VILNIUS UNIVERSITY

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PHYSIOLOGICAL MODULATION OF AUDITORY STEADY-STATE RESPONSES: AROUSAL, ACTIVATION AND ATTENTION

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VILNIAUS UNIVERSITETAS

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FIZIOLOGINĖ KLAUSOS SUKELTO PASTOVAUS ATSAKO MODULIACIJA: BUDRUMAS, AKTYVUMAS IR DĖMESYS

Daktaro disertacija
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A science imposes limits on itself and makes its progress by attacking only those problems that it is fitted to attack by existing knowledge and methods (D.O. Hebb, 1974)

CONTENTS

ABBREVIATIONS

Ach-acetilcholine
ANOVA-analysis of variance
ASSR-auditory steady-state response
avWT- phase-synchronized WT amplitude measure, evoked amplitude
C3, Cz, C4- central electrodes
DA-dopamine
EEG-electroencephalography
ERPs-event-related potentials
F3, Fz, F4-frontal electrodes
FFT-Fast Fourier transform
GBR-gamma band response
ICA-independent component analysis
ITPC-intertrial phase coherence, phase precision
MEG-magnetoencephalography
MOI – measure of interest
N1-first negative deflection of slow ERPs
NA-noradrenalin
NMWF- non-negative multi-way factorization
P1-first positive deflection of slow ERPs
P2-second positive deflection of slow ERPs
P3, Pz, P4-parietal electrodes
PARAFAC- Parallel Factor
PCA-principal component analysis
ROI-region of interest
SD-standard deviation
SPL-sound pressure level
TF-time-frequency
WTav- average amplitude of the oscillation, total intensity
WT-wavelet transform

1. INTRODUCTION

Thirty years ago, Charles Shagass had already noted that electrophysiology stands at the interface between behavioral and neuronal events and that it offers some accessible means of bridging part of the gap between them: ''Rapid and complete data reduction will permit analysis of recordings of many different kinds of electrical events from numerous sites. More important, it should become possible to extract the mathematical properties of these signals and the relationships between them. It is my hope that these properties will both reflect underlying processes and be of diagnostic utility'' (Shagass, 1976).

This mainly refers to the analysis of brain activity that is non-invasively recorded from the scalp-electroencephalogram (EEG). When some kind of stimulus is presented, sequences of neural responses that are related to the processing of this stimulus are evoked and called event-elated potentials (ERPs). Processing stages are manifested either in parallel or in a series as an activity of several brain regions. Neural activities generate the flow of electric current in the brain, and all processing occurs as a spatiotemporal alteration in global current. The momentary electric field, i.e. the electric potential distribution on the scalp, shows us the sum of all currents at any given moment in time. In a sense, the EEG is the recording of these electric field potentials in discrete scalp positions (Michel et al., 1999). This can be applied to the study of both sensory and cognitive aspects of the brain's information processing.

The EEG/ERP recording procedure is rather routine and highly standardized in respect to electrode placement, recording conditions and equipment. However, a variety of stimulation/behavioral protocols exists. It is pertinent to know the best recording conditions for each type of the ERPs to ensure correct and stable results. Several types of ERPs elicited by auditory stimuli have found widespread application in practice. For example, P300 potential requires direct subjects' participation during recording procedure, as the subject has to count/respond with button press to a particular kind of

stimuli. However, for clinical populations passive paradigms are occasionally more appropriate.

Recently so called auditory steady-state response (ASSR) became very popular. ASSR is recorded when stimuli are presented periodically and demonstrate how the brain „follows“ a stimulus or how the stimulus „drives“ a response. ASSR is a complex potential with transient response comprised of P1-N1-P2 complex in the beginning of stimulation and steady-state phase during entrainment. While transient responses are most appropriately measured by selecting peaks and throughs and calculating their amplitudes, ASSRs are most appropriately measured in the frequency domain – determining phase and amplitude of the oscillation.

The ASSR has been shown to be impaired in schizophrenia and other disorders. As the eliciting paradigm of the potential is passive-subject does not have to respond in either way to the stimulation - the background behavioral conditions during recordings have to be standardized. However, it has not been done yet.

ASSR recording conditions vary in the level of arousal/activation and attentional demands towards stimulation. Both attention and arousal are multi-dimensional psychological processes that interact closely with one another (Hebb, 1955). Moreover, it has been suggested that arousal and activation reflected separate processes of the attention system: ‘‘arousal’’ defined as the current energetic level of the organism, i.e. a non-specific activation of cerebral cortex in relation to sleep wake states, ‘‘activation’’ defined as a separable tonic measure reflecting the task-related mobilisation of energy (relative to some baseline level of arousal, and thus activation is not easy to separate experimentally from arousal and referred to as “activation/arousal”), which is needed to perform a task and “attention” implying a more focused activation of the cerebral cortex that enhances information processing (Oken et al., 2006). Thus, tasks involving focused attention also involve tonic mobilization depending on the level of effort, therefore leading to increased activation level and increased arousal level during the task processing (Barry et al., 2007).

There is a great lack of information on the optimal recording states of ASSRs and there are no systematic researches up to date to uncover the effect of arousal/activation and attentional demands on this ERP.

1.1. Aim and objectives

The aim of the study was to investigate the effects of varying arousal, activation and attention level on the auditory steady-state responses.

The following objectives were formulated:

1. To assess and compare auditory steady-state responses in:
   - high vs. low arousal level conditions;
   - high vs. low activation level conditions;
2. To assess the effects of:
   - attentional demands, varying from focused attention on stimulation to distraction from stimulation, on auditory steady-state responses.
   - arousal/activation and attentional manipulations on transient auditory response.

1.2. Actuality and scientific novelty

For the fist time focused attention, unfocused state and distraction tasks were applied during auditory steady-state response in one experimental setup.

For the first time concept of “activation” was introduced to auditory steady-state response experiment and the complex effect of attention and activation on auditory steady-state responses was shown.

For the first time differential modulation of 20Hz auditory steady-state response and 20Hz ASSR-related 40Hz activity by arousal/activation and attention was shown.

For the first time multichannel auditory steady-state responses data was decomposed with non-negative multi-way factorization (NMWF), validating this method as suitable for steady-state response analyses.

1.3. Application

The results contribute to 1) the clarification of the modulatory effects of arousal/activation and attentional demands on both auditory steady-state responses and auditory transient responses; 2) the selection of appropriate recording conditions for auditory steady-state responses and transient event-related potentials; 3) the correct data interpretation from auditory steady-state response experiments; 4) the application of NMWF decomposition for multichannel auditory steady-state response data.

1.4. Defended statements

1) The lower arousal level increases the total neuronal resources (measured as total intensity) leaving the proportion of phase-aligned neuronal recourses (measured as evoked amplitude and phase precision) of the 40Hz ASSR unchanged.
2) The differences between tasks varying in arousal/activation and attention level are the result of the counterbalancing effects of the attention and arousal/activation induced processes:
    - the lower arousal/activation level increases the proportion of phase-aligned neuronal resources (measured as evoked amplitude and phase precision) of the 40Hz ASSRs and 20Hz ASSR-related 40Hz activity leaving the total amount of neuronal resources (measured as total intensity) of the oscillation unchanged;
    - the lower attention level paid to the stimuli reduces the proportion of phase-aligned neuronal resources (measured as evoked amplitude and phase precision) of the 40Hz ASSRs

leaving the total amount of neuronal resources (measured as total intensity) of the oscillation unchanged.

3) Neither total amount of neuronal resources (measured as total intensity) nor the proportion of phase-aligned neuronal resources (measured as evoked amplitude and phase precision) of the 20Hz ASSR are sensitive to the levels of arousal, activation and attention.
4) The lower arousal level increases the neuronal resources (measured as the amplitude of P1 wave) for preattentive auditory stimulus processing.
5) The lower arousal/activation and attention levels enhance processes that trigger allocation of attention to the stimulus, as reflected by N1 wave amplitude.

2. LITERATURE REVIEW

2.1. Auditory steady-state responses

Electroencephalographic responses to acoustic stimuli in general have been recorded for about 70 years (Davis, 1939). When high-frequency sinusoids modulated with a lower modulation frequency are applied to the ear, the auditory neuron fibres fire synchronously to the envelope (Moller, 1974). This can be recorded with EEG and the first significant auditory potential study with modulated tones was conducted by Galambos et al. (1981) on modulation frequencies of 40 Hz. There are several definitions being used to refer to periodic electroencephalographic response: 1. the *amplitude modulation following response* (AMFR by (Kuwada et al., 1986)), 2. the *steady-state evoked potential* (SSEP by (Cohen et al., 1991)) and 3. the *envelope following response* (EFR by (Dolphin and Mountain, 1992)). The currently most popular term is the *auditory steady-state response* (ASSR) and it will be used throughout this manuscript.

A steady-state response is a response `whose constituent discrete frequency components remain constant in amplitude and phase over an infinitely long time period (Regan, 1989). The ASSR may reflect the driving of smaller populations of neurons compared with larger network. Whereas transient responses (e.g. auditory brainstem responses) are evoked by stimuli that do not have a periodic nature, steady-state responses are evoked by repetitive stimuli that are presented continuously or at a stimulus rate that is fast enough to cause an overlap of the consecutive responses.

Typical stimuli for eliciting auditory steady-state responses (ASSR) are sequences of clicks (Galambos et al. 1981; Hari et al., 1987), Gaussian tone pulses (Pantev et al., 1996), or amplitude modulated tones (Linden et al., 1987)

2.1.1. ASSR structure

Four intervals can be distinguished in the ASSR (Figure 2.1):

1) The onset of the tone elicits P1-N1-P2 complex that lasts for about 100ms, and then a sustained potential (SP) lasting through the tone (Figure 2.1B). This part is similar to a conventional P1-N1-P2 complex of auditory ERPs (Ross et al. 2005). Riding on the SP there is a small ASSR (Figure 2.1A). This can be seen when analyzing in the time-voltage domain. When analyzing in time-frequency domain, the beginning of ASSR is preceded by a brief burst (about 50ms) of gamma band activity, gamma band response (GBR) that is evident after high pass filtering (Figure 2.1C).
2) During 100-250 ms ASSR amplitude increases linearly and reaches its maximum.
3) Up to the end of stimulation (800 ms), the ASSR signal with stable amplitude is registered.
4) The response falls back within 50 to 100 ms.

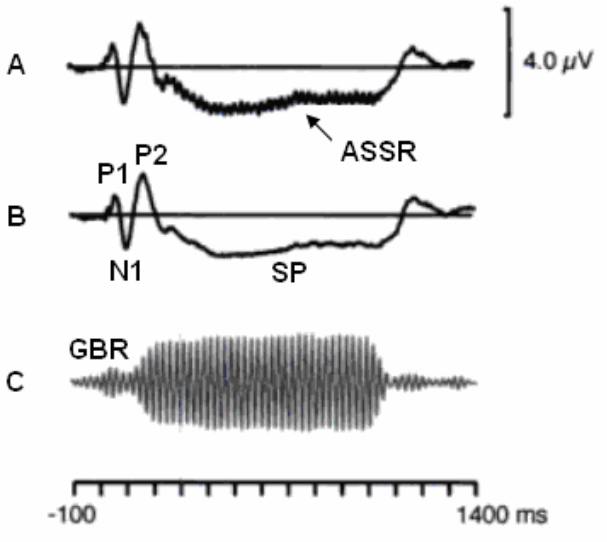

Figure 2.1. The constitutive parts of ASSR. The data from eight subjects having 200 responses is presented. The stimuli were 1000Hz tones lasting 1 second presented binaurally every 2 sec at 60 dB HL. (A) Transient response in the beginning of stimulation and ASSR riding on the sustained potential; (B) constitutive parts of transient response (P1-N1-P2 complex) and sustained potential (SP), seen when data filtered low pass at 24Hz; (C) gamma band response (GBR) in the beginning of stimulation followed by ASSR are evident when data band passed at 32-60Hz. Figure based on Picton (2007) and Ross et al. (2005).

2.1.2. SSR generation theories

Currently there are three theories seeking to explain the generation of SSRs.

