shifts from the upper to the lower portion during the downward movement of the upper eyelid over the cornea. The eyelid acts as a 'sliding-resistor' creating a positive standing potential between the positively charged cornea and the fundus. Hence during the downward movement of the lid positivity is increased and contrary during refraction negativity (Stern et al, 1984).

For measuring the movements of an eye in vertical as well as in horizontal plane and additionally the amplitude of eye-blinks four electrodes equally placed around the eye are necessary (Morris and Miller, 1996). But for the simple detection of ocular- and blink-activity two staggered electrodes attached at the outer canthi of the eyes are sufficient. According to the standards of the AASM (American association of sleep medicine; Iber et al, 2007) the electrode at the left eye is secured 1 cm below the outer canthus, on the right eye 1 cm above. The additional ground electrode is usually placed on the mid-forehead and the reference electrodes on the earlobes.

To facilitate the analysis of EOG-data, standardize the evaluation and gain the results immediately it is tried to automatizate the technique by algorithms. Until now there are no common agreements between institutions about criteria of manual detection during wakefulness (Hyoki et al.,1998). Jammes et al. (2008) i.e. introduced an algorithm for blink characteristics to automatically score the EOG-signal and described difficulties caused by the great variation of EB and changes of the signal due to gaze direction: The algorithm could not distinguish whether the eyes have been closed or the subject looked down. The work on this technique is still in progress focussing on improvements of establishing the baseline and on the question, which blinks are considered as representative for fatigue.

Oculographic measurements and driving:

An advantage of PERCLOS is that it is a contact free method allowing completely free movements during driving. But Johns and colleges (2007) pointed out that the driving-off-road events occurred also with wide-open eyes, staring straight ahead that would not be detected by the criteria of this method. Head movements, and in particular sun glasses could impede the evaluation of the eye-lid activity. Furthermore environmental light could have an impact on the closure-rate. Finally the technical requirements are an impediment to its use– complex software and cumbersome hardware are necessary (Lo Castro; 2008). Johns and colleagues (2007, 2008) have developed an infrared-oculography that neither restricts the driver's vision nor interferes with driving. As IR is invisible it causes no distraction. They report as a further advantage that there are fewer inter-individual differences in amplitude-velocity-ratios and that individual adjustment was not necessary. Though Lo Castro (2008) is critical concerning a possible hazard of radiation by IR-techniques used so close to the eyes.

Most of the studies verifying these devices however have not been performed in a realistic driving situation until now but just in a simulator.

1.5 Aims of this study

The analysis of the EOG-signals during driving, presented in this thesis, was part of a project coordinated among the University of Vienna, the Medical University of Vienna, the ÖAMTC, the Section for Traffic Security and Traffic Psychology of Vienna, the AKH of Vienna, and the Institute for Sleep-Wake-Research of Vienna. In it, several possible indicators of sleepiness were examined that connected the subjective, the behavioural and the physiological levels of sleep and wakefulness. The novelty here was the analysis of the phenomena on all three levels in a naturalistic driving setting. Most of the driving related studies investigating the impact of fatigue on physiological and behavioural correlates have been performed until now in simulators.
As shown in the model (Fig. 1) we set the sleep pressure to a maximum regarding the circadian component. Subjects were not allowed to sleep in the hours before the experiment to enhance the contribution by the homeostatic process. Additionally the subjects were expected to be fatigued by the driving task itself (time-on-task) and we choose a monotone surrounding regarding the street that round for round has been the same. We assumed that the motivation to combat sleepiness on the other side would be very high as the subjects drove their own cars and were aware of possible consequences when falling asleep. This extreme condition of struggling may also affect physiological changes. The aim of this thesis and its contribution to the study was to focus on changes in the occurrence of eye-movements and eye-blink patterns. As visual input is absolutely necessary for driving interruptions in information uptake caused by blinks should be suppressed to a minimum and REM to scan the environment should occur frequently. The eyes should never be motionless for a longer time. With increasing sleepiness however REM-activity was expected to decrease, resulting in more periods without EM and EB to increase in their frequency

Furthermore the impact of a powernap on sleepiness-associated eye-movements was assessed. REM, NEM and EB were analysed visually. Trends were found, but the result did not actually support predictions from the literature. Nap did not produce positive effects on driving performance and sleepiness as assess with associated eye movements.

In line with the model described above the following hypotheses were formulated:
1. Fatigue increases are accompanied by decreasing REM- and increasing NEM- and EB-activity
2. Positive nap-effect would produce an increased REM and decrease in NEM und EB
3. Subjects that have been longer awake and slept less would have higher homeostatic sleep pressure and hence show more sleepiness-associated ocular features.
4. Based on the literature EB would be more frequent in women
5. Due to differently coping strategies and stress responses, assessed by cortisol patterns, there would be variation among subjects with regard to sleepiness and ocular parameters during the drive.

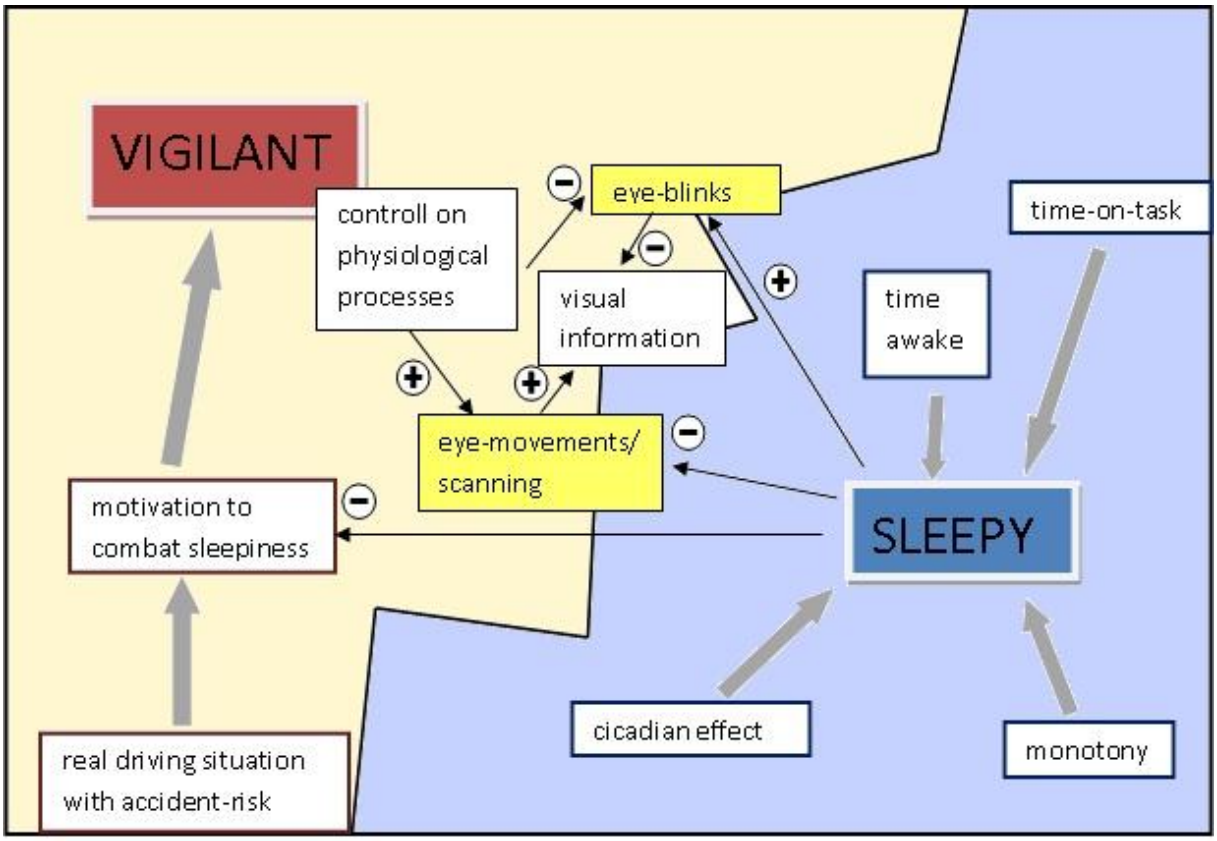

Figure 1: Different factors that contribute to (blue frame) and oppose (red frame) sleepiness and the measured physiological outcome as assessed in ocular activity (yellow).

2 METHODS

2.1 Study design and participants

The experiment was carried out on 13 weekend-nights in October/ November 2010 on a test-route of the ÖAMTC in Teesdorf, Austria (Fig. 2). The approximately 2.5 km long stretch consisted of curves, little hills as well as flat parts. For the analysis of the EOG-signal only data of an approximately 500 m long straight section of the track (marked in orange in Fig. 2) was chosen for the analyses described below. It was the most monotonous part of the stretch. 60 healthy subjects between 30 and 54 years of age, 30 men and 30 women, were paid to participate with their own cars in the experiment. Amongst the criteria for participation was the possession of the driving licence for over seven years, driving experience of 7 000 to 12 000 km per year and no previous involvement in serious accidents. These prerequisites guarantee similar driving experience among the subjects. Professional drivers were however also excluded as the subject's reaction to the task was planned to represent that of an 'average driver' and not one trained to drive at extraordinary times and for extended distances. All subjects reported normal sleep behaviour. The chronotype was established using the morningness-eveningness questionnaire (Horne & Östberg, 1976). Wake- and sleep-times as well as self-evaluated quality of the day before the experiment were determined via sleep diaries. All subjects were informed about the goals of the experiment. In line with the Guidelines for Good Clinical Practice (E6[R1], 1996) and procedures described in the Declaration of Helsinki (1964) they were asked to sign a consent form in advance for their participation and the public use of the data gathered.

Figure 2: Test area in Teesdorf; analysed section marked in orange

2.2 Study procedure

Participants were randomly divided into two groups. Half of them were allowed to take a break with the opportunity to nap after 1.5 h of driving (‘*napper*’) before completing the last 30 min. The other half (‘*non-napper’*) drove continuously for two hours. The analysed drive started at 2.00 a.m., after three training rounds in which subjects followed the instructors to get familiar with the stretch. In order to guarantee that the drivers had not slept before the experiment they were asked to be at the experimental side from 10.00 p.m. onwards. To avoid distraction through other drivers and to decrease the risk of a rear end collisions only five or six subjects were allowed to participate on a particular day. The distance to be maintained between the cars during the driving task was chosen in a manner that subjects did not see the backlights of the car in front of them. Due to characteristics of the curve-rich driving side the speed was limited to 70 km/h. The whole experimental area was monitored by trained inspectors, connected via walky-talkies to the drivers. They were able to pass instructions on to the drivers and - if it had been necessary - to interrupt the experiment. Participants always had the opportunity to abort. During the experiment the use of chewing gums, listening to the radio as well as the consumption of stimulants like coffee, coke or red bull and cigarettes were forbidden.

2.3 Equipment and measures

On-line detection during driving:
All cars were equipped with on board cameras pointing onto the driver and street and a GPS-transmitter that recorded the rough spatial positions of the car on the driver’s side. For polysomnographic recordings at least three gold cup electrodes (Grass Inc.) were used for the EEG (electrode positions: C3, F3, O1 according the 10-20 system) with A1 and A2 as the common reference. Two were mounted for the EOG, and one bipolar EMG as well as a grounding electrode (forehead) according to the AASM-criteria published in 2007 (see Iber et al.,2007).
The two EOG- electrodes were attached on the outer canthi of the eyes. To record vertical eye-movements along with the horizontal movement, the electrode of the left eye was secured one cm below the outer canthus and on the right eye one cm above. The data for the analysis of EM was produced by three different PSG devices: SOMNOwatch™, SOMNOscreen™ and an ambulant recording device of Alpha-mobil™. The data of all channels have been stored on a computer device carried along in the car.
The technical data is shown in Table 1.