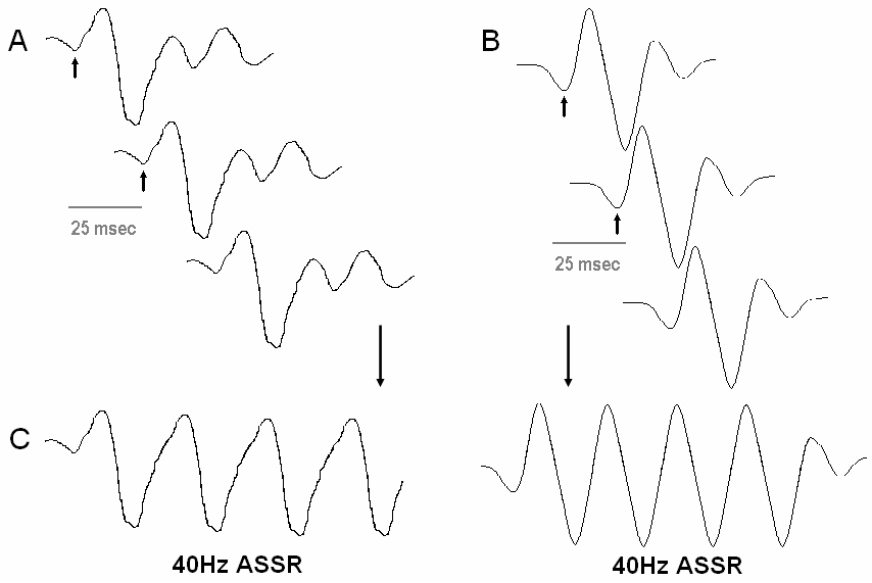

Figure 2.2. SSR response generation theories. (A) a model proposed by Galambos et al (1989) where SSR is the result of superposition of transient responses; (B) SSR as a result of superposition of transient gamma band responses and (C) SSR is a separate stimulus-driven oscillatory activity.

In the initial study Galambos et al. (1989) suggested that the auditory 40Hz ASSR represented the superimposition of the transient middle-latency responses to each of the stimuli. Since the waves of the transient responses reoccurred at inter-peak intervals of about 25ms, the superimposition was most effective at stimulus rates of 40Hz (Figure 2.2 A) (Stapells et al., 1987).

Another proposition was that SSRs result from superimposition of transient gamma band responses that are evoked with every auditory stimulus. However this hypothesis was rejected as GBR and SSR were generated by different generators (Figure 2.2B) (Pantev et al., 1993; Pantev et al., 1996).

However, several findings indicate that the ASSR in the gamma range represents a resonance response or oscillation, rather than a simple superposition of discrete evoked potentials to individual clicks. Ross et al.

(2005) provided evidence that the SSR is a type of stimulus–driven oscillatory brain activity rather than an evoked response that is generated in addition to ongoing spontaneous activity (Figure 2.2C). It is possible that the ASSR reflects a combination of the initial response to the individual stimulus and the output of a resonance response (Ross et al., 2005).

2.1.3. ASSR sources

Animal studies

ASSRs have been elicited from many nonhuman mammalian species, including primates (Burton et al., 1992; Steinschneider et al., 1998; Harada et al., 1994), cats (Hari et al., 1987; Karmos et al., 2002); rabbits (Kuwada et al., 2002) and rodents (Conti et al., 1999; Franowicz and Barth, 1995; Santarelli et al., 2003; Szalda and Burkard, 2005). As with human recordings, there is growing evidence that the ASSR in animals is primarily generated in auditory cortex and may be related to local field potentials. Kuwada et al. (2002) showed that while the cortex is optimally activated by low frequencies (<20 Hz) of stimulation, subcortical structures are likely involved in generating responses at higher frequencies of stimulation (<90 Hz). This pattern has been supported by source localization analysis of EEG data in humans (Herdmann et al., 2002). However, in cats, ablation of lower auditory structures, such as the inferior colliculus, has been shown to decrease the phase synchrony of the ASSRs to frequencies of stimulation within the gamma range (20–80 Hz) suggesting that this structure is a primary generator of the response (Kiren et al., 1994). The role of the cortex is greatly diminished under anaesthesia compared with that of the inferior colliculus (Szalda and Burkard, 2005). These data indicate that ASSRs, especially at frequencies below 80 Hz, likely reflect contributions from multiple brain generators with the primary auditory cortex and the inferior colliculus playing important roles.

Human studies

Intracranial recordings from patients with epilepsy indicate that the response is generated in medial Heschl's gyrus (Bidet-Caulet et al., 2007), which is consistent with several source analysis studies of the response (Gutschalk et al., 1999; Pantev et al., 1996; Ross et al., 2004; Schoonhoven et al., 2003) . It has therefore been suggested that the response is primarily generated in the auditory core area and in particular in the more medial field that would correspond to A1 in primate auditory cortex (Hackett et al., 2001). This conclusion is also supported by tonotopy studies (Pantev et al., 1996; (Romani et al., 1982; Wienbruch et al., 2006), which indicate that the ASSR generator is located more medial for higher and more lateral for lower frequencies.

2.1.4. Transient auditory responses

The arrival of auditory stimulus evokes a sequence of waves that are classified as fast (2-20ms), middle (10-100ms) and slow (50-300ms). The slow components are of interest in the current work as they are constitutive part of ASSRs (Martin et al., 2007).

Davis in 1939 was the first, who described changes in electroencephalogram in response to sound. He also described the P1-N1-P2 complex (Davis, 1939).

P1-N1-P2 component is generally considered to be comprised of slow components (50-300ms) (Martin et al., 2007). The P1-N1-P2 complex while composed of sensory evoked responses is not purely sensory. This component is not specific to the auditory modality: similar responses with different cortical sources are seen in other sensory modalities.

The P1-Nl-P2 complex is considered to index the arrival of stimulus information to the auditory cortex and the initiation of cortical sound processing, i.e. it represents the capacity for stimulus discrimination. While it is possible to dissociate them, P1, N1 and P2 are typically recorded together (Martin et al., 2007).

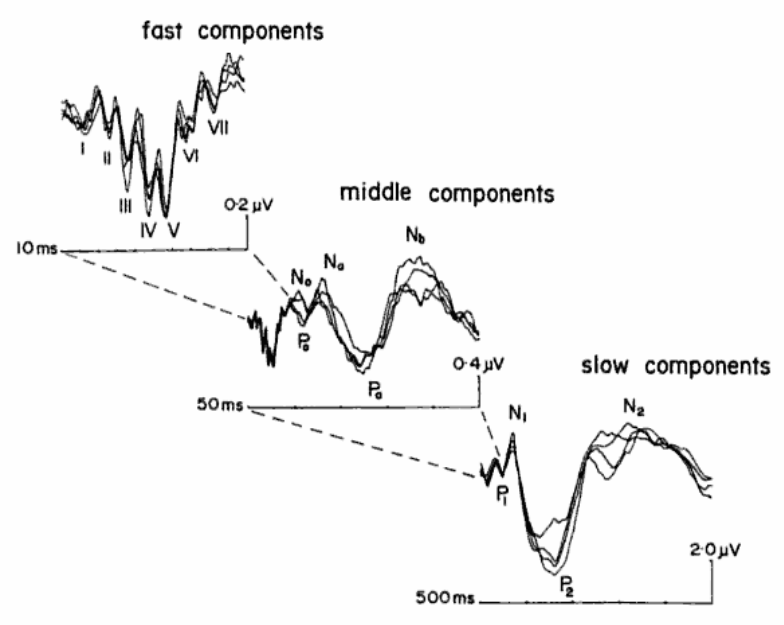

Figure 2.3. The sequence of waves evoked by the auditory stimulus. Fast components elicited within 0-10ms after the stimulus arrives, middle components elicited within 10-50ms after the stimulus presentation and slow components elicited within 50-500ms after the stimulus presentation. Modified from Naatanen (1992).

P1 is the first major component of the P1-N1-P2 complex. It is a vertex-positive voltage deflection that occurs approximately 50ms after the sound onset and thus it is often referred to as P50 potential. Generators have traditionally been identified in the primary auditory cortex and Heschl's gyrus. However additional regions contributing to P1 generation have been suggested: hippocampus, planum temporale, and lateral temporal cortex, neocortex (Ponton et al., 2002; Reite et al., 1987). P1 potential reflects a predominantly preattentional stimuli processing (Naatanen, 1992).

N1 appears as a negative peak at approximately 100ms after the stimulus onset. It is often referred to as N100 potential. N1 is known to have multiple generators in the primary and secondary auditory cortex. In general N1is maximally recorded from midline central scalp locations (Howard et al., 2000; Liegeois-Chauvel et al., 2004; Naatanen and Picton, 1987; Steinschneider et al., 1999; Vaughan and Ritter, 1970). The N1 evoked by

auditory stimuli in passive listening tasks could reflect a trigger to allocate attention to the stimulus (Naatanen, 1992).

P2 is a positive waveform that occurs approximately 180ms after the stimulus presentation; may be referred to as P200. P2 is not as well understood as P1 and N2. It appears to have multiple generators in auditory areas including the primary auditory cortex, the secondary cortex, and the mesencephalic reticular activating system (Hari et al., 1987; Rif et al., 1991; Scherg and Von Cramon, 1986). The P2 evoked by auditory stimuli in passive listening tasks could reflect early allocation of attention and initial conscious experience of the stimulus (Naatanen, 1992).

2.1.5. ASSR application

Currently ASSRs are widely applied in audiology, anaesthesiology, neurology and in psychiatry.

Audiology

In audiometry ASSRs are widely applied to detect hearing loss, to estimate hearing thresholds in hearing impaired subjects; to investigate discrimination of speech sounds (Dimitrijevic et al., 2004; Swanepoel and Erasmus, 2007). The application is especially widespread and informative in the subjects that cannot give adequate response like newborn infants (Rickards et al., 1994). Additionally, The 40Hz and 80Hz stimulation is used to check if the deaf-aid functions correctly (Picton et al., 2003a)

Anaesthesia

ASSRs are applied to test the depth of anaesthesia: the deeper is the level of anaesthesia, the lower is the ASSR (Plourde, 1996; Plourde and Boylan, 1991; Plourde and Picton, 1990; Plourde et al., 1998). However, the exception is the ketamine anaesthesia that increases ASSR amplitude (Plourde et al., 1997)

Neurology

The amplitude of ASSR is reduced in patients with impairments of brain stem and hippocampus (Picton et al., 2003a), and in patients with dyslexia (Poulsen et al., 2009). During coma and brain death ASSR is not present (Bonhomme et al., 2000). Likewise, ASSR is delayed during hypoxia (Lucertini et al., 2002) and changed during encephalopathy and auditory neuropathy (Harada et al., 1994; Santiago-Rodriguez et al., 2005; Shinn and Musiek, 2007).

Psychiatry

ASSRs are used to test the ability of the brain to follow incoming stimulation in psychiatric populations. There is a number of studies that showed reduced and less synchronized ASSR responses in schizophrenic patients (Brenner et al., 2009; Hong et al., 2004; Krishnan et al., 2009; Kwon et al., 1999; Light et al., 2006). ASSR amplitudes are also reduced in bipolar disorder (O'Donnell et al., 2004). However, it has been shown that ASSR amplitude is increased in Alzheimer disease patients (Osipova et al., 2006).

2.1.6. SSR measures

The signal-to-noise ratio of steady-state responses can be enhanced by averaging in the time or frequency domain (Stapells et al., 1984) because of their strict phase locking to the eliciting stimulus.

In the time domain, a recording can be measured by selecting peaks and throughs and calculating their amplitudes and latencies. In the frequency domain, activity is measured as the amplitudes and phases at particular frequencies. Both transient and SSRs can be measured in either the time or frequency domain. Although the frequency spectrum of a transient response may be evaluated over the whole duration of the response, these responses are more appropriately measured in the time domain (as peaks and throughs) The SSRs are most appropriately measured in the frequency domain, since one need only consider the peaks in the spectrum at the stimulus rate or its

harmonics. SSRs are therefore generally measured in the frequency domain (Picton et al., 2003a).

2.2. Time-frequency analysis

The assessment of the frequency component (oscillation) often yields insight into the functional sensory and cognitive correlations of the signal. Actually, oscillations were the first to be detected at the very beginning of EEG research. The German neurophysiologist Berger first observed the dominant oscillation recorded from the human scalp at 10 Hz and named it alpha (reviewed by Hermann et al., 2004.)

Oscillations are characterised by their amplitude and phase. At every point in time, the amplitude and phase of an oscillation can be determined. Oscillatory activity, based on the degree of phase-locking to the stimulus, is classified as 1) spontaneous (completely uncorrelated with the occurrence of the experimental condition), 2) induced (is correlated with a particular condition but not strictly phase-locked to the onset of stimulus and thus it is not apparent when averaged), 3) evoked (is strictly phase-locked, i.e. has the same phase in every stimulus repetition and thus it is evident when averaged) (Morup et al., 2006).

Several methods exist to extract and characterize the oscillations of a specific frequency from ERP data. Every signal can be decomposed into sinusoidal oscillations of different frequencies and the contribution of these frequencies to the signal can be evaluated. Among the most popular methods to do that are filtering, Fourier transformation and wavelet analysis (Herrmann et al., 2004).

2.2.1. Wavelet analysis

Wavelet analysis allows the representation of evoked activity in the time and frequency domain, i.e. to create a time-frequency (TF) map of the response (Hermann et al., 2004). Wavelet analysis makes it possible to evaluate specific

characteristics of the signal (time course, wave shape, fine structure details, frequency contents related to time). When evoked activity is analysed (like ASSRs), the TF representation of the ERP contains only that part of the activity that is phase-locked to the stimulus onset. The attractive property of the wavelet transform in accessing the oscillatory activity of EEG is that high frequency burst are believed to vary more rapidly in time than low frequency activities. Since the wavelet length for high-frequency analysis are shorter than low frequency analysis the wavelet gives a good trade-off between precision in time and in frequency of the EEG/MEG signals (Hermann et al., 2004).

Wavelet transform (WT) yields several measures that describe oscillatory activity form different perspectives. For example, the inter-trial phase coherence (ITPC) that is phase synchronization index (Morup et al., 2007). It is best conceptualized as phase precision or synchronization of the evoked oscillations from trial to trial ranging from 0 (random phase) to 1 (nearly identical phase) and total intensity (WTav), representing the average amplitude of the oscillation (both non-phase-locked and phase-locked). Let $X(c, f, t, n)$ denote the time–frequency coefficient at channel c, frequency f, time t and epoch n of the EEG/MEG signal given by $x(c, t, n)$, then (Morup et al., 2007)

$$\mathrm{ITPC}(c, f, t) = \frac{1}{N}\sum_{n}^{N}\frac{X(c, f, t, n)}{|X(c, f, t, n)|},$$

2.2.2. Data decomposition

A further and very novel step in data analysis is to apply complex mathematical methods to determine wavelet transformed coefficients that yield significant activity at the frequency of interest (Herrmann et al., 2004).

Most analyses are based on the time–frequency representation of single channels – they apply information based signal processing on channel × time decompositions, like the principal component analysis (PCA) or independent component analyses (ICA) previously reported (Delorme and Makeig, 2004; Rogers, 1991). The two-way analyses have to collapse dimensions of the data

when more than one data set is analyzed (as the data already have three dimensions: space, time and frequency). This problem is avoided in multi-way analyses like the parallel factor analysis, where all dimensions of the data can be explored. The multiway decomposition method Parallel Factor (PARAFAC) (one of the non-negative multi-way factorization methods), also named Canonical Decomposition (CANDECOMP), was recently used to decompose the wavelet transformed ongoing EEG of channel×frequency×time (Morup et al., 2006). In 2006, PARAFAC was used for the first time to decompose wavelet transformed event-related EEG given by the inter-trial phase coherence (ITPC) (Morup et al., 2006). The resulting component is then described, not only by the topography and the time-frequency signature but also by the relative contribution from the different subjects or conditions (Morup et al., 2006). Several reports appeared validating this way of data processing and showing it being useful (Arnfred et al., 2008; Arnfred et al., 2010). Moreover, open source Matlab based toolbox 'ERPWAVELAB' was developed for multi-channel time–frequency analysis of event related activity of EEG and MEG data (Morup et al., 2007). The flow chart of data processing encompassing this type of decompositions is given in Figure 2.4.

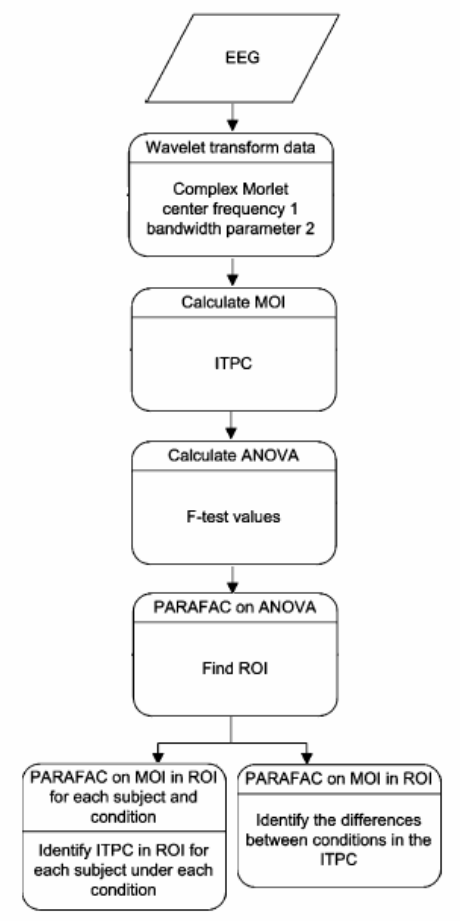

Figure 2.4. Flow chart of the analysis. The ERP was wavelet transformed using a complex Morlet wavelet with centre frequency 1 and bandwidth parameter 2. The measure of interest (MOI), i.e. the ITPC, was calculated for each subject and each condition according and a 3-way array of the ANOVA F test value calculated. The F test array was analyzed using PARAFAC and the region of interest (ROI, here in the time– frequency domain) of most difference between the two conditions identified. The ITPC in the ROI was then analyzed using PARAFAC on the 5-way array given by channel×frequency×time×subject×condition of ITPC values. Finally, the ITPC of the ROI of each subject under each condition given by the 3-way array of channel×frequency×time was also analyzed by PARAFAC and the individual peaks of the ITPC identified under each condition. Adapted from Morup et al. (2006).

2.3. Arousal, activation and attention

Arousal, activation and attention are closely interconnected processes that have effects on sensory and cognitive functioning of the subject.

2.3.1. Arousal

Since Yerkes and Dodson (1908), the relationship between arousal and performance has been repeatedly described as an inverted U-shaped function (Yerkes and Dodson, 1908). The early concept of arousal was one-dimensional: Arousal was assumed to reflect the general state of activity of the central nervous system ranging from deep sleep to excitement (Hebb, 1955). Hebb (1955) was also one of the first who ascribed a basic attentional function to arousal, modulating the processing of sensory input.

More elaborated views of arousal hold that ‘‘general arousal’’ is (a) supported by several distinct arousal systems that are, in turn, influenced by both external and internal factors, and (b) a prerequisite for, and a modulator of, the activation of specific attentional, cognitive, and motor systems (Garey et al., 2003).

Early electrophysiological studies located the neural substrate of cortical arousal regulation in the reticular formation, thought to exert bottom–up control over the arousal level of the entire cortex via widespread unspecific projections (Lindsley, 1968; Moruzzi and Magoun, 1949). However, the notion of a unitary arousal system had to be abandoned in favour of several neurochemically distinct arousal systems ascending from different subcortical regions (McCormick, 2003; Robbins, 1997; Robbins and Everitt, 1996).

2.3.2. Activation

Barry et al. (2005) recently argued that inconsistency in the literature arose from confusion between arousal (as discussed above) and activation and the distinction between ‘‘arousal’’ and ‘‘activation’’ has not been made explicit. Traditionally, these terms have been used interchangeably to refer to the energetics of a physiological system and the behavioural intensity associated with a response (Duffy, 1957; Malmo, 1959). Barry et al. (2005) demonstrated a clear separation between arousal, considered as a state variable reflecting current energetic factors, and task-related activation, considered as

the task related change in arousal (from a resting baseline to the task). Behavioural performance reflected activation (but not arousal), whereas phasic electrodermal stimulus-elicited responses reflected arousal (but not activation). This is generally compatible with Pribram and McGuiness (1975, 1992), who identified arousal and activation systems with different neural substrates (Pribram and McGuinness, 1975; Pribram and McGuinness, 1992).

Eyes-closed resting condition resembles the lowest level of arousal commonly accessible in the laboratory. With no task, it also represents the lowest level of activation achievable. Barry et al. (2007, 2009) suggested that the eyes-closed resting condition may be better identified as a convenient arousal baseline, with the eyes-open testing condition serving as a convenient activation baseline, particularly for tasks that involve visual processing. The latter is particularly important for those ERP studies involving the use of a visual fixation point to reduce ocular artefact (Barry et al., 2009a; Barry et al., 2007; Barry et al., 2009c).

During specific tasks such as reading, increased activity is found in the parietal areas of both hemispheres, focussed over language areas bordering the Sylvian fissure (Harmony et al., 1990; Rebert, 1978) where processing of the written word takes place. However, there are also changes found across the entire scalp during reading (and other cognitive tasks), which may indicate global arousal effects. That is, focal EEG changes associated with processing may occur with or without arousal changes.

In the present thesis ‘‘arousal’’ is defined as the current energetic level of the organism, and ‘‘activation’’ as a separable tonic measure reflecting the task-related mobilisation of energy (relative to some baseline level of arousal), which is needed to perform a task and to emphasize that the term “arousal/activation” is used. This conceptualisation is represented in Figure 2.5. Importantly, the two different aspects of the energetic dimension differentially influence physiological responding and behaviour, suggesting the usefulness of their separation and the need for further research.

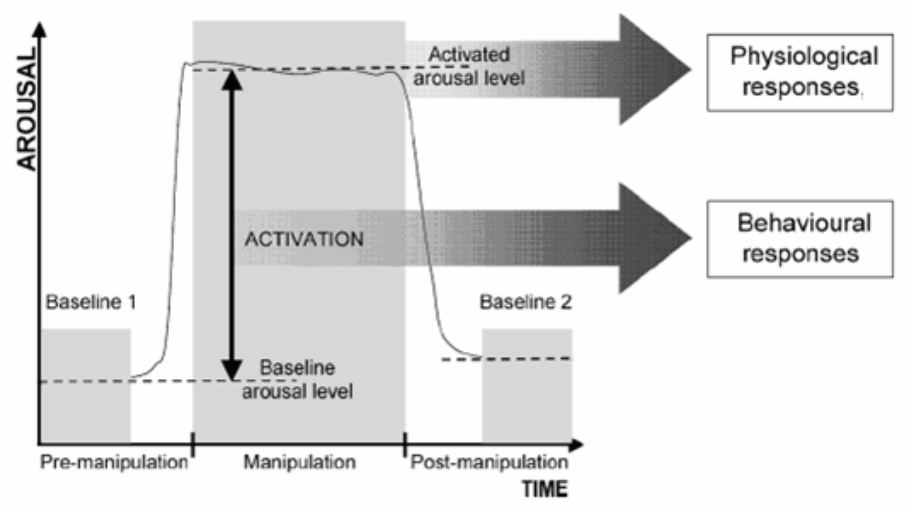

Figure 2.5. Separation of energetic effects on behavioural and physiological aspects of performance. "Arousal" refers to the individuals' energetic state at any moment and is measured as some physiological parameter.. Task requirements result in changes in arousal level. The task-related activation is defined as the change in arousal from resting baseline to the task. Current activated arousal level during the task affects physiological responses to stimuli presented in the task, while the activation affects task performance. Adapted from Barry et al. (2007).

2.3.3. Attention

Attention usually refers to a more focused activation of cerebral cortex that enhances information processing (Mesulam, 1981; Mountcastle, 1978; Posner et al., 1988).

The attention system of the brain is anatomically separate from the data processing systems that perform operations on specific inputs even when attention is oriented elsewhere. In this sense, the attention system is like other sensory and motor systems. It interacts with other parts of the brain, but maintains its own identity (Coull, 1998).

Attention is carried out by a network of anatomical areas. It is neither the property of a single centre, nor a general function of the brain operating as a whole (Mesulam 1981). The areas involved in attention carry out different functions, and these specific computations can be specified in cognitive terms (Posner et al., 1988). Three major functions are prominent in cognitive accounts of attention (Posner, 1980; Posner, 1987; Posner et al., 1982; Posner

et al., 1980): (a) orienting to sensory events; (b) detecting signals for focal (conscious) processing, and (c) maintaining a vigilant or alert state.

Attention can be fractioned into the following subprocesses: attentional orientation (the simple direction of attention to a particular stimulus); selective (or focused) attention (giving attentional priority to one stimulus in favour of another); divided attention (dividing attention between two or more different stimuli); sustained attention (attending to one stimulus over a prolonged period of time) (Table 2.1.).

Table 2.1. Characteristics and possible neural substrates of subtypes of attention (adapted from Perry and Hodges (1999)).

Attentional subtype	Defusing characteristics	Possible neural substrates
Selective attention	Focusing on single relevant stimulus or process at one time while ignoring irrelevant or distracting stimuli	Posterior parietal systems for orienting and shifting modulated by anterior midline and basal ganglia systems for response selection
Sustained attention	Maintenance of abilities to focus attention over extended periods of time	Right-sided frontoparietal system
Divided attention	Sharing of attention by focusing on more than one relevant stimulus or process at one time	Dorsolateral prefrontal cortex and anterior cingulated gyrus

In principle, the entry of information to the limited capacity system is controlled by two types of processes: active selection (focussed attention) and breakthrough of the unattended (passive attention) (James, 1890). The first is a top-down process, in which channels of information are selected or rejected under the direction of the central mechanisms of behaviour control. Certain

subfunctions of the selection processes might be automatic; for example, auditory streaming is largely automatic (Bregman et al., 1990; Sussman et al., 1999), but, at least in ambiguous cases, it is affected by top-down control (Sussman et al., 1998). The second is a bottom-up process that enables the conscious evaluation of those potentially important events that are not currently selected by the first mechanism. Without a good balance between the two, one cannot behave adequately in many situations (Perry and Hodges, 1999).