Table 1: The technical data of the three PSG devices used to gain the EOG-data analyzed in this study

	SOMNOscreen™	SOMNOwatch™	Alpha-mobil™
Impedance	> 100 MΩ	> 100 MΩ	> 100 MΩ
Sampling rate	128 Hz	256 Hz	128 Hz
Resolution (digital)	16 bit/channel	16 bit/channel	13 bit/channel
Number of used channels	8 EEG	4 EEG	8 EEG
	2 EOG	2 EOG	2 EOG
	1 EMG	1 EMG	1 EMG

Off-line measurements of sleepiness and vigilance before and after driving:
The participants rated their sleepiness by means of the Karolinska Sleepiness Scale (KSS). The nap group also recorded subjective sleepiness additionally before and after the mid-drive break. Furthermore a test on attention, the AD-test, was performed ('Alphabetischer Durchstreich-Test'). The subjects were asked to find certain combinations of letters in a text under time pressure. Preciseness (missed ones and false positives) as well as speed were evaluated and scores/line of letters calculated. To estimate the stress response to the drive, changes in saliva cortisol levels from before the drive to afterwards were analysed using an enzyme immune assay system developed at the University of Veterinary Medicine, Department of Biochemisty.

Categorization of eye-movements and –blinks:
Of all data EOG-signals of only 20 subjects could be included in the analysis due to technical problems like the complete drop-out of the registration of the EOG-channels or an exclusion due to artefacts in more than 80% of the data. All analyzed subjects are listed in the appendix. The eye-characteristics were analysed visually in 30 s epochs in two rounds – first without a filter and then some weeks later in a second analysis with a low pass 20 Hz filter using the software "SleepExplorer" (EDF-viewer, Vers 1.0.0., freeware distributed by Th. Nössler, 2004). Parallel I used the software 'Arteviewer' (EDF-viewer distributed by A. Flexer et al. 1999) to label each second of the signal. The two analyses were done in a random order with visible EEG and EMG- traces on the screen during analyses to identify artefacts and body movements (Fig. 4). The amplitude of the different signals I adapted to the standardized amount of space for the signal in a manner that it was completely used when the maximum deflections of the signal occured. From this highest deflections of the signal I took ratios to estimate thresholds for the diverse categories (Tab. 2) of ocular activity.
Each single 1s epoch of the analyzed data of each round was categorized as containing eye movement (EM), no eye movement (NEM) or an eye blink (EB).

Table 2: The diverse criteria of ocular activity analyzed in this study ordered in the hierarchical process of exclusion that was used for every single 1 s epoch.

Hierarchical order & name	Criteria
1. REM (rapid eye-	Epochs containing irregular, continuous, opposed-phase movements (max 1/3 deviation in amplitude between the two EOG-channels) with a sharp peak, initial

movements)	deflection that amounted to at least 20% of the highest deflections of the respective subject with a duration shorter than 0.5 s (Fig. 3a).
2. EM (eye-movements)	Epochs containing eye-movements with a deflection of at least 10 % of the maximal one in the respective signal and that did not meet the criteria of REM (i.e. of being opposed) formed together with REM the category eye-movements (EM)(Fig. 3b).
3. EB (eye-blinks)	Epoch without EM but a single deflection lasting less than 0.5 s, preceded and followed by 0,5 s without EM (Fig. 3c). The amplitude of the deflection was highly variable both between and within subjects. The individual minimal threshold was set to approximately 20% of the largest deflections of the signal. The event occurred partly in both EOG-channels (parallel/ opposed-phase deflections) and also occasionally in only one.
4. SEM (slow eye-movements)	Epochs with no EM or EB, preceded by at least 1 s without EM that show relatively regular, sinusoidal, opposed-phase deflections that lasted several seconds (> 1 s) and had an amplitude of 20-200µV (following the conventional criteria of the AASM; Iber et al., 2007; Fig. 3d). In the segments analyzed, however, no SEM were found to occur. The absence was noted but the SEM category was excluded from further analysis.
5. NEM (no eye-movements)	Epochs that did not contain any of the described events (change in amplitude from 0 to 10 % of the signal's amplitude range)

EB and EM actually did not exclude each other; quite the opposite was true. EB were often integrated into EM - but as EB were not clearly visible parts of the EM I analysed them on the basis of their expression in the absence of other EM. Hence a hierarchical order of categorization for the analyzable single 1-s epochs emerged.

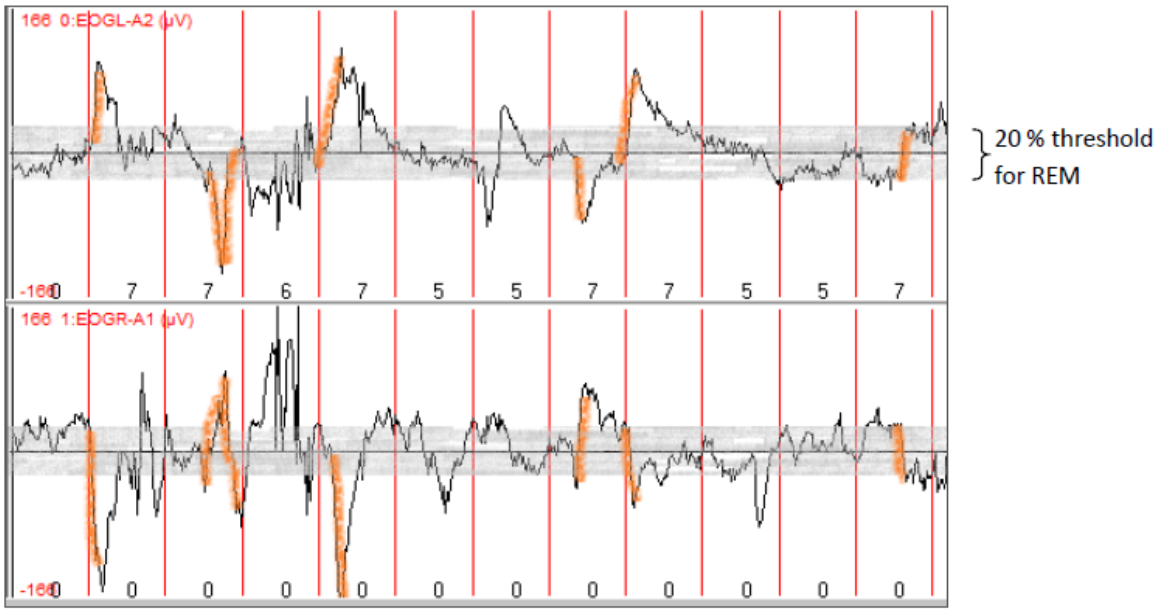

Figure 3a: Example for REM in the left and right EOG channels (EOGL & EOGR): REM marked in orange; grey shaded areas show the 20 % threshold as a criteria of REM; each channel shows the range of 332 µV, red lines divide the sequence into 1-s-epochs, unfiltered raw signal.

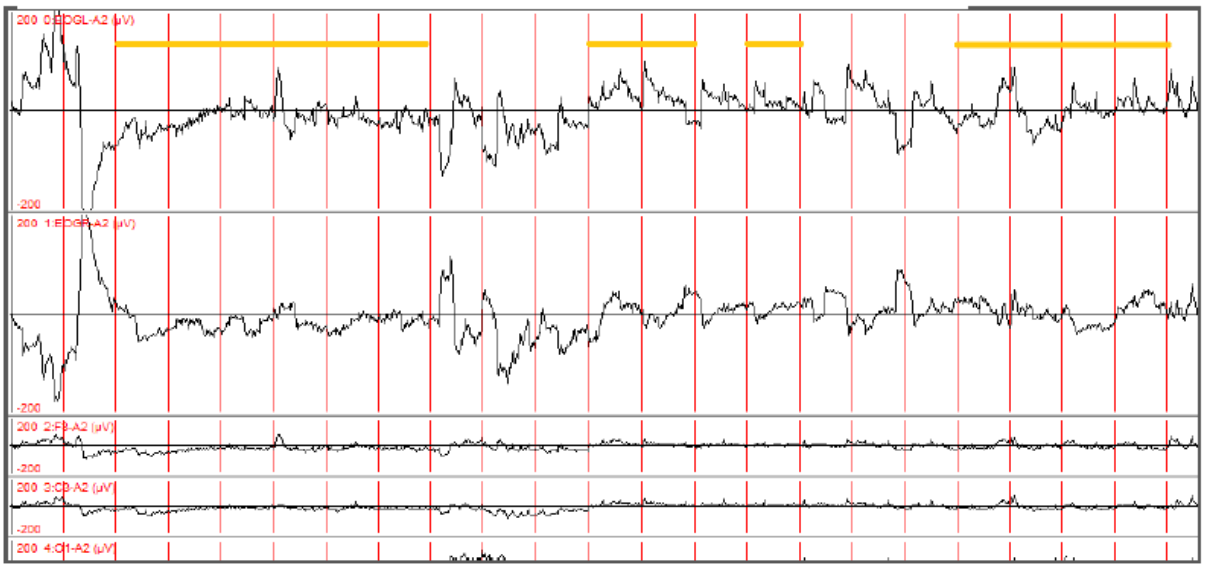

Figure 3b: An example of EM's: All epochs of the 2 EOG channels were recorded. EM- epochs that are no REM are labeled by the orange bar; each channel shows the range of 400 µV, red lines divide the sequence in 1-s-epochs, unfiltered raw signal.

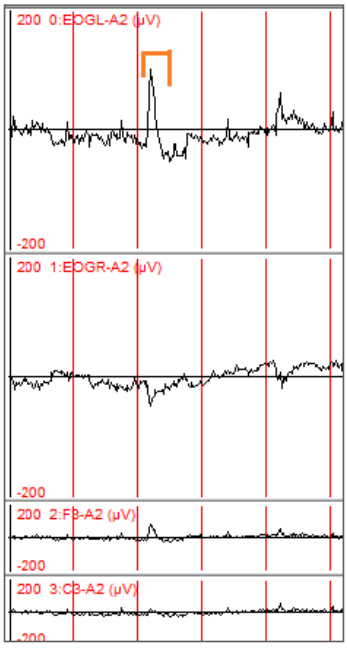

Figure 3c: An example of an EB that was more pronounced in the EOGL than EOGR. The EB is indicated by the orange lines. The frontal electrode of the EEG showed a deflection originating from the EOG event. In the central electrode of the EEG the signal is almost not visible; each channel had a range of 400 µV, red lines divide the sequence in 1-s-epochs, unfiltered raw signal.

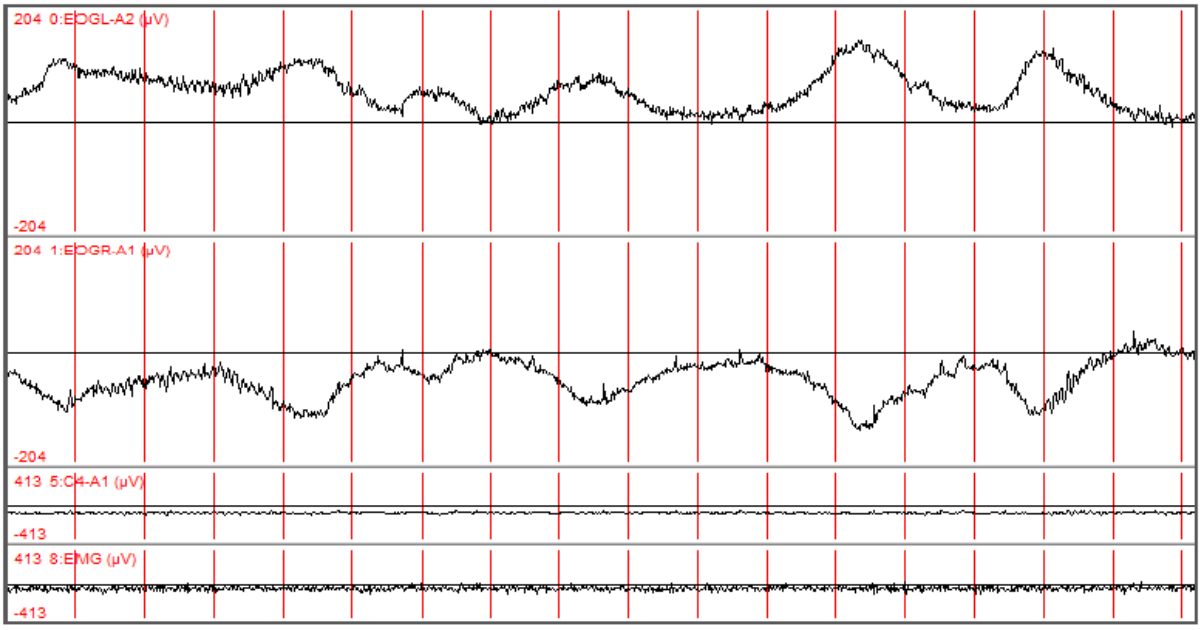

Figure 3d: The graph demonstrates continuous SEM in the EOG channels during a nap-break; each channel had a range of 408 µV, red lines divide the sequence in 1-s-epochs, unfiltered raw signal.

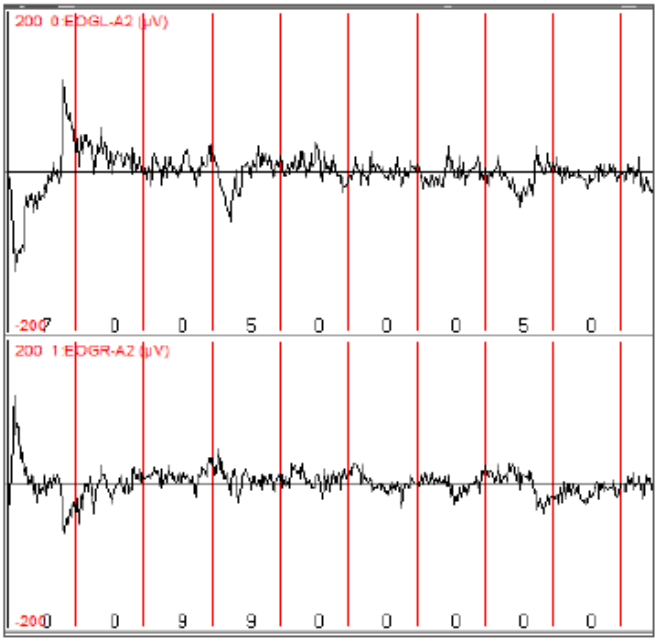

Figure 3e: NEM's in the graph are marked with zeros; the EOG-channels had a range of 400 µV, red lines divide the sequence in 1-s-epochs unfiltered raw signal.

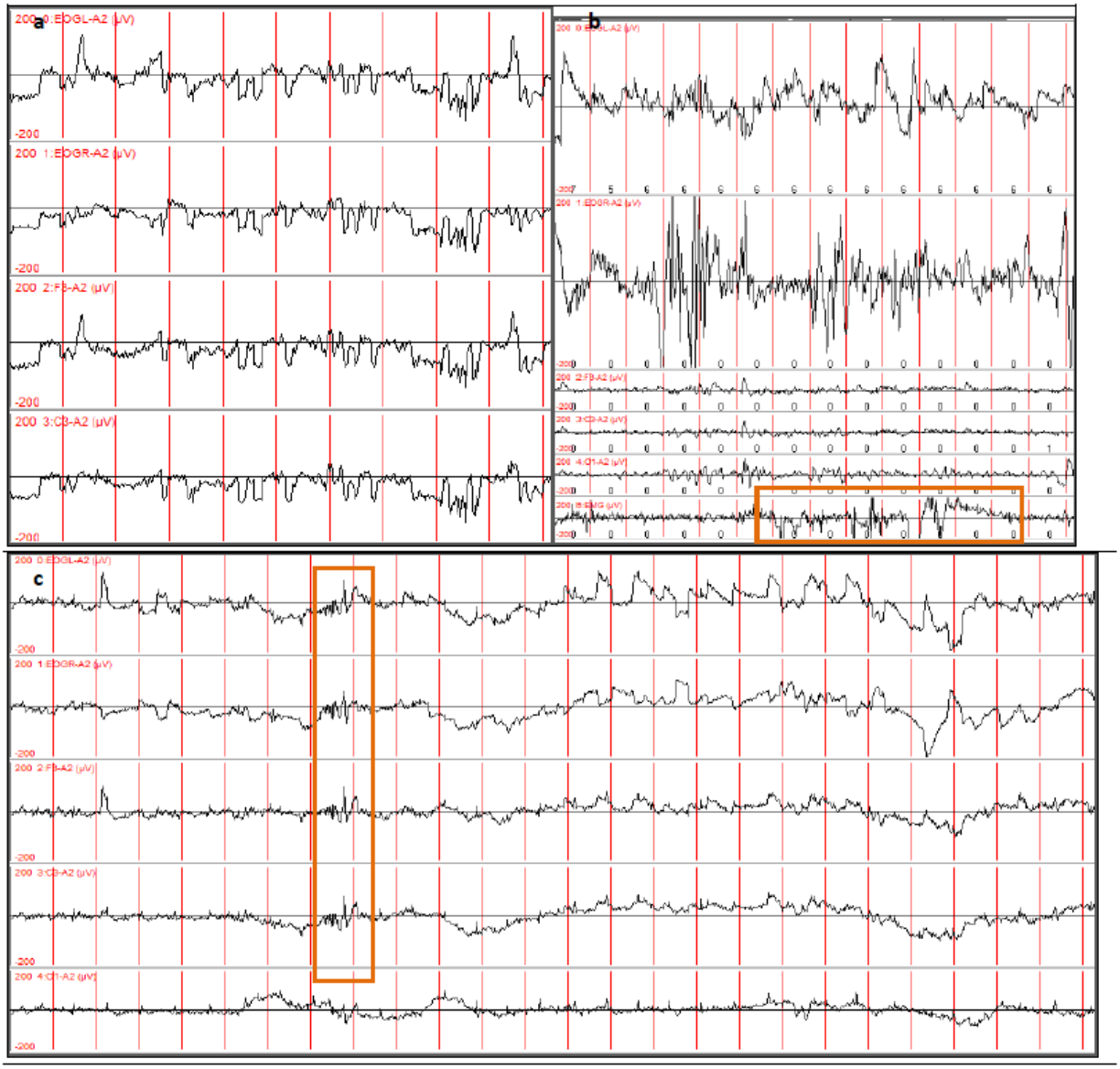

Figure 4a-c. Different types of artifacts: **a.** artifacts over all channels; **b.** body movements, visible in the EMG trace (highlighted by the orange box) produced artefacts in the two EOG channels (first two traces); **c**. short lasting artefacts, visible in all EOG and EEG channels (indicated by the orange box); channels had a range of 400 µV, red lines divide the sequence in 1-s-epochs, unfiltered raw signal.

Artefacts:

All epochs in which one of the following features was found and lasted for more than 0.5 s were excluded from the analysis. The features were:

1) deflections that occurred in several channels of the EOG and EEG with the same intensity (Fig. 4a)

2) continuous differences between two EOG-channels (Fig 4b)

3) high frequent noise that impeded an identification of the signal (Fig 4c)

4) body movements that impacted the EOG-channels (Fig 4b)

5) parallel drift of channels from baseline (Fig 4c)

Additionally, epochs containing shorter disruptions were excluded when the disrupted impeded a clear identification of the EOG-signal.

2.4 Statistical analysis

Before statistical tests could be performed, the data of the different subjects was standardized for the following reasons according to the procedure described: Depending on the driving speed each analyzed segment differed in duration and hence the number of possible epochs per round. To make the distribution of the different events between rounds comparable, within an individual and between individuals, the sum of epochs with specific parameters was divided by the total duration of the analyzed segment. All epochs that have been labelled as artefacts were subtracted from the total duration. As EB could not be identified in epochs with EM these were also subtracted from the total duration of the analyzed segment. The drivers did not pass the start and endpoints of the curve-less stretch at the same time. Hence it was not possible to compare time points and also rounds proved to be difficult as the amount of analyzable rounds differed among subjects. In order to have enough data for statistical analysis I choose time intervals of 15 or 30 minutes during the task to sum the information. The averages of all analyzed segments were calculated for each category and subject per time interval. As the different devices diverged in their time codes I did not use real time. Driving time or 'time on task' described the temporal course. Hence zero terms the subjects start of the two hour drive. The sample size of the 20 analyzed subjects was further reduced in the statistical analysis as the data sets have not been complete. To include nevertheless also the subjects with missing values I calculated the means and standard-deviations for the graphs from all analyzed data. Hence they deviate in some cases from the values shown in the ANOVA-tables.

The *effect of time-on-task* on the different eye-characteristics was tested utilizing univariate repeated measures ANOVA's. To avoid further loss of data due to augmented incompleteness of data sets in smaller time intervals 30-minute intervals were used in the statistical analysis, when necessary. The data of the first 90 minutes of both groups (napper & non-napper) were pooled and the last 30 minutes excluded. The category EM, in contrast to the others, was not spherical in a statistical sense and hence the degrees of freedom were corrected after Greenhouse Geisser. With the Friedman-Test it was possible to compare also 15' intervals if the first interval was excluded (most PSG-devices did not function initially). Also in this analysis the data of both groups were pooled and the last 30 min excluded. As EB were in contrast to EM, REM and NEM not normally distributed the Friedman-Test was preferable and used for this particular category.

The *effect of a nap* on ocular activity I tested with one way ANOVA's between the non-napping and the napping group. Data of the last 30 minutes was analyzed by the subjects' means of the whole interval as well as split in two 15 minute intervals. Furthermore the 15 minutes previously to the nap have been compared with those following the break by means of the Wilcoxon and the t-Test for paired samples within the nap-group. Only four out of nine subjects belonging to the 'nap-group' actually slept during the nap-break (according to PSG-data, categorized following the standards of the AASM, see Iber et al, 2007). Hence to

evaluate the *effect of sleep* I performed the same procedure for the groups of non-napper and sleeper.

Further effects like individual factors (age, chronotype and sex) and factors contributing to the homeostatic sleep pressure (time awake and duration of sleep on the day preceding the experiment) as well as differences in cortisol-rise and the performance of the test on attention have been analyzed by repeated measures ANOVA's like those used for investigating the time-on-task effect but with a split design including also the evaluation of between-group effects. Due to the small sample size I split the subjects just in two groups for running the ANOVAs. Furthermore I could run each analysis only with one group-factor.

Regarding the *chronotype* I had only subjects with the indifferent and the slightly pronounced morning type among the 20 subjects apart from one with the strongly pronounced morning type according to self-ratings. The data of the latter have been too incomplete for being included in the statistical analysis. In *age* I split the group at the median of 40 for having two groups with the same sample size. In the same manner I dealt with the factors *sleep duration* and *time awake* previously to the experiment. In the former the threshold between groups was 8 h of sleeping and regarding time awake 19,5 h. In the *AD-Test on attention* I grouped subjects accordingly whether they showed an increase or a decrease in maximum scores/line from the measurement before driving to the succeeding one. The level of saliva-*cortisol* abated in some of the subjects with driving time. They have however been too less to form a group. Hence I choose an increase of 2000 µl in cortisol between the two measurements as threshold and compared participants with a falling or slightly increasing level with those of a more pronounced rise in cortisol. The range of the diverse parameters is shown in the list including all analyzed subjects in the appendix.

The statistical analyses were done with SPSS (Vers. 11.5.1; 2002) and Statgraphics (Centurion XVI; Vers 16.1.11).