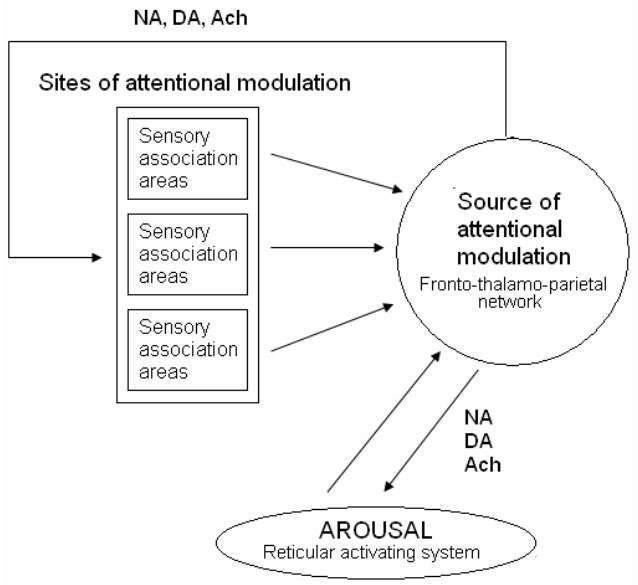

Figure 2.6. Hypothetical model of the neuroanatomical and neurochemical modulation of attention and its interaction with arousal. NA- noradrenaline, DA-dopamine, Ach-acetylcholine. Adapted from Coull (1998).

One of the models relating attention and arousal was proposed by Coull (1998) (Figure 2.6). The primary attentional generators feed back to sensory association areas in order to modulate neuronal firing in areas specifically recruited to process particular types of information so that relevant stimuli can be selected for further processing at an early stage of stimulus selection. Reciprocal connections between the attentional generators and the areas which express the attentional modulation are crucial to a comprehensive model of the neural correlates of attention. These connections may be modulated by activity

in neurotransmitter systems: directly via cortico-cortical projections and indirectly via modulation of arousal system (based on reticular activating system) (Coull, 1998).

2.3.4. ASSRs: arousal and attention

There is a limited number of studies that evaluated effects of changing arousal level on ASSRs. Most of the previous ASSR studies investigating the effect of arousal level evaluated ASSR during sleep. The main finding being diminished ASSR amplitude with stimulation frequencies up to 70Hz (Cohen et al., 1991; Jerger et al., 1992; Linden et al., 1985). However, Pockett and Tan (2002) showed an increased amplitude of the ASSR during low arousal state (Pockett and Tan, 2002).

ASSRs seem to be modulated by attentional demands. However the results within this field are conflicting: some authors reported enhancement of ASSRs with increasing attention to stimulation (Ross et al., 2004; Skosnik et al., 2007) and others found no effect of attention (de Jong et al., 2009; Linden et al., 1987). All the authors implied highly different study designs and experimental tasks. The summary of available studies is given in Table 2.2.

Table 2.2. A summary of currently available studies dealing with attentional modulation of ASSRs.

Study	Experimental tasks	Result
Linden et al. 1987	Unfocused closed eyes vs. reading; Counting vs. reading	No differences in amplitude and phase
Ross et al. 2004	Auditory stimulus modulation discrimination task vs. picture counting task	Largely enhanced sustained responses under the attention condition; N1 responses were not affected by the different states of attention

Gander et al. 2007	40Hz target vs. watching a silent movie	ASSR amplitude increased when the discrimination task was performed; Enhancement of N1 and P2 by attention
Skosnik et al. 2007	Classical oddball paradigm with 20Hz and 40Hz stimuli	Selective attention enhanced signal power and phase-locking of the 40Hz ASSR
Alegre et al. 2008	Attend to the sound vs. reading a novel	No differences in the 40-Hz phase precision and power values
Lazzouni et al. 2009	To detected a carrier frequency change in amplitude-modulated tones vs. ignoring tones; 40Hz stimuli	Attending to carrier frequency change enhanced the right hemisphere ASSR amplitude for dichotic stimulation
Muller et al. 2009	a dichotic listening experiment with a task to detect target tones in a cued ear; 20Hz and 40Hz stimuli	modulation of the ASSR by attention restricted to the left hemisphere and 20 Hz responses
Saupe et al. 2009	Attend auditory vs. Attend visual stimuli	Attention to the sounds led to a significant enhancement of the ASSR
De Jong et al. 2010	Intermodal detection and discrimination task	ASSR showed no effects of attention at frontocentral locations, but did so at occipital locations - evident only when attention was divided between audition and vision

EXPERIMENT I: AROUSAL/ACTIVATION EFFECTS

3. METHODS

3.1. Subjects

Eleven healthy subjects (six females) who reported normal hearing were included into the study. Written informed consent was obtained, as approved by the Ethics Committee of Copenhagen University Hospital (Copenhagen, Denmark), and the subjects were paid for the participation. Due to technical reasons, data of two subjects could not be used. The final sample consisted of nine subjects (four females). The mean age of the sample was 23.1 years (standard deviation (SD) 1.6).

3.2. Stimulation

A stimulus train lasted 1s and consisted of 40 and 20 clicks each being identical 1.5 ms burst of white noise. The stimuli were delivered through Sennheiser HD 565 Ovation© headphones at peak SPL of 60 dB. 72 presentations of the 40Hz and 20Hz trains were interspersed with trains of other frequencies (8, 10, 12, 30, 46 and 60Hz not reported here) with an inter-train interval of 1s. Two runs lasting 22 minutes were recorded in each of two conditions used. Conditions were defined as follows: “high activation” condition when subject was sitting upright and reading a self-selected book (the attention to reading was not controlled); “low activation” condition when subject was sitting much reclined with closed eyes and the lights turned down. In general, the instruction given to the subjects was to let their thoughts wander and not pay attention to the stimulation. The order of stimulation conditions was not counterbalanced across subjects.

3.3. EEG recordings

EEG was recorded with 61 scalp electrodes (BioSemi Active electrodes system) arranged according to the International 10-10 system. Digitally linked

earlobes electrodes were used as reference. The active recording reference electrodes (CMS-DRL) were placed centrally, close to POz. Data was recorded continuously at 2048 Hz/channel and band passed at 0.1-760 Hz by a LabView© application (ActivView©) on a Windows© based PC.

3.4. Data analysis

3.4.1. ERPs

15% of the epochs with the largest variability were rejected automatically. Grand averaged evoked potentials of ASSR for both conditions were created (individual averages based on 122 epochs, referenced to digitally linked earlobes, cut into epochs (-500 to +1500 ms), filtered band pass 10-50Hz).

3.4.2. Baseline EEG

Baseline EEG power was measured at 7 electrodes (F3, F4, Fz, C3, C4, Cz, Pz), through the fast Fourier transformation (FFT) of the 1000ms inter-trial interval of each trial.

For the 40Hz stimulation the averaged power in the beta range (12-32 Hz) was divided by the total power in the 1-32Hz range then multiplied by 100 to get a percentage estimate for each electrode and the electrode percentages were then averaged (Cardenas et al., 1997).

For the 20Hz stimulation baseline EEG theta (4-7Hz), alpha (8-12Hz) and beta (13-30Hz) power was measured to assess the level of arousal/activation: left frontal (F3, F5), midline frontal (Fz), right frontal (F4, F6), left central (C3, C5), midline central (Cz), right central (C4, C6), left posterior (P3, P5), midline posterior (Pz) and right posterior (P4, P6). Subjects were observed during the experiment and interviewed about their state of activation after it.

3.4.3. Wavelet transform

Off-line processing was performed in EEGLAB and ERPWAVELAB for MatLab© (Delorme and Makeig, 2004; Morup et al., 2007). Wavelet transformation (WT) was performed on the raw EEG data after the rejection of 15% of the epochs with the largest variability. WT (complex Morlet wavelet from MatLab© Wavelet Toolbox; the wavelet $\tilde{\varphi}(t) = \frac{1}{\sqrt{2\pi}} \exp(i2\pi t) \exp\left(-\frac{t^2}{2}\right)$) was performed with frequencies represented from 4 to 70 Hz, 2 Hz intervals between each frequency) was performed. This yielded the following measures: 1) the wavelet transformed evoked potential measure (avWT, evoked amplitude - corresponding to phase-synchronized WT amplitude measure), 2) phase synchronization index (inter-trial phase coherence, ITPC – phase precision), that is best conceptualized as phase precision or synchronization of the evoked oscillations from trial to trial ranging from 0 (random phase) to 1 (nearly identical phase) and 3) total intensity (WTav), representing the average amplitude of the oscillation (both non-phase-locked and phase-locked) (Morup et al., 2006).

The calculations were made based on Delorme and Makeig, 2004; Herrmann et al., 2005 in the following way: Let $X(c, f, t, n)$ denote the time–frequency coefficient at channel c, frequency f, time t and epoch n of the EEG/MEG signal given by $x(c, t, n)$.

$$\mathrm{ITPC}(c, f, t) = \frac{1}{N} \sum_{n}^{N} \frac{X(c, f, t, n)}{|X(c, f, t, n)|},$$

$$\mathrm{avWT}(c, f, t) = \frac{1}{N} \sum_{n}^{N} X(c, f, t, n).$$

$$\mathrm{WTav}(c, f, t) = \frac{1}{N} \sum_{n}^{N} |X(c, f, t, n)|.$$

These measures describe evoked activity dealing with different domains of the signal – amplitude and phase - that allows more detailed description of the response.

3.4.4. Decomposition

Individual time-frequency representation of avWT, ITPC and WTav across all channels was created. The avWTs, ITPCs and WTavs were decomposed through non-negative multi-way factorization (NMWF) (Morup et al., 2006).

The application of NMWF creates time-frequency plots of the avWT and ITPC while indicating how the parameter varies with experimental manipulation. In other words, the multi-subject NMWF analysis of the 3-way array of channel x time - frequency x subject – condition gives the subject-specific strength to the activity that is most common across subjects, conditions and runs, i.e. creates a subjects-weighted collapse and makes it possible to quantify (by giving the single estimation of the measure of interest) how the measure of interest varies with experimental manipulation for all the subjects in all conditions (Morup et al., 2006). This has proven being useful in the analysis of event-related potentials (Arnfred et al., 2008; 2010). Prior to NMWF analysis, random avWT and ITPC activity, estimated by calculating the mean of an artificially generated random avWT and ITPC samples, was extracted (Morup et al. 2006).

The primer window for mathematical decomposition was set as 10-70Hz and -500 to +1500 ms. As explained in the results section, further analyses were performed on more narrow time-frequency windows to focus on beta and gamma range activities separately: for beta range analyses (in case of 20Hz stimulation) a window of 16-26Hz and -10 to +1200 ms was used; for gamma range analysis a window of 30-46Hz and -10 to +1200 ms was selected.

3.4.5. Statistical analysis

The results of the NMWF decompositions were normally distributed (as indicated by Shapiro-Wilk test) and were further tested in repeated measures analysis of variance (r.m. ANOVA) (SPSS© v. 9.1) for effects of “condition”, “run” and “condition * run” interaction. Baseline power measures for 40Hz

stimulation were tested by Student's t-test; baseline power measures for 20Hz stimulation were tested by r.m. ANOVA for effects of "condition" and "region" and "condition * region" interaction.

4. RESULTS

4.1. 40Hz stimulation

ASSRs in response to 40Hz stimuli were detected for all subjects in both conditions. Grand averaged steady-state evoked potentials for low arousal/activation and high arousal/activation conditions are presented in Figure 4.1 A.

4.1.1. NMWF scores of avWT

The NMWF decomposition of avWT resulted in the observation of two distinct components: a component at the frequency of stimulation (Figure 4.1 B) and a noise component (Figure 4.1 E). Time-frequency plots as a weighted collapse (relative weighting strength is given by the subject scores) across subjects and electrodes for the ASSR and noise component in both conditions are presented in Figure 4.1 C and F. The r.m. ANOVA demonstrated that avWT of the focal 40Hz ASSR component is significantly larger in the low arousal condition ($F(1,8)=7.463$, $p=0.026$) (Figure 4.1 D). Whereas avWT of the noise component was significantly larger in the alert reading condition ($F(1,8)=30.488$, $p=0.001$) (Figure 4.1 G).

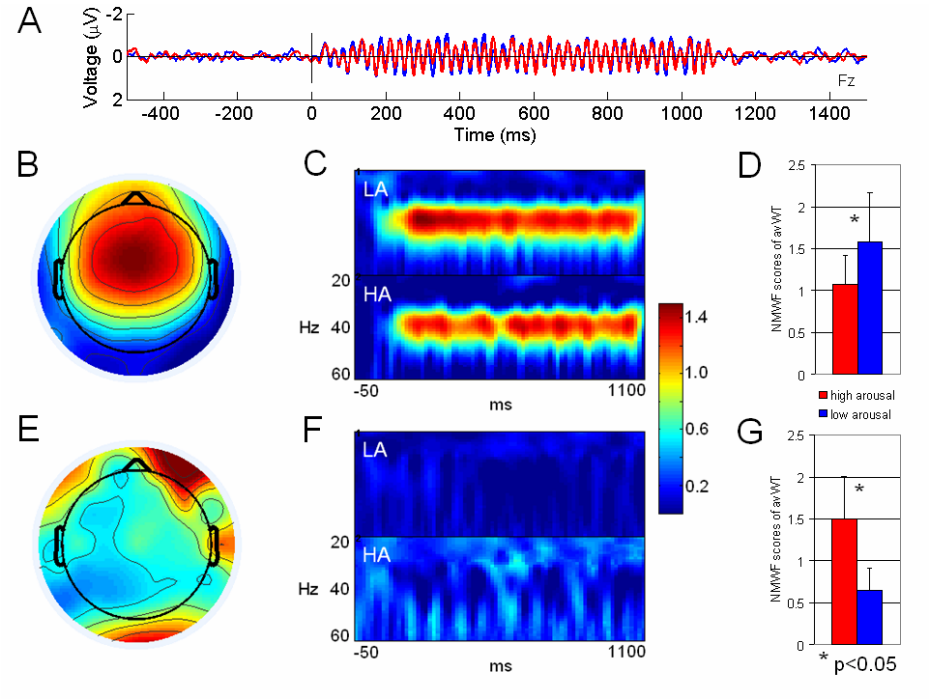

Figure 4.1. avWT results of 40Hz stimulation. A Grand averaged evoked potential for 40Hz ASSR in low arousal/activation (LA) and high arousal/activation (HA) conditions, N=9. Headplots of avWT collapsed across subjects for the ASSR (B) and noise component (E). Time-frequency plots as a weighted collapse across subjects and electrodes for the ASSR (C) and noise (F) components in low arousal/activation (LA) and high arousal/activation (HA) conditions. Means and standard deviations of NMWF scores of avWT for the ASSR (D) and noise (G) components in both experimental conditions.

4.1.2. NMWF scores of ITPC

The NMWF decomposition of ITPC resulted in the detection of one component (Figure 4.2 A). Time-frequency plots of ITPC as a weighted collapse across subjects and electrodes for 40Hz in both conditions are presented in Figure 4.2 B. The r.m. ANOVA demonstrated significant effect of condition on the ITPC: ($F_{1,8}$ = 15.391, p = 0.004), but no effect of run and no interaction effect. NMWF scores of the ITPC were higher in the low arousal/activation condition (Figure 4.2 C).

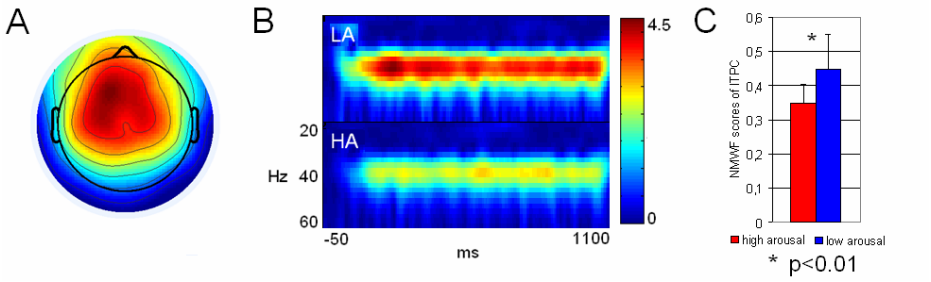

Figure 4.2. ITPC results of 40Hz stimulation. A Headplot of ITPC collapsed across subjects reveals a single component, N=9. B Time-frequency plots of ITPC as a weighted collapse across subjects and electrodes for 40Hz in low arousal/activation (LA) and high arousal/activation (HA) conditions. C Means and standard deviations of NMWF scores of ITPC for both conditions.

4.1.3. Baseline Beta Power

The percentage of beta power was significantly lower in the low arousal/activation condition (22.11% (SD 3.59) vs. 19.45% (SD 4.62); t=-2,833; df=17, p=0.017).

4.2. 20Hz stimulation

ASSRs in response to 20Hz stimulation were detected for all subjects in both conditions. The grand averaged steady-state evoked potential from Fz electrode for low arousal/activation and high arousal/activation conditions is given in Figure 4.3 A.

The NMWF decomposition of avWTs and ITPCs resulted in the observation of two distinct components: a component at the frequency of stimulation (in the following named 20Hz ASSR) and a component emerging at 40Hz (named 20Hz ASSR-related 40Hz activity). Full range time-frequency plots of ASSR as a weighted collapse (relative weighting strength is given by the subject scores) across subjects, electrodes and conditions are presented in Figure 4.4 A and Figure 4.5 A. As the detected components were not separated by the decomposition, each subcomponent was separately analyzed in a narrowed time-frequency window. Means and SDs of avWTs, ITPCs and WTavs are given in Table 4.1.

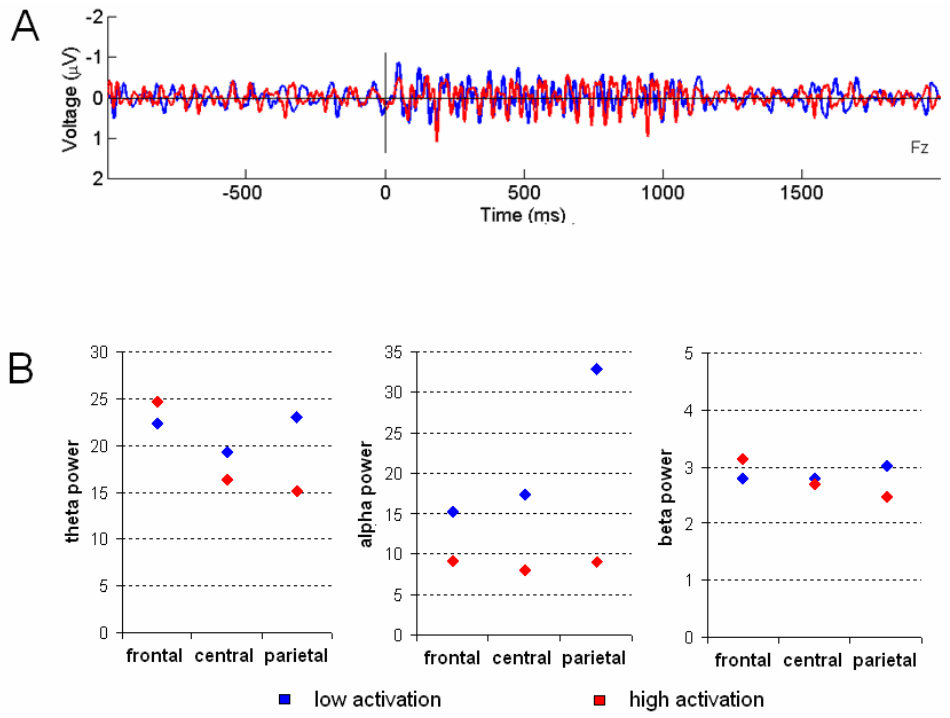

Figure 4.3. 20Hz stimulation: ERP and background EEG power results. A Grand-averaged ERPs for both experimental conditions from Fz electrode are depicted (N=9; individual averages based on 122 epochs). B Powers of theta, alpha and beta bands averaged for frontal, central and parietal conditions are presented. Alpha power significantly differed between two experimental conditions ($p<0.05$, N=9).

Table 4.1. Means and SDs of ITPC, avWT and WTav of 20Hz ASSRs and 20Hz ASSR-related 40Hz activity from 20Hz stimulation.

	Condition	ITPC		avWT		WTav	
		Mean	SD	Mean	SD	Mean	SD
20Hz ASSR	Unfocused closed eyes	0,144	0,010	0,192	0,018	0,348	0,023
	Reading	0,145	0,010	0,196	0,014	0,366	0,020
20Hz ASSR-related 40Hz activity	Unfocused closed eyes	0,165	0,009	0,202	0,013	1,065	0,006
	Reading	0,157	0,012	0,192	0,015	1,070	0,011

4.2.1. 20Hz ASSR

As assessed by r.m. ANOVA, neither run nor condition (level of activation) had significant effects on avWTs and ITPCs of 20Hz ASSR ($F(1,8)=1.018$, $p=0.343$ and $F(1,8)=0.681$, $p=0.433$ respectively) (Figure 4.4 B and 4.5 B). There was no interaction of the factors observed. There was a trend for higher WTavs of 20Hz ASSR in high arousal/activation condition, however it did not reach statistical significance ($F(1,8)=4.719$, $p=0.062$). The effect of run and the interaction of analyzed factors were not significant.

4.2.2. 20Hz ASSR-related 40Hz activity

The r.m. ANOVA indicated significant difference between two conditions for both avWTs and ITPCs of 20Hz ASSR-related 40Hz activity ($F(1,8)=7.970$, $p=0.022$ and $F(1,8)=5,941$, $p=0.041$ respectively), higher scores being obtained in the low arousal/activation condition (Figure 4.4 C and 4.5 C). No effect of run and interaction of run*condition factors emerged. There were no effects of either run or condition ($F(1,8)=1.357$, $p=0.278$), as well as their interaction, observed for WTavs of 20Hz ASSR-related 40Hz activity.

4.2.3. Baseline theta, alpha and beta power

The r.m. ANOVA failed to demonstrate any significant effect of condition and region for theta power, but there was a trend for condition*region interaction to be significant ($F(1,8) = 158.354$, $p = 0.061$), indicating higher frontal midline theta dominance in a high activation condition. Significant effect of condition on alpha power emerged ($F(1,8)= 13.310$, $p = 0.007$), alpha power being higher in the low arousal/activation state, but no effect of region and interaction of the factors was observed. Neither condition nor interaction of condition*region factors had an effect for beta power, meaning that beta power did not differ between conditions. However, a nearly significant effect of region was observed ($F(1,8)=181.789$, $p=0.057$), pointing to larger beta power over right frontal area. Means of theta, alpha and beta powers from both conditions are given in Figure 4.3 B.

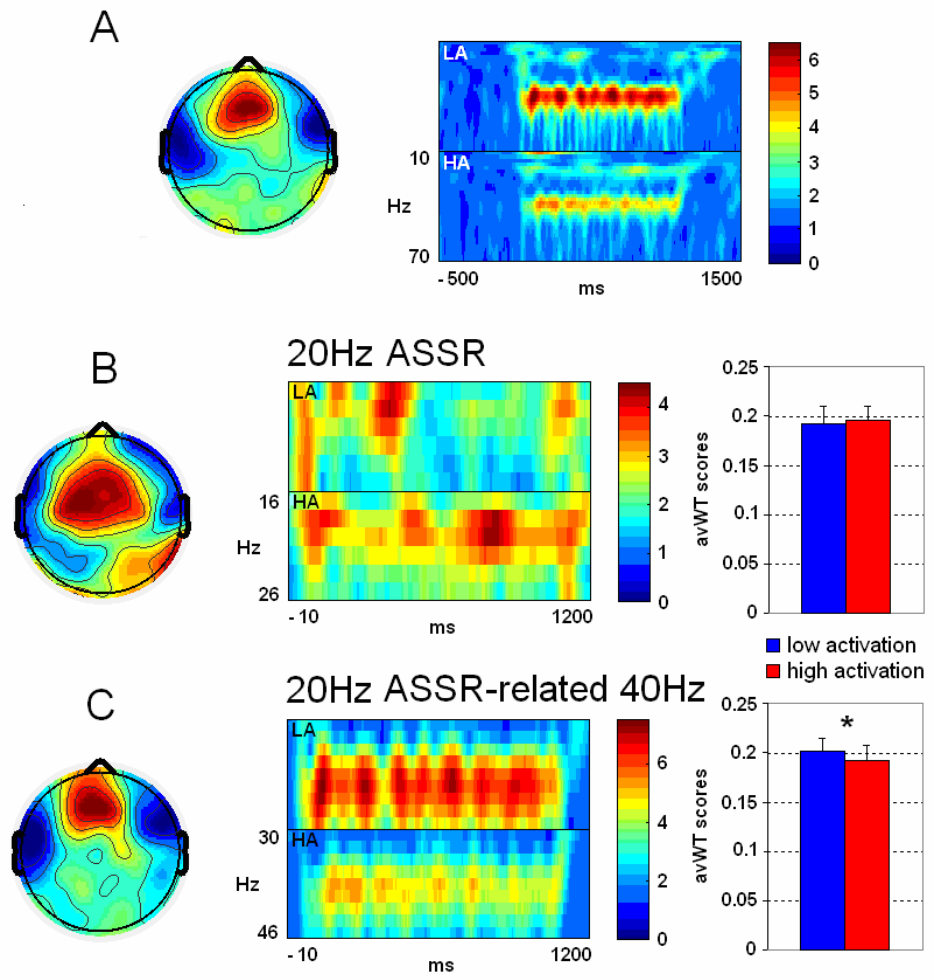

Figure 4.4. avWT results of 20Hz stimulation. Headplot and broad time-frequency plot (10-70Hz, -500 – 1500ms window) of avWT (A) as a weighted collapse across subjects and electrodes for the ASSR in low arousal/activation (LA) and high arousal/activation (HA) conditions. Note appearance of pronounced activity around 40Hz. Headplots and time-frequency plots of avWTs were created as a weighted collapse across subjects and electrodes for 20Hz ASSR (B) and for 20Hz ASSR-related 40Hz activity (C) in both experimental conditions (LA – low arousal/activation, HA – high arousal/activation). As shown by means and standard deviations of the scores, avWTs of 20Hz ASSR did not differ between conditions whereas avWTs of 20Hz ASSR-related 40Hz activity were higher during low arousal/activation condition ($p < 0.05$, N=9).