3 RESULTS

3.1 Ocular changes with time-on-task

Eye Movements

The pattern of change in the frequency of all EM is shown in Fig. 5a. As seen in the graph EM decreased slightly with time on task. The effect of time was apparent but not statically significant when analyzed (Tab. 3a&b; see appendix). The REM-component of EM showed the same trend although an increase was found in the last 15 min of driving compared to the previous time interval in the non-nappers as well as the nappers. In contrast, NEM showed the opposite trend. They increased a little over time on task. Statistically, when comparing the first 90 min of driving in 30-minute intervals with a one-way repeated measures ANOVA, I did not find any significant patterns in EM or its REM-component and also no pattern for epochs without any eye-movements (NEM). In this analysis, only complete data sets could be used. Hence for the analysis fewer subjects were included in the ANOVA and one can see that the means were not the same like those of the graphs in that all subjects have been included. In the analysis, however, the differences among subjects was shown to be significant (Tab. 3a). As an additional test, I compared the pattern using smaller 15-minute intervals and the Friedman-Test. Here again the temporal patterns of the three categories were not significant over time-on-task (Tab. 3b).
SEM have not been detected during the participants were driving.

Eye Blinks

The pattern of eye blinks (EB) over time-on-task is shown in Fig. 5b and the statisticak analysis in Tab. 3a&b (see appendix). Here the pattern was more complex than that we had been seen for EM. There were many EB in the first 15 min on task. The number then dropped in the next 15-minute interval and thereafter increased gradually until the end of the drive. Despite of these trends, neither in the r-ANOVA nor in the Friedman-Test the time showed a significant effect of time-on-task for EB. As EB were not normally distributed the Friedman-Test was actually the test of choice. Lastly, as with the EM-data the GLM analysis showed a significant effect between subjects in the temporal pattern.

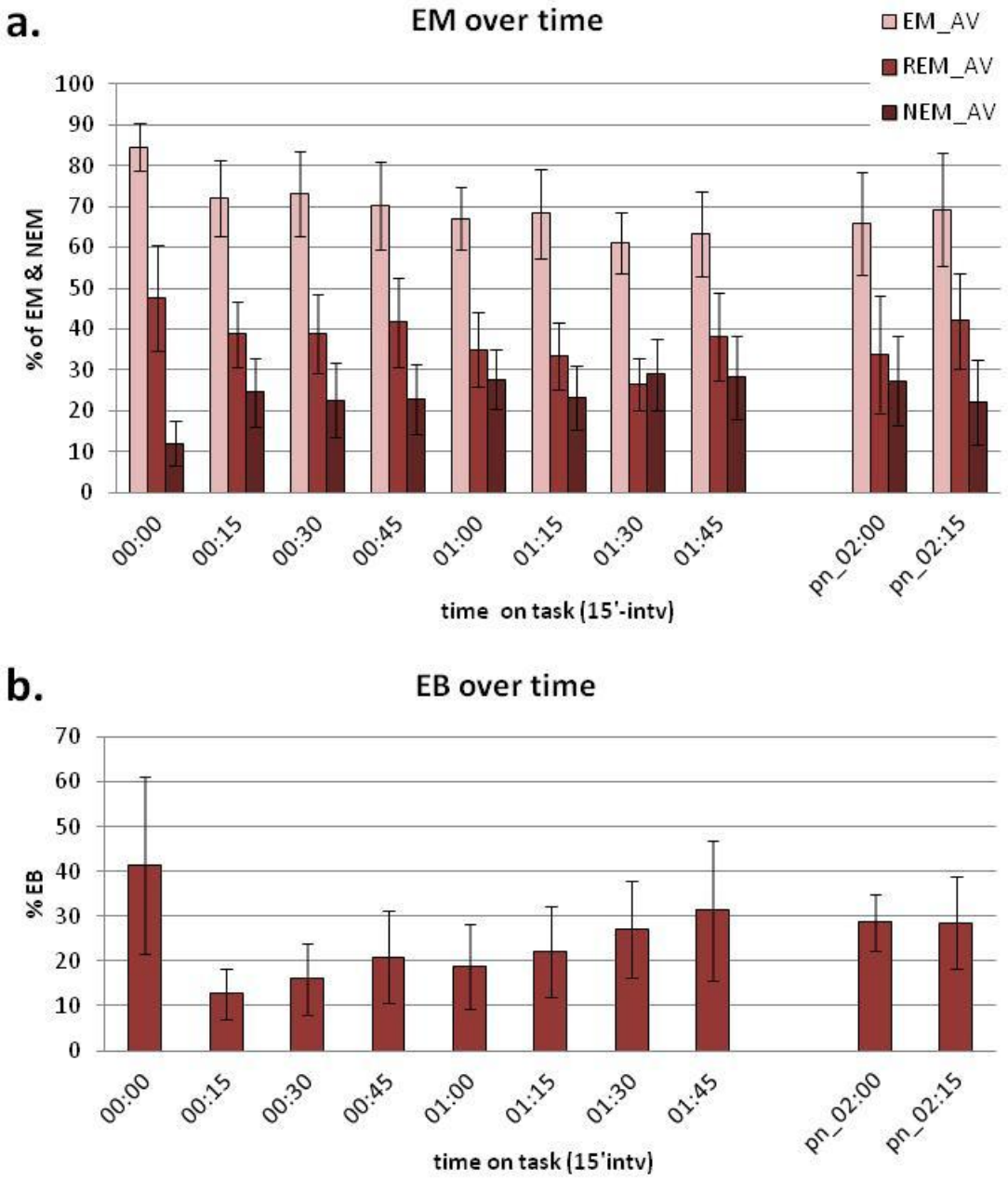

Figure 5a & b: Variation of ocular activity with driving time (mean and standard-deviation) in 15-min intervals. **a**. EM (eye-movements; brightest red), the EM-compound REM (rapid eye-movements; medium red) and NEM (no eye-movements; darkest red); **b**. EB (spontaneous eye-blinks); from 1:30 to 2:00: only non-napper; from 2:00 to 2:30 only napper; pn =postnap; first 90 min of driving; the data of all analyzed subjects is included.

3.2 Effect of a nap and sleeping

Napping:

The potential effects of having a break with or without sleeping during the driving task are shown in Fig. 6a-c and Tab. 4a-d (see appendix). The break did not seem to have a pronounced impact on the sleepiness-associated EM and EB. Neither comparing the last 30 minutes of driving of both groups with one-way ANOVA's in one interval (Tab. 4a) or when splitted in two 15-minute intervals (Tab 4b&c) nor when I compared the ocular activity with a T- and a Wilcoxon-Test within the napping group of the 15 minutes before and those after the nap (Tab. 4d). In the latter mentioned analysis the only trend occurred in EB that tended to increase but not properly significant (Wilcoxon: Z = -1,48; p = ,14).

Effects of Sleep:

Comparing the four out of nine subjects of the napping group that slept (according to the PSG data) with the non-napping group I also did not find any difference like shown in Fig. 6a-c and Tab. 5a-d (see appendix). Finally, also the comparison of the pre- and post-nap intervals (15 min) within this sub-sample did not demonstrate any differences or clear trend in the three analyzed categories of ocular activity.

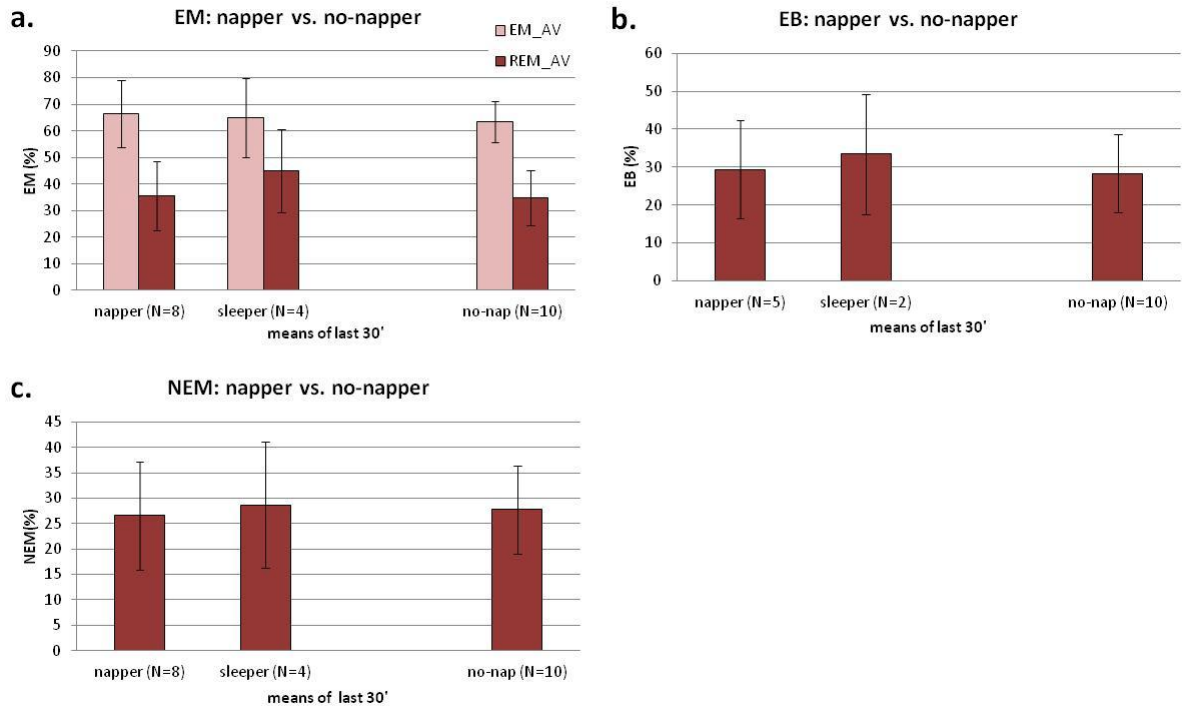

Figure 6a-c: Ocular activity (mean and standard-deviation) of the last 30 min of driving after the 30-min-break (napper), a break with sleeping (sleeper) and after 90 min of driving without break (no-nap); **a.** EM (eye-movements; bright red) & the EM-compound REM (rapid eye-movements, dark red); **b.** NEM (no eye-movements); **c.** EB (spontaneous eye-blinks); the data of all analyzed subjects is included.

3.3 Impact of individual characteristics (age, chronotype, sex)

None of these three characteristics were found to have a significant effect on the temporal patterns of REM, NEM and EB within subjects or between subjects as shown in Tab. 6a-c (see appendix). The only tendency, that can be described occurred regarding the chronotyp (Tab 6a). In order to demonstrate this trend, the ocular activity of the two classes of chronotype that have been represented in the analyzed data have been plotted in Fig. 7a-c. Means of REM, and EB were slightly higher in subjects with the indifferent chronotype (CT3) in all three 30-minute intervals than those of the weak morning types (CT4; r-ANOVA between groups: REM: $F_{(1)}$= 3,19; p = ,10; EB: $F_{(1)}$= 2,84; p = ,13). In EB the difference was most pronounced in the first interval. In the subsequent hour the levels of EB have been more similar between groups. In CT4 drivers EB tended to increase. Variation in particular in CT3 drivers was however high regarding EB. NEM tended to occur more often in CT4 (r-ANOVA between groups: $F_{(1)}$= 2,28; p = ,16).