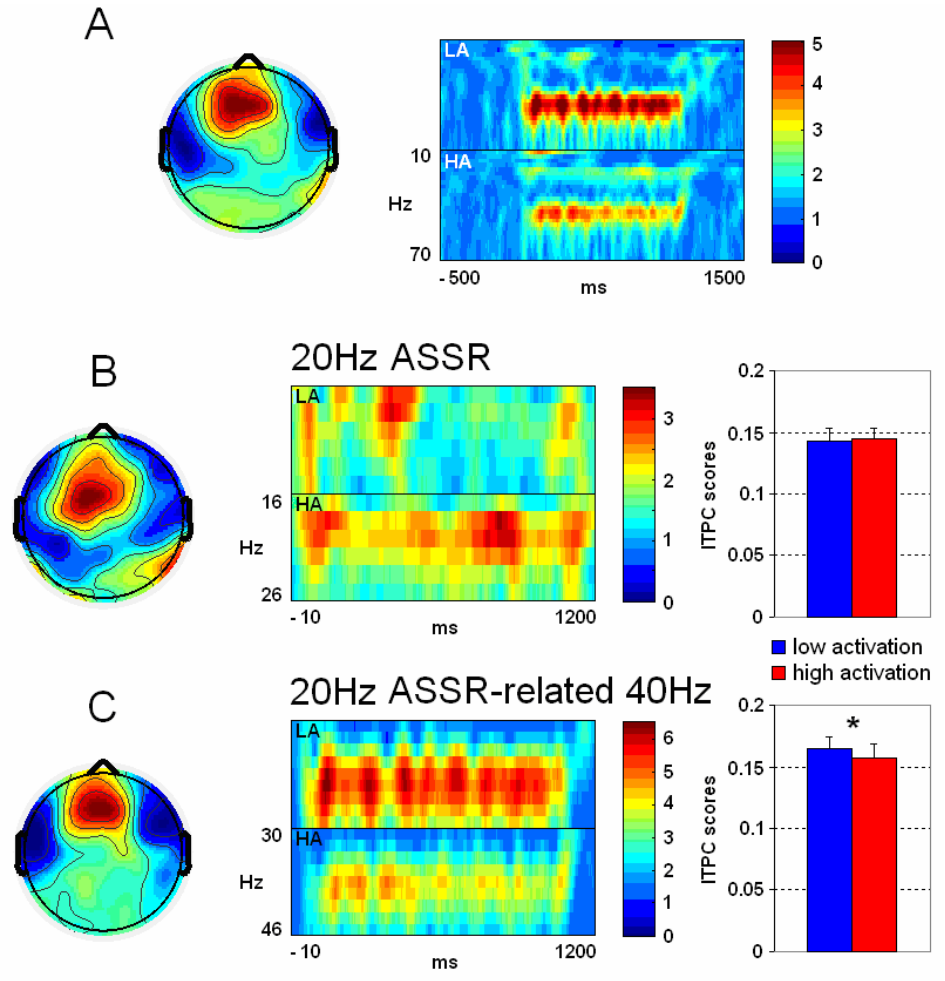

Figure 4.5. ITPC results of 20Hz stimulation. Headplot and broad time-frequency plot (10-70Hz, -500 – 1500ms window) of ITPC (A) as a weighted collapse across subjects and electrodes for the ASSR in low arousal/activation (LA) and high arousal/activation (HA) conditions. Note appearance of pronounced activity around 40Hz. Headplots and time-frequency plots of ITPCs were created as a weighted collapse across subjects and electrodes for 20Hz ASSR (B) and for 20Hz ASSR-related 40Hz activity (C) in both experimental conditions (LA – low arousal/activation, HA – high arousal/activation). As shown by means and standard deviations of the scores, ITPCs of 20Hz ASSR did not differ between conditions whereas ITPCs of 20Hz ASSR-related 40Hz activity were higher during low arousal/activation condition ($p < 0.05$, N=9).

EXPERIMENT II: AROUSAL, ACTIVATION AND ATTENTION

5. METHODS

5.1. Subjects

Sixteen healthy subjects (eight females) participated in the study. Eleven subjects were included in ”Arousal” part (mean age 22.27, SD 2.20; seven females) and eleven subjects were included in “Arousal/activation and attention” part (mean age 22.82, SD 2.18; five females); 6 subjects participated in both parts. All subjects reported normal hearing. Written informed consent was obtained, as approved by the Ethics Committee of the Republican Vilnius Psychiatric Hospital (Vilnius, Lithuania).

5.2. Stimulation

This section, except for the part described below is identical to the ‘Stimulation’ section of Experiment 1 (p. 34).

Stimuli were generated with Adobe Audition 3.0 and presented through custom EEG apparatus stimulation system. 60 presentations of the 40Hz train and 60 presentations of the 20Hz train were presented in a pseudo-random order with an inter-train interval of 3s. One stimulation trial lasted about 8 minutes.

5.3. Procedures

5.3.1. Arousal

The first part was designed to explore the modulation in the arousal level and check how it affects ASSRs. There were two experimental conditions with two runs each, where an auditory stimuli (20Hz and 40Hz) were presented: 1. the first run with open eyes and fixation at the cross on the computer screen in front of the subject; 2. the first run with closed eyes and lights turned off, followed by 10 minutes of silence with closed eyes and lights turned off; 3. the second run with closed eyes and lights turned off; 4. the

second run with open eyes fixated at the cross. The instruction given to the subjects was to let their thoughts wander and pay no attention to the stimulation. At the end of experiment subjects were enquired about their state during the experiment. Summing up, the level of arousal was diminishing in the following way: 1) the first run of eyes open condition, 2) the second run of eyes open condition, 3) the first run of eyes closed condition, 4) the second run of eyes closed condition. The order of runs was not counterbalanced and the 10 minutes period without auditory stimulation was introduced to allow more substantial drop in arousal from the first run with eyes closed to the second run with eyes closed.

5.3.2. Arousal/activation and attention

The second part of the experiment was designed to achieve the modulation of the ASSR by attention paid to the stimuli and by the change in the level of activation. Six tasks that differed by the level of focused attention were selected: counting 20 Hz and 40Hz stimuli, sitting with closed eyes, sitting with opened eyes, reading an article, and performing a search task. The order of tasks was counterbalanced across subjects and auditory stimulation was present during all tasks.

In the counting task subjects were instructed to count 40Hz stimuli in one recording run and 20Hz stimuli in another run. The subjects were asked to report the answer after each counting run. It was expected that attention would always be focused on all the stimuli but with enhanced processing of the counted targets. Thus, counting condition was referred to as “focused attention” and was defined by high attentional demands to stimulation. The counting target condition further is referred to as “focused on target”; corresponding response to non-target is further refer to as ”focused on non-target”.

Sitting with closed eyes and sitting with open eyes fixated conditions were referred to as ”unfocused attention” conditions, that were defined by no

specific attentional demands and further will be referred to as “unfocused closed eyes” and “unfocused open eyes”.

During reading task subjects read a magazine article and were informed about the enquiry on the context after the task completion.

The search task consisted of a visual search for Landolt rings having a gap at various positions (left, right, bottom, top and the 45° positions in between) with appropriate (1 p.m. and 9 p.m for all the subjects) gap orientation printed on the paper (Bourdon 1895).

Reading and performing a searching task - were assigned to as ”distraction” and these were conditions defined by low attention to stimulation as the subjects were absorbed in the distracting task. Conditions further are referred to as “reading” and “the search task”.

Summing up, the level of attention paid to the stimuli was diminishing in the following way: 1) focused on targets, 2) focused on non-targets, 3) un-focused closed eyes; 4) un-focused opened eyes, 5) reading (easy task) and 6) the search task (difficult task). The level of arousal/activation (i.e. mobilization of energy resources for task completion) was increasing in the following manner: 1) unfocused closed eyes, 2) unfocused open eyes condition, 3) focused on non-target, 4) focused on target, 5) reading and 6) the search task (Figure 5.1).

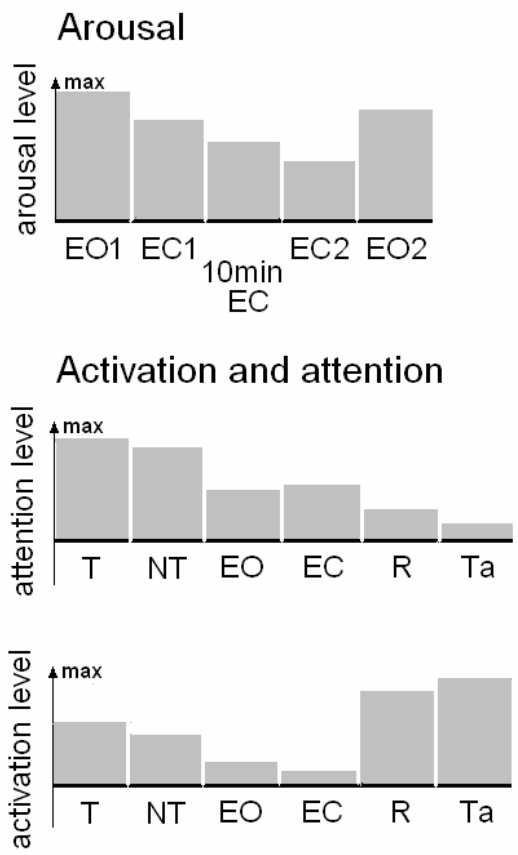

Figure 5.1. A schema of experimental design and expected arousal, attention and activation levels. In "Arousal" part, during auditory stimulation, subjects were instructed to sit with open eyes (EO1), then with closed eyes and light turned of (EC1). This was followed by 10 minutes of silence with closed eyes (10 min EC); recording run with closed eyes (EC2) and with eyes open (EO2). Expected arousal level and its changes are depicted in grey. There were 6 recording conditions in "Arousal/activation and attention" part, when subject had to count 40Hz or 20Hz stimuli (for 40Hz ASSR, targets (T) were 40 Hz stimuli, non targets (NT) were 20Hz stimuli; for 20Hz ASSR targets were 20Hz stimuli and non targets were 40Hz stimuli), stay with unfocused eyes open (EO), stay unfocused with eyes closed (EC), reading (R) and perform Landolt rings searching task (Ta). Note, that the order of conditions was counterbalanced between subjects. Expected level of activation and stimulus-focused attention is depicted in grey.

5.4. EEG recordings

The EEG was recorded with a 32- channel EEG device (Galileo NT, by EBNeuro, Italy) from F3, Fz, F4, C3, Cz, C4, P3, Pz and P4 sites (according to the 10/20 International system) using Ag/AgCl electrodes. Averaged earlobe

electrodes served as a reference for all electrodes and the ground electrode was attached to the forehead. The impedance was kept below 5 kΩ. Data was recorded at 512 Hz and band passed at 0.1-760 Hz.

5.5. Data analysis

5.5.1. ERPs

This section is identical to the 'ERPs' section of Experiment I (p. 35).

10% of the epochs with the largest variability were rejected automatically.

5.5.2. Baseline EEG

For the evaluation of arousal level baseline EEG alpha (8-12Hz) activity was monitored in both parts. Likewise, gamma (30-40Hz) power was monitored to evaluate the intervention of muscular noise in the frequency range of activity of interest. These measures were obtained for 3 regions through averaging the fast Fourier transformation results of the 1000ms inter-trial interval: frontal (F3, Fz, F4), central (C3, Cz, C4) and parietal (P3, Pz, P4).

5.5.3. Wavelet transform

This section is identical to the 'Wavelet transform' section of Experiment I (p. 36).

5.5.4. Decompositions

This section, except for the part described below is identical to the 'Decompositions' section of Experiment I (p.37).

Prior to WT, 10% of the epochs with the largest variability were rejected automatically. The primer window for mathematical decomposition of ASSRs was set as 10-80Hz and -500 to +1500 ms to define windows of the activity of interest. Further analyses were performed on narrow time-frequency windows: for beta range analyses (in response to 20Hz stimuli) a window of

16-26Hz and -10 to +1100 ms was used; for gamma range analysis (in response to 20Hz and 40Hz stimuli) a window of 30-46Hz and -10 to +1100 ms was selected. In order to evaluate temporal dynamics of task effect on ASSRs, separate analyses were conducted on 100ms epochs starting with 0 ms and ending with 1100 ms.