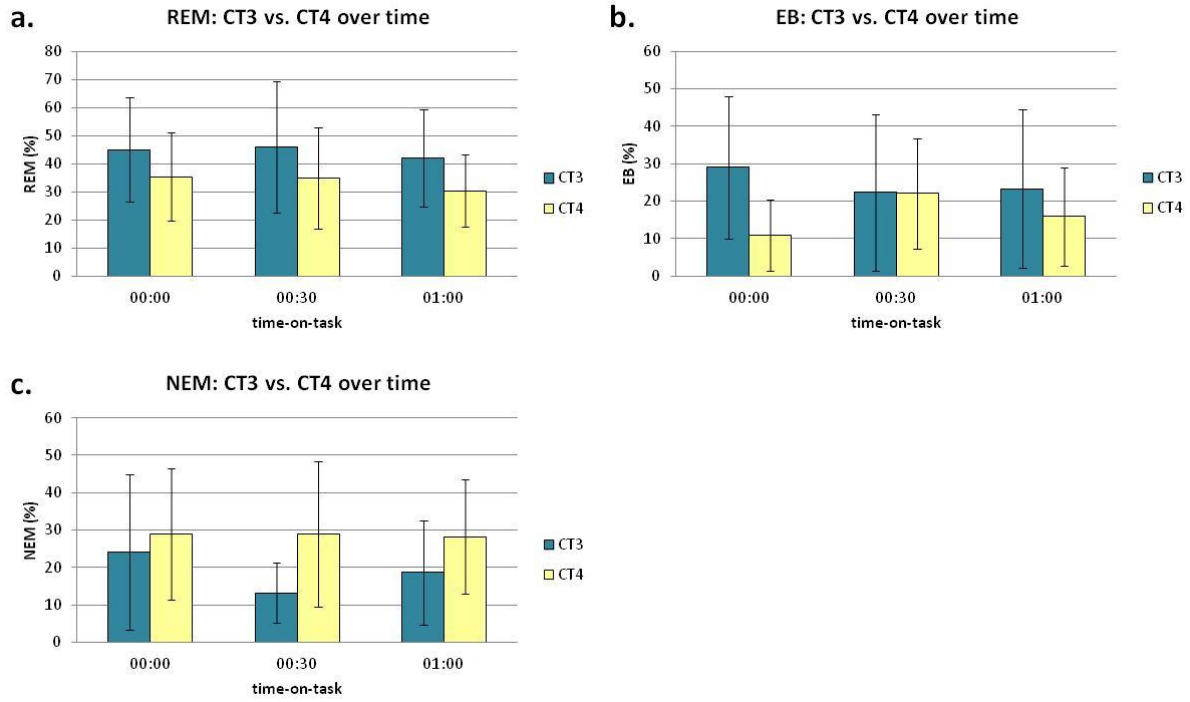

Figure 7a -c: Comparison of the ocular activity (mean and standard-deviation) between the two represented chronotypes (CT): indifferent type (CT 3) and weak morning type (CT 4) over the first 90 minutes of driving time. **a**. REM (rapid eye-movements), **b**. EB (spontaneous eye-blinks) and **c**. NEM (no eye-movements); the data of all analyzed subjects is included.

3.4 Impact of the homeostatic sleep pressure (time awake and sleep-duration)

This analysis was run in order to examine the potential influence of sleep and wake patterns before the nocturnal drive on sleepiness associated ocular activity. Sleep duration of the

previous night and time awake before the onset of the drive differed between subjects (see list of participants in appendix). Correspondingly the parameters were found to impact at least partly the temporal patterns of ocular activity during the drive as shown in Tab. 7a&b (see appendix). In the analyses two classes of previous sleep durations, more or less than 8 hours, and wake periods more or less than 20 hours, were compared.

Wake duration:

In REM there was a clear effect of time awake regarding the occurrence of activity over time ($F_{(2)}$ = 7,11; p = 0,005; Tab. 7a). As graphically shown in Fig. 8 subjects that had been awake less than 20 hours had the highest mean REM-activity in the first 30 minutes of driving followed by a gradual drop in the second and third period. In contrast, those subjects who had been awake longer began the drive with a lower mean of REM frequency that was increased in the second time period. There were no clear differences in EB and NEM that could be related to previous time awake.

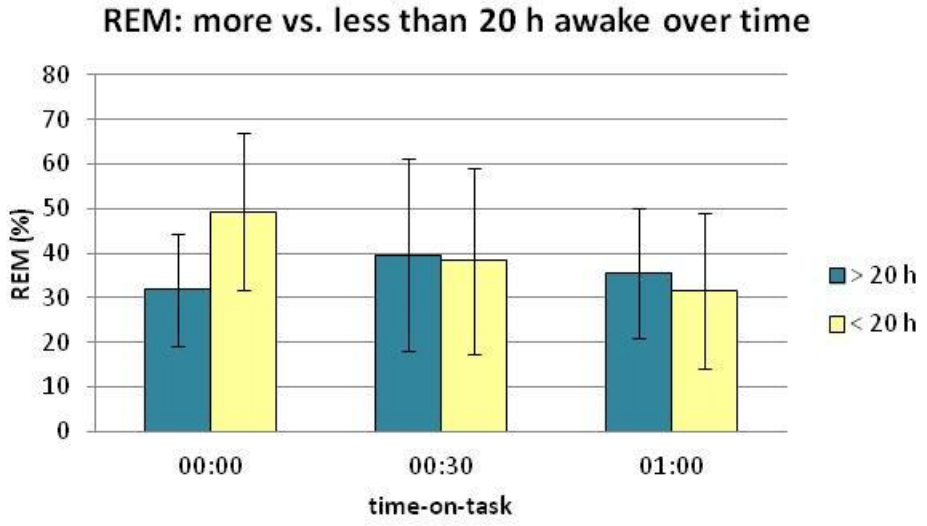

Figure 8: Comparison of REM frequencies (mean and standard-deviation) in participants with wake durations longer (blue) and shorter (yellow) than 20 hours over time; the data of all analyzed subjects is included.

Sleep duration:

Previous-night sleep duration did not have a significant effect on the frequency of ocular activity (Tab 7b & c; see appendix). A trend, was however present in EB (r-ANOVA, wake-duration*time: $F_{(2)}$ = 3,26; p = ,065). They occurred more frequently in the first time-interval in the group that slept less than eight hours. The first 30 minutes were followed by a drop in this group and a slight increase. In the group that slept more, EB gradually increased over the first 90 minutes of driving producing a recognizable different trend in the data.

3.5 Changes in saliva cortisol versus ocular activity

Changes in the cortisol pattern between the measurements before and after driving have been analyzed as they possibly reflect differences in stress responses combined with the circadian pattern of adrenal activity. As shown in Tab. 8 (see appendix) participants with a weaker increase in cortisol (< 2000 µl) or even decreasing levels had significantly different temporal patterns in REM-frequency than the group with a stronger increase in cortisol (r-ANOVA, time*cortisol: $F_{(2)}$ = 12,07; p = ,000). In the former group there was less REM in the first 30 minutes of driving. The mean of the second period was then higher. In the third period the levels decreased relative to the second in both cortisol groups. This tendency is graphically illustrated in Fig.9. There were no differences in the NEM and EB associated with the two groups diverging in the change of the cortisol level over driving time.

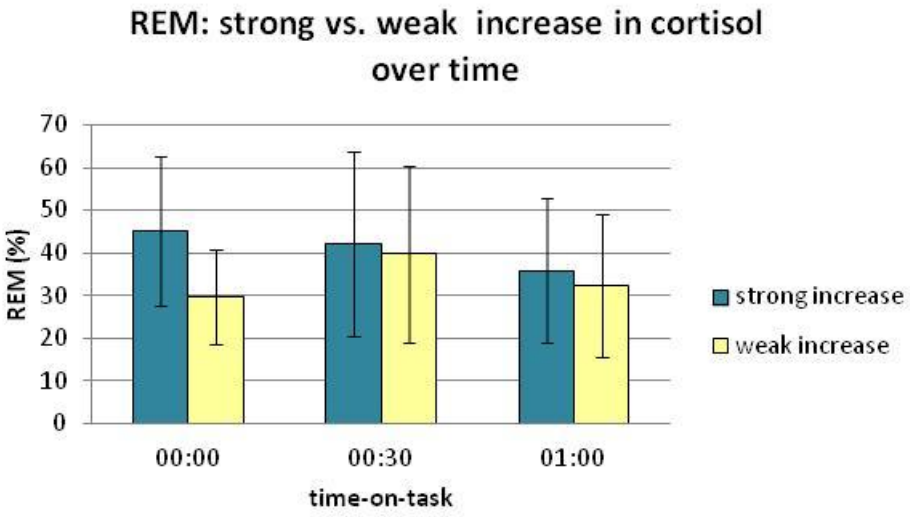

Figure 9: Comparison of REM frequencies (mean and standard-deviation) in participants with a strong cortisol increases over the night-drive (blue) and those with a less-pronounced one (yellow); the data of all analyzed subjects is included.

3.6 Performance in the AD-test on attention versus ocular activity

The performance of the AD-Test as measurement of attention decreased significantly (paired sample T-test, $T_{(57)}$= 4,33; p = ,000) from the performance before to the one after driving. This indicator of increasing sleepiness should be compared with ocular activity. The ocular activity pattern of individuals that did not change or even improved their performance was compared with the ones whose performance worsened. In Fig 10a and Tab. 9 (see appendix) one can see that NEM occurred in a different pattern over time between the groups (r-ANOVA, time*AD-performance: $F_{(2)}$ = 3,65; p = ,041): Drivers with a decrease in performance had more NEM in the first 30 minutes of driving than in the following hour and compared to the other group. Another effect that marginally failed to be significant was found for EB (r-ANOVA, AD-effect: $F_{(1)}$ = 4,70; p = ,062) as shown in Fig 10b. Here, levels were higher in all three 30-minute intervals in the group with decreased performance

throughout the drive. A slight trend (r-ANOVA, time*AD-performance: $F_{(2)}$ = 2,12; p = ,14) occurred in REM-activity that tended to be more pronounced in subjects with equal or improved performance in the first 30 minutes followed by a drop. In the other group the activity augmented resulting in similar means after the first 30 minutes.

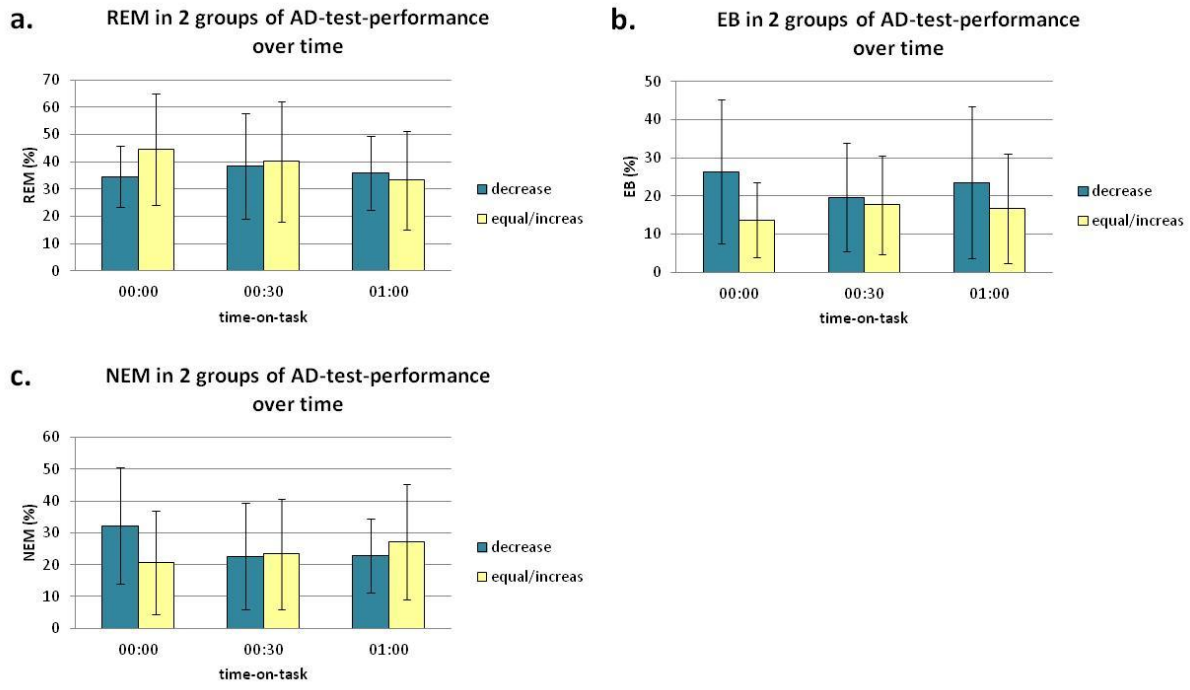

Figure 10a-c: Comparison of the ocular activity (mean and standard-deviation) between subjects with decreasing (blue) and increasing/equal (yellow) performance in the AD-test on attention over the first 90 min of driving time. **a**. REM (rapid eye-movements), **b**. EB (spontaneous eye-blinks) and **c**. NEM (no eye-movements); the data of all analyzed subjects is included.

4 DISCUSSION & CONCLUSIONS

Variation of ocular activity with time-on-task

Means of EB and NEM increased over time and REM means dropped as one would have expected. These trends were however not a clear indication of the increase in sleepiness expected over drive on the basis of other studies summarized in the introduction. This might have been due to the quite rough statistical analysis of the data in 30-minute intervals. This strategy was chosen because the data sets were too incomplete for a more precise temporal analysis. Nonetheless, we assume that even with this low level of precision, clear linear trends should have been found in ocular movements, if current conclusion of their function are correct.

The duration of driving is comparable to other studies and should show a time-on-task effect and increasing sleepiness

During driving we did not find any SEM as it has been reported in other studies investigating ocular activity in simulated driving (i.e. Shin et al., 2011). Furthermore, in contrast to simulator tests all of our drivers were able to maintain driving performance without mishaps until the end of the two-hour drive. No one aborted the experiment or drove off the road. This indicates that the participants were either not pushed completely to their limits or that the threshold for sleep on a simulator was simply much lower. The duration of the drive was comparable to other studies (Campagne et al, 2005; Schleicher et al, 2008) in that an effect of time-on-task on ocular activity was found. Regarding other correlates of decreased vigilance we found however absolutely an effect of sleepiness: most subjects did report being very exhausted after the drive. This was in line also with the analysis of EEG-data, behavioural changes an performance decrease in the tests on attention. In our experimental setting for reasons of security it was not possible to further increase the amount of previous sleep-deprivation in the participants. We assume the sleep propensity in our experimental design however as quite representative for a situation with that the average driver might be confronted. Furthermore we suppose that the lack of a clear association between ocular activity and sleepiness in our data is caused by the high motivation of the participants not to fall asleep during driving. For the safe performance of this particular task visual input is absolutely essential and probably is, and in this case was, compensated for as long as possible. In this light we would also interpret the rise in REM frequency in the last 15-minutes of driving as a kind of last effort in ocular activity as the subjects knew they would finished the task soon. It occurred in both groups although it was less pronounced in nappers. At the other end of the task, the EB – peak in the first interval was mainly caused by technical difficulties that reduced the sample size in the first 15 minutes (N=4) compared to other intervals (N > 14). Many devices did not work properly yet. One of the four participants had high EB levels in all intervals. When this subject is excluded the resulting mean of the remaining three subjects were similar to the other time periods (21,79 ± 6,52). It remained, however, slightly above the following 15 minute intervals as subjects possibly already had been sleepy when the drive started at 2 pm and then inhibited EB activity to concentrate on driving.

The effect of a nap and sleeping

A nap did not show a clear positive effect on sleepiness associated ocular activity. The differences between the napping and non-napping group and of the sleeping individuals with the non-napping group were marginal. Within the napping group, however, a comparison of the 15 minutes before and after the nap, showed that EB-frequency-means had doubled but failed to be significant as the variation between subject was high. This trend actually would support a proposed negative effect of the nap that was also documented in the analysis of the behavioural correlates of this study. Surprisingly the negative effect contradicts the subjective perception as in the self evaluation subjects indicated less sleepiness. These types of findings are consistent with other studies. For instance, Di Stasi et al (2012) showed that a nap-break after two hours of driving performance improved only the subjective rating but not the ocular correlate of vigilance thereafter.

The impact of individual factors

The high inter-individual differences we found in the occurrence of the diverse ocular patterns over time could not be sufficiently explained by the factors age and sex as other studies, like those of Caffier et al.(2003) and Campagne et al. (2004) had proposed. EB in the former study, however, were analysed after a normal workday and the ocular activity in the latter one was analysed in a simulated-drive in the early evening. This situations for measurement were different from those used here. Additionally with regard to the assessment of sleepiness, the subjects in both studies were not in a comparable situation to the ones analysed in our study when indicating fatigue. Another constraint was the variation in chronotype among the subjects. Unfortunately the dataset used for the analysis of ocular activity included only individuals with the indifferent type and the weak morning type according to self-ratings. We would have expected a morning type to suffer more from the lack of sleep in the early night than the indifferent or even the evening type (i.e. Taillard et al, 1999). This idea could be in line with the finding of more REM-activity in the indifferent and more NEM in the weak morning type but cannot be analyzed sufficiently by our limited dataset.

Homeostatic sleep pressure

None of the subjects had slept in the evening before driving. Hence there was no reduction in the homeostatic sleep pressure. The total *time awake* since the last sleep previously to the experiment differed among subjects between 17 to 22 hours. In REM we found an impact of time awake. It was more frequent in the first 30 minutes in participants that had been awake less than 19,5 hours. We supposed that they were better able to compensate for sleepiness at the beginning of the drive before being similarly exhausted like the other group struggling to maintain ocular activity. The *duration of sleeping* from the night previously to the experiment did not clearly influence ocular activity. If any, there was a trend in frequency of EB. They tended to be higher in individuals that had slept less than 8 hours confirming that this group was probably more sleepy. The effect was small. This is not surprising as it is questionable whether a previous prolongation of sleep could have a

positive effect. Sleep effects cannot be 'stored' above those related to the actual sleep used to saturate the need. Sleep-needs diverge between individuals but in the data included for the analysis no participant slept extremely short and hence could have been considerably more deprived than others (range 6,5 – 10 h).

Comparisons of ocular activity with other correlates for stress and the attentional state

Participants with low increases in the salvia-*cortisol* had significantly less REM in the first 30 minutes than those with a stronger pronounced rise. In the following hour of driving, however, REM activity was the same in both groups. Individuals with a higher increase were either more stressed by the driving task or alternatively simply showed a different circadian pattern of adrenal activity. It is known that the circadian clock stimulates increases in cortisol secretion at the end of sleep. These patterns are also found during sleep deprivation (Salín-Pascual et al., 1988) If the stress factor were true, subjects with more pronounced stress responses, possibly tried to combat sleepiness more, resulting in higher REM-activity during the first 30 minutes of driving. Thereafter, they could not or at least did not keep the activity at the same level. With increasing exhaustion REM-activity dropped to the same level as subjects in the other group. This explanation is, nonetheless, highly speculative. To address the question whether these changes in cortisol are caused by the circadian oscillation or stress, reference-samples of the same subjects of nights without stressing context should be analyzed for instance.

The group showing a decrease in performance in the *AD-Test on attention* had also more epochs without any EM. Significant differences were however only found in the first 30 minutes of driving. Additionally EB were more frequent in this group than in the one with no performance decrease. The EB-activity was also most pronounced in the first 30 minutes. The AD-test measured temporary attention. Hence the group with better test-results after driving compared to the other group was presumably better in combating sleepiness over a short time but could not maintain a higher level of sustained attention over the first 90 minutes of driving. In a moment of particularly required attention (like during the test performance) however, they were capable to activate themselves in a way to compensate for the sleep-drive.

Environmental influences on ocular activity

As described in the introduction EB's are influenced by several factors and are probably the most sensitive to environmental factors of the ocular characteristics analysed in this study. One of the drivers had an extremely high blink frequency that was probably caused by contact lenses and dry eyes. Dazzling light that could affect the blink activity did not occur as far as we know during the experiment but in usual night-time conditions it also has to be taken into consideration. The data analysed was just taken of the straight section of the street. Hence eye-movements as a response due to different curved parts of the street could be excluded.

Conclusions

Findings of other studies showing a clear association of increasing sleepiness and changes in ocular patterns could not be confirmed in this study. This is probably due to the extremely high motivation to combat sleepiness caused by the real driving situation. This was a conflicting condition as described by the two-dimensional arousal model of Noguchi and colleagues (2009; see introduction). Most studies investigating eye-tracking systems have been performed in simulators. But it seems that falling asleep unwillingly during real driving is a more complex phenomenon. Visual input is absolutely essential for maintaining the performance. Hence ocular activity is probably used to compensate until the very end when the biological system cannot cope anymore with the situation and the subject falls asleep. According to the data gained in this project we assume that the break-down of the control of ocular activity is one of the very last steps before falling asleep. The registration of ocular activity could help to prove whether an accident was caused by sleepiness but it might be questionable whether it can warn the driver in time of the risky situation. High inter-individual differences in ocular activity have been also reported in other studies (i.e. Jammes et al., 2008; Schleicher et al, 2008). None of the investigated individual factors could explain the vast variation between the 20 individuals analysed in this study. It seems that sleepiness-regulating factors like the time awake that diverged between subjects can partly account for it. We found an association between decreased performance in a test on attention and more NEM and EB. Furthermore the participants also reacted with different levels of adrenal activity as indicated by the changes in cortisol to this struggling situation. This difference was also partly reflected in ocular activity. Hence, it seems that the motivation not to fall asleep during driving and the individual's capacity to maintain attention at least temporarily were factors that affected subjects also on a physiological level. These may then have been more decisive in this extreme driving situation than factors like sex and age. Individually different stress responses and coping mechanisms with such an extreme condition however are difficult to control and it is questionable whether a clear associations of ocular activity with a certain 'stage' of sleepiness are possible. Even individually calibrated systems could be vulnerable to motivational changes affecting the physiological level. As indicated by this study it is important to take motivational factors in consideration. During a fictive driving task this is however unlikely in a proper manner. According to our results a nap in a situation of high sleep-propensity may have a negative effect on physiological and behavioural correlates of sleepiness and could be dangerous due to its deluding effect on the driver's perception to feel subjectively less sleepy.

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6 APPENDIX

6.1 English and German summary

Abstract

Introduction: Sleepiness during driving is seen as one of the main causes for severe and often lethal accidents. Amongst different kinds of alert systems eye-tracking has been assumed to be a convenient method to detect fluctuations in vigilance. These systems are easy to handle and mount in vehicles. Most experiments investigating eye-characteristics during driving have been conducted in simulators. We assume however that the motivation to combat sleepiness may be very different in a realistic driving situation. Hence data were collected on eye movements and driving in the field. We also assume that if differences are found that they would reflect how the psychological environment of driving might interact with physiological markers of sleepiness.

Methods – Subjects: The aim of our study was to investigate changes in the occurrence of eye-movements and eye-blink patterns in subjects driving their own cars late at night. This was a situation with high sleep pressure. In addition to this, we examined the impact of a powernap on sleepiness-associated eye-characteristics. 60 healthy subjects drove for two hours starting at two am in the night. Half of the subjects drove two hours continuously whereas the other half had a 30-minute break before completing the last 30 minutes of the two hour driving time. The cars were equipped with video cameras and GPS transmitters to detect driving performance and behavior. The eye-activity was recorded by EOG-electrodes. Rapid eye movements, eye-blinks and periods without eye movements were analyzed visually.

Results: EB and epochs without EM became more frequent with driving time and rapid EM as indicators of vigilant scanning activity decreased but these trends have been slight and statistically not significant in contrast to the high variation between participants. A nap did not produce a positive effect on driving performance and sleepiness associated eye characteristics.

Conclusion: The close relationship of sleepiness and changes in ocular activity described in other studies could not be confirmed in this study. As visual input is absolutely essential in driving it is probably compensated as long as possible. A breakdown of ocular control occurs

presumably as one of the very last steps before sleep-onset in the struggling situation between awake and falling asleep. Hence it is questionable whether changes in ocular activity can be used to warn drivers in time about the risky situation. High variation between subjects could not be explained by individual factors like age, chronotype or sex and possibly reflects different stress responses and coping mechanisms with this extreme condition. A nap seems to have a deluding effect as the subjective perception to feel less tired afterwards was not confirmed by any of our behavioural and physiological correlates.