5.5.5. Statistical analysis

The results of the NMWF decompositions were normally distributed (as indicated by Shapiro-Wilk test, SPSS© v. 9.1). avWT, ITPC and WTav data (both full time window and each interval) from ”Arousal” part were analysed in repeated measures analysis of variance (r.m. ANOVA) with factors “condition” (open eyes vs. closed eyes) and “run” (run 1 vs. run 2), and the interaction of factors. The avWT, ITPC and WTav (both of full time window and each interval) from ”Arousal/activation and attention” part were analysed in univariate analysis of variance (uANOVA) for the effect of “task” (focused on target vs. focused on non-target vs. eyes closed vs. eyes open vs. reading vs. search task). Baseline power measures were tested by repeated measures ANOVA for effects of “condition” (eyes open vs. eyes closed), “run” (run 1 vs. run2) and “region” (frontal vs. central vs. parietal) and interaction of these factors in ”Arousal” part. In ”Arousal/activation and attention” part, alpha and gamma baseline measures were tested in univariate ANOVA for effect of task (focused on target vs. focused on non-target vs. eyes closed vs. eyes open vs. reading vs. search task) and “region” (frontal vs. central. vs. parietal) and interaction of these factors. All post hoc analyses were performed using Least Significant Difference (LSD) test.

Additionally, some paired T-Tests were performed on the data from ”Arousal/activation and attention” part: avWT and ITPC of unfocused closed eyes condition was compared with reading condition to assess the repeatability of results of Experiment I; WTav of unfocused closed eyes and unfocused open eyes were compared to uncover the effect of closed/open eyes without

significant modulation of arousal and to compare it with results of "Arousal" part.

6. RESULTS

ASSRs were detected for all subjects. In line with Experiment I, the non-negative multi-way factorization decomposition of selected measures of ASSRs resulted in the observation of a single frontal component at 40Hz following 40Hz stimulation (40Hz ASSR) and two distinct frontally distributed time-frequency subcomponents, that could not be separated via decomposition (20Hz ASSR and 20Hz ASSR-related 40Hz activity, maximal over Fz electrode) following 20Hz stimulation.

6.1. Arousal

6.1.1. 40Hz ASSR

WTav of 40Hz ASSR ($F(1,10)=5.076$, $p=0.048$) was significantly higher in the low arousal closed eyes condition. This effect was present in nine out of eleven subjects. Grand averaged time-frequency plot of WTav in closed eyes and open eyes conditions together with means and SDs are given in Figure 6.1 A and B. Run or interaction effects were not significant. Analysis of time intervals revealed that effect of condition was significant at 100-200 ms ($F(1,10)=5.581$, $p=0.04$) and nearly significant at 600-700 ms ($F(1,10)=4.789$, $p=0.053$). No effects were observed on ITPCs and avWTs.

6.1.2. 20Hz ASSR and 20Hz ASSR-related 40Hz activity

There were no effects of either run or condition on avWTs, ITPCs and WTavs of ASSRs.

6.1.3. Baseline alpha and gamma power

As expected, alpha power was higher in the low arousal closed eyes condition ($F(1,10) = 7.912$, $p = 0.018$) and during second runs ($F(1,10)=10.998$, $p=0.008$). Moreover, significant effect of region was observed ($F(2,9)=6.635$, $p=0.017$), pointing to overall larger alpha power over parietal area as compared to frontal and central regions. Significant effect of region was observed for gamma power ($F(2,9)=4.959$, $p=0.035$), pointing to larger gamma power over parietal area as compared to frontal and central regions. No interaction of the factors was observed. Means of alpha and gamma power over frontal, central and parietal regions are given in Figure 6.1 C.

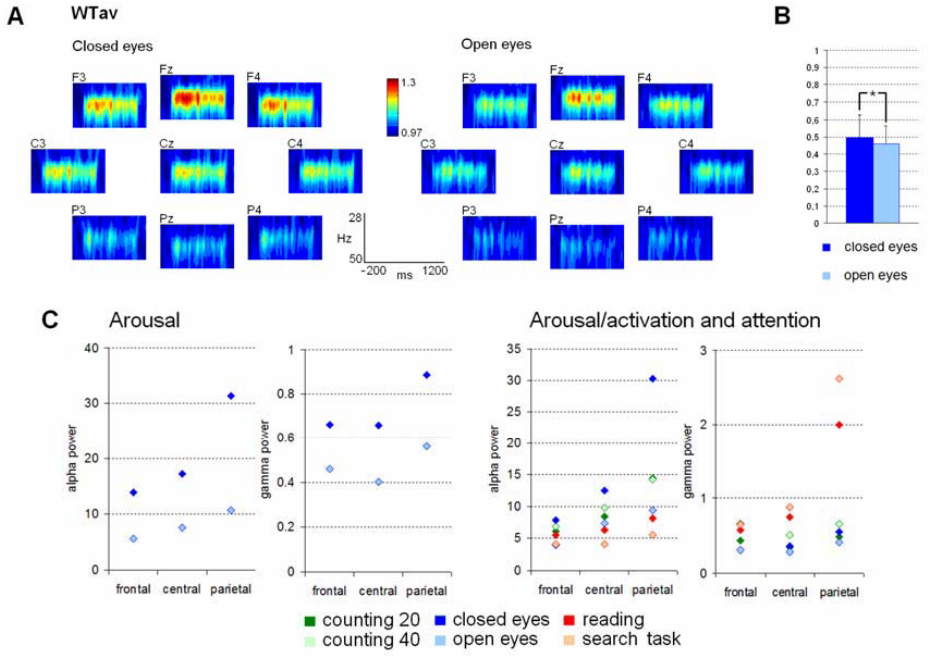

Figure 6.1. WTav results of 40Hz stimulation in "Arousal" part and baseline EEG data from Experiment II. Grand averaged time-frequency plots (A) (for graphical representation 28-50 Hz, -200 – 1200ms window) of 9 EEG channels in a topographical representation of WTav in eyes closed and eyes open conditions of Experiment 1. Means and SDs from Fz electrode (B) - higher WTav at Fz electrode in closed eyes condition as compared to open eyes condition (*$p<0.05$, N=11). (C) Means of alpha and gamma power over frontal, central and parietal regions for both "Arousal" and "Arousal/activation and attention" parts.

6.2. Arousal/activation and attention

Means and SDs of avWT, ITPC and WTav of 40Hz ASSR, 20Hz ASSR and 20Hz ASSR-related 40Hz activity in all conditions are given in Table 6.1. Grand averaged time-frequency plots of 40Hz ASSR avWT and ITPC from Fz electrode of six experimental conditions are given in Figure 6.2 A and B together with corresponding means and SDs (Figure 6.2 C and D).

Table 6.1. Means and SDs of ITPCs, avWTs and WTavs of 40Hz ASSR, 20Hz ASSRs and 20Hz ASSR-related 40Hz activity.

	Condition	ITPC		avWT		WTav	
		Mean	SD	Mean	SD	Mean	SD
40Hz ASSR	Focused on target	0,424	0,135	0,596	0,225	0,376	0,067
	Focused on non-target	0,430	0,138	0,621	0,230	0,395	0,068
	Unfocused closed eyes	0,477	0,154	0,696	0,261	0,417	0,069
	Unfocused open eyes	0,441	0,150	0,637	0,247	0,391	0,075
	Reading	0,337	0,101	0,459	0,157	0,360	0,035
	The search task	0,326	0,074	0,447	0,113	0,362	0,032
20Hz ASSR	Focused on target	0,184	0,016	0,267	0,053	1,069	0,031
	Focused on non-target	0,185	0,018	0,268	0,049	1,072	0,023
	Unfocused closed eyes	0,177	0,019	0,286	0,060	1,100	0,041
	Unfocused open eyes	0,184	0,016	0,296	0,067	1,076	0,022

	Reading	0,187	0,023	0,238	0,023	1,079	0,024
	The search task	0,187	0,027	0,257	0,051	1,073	0,036
20Hz ASSR-related 40Hz activity	Focused on target	0,189	0,016	0,239	0,019	0,281	0,029
	Focused on non-target	0,190	0,017	0,237	0,026	0,277	0,024
	Unfocused closed eyes	0,182	0,019	0,235	0,021	0,289	0,021
	Unfocused open eyes	0,189	0,015	0,240	0,028	0,292	0,024
	Reading	0,191	0,022	0,244	0,039	0,281	0,028
	The search task	0,192	0,026	0,247	0,040	0,297	0,009

6.2.1. 40Hz ASSR

The analysis of average ITPCs and avWTs showed that they were similarly affected by the tasks: the main effect of task was significant (ITPC: $F(5,60)=2.424$, $p=0.046$; avWT: $F(5,60)=2.482$, $p=0.041$). Likewise the post-hoc tests showed that both measures were attenuated during the reading as compared to the unfocused closed eyes condition ($p=0.011$ and $p=0.014$). This was substantiated by planned paired T-Test ($t=4.210$, $df=10$, $p=0.002$ and $t=4.326$, $df=10$, $p=0.001$). avWTs and ITPCs were lower during the search task as compared to the unfocused closed eyes ($p=0.008$ and $p=0.008$) and unfocused open eyes ($p=0.04$ and $p=0.041$) conditions. There were no significant effects on WTavs of 40Hz ASSRs.

Analyses of intervals showed significant effect of task on avWTs and ITPCs at 400-500ms ($F(5,60)=2.500$, $p=0.04$ and $F(5,60)=2.533$, $p=0.038$), at 500-600ms ($F(5,60)=2.520$, $p=0.039$ and $F(5,60)=2.524$, $p=0.039$), at 700-800ms ($F(5,60)=2.909$, $p=0.02$ and $F(5,60)=2.554$, $p=0.037$) and at 800-900ms only for avWT ($F(5,60)=2.524$, $p=0.039$). Post hoc testing indicated that at

400-500ms avWT and ITPC were larger during focused on target (p=0.044 and p=0.039) and focused on non-target (p=0.048 and p=0.032) than during the search task. Moreover, the measures were attenuated during reading (p=0.014 and p=0.017) and the search task (p=0.007 and p=0.007) as compared to unfocused closed eyes condition. At 500-600ms avWT and ITPC were attenuated during reading as compared to focused on target (p=0.025 and p=0.021), unfocused closed eyes (p=0.025 and p=0.027) and unfocused open eyes (p=0.031 and p=0.035). Similarly, avWTs and ITPCs during the interval of 500-600ms were attenuated during the search task as compared to focused on target (p=0.023 and p=0.021), unfocused closed eyes (p=0.023 and p=0.022) and unfocused open eyes (p=0.029 and p=0.03) conditions. At 700-800ms the measures were lower during reading (p=0.005 and p=0.009) and the search task (p=0.002 and p=0.005) as contrasted to unfocused closed eyes state and lower during the search task as compared to unfocused open eyes state (p=0.036 and p=0.043). Means of ITPC and avWT of the intervals are given in Figure 6.3.

Paired T-Test revealed that both avWTs and ITPCs (t=4,210, df=10, p=0.002 and t=4,326, df=10, p=0.001) during unfocused closed eyes condition were significantly higher than during reading. WTav during unfocused closed eyes condition was larger as compared to unfocused open eyes condition (t=4,421, df=10, p=0.001).

6.2.2. 20Hz ASSR and 20Hz ASSR-related 40Hz activity

No task effects were observed on ITPC, avWT and WTav of 20Hz ASSR or 20Hz ASSR-related 40Hz activity. But paired T-Test revealed that avWT and ITPC of 20Hz-related 40Hz activity (t=2.758, df=10, p=0.02 and t=2.632, df=10, p=0.025) were larger in unfocused closed eyes condition as compared to reading condition. There were no differences obtained by paired T-Test for 20Hz ASSRs.

6.2.3. Baseline alpha and gamma power

Task and region factors and their interaction had significant effect for alpha power ($F(5,180)=6.926$, $p<0.001$; $F(2,180)=12.262$, $p<0.001$; $F(10,180)=2.023$, $p=0.033$). Post hoc test revealed that during closed eyes condition alpha power was significantly larger ($p<0.05$) than during all other conditions and that alpha power was significantly larger over parietal region as compared to frontal and central regions ($p<0.05$) in general. The interaction analysis revealed the following ($p<0.05$ in all below mentioned cases): parietal alpha power in focused on target condition (counting 40Hz stimuli) was significantly larger than frontal alpha during unfocused open eyes condition; than frontal and central alpha power during reading; than frontal, central and parietal alpha during the search task; and than frontal alpha power in focused on target (counting 20Hz stimuli). Also, parietal alpha power during focused on target (when counting 20Hz stimuli) was larger than frontal alpha power during unfocused open eyes condition; than frontal and central alpha power during reading; than frontal, central and parietal alpha power during the search task.

Significant effect of task ($F(5,180)=11.962$, $p<0.001$), significant effect of region ($F(2,180)= 16.127$, $p = 0.001$) and significant interaction of the factors ($F(10,180)=4.443$, $p<0.001$) was observed for gamma power. Post hoc testing pointed to the highest gamma power over parietal region during reading (p values 0.001-0.04) and performing the search task (p values 0.001-0.04).

Means of alpha and gamma power over frontal, central and parietal regions are given in Figure 6.1 C.