Zusammenfassung der Arbeit zum Thema Augenbewegungen und Lidschlag als mögliche Indikatoren für Schläfrigkeit beim Autofahren in der Nacht

Übermüdung am Steuer stellt eine der Hauptursachen für schwere und nicht selten tödliche Unfälle während der Nacht dar. Bis heute gibt es kaum zuverlässige Warnsysteme oder Möglichkeiten, Müdigkeit als Unfallursache festzustellen. ‚Eye-tracking'-Systeme gewinnen auf Grund meist einfacher Handhabung, geringer Größe und keinerlei Beeinträchtigung der Fahrtätigkeit zunehmend an Beliebtheit. Die meisten Studien, die Vigilanzschwankungen anhand von Augenbewegungen und Lidschlag-Charakteristika analysierten, wurden in Fahrsimulatoren durchgeführt. Wir nehmen jedoch an, dass die Motivation, Aufmerksamkeit aufrecht zu erhalten, eine entscheidende Rolle in der Schläfrigkeitsresistenz spielt, die sich dann auch auf physiologischer Ebene wiederspiegelt und in einer fiktiven Situation nicht ausreichend untersucht werden kann. Neben der Analyse von Augencharakteristika in einem realistischeren Versuchsdesign wollten wir in unserer Studie untersuchen, wie sich ein ‚Powernap' auf die schläfrigkeitsassoziierten Augencharakteristika und das Fahrverhalten auswirkt. 60 gesunde Probanden fuhren um zwei Uhr morgens in ihren eigenen Autos für zwei Stunden auf einer Übungsstrecke des ÖAMTC. Die eine Gruppe, welche ein Nickerchen machte, legte vor Beendigung der letzten halben Stunde eine Pause von 30 Minuten ein; die andere Gruppe fuhr zwei Stunden ohne Unterbrechung. Die Autos waren zur Aufnahme des Fahrverhaltens mit Kameras und GPS-Transmittern ausgestattet. Die Augenbewegungen wurden über EOG-Elektroden aufgezeichnet. Das Auftreten von Lidschlag-Ereignissen, schnellen Augenbewegungen und Epochen ohne Augenbewegungen wurde visuell ausgewertet. Der Lidschlag und Epochen ohne Augenbewegungen zeigten die Tendenz, mit der Zeit zuzunehmen; schnelle Augenbewegungen, ein Indikator für vigilantes Scanverhalten, nahmen hingegen ab. Allerdings waren diese Veränderungen klein und konnten die enge Assoziation von Schläfrigkeit und Veränderungsmustern der Augenbewegungen und des Lidschlages, die in anderen Studien gefunden wurden, nicht bestätigen. Wir fanden jedoch signifikante Unterschiede zwischen den Probanden. Langsam rollende, mit dem Einschlafen assoziierte Augenbewegungen, die als Indikator für den Zusammenbruch der Kontrolle über die Augenbewegungen gelten, wurden nicht gefunden.
Ein Powernap zeigte bei unseren Probanden keinen deutlichen Einfluss auf das Fahrverhalten und die müdigkeitsassoziierten Augencharakteristika.

Wir nehmen an, dass die Motivation, wach zu bleiben, bei den Fahrern unserer Studie durch das realistische Versuchsdesign extrem hoch war. Um die Fahrleistung aufrecht zu erhalten, ist visuelle Information absolut essenziell. Möglicherweise spiegelt sich Schläfrigkeit erst deutlich in der Augenaktivität wieder, wenn die Kapazität, gegen den Schlafdruck anzukämpfen, erschöpft ist. Die große Variationsbreite der untersuchten Charakteristika bei den Probanden ließ sich nicht ausreichend durch Faktoren wie Geschlecht, Alter oder Chronotyp erklären und spiegelt möglicherweise individuelle Stressreaktionen und Adaptionsstrategien wieder. Ein Powernap führt möglicherweise bei dem großen Schlafdruck, dem die Probanden ausgeliefert waren, zu Fehleinschätzungen des eigenen Vigilanzstadiums. Die subjektive Wahrnehmung, sich danach wacher zu fühlen, stand im Gegensatz sowohl zu den Ergebnissen von physiologischen Messvariablen, die mit Schläfrigkeit assoziiert sind, als auch denen auf der Verhaltensebene.

6.2 Tables

List of participants that have been included in the analysis:

ID	group	age (years)	sex	CT	SD (min)	WD (min)	AD-P_dif	CTL_dif (µl)
04c	nn	41	w	4	600	1020	0	17986
12c	nn	50	m	4	490	1180	-1	32781,5
14c	nn	42	m	3	400	1140	-1	*nd*
16c	nn	40	w	3	480	1140	-3	*nd*
17c	nn	49	m	5	480	1170	1	1227
22c	nn	42	w	3	410	1140	0	-510,5
23n	bs	30	w	3	360	1200	2	2266
25n	nb	53	m	3	375	1155	1	-3309
26n	bs	40	m	*nd*	420	1285	0	4741,5
29n	bs	31	m	4	540	1020	0	-3853
30n	nb	54	m	4	*nd*	*nd*	-5	4902
32c	nn	30	w	4	495	1215	0	15679
36c	nn	31	w	3	450	1160	-8	9048,5
38n	bs	47	m	4	*nd*	*nd*	0	8382,5
47c	nn	46	w	4	460	1100	-11	20740,5
48c	nn	44	m	3	580	1080	-5	1084
53n	nb	33	m	4	420	1230	-1	*nd*
55n	bs	39	m	4	460	1205	-1	2401,5
59c	nn	34	m	4	480	1140	1	1987,5
66n	nb	42	w	3	440	1100	-7	-1484

List of all participants whose ocular activity has been analyzed, grouped in ‘non-napper’ (nn) and ‘napper’ consisting of those who did not sleep (nb) and those who slept (bs). AD-P_dif = difference in max. credits/line in test on attention between performance before and after driving ; CT = chronotype (3 = indifferent type; 4 = slightly pronounced morning type; 5 = strongly pronounced morning type); CTL_dif = difference in level of saliva-cortisol between measurement

before and after driving; m = man; nd = not detected; SD = sleep-duration in night previously to experiment; w = woman; WD = time awake previously to experiment

ANOVA-tables

Table 3a & b: Variation with time on task in EM, the EM- compound REM, EB and NEM, **a**: repeated measures ANOVA: first 90' of driving compared in three 30'-intervals; (**b**: Friedman-Test: First 90' compared in 15'-intervals, first 15 min excluded due to many missing values; I/Intv. = interval; (ns) = not spherical (df of test on within-subjects effect with Greenhouse Geisser correction); TI = time-interval.

a. Variation over time, 30'-intv. (r-ANOVA)

	mean & STDEV							within-subject-effects			between-subject-effects		
	I_1		**I_2**		**I_3**		**N**	**p**	**F**	**df**	**p**	**F**	**df**
EM*(ns)*	55,4	± 37,46	70,34	±20,88	65,52	±23,01	20	2,59	0,11	1,46	**0,00**	165,9	1
REM	39,96	±16,83	44,59	±19,00	40,13	±14,11	14	0,20	1,33	2	**0,00**	104,0	1
EB	20,1	±18,54	11,7	±9,86	18,27	±19,41	10	0,39	0,99	2	**0,00**	19,8	1
NEM	26,95	±18,42	22,7	±18,05	21,72	±13,62	14	0,31	1,21	2	**0,00**	35,4	1

b. Variation over time, 15'-intv.

	mean & STDEV										Friedman-Test			
	I_2		**I_3**		**I-4**		**I_5**		**I_6**		**N**	**Chi²**	**df**	**p**
EM	72,46	±19,38	72,13	±23,37	72,02	±19,3	72,76	±12,93	69,5	±17,18	12	2,53	4	0,64
REM	40,09	±17,59	42,62	±19,30	44,48	±18,58	43,39	±16,75	36,78	±16,64	12	5,33	4	0,25
EB	11,24	±10,98	10,16	±11,18	11,83	±11,9	18,31	±21,37	19,13	±18,4	10	3,77	4	0,44
NEM	24,52	±17,46	24,31	±20,31	24,05	±17,71	22,75	±13,96	23,22	±12,88	12	0,27	4	0,99

Table 4a-d: Effect of a nap; **a**: Comparison of the last 30 min-interval between nap- and non-napping group using a one-way ANOVA; **b** & **c**: Comparison of the first respectively the second 15 min-interval of the last 30 min of driving between nap- and non-napping group using a one-way ANOVA; **d**: within-nap-group comparison of the 15 min before and 15 min after the break with a paired sample T-Test and the Wilcoxon- test; intv = interval.

a. 30'-intv (one-way ANOVA; nap-effect)

	no-napper			napper			ANOVA		
	mean	STDEV	N	mean	STDEV	N	df	F-ratio	p-value
EM	63,34	15,61	10	66,36	25,18	8	1	0,10	0,76
REM	34,77	20,48	10	35,53	26,00	8	1	0,00	0,95
EB	28,09	23,06	10	29,39	15,78	5	1	0,01	0,91
NEM	27,72	17,48	10	26,57	21,14	8	1	0,02	0,90

b. 1st 15'-intv of last 30' (one-way ANOVA; nap-effect)

	no-napper			napper			ANOVA		
	mean	STDEV	N	mean	STDEV	N	df	F-ratio	p-value
EM	61,13	15,12	9	65,75	25,23	8	1	0,22	0,65
REM	26,51	13,06	9	33,84	28,99	8	1	0,47	0,50
EB	27,22	21,69	9	28,62	12,84	5	1	0,02	0,90
NEM	28,97	17,32	9	27,37	21,65	8	1	0,03	0,87

c. 2nd 15'-intv of last 30' (one-way ANOVA; nap-effect)

	no-napper			napper			ANOVA		
	mean	STDEV	N	mean	STDEV	N	df	F-ratio	p-value
EM	63,33	20,56	10	69,27	27,56	7	1	0,26	0,62
REM	38,13	21,73	10	42,12	23,28	7	1	0,13	0,72
EB	33,12	29,78	10	30,70	17,57	5	1	0,03	0,87
NEM	28,18	20,36	10	22,17	20,49	7	1	0,36	0,56

d. Within-group-comparison (nap-effect)

	pre-nap (15')		post-nap (15')			T-Test (paired samples)			Wilcoxon	
	mean	STDEV	mean	STDEV	N	T	df	p (2 tailed)	Z	p (asym)
EM	71,43	27,86	65,75	25,24	8	1,15	7	0,29	-0,84	0,40
REM	36,33	19,45	33,84	28,99	8	-0,4	7	0,70	-0,42	0,67
EB	14,05	15,99	28,62	12,84	5	1,59	4	0,19	-1,48	0,14
NEM	22,53	18,87	27,37	21,65	8	0,86	7	0,42	-1,26	0,21