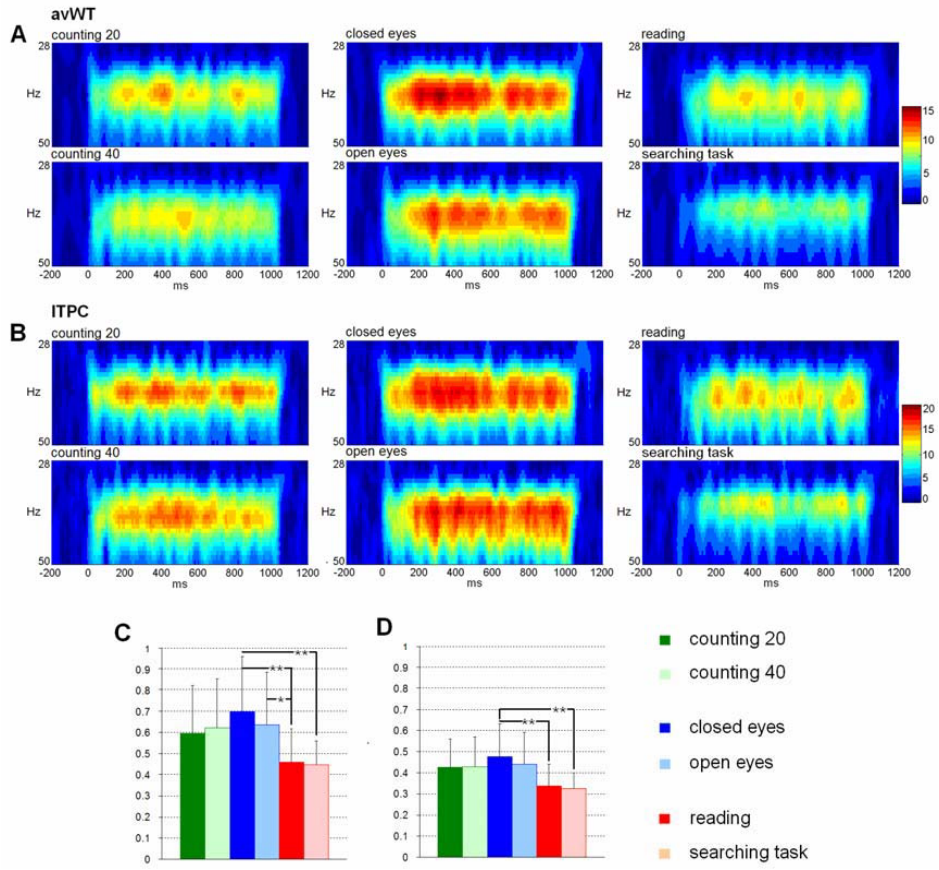

Figure 6.2. avWT and ITPC results of 40Hz stimulation in "Arousal/activation and attention" part. Grand-averaged time-frequency plots, means and SDs of avWT (A, C) and ITPC (B, D) of 40Hz ASSR (for graphical representation 28-50 Hz, -200 – 1200ms window) from Fz electrode for six experimental conditions: focused on target (counting 40), focused on non-target (counting 20), eyes closed, eyes open, reading and performing a searching task. Both avWT and ITPC were significantly attenuated during performing a searching task as contrasted to eyes closed and eyes open conditions and during reading as contrasted to eyes closed condition (* $p < 0.05$, ** $p<0.01$).

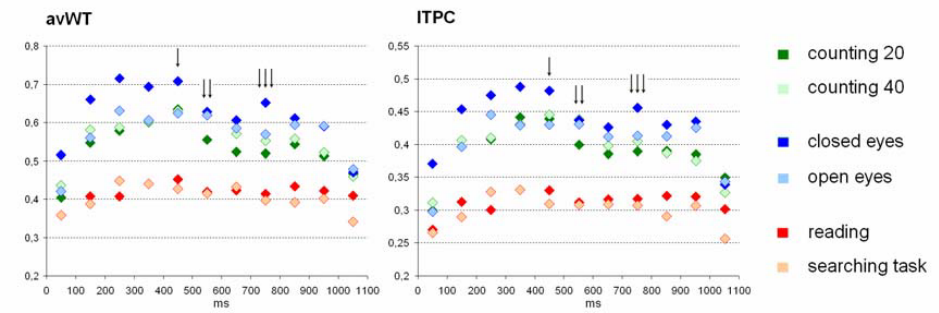

Figure 6.3. Means of avWT and ITPC for 100ms intervals for time period 0-1100ms. Significant differences at $p<0.05$ are marked with arrows: ↓ - counting 40 vs. searching task, counting 20 vs. searching task, eyes closed vs. reading and searching task; ↓↓ - counting 40 vs. reading and searching task, eyes closed vs. reading and searching task, eyes open vs. reading and searching task; ↓↓↓ - eyes closed vs. reading and searching task, eyes open vs. searching task.

EXPERIMENT III: P1-N1-P2 – EFFECTS OF AROUSAL, ACTIVATION AND ATTENTION

7. METHODS

7.1. Subjects

Twenty one healthy subjects (11 females) participated in the study. Eleven subjects were included in “Arousal” part (mean age 23.6 years, SD 2.7; five females) and ten subjects (six females) were included in ”Arousal/activation and attention” part (mean age 22.6 years, SD 1.5). Written informed consent was obtained, as approved by the Ethics Committee of the Republican Vilnius Psychiatric Hospital.

7.2. Stimulation

Sixty stimulus pairs (generated with Adobe Audition 3.0) were presented through the headphones with an inter-stimulus interval (ISI) of 500 ms. All stimuli were clicks of 3 ms duration, delivered binaurally at peak SPL of 100 dB. The pairs were repeated every 10 s. One stimulation trial lasted about 8 minutes.

7.3. Procedures

7.3.1. Arousal

This section, except for the part described below is identical to the ‘Procedures, Arousal’ section of Experiment II (p. 45).

Before the second run of low arousal eyes closed condition there was a brief period of uplifting music listening (Vivaldi Spring).

7.3.2. Arousal/activation and attention

This section, except for the part described below is identical to the 'Procedures, Arousal/activation and attention' section of Experiment II (p. 46), except that the unfocused closed eyes condition was not used.

7.4. EEG recordings

This section is identical to the 'EEG recordings' section of Experiment II (p. 48).

7.5. Data analysis

7.5.1. ERPs

Evoked potentials from Fz electrode for all conditions were created as follows: continuous EEGs were cut into epochs from -100 to +400 ms separately for first stimulus of the pair (S1) and second stimulus of the pair (S2) and filtered at 10-50Hz. S2 data is not presented in this thesis. 10% of epochs with the largest variability were rejected. P1, N1 and P2 components were identified and measured manually, blind to the condition: P1 was the positive deflection at 40-80 ms; N1 was the negative deflection at 60-170 and P2 was the positive deflection at 100-260ms. The amplitude was measured as peak-to-peak difference. Grand averaged evoked potentials for both conditions were created from 500 epochs.

7.5.2. Baseline EEG

This section is identical to the 'Baseline EEG' of Experiment II (p. 49), except that only alpha power was estimated in "Arousal" part and no baseline EEG measures were conducted in "Arousal/activation and attention" part.

7.5.3. Statistical analysis

Non-parametric statistical tests were used (SPSS© v. 9.1). The effect of "open/closed eyes" (two runs of each condition were merged together) and the baseline alpha power measures (runs merged) of "Arousal" part were tested by

Wilcoxon test. The effect of task in ”Arousal/activation and attention” part was tested with Friedman test, and where appropriate pair-wise comparisons of task effect were tested with Wilcoxon test.

8. RESULTS

8.1. Arousal

8.1.1. ERPs

Grand averaged waveforms of P1 potential from Fz site are given in Figure 8.1 A. Wilcoxon Signed Ranks test revealed that P1 amplitudes were significantly enhanced in closed eyes condition as compared to open eyes condition (Z=-2.44, p=0.04). There was no condition effect on the amplitudes of N1 and P2. Percentiles of P1, N1 and P2 amplitudes from ”Arousal” part are given in Table 8.1.

Table 8.1. Percentiles of P1, N1 and P2 amplitudes in closed eyes and open eyes conditions. N=11.

Component	Condition	Percentiles		
		25th	50th (Median)	75th
P1 amplitude	Eyes closed	2,185	2,539	3,527
	Eyes open	1,461	1,860	3,081
N1 amplitude	Eyes closed	4,975	6,030	8,457
	Eyes open	5,659	6,250	8,422
P2 amplitude	Eyes closed	7,700	10,347	16,711
	Eyes open	8,844	10,359	17,912

8.1.2. Baseline EEG

Alpha power was larger in closed eyes condition over frontal (Z=-2.223, p=0.026), central (Z=-2.490, p=0.013) and parietal (Z=-2.756, p=0.006) regions. Percentiles of alpha power estimations are given in Table 8.2.

Table 8.2. Percentiles of alpha power values in closed eyes and open eyes conditions. N=11.

Condition	Region	Percentiles		
		25th	50th (Median)	75th
Eyes open	Frontal	2,045	3,601	5,450
	Central	2,686	5,767	7,454
	Parietal	3,783	7,754	14,258
Eyes closed	Frontal	5,247	6,071	13,082
	Central	5,553	9,972	19,460
	Parietal	12,231	23,605	38,136

8.2. Arousal/activation and attention

8.2.1. ERPs

Grand averaged ERPs from Fz site are given in Figure 8.1 B. Percentiles of P1, N1 and P2 amplitudes are given in Table 8.3.

Friedman test showed that the effect of task was significant only for N1 potential amplitude (df=3, p=0.013) that was significantly higher during open eyes condition as compared to counting (Z=-2.29, p=0.02) and performing the task (Z=-2.60, p=0.009) and nearly significantly increased in comparison to reading (Z=-1.89, p=0.059). Task had no effect on P1 and P2 amplitudes.

Table 8.3. Percentiles of P1, N1 and P2 amplitudes in counting, unfocused open eyes, reading and the search task conditions. N=10.

Component	Condition	Percentiles		
		25th	50th (Median)	75th
P1 amplitude	Counting	1,691	2,438	4,730
	Unfocused open eyes	0,826	3,194	4,866
	Reading	1,322	1,835	3,209
	The search task	0,809	2,484	4,181

N1 amplitude	Counting	5,188	7,316	10,238
	Unfocused open eyes	6,945	9,161	15,215
	Reading	6,193	8,293	12,446
	The search task	2,498	6,320	7,916
P2 amplitude	Counting	9,961	11,187	19,415
	Unfocused open eyes	11,696	16,943	22,363
	Reading	9,452	11,244	20,561
	The search task	6,268	9,781	18,067

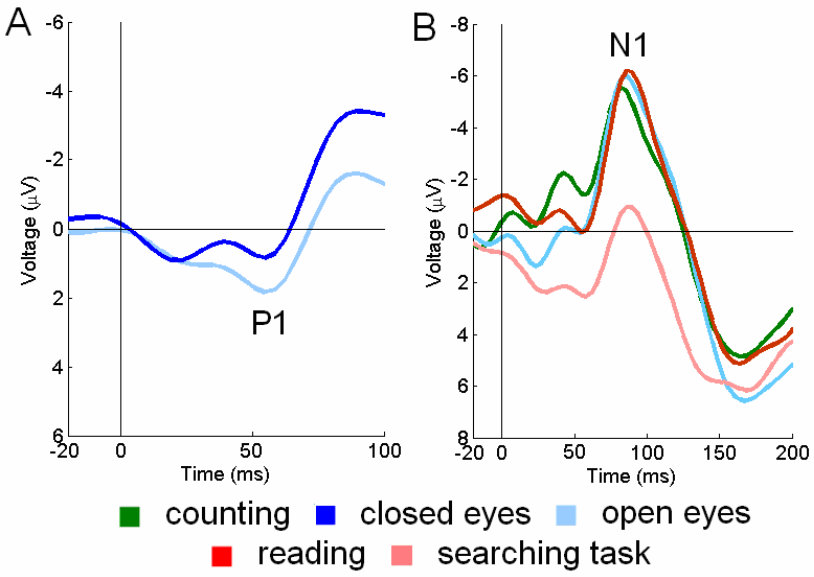

Figure 8.1. Grand averaged ERPs from Experiment III. (A) Grand averaged evoked potentials from Fz electrode of eyes open high arousal level condition and eyes closed low arousal level condition from ”Arousal” part (N= 11). Two runs of each condition were merged together so that each potential is an average of 1188 epochs. (B) Grand averaged evoked potentials from Fz electrode of counting, unfocused open eyes, reading and the search task conditions from “Arousal/activation and attention” part. Each potential is an average of 500 epochs, N=10.

9. DISCUSSION

9.1. Eyes closed vs. eyes open: arousal?

9.1.1. 40Hz ASSRs

Neither the phase precision nor the evoked amplitude of 20Hz ASSR and 40Hz ASSR were affected by arousal level. These parameters did not differ between eyes open and eyes closed conditions. However, total intensity of 40Hz ASSR was higher in closed eyes condition. This effect was most pronounced at 100-200ms.

With eyes open, the subject is more aroused than with the eyes closed (Barry