Table 5a-d: Effect of sleeping; **a**: Comparison of the last 30'- interval between sleepers and non-napping group using a one-way ANOVA; **b** & **c**: Comparison of the first respectively the second 15'-interval of the last 30' of driving between sleeping and non-napping group using a one-way ANOVA; **d**: within group of sleepers comparison of the 15' before and 15' after the break with a paired sample T-Test and the Wilcoxon- test; intv = interval

a. Sleep Effect (last 30'-intv)

	no-nap			sleeper			ANOVA		
	mean	STDEV	N	mean	STDEV	N	df	F-ratio	p-value
EM	63,34	15,61	10	64,79	29,74	4	1	0,01	0,91
REM	34,77	20,48	10	44,94	31,57	4	1	0,52	0,48
EB	28,09	23,06	10	33,40	13,80	2	1	0,09	0,76
NEM	27,72	17,48	10	28,70	24,71	4	1	0,01	0,93

b. Sleep Effect (1st 15'- intv of last

	no-nap			sleeper			ANOVA		
	mean	STDEV	N	mean	STDEV	N	df	F-ratio	p-value
EM	61,13	15,12	9	65,28	30,64	4	1	0,11	0,74
REM	26,51	13,06	9	45,09	37,28	4	1	1,90	0,20
EB	27,22	21,69	9	21,49	3,87	2	1	0,13	0,73
NEM	28,97	17,32	9	29,98	26,14	4	1	0,01	0,93

c. sleep-effect (2nd 15'- intv of last 30')

	no-nap			sleeper			ANOVA		
	mean	STDEV	N	mean	STDEV	N	df	F-ratio	p-value
EM	63,33	20,56	10	56,12	33,14	3	1	0,22	0,65
REM	38,13	21,73	10	35,10	22,77	3	1	0,04	0,84
EB	31,34	31,30	10	39,11	22,97	2	1	0,11	0,75
NEM	28,18	20,36	10	33,46	26,80	3	1	0,14	0,72

d. sleep-effect within nap-group

	pre-nap (15')		post-nap (15')			T-Test (paired samples)			Wilcoxon	
	mean	STDEV	mean	STDEV	N	T	df	p (2 tailed)	Z	p (asym)
EM	67,95	32,75	65,28	30,64	4	0,32	3	0,77	0,00	1,00
REM	40,87	24,78	45,09	37,28	4	0,49	3	0,66	-0,37	0,72
EB	19,38	27,41	21,49	3,87	2	0,13	1	0,92	-0,45	0,65
NEM	23,35	18,22	29,98	26,14	4	1,00	3	0,39	-0,73	0,47

Table 6a-c: Effect of diverse individual factors on REM, EB and NEM analyzed with repeated measures ANOVAs ; **a**: effect of chronotype (CT); CT 3 = indifferent type; CT 4 = slightly pronounced morning type; **b:** between subjects having more and less than 40 years; **c**: comparison between men and women; ic = intercept; (ns) = not spherical (df of test on within-subjects effect with Greenhouse Geisser correction); t = time.

a. CT (r-ANOVA, split design)

		CT 3			CT 4			within-subjects				between subjects			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	45,06	18,60	6	36,44	16,70	7	time	2,16	0,14	2	ic	125,18	0,00	1
	2	55,54	14,30	6	38,89	18,60	7	t*CT	0,94	0,41	2	CT	3,19	0,10	1
	3	50,13	9,90	6	33,87	12,70	7								
EB*(ns)*	1	29,01	19,10	6	6,72	5,00	4	time	0,60	0,49	1,24	ic	19,72	0,00	1
	2	12,20	10,40	6	10,95	10,40	4	t*CT	1,41	0,27	1,24	CT	2,84	0,13	1
	3	22,98	23,60	6	11,19	9,50	4								

NEM	1	24,12	20,90	6	29,55	18,90	7	time	1,79	0,19	2	ic	31,18	0,00	1
	2	11,89	8,70	6	30,04	20,80	7	t*CT	1,52	0,24	2	CT	2,28	0,16	1
	3	13,90	6,90	6	27,37	16,00	7								

b. Age (r-ANOVA, split design)

		<40 y			**>40 y**			**within-subjects**				**between subjects**			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	41,56	22,70	6	38,76	12,40	8	time	1,05	0,37	2	ic	93,91	0,00	1
	2	40,38	19,90	6	47,76	19,00	8	t*age	1,27	0,30	2	age	0,10	0,76	1
	3	38,14	17,40	6	41,63	12,10	8								
EB	1	20,78	23,30	5	19,41	15,10	5	time	0,88	0,43	2	ic	17,64	0,00	1
	2	11,73	12,40	5	11,67	8,10	5	t*age	0,03	0,97	2	age	0,04	0,84	1
	3	20,01	27,10	5	16,52	10,30	5								
NEM	1	33,04	18,60	6	22,38	18,10	8	time	1,16	0,33	2	ic	38,90	0,00	1
	2	29,98	18,80	6	17,24	16,50	8	t*age	0,24	0,79	2	age	1,73	0,21	1
	3	26,06	15,80	6	18,46	11,80	8								

c. Sex (r-ANOVA, split design)

		men			**women**			**within-subjects**				**between subjects**			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	36,01	13,40	7	43,90	19,90	7	time	1,27	0,30	2	ic	98,05	0,00	1
	2	43,53	17,20	7	45,66	22,00	7	t*sex	0,45	0,64	2	sex	0,26	0,62	1
	3	38,71	11,30	7	41,56	17,30	7								
EB	1	9,87	7,40	4	26,91	21,10	6	time	0,74	0,49	2	ic	17,54	0,00	1
	2	9,06	10,30	4	13,46	10,10	6	t*sex	0,65	0,53	2	sex	1,24	0,30	1
	3	15,98	13,30	4	19,79	23,80	6								
NEM*(ns)*	1	25,89	14,40	7	28,00	22,90	7	time	1,20	0,31	1,35	ic	32,66	0,00	1
	2	21,39	12,90	7	24,01	23,20	7	t*sex	0,87	0,40	1,35	sex	0,00	0,97	1
	3	24,62	8,00	7	18,81	17,80	7								

Table 7a& b: Effects of time awake over driving time (**a.**; awake shorter and longer than 19,5 hours) and previous night sleep-duration (**b.**; more and less than 8 h of sleep) on REM, EB and NEM; ic = intercept; t = time; sd = sleep-duration; wd = time awake

a. Wake-duration (r-ANOVA, split design)

		< 19,5 h			>19,5 h			within-subjects				between subjects			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	49,34	17,60	7	32,21	13,60	5	time	1,68	0,21	2	ic	78,68	0,00	1
	2	43,42	21,40	7	46,28	19,50	5	t*wd	7,11	**0,01**	2	wd	0,19	0,67	1
	3	38,58	19,20	7	40,65	12,10	5								
EB	1	19,71	15,90	6	20,67	24,60	4	time	0,83	0,45	2	ic	16,55	0,00	1
	2	11,47	11,20	6	12,04	9,00	4	t*wd	0,31	0,74	2	wd	0,08	0,78	1
	3	21,62	24,60	6	13,24	7,80	4								
NEM	1	27,87	19,80	7	29,23	19,20	5	time	1,34	0,29	2	ic	27,84	0,00	1
	2	19,22	17,90	7	27,32	21,60	5	t*wd	0,52	0,60	2	wd	0,44	0,52	1
	3	17,59	14,00	7	26,77	14,70	5								

b. Sleep-duration (r-ANOVA, split design)

		< 8 h			> 8 h			within-subjects				between subjects			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	48,40	17,70	5	32,88	14,40	7	time	1,26	0,30	2	ic	95,99	0,00	1
	2	51,45	19,70	5	40,55	19,30	7	t*sd	0,25	0,78	2	sd	2,03	0,18	1
	3	45,96	15,70	5	35,38	13,20	7								
EB	1	32,51	22,80	4	11,82	10,00	6	time	1,86	0,19	2	ic	18,13	0,00	1
	2	8,60	3,80	4	13,77	12,40	6	t*sd	3,26	0,07	2	sd	0,23	0,65	1
	3	15,83	10,20	4	19,89	24,70	6								
NEM	1	20,86	22,20	5	33,84	15,10	7	time	1,49	0,25	2	ic	27,61	0,00	1
	2	14,33	13,50	5	28,50	21,10	7	t*sd	0,35	0,71	2	sd	1,72	0,22	1
	3	16,98	9,80	5	24,59	16,90	7								

Table 8: Comparison of participants with a high cortsiol increases over during driving time and those with a slight increase with regard to patterns in REM, EB and NEM. Analysis was a repeated measures ANOVA: first 90' of driving compared in three 30'-intervals; crl = cortisol; ic = intercept; (ns) = not spherical (df of test on within-subjects effect with Greenhouse Geisser correction); t = time.

Cortisol (r-ANOVA, split design)

		strong increase			slight increase			within-subjects				between subjects			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	45,06	17,57	9	29,97	13,47	3	time	6,17	**0,01**	2	ic	55,82	0,00	1
	2	42,21	21,66	9	52,84	13,23	3	t*crl	12,07	**0,00**	2	crl	0,01	0,93	1
	3	38,15	16,39	9	45,67	12,49	3								
EB*(ns)*	1	18,89	21,69	5	20,06	23,45	3	time	1,40	0,28	1,01	ic	16,92	0,01	1
	2	12,49	7,86	5	6,14	3,91	3	t*crl	0,23	0,65	1,01	crl	0,46	0,52	1
	3	13,30	6,79	5	5,89	2,69	3								
NEM*(ns)*	1	27,38	20,16	9	29,00	20,69	3	time	1,37	0,27	1,25	ic	18,82	0,00	1
	2	26,22	20,19	9	16,24	15,29	3	t*crl	0,81	0,41	1,25	crl	0,14	0,72	1
	3	23,98	14,68	9	20,22	15,42	3								

Table 9: Comparison of participants with decreasing or equal and slightly increasing performance in REM, EB and NEM with a repeated measures ANOVA: first 90' of driving compared in three 30'-intervals; AD = performance in AD-test; ic = intercept; t = time.

AD-Test_max/line (r-ANOVA, split design)

		decrease			equal/increase			within-subjects				between subjects			
	I_30'	mean	STDEV	N	mean	STDEV	N		F	p	df		F	p	df
REM	1	34,55	11,27	8	47,17	21,20	6	time	1,33	0,28	2	ic	99,22	0,00	1
	2	42,92	18,01	8	46,83	21,78	6	t*AD	2,12	0,14	2	AD	0,43	0,52	1
	3	40,12	11,26	8	40,15	18,45	6								
EB	1	26,33	18,86	7	5,56	5,09	3	time	0,40	0,67	2	ic	15,76	0,00	1
	2	14,18	10,53	7	5,92	5,72	3	t*AD	0,39	0,68	2	AD	4,70	0,06	1
	3	23,02	21,58	7	7,18	5,89	3								

NEM	1	32,15	18,29	8	20,02	17,70	6	time	0,87	0,43	2	ic	31,76	0,00	1
	2	20,98	15,36	8	24,99	22,48	6	t*AD	3,65	**0,04**	2	AD	0,05	0,82	1
	3	20,69	9,92	8	23,09	18,46	6								

6.3 Curriculum Vitae

Miriam Kirsch

Education:

3/2010 – 4/2013	University of Vienna (master: behaviour, neurobiology and cognition; master thesis: 'Ocular indicators for sleepiness during night-time driving'; 2013, Prof.Dr. J. Dittami)
3/2012 – 7/2012	University of Trieste, Italy; Erasmus-exchange Courses of the international Master of Neuroscience
2/2012 – 3/2012	Ca'Foscari University, Venice, Italy Italian course
8/2009 – 9/2009	Limnology-summerschool in Romania
10/2006 – 7/ 2009	University of Constance, Germany (B. Sc.) (Bachelor thesis: 'Inter- and intraspecific comparison of Distresscall-elements of tropical bat species in a phylogenetic context'; 2009, Prof. Dr. M. Wikelski)
2/2009 – 4/2009	Courses in marine ecology and vertebrates in the rainforest in Panama (project of the university of Constance, the Princeton university and the Smithsonian Tropical Research Institute of Gamboa)

Other experiences and activities:

7/2011 – 9/2011	Internship in the sleep laboratory of the state hospital of Melk, Austria (Dr. J. Lechner)
8/2009	Participation in a gene sequencing project of Lycaenidae (University of Constance, Germany)
2/2008 – 8/2009	Work in labs of university of Constance, Germany (limnology; evolutionary biology)
8/2005 - 8/2006	Ecological volunteer service (in the regional-park of Kurtuvenai, Šiauliai, Lithuania)

Knowledge of language

- German (mother tongue)
- English (fluently)
- Italian (fluently)
- Spanish (basic)

EDP skills:

- Office (Word, Excel, Powerpoint)
- SPSS, Statgraphics
- Programs for categorization of sleep stages (RemLogic, SleepExplorer)
- Avisoft (analysis of acoustic biosignals)

Miriam Kirsch, 17/4/2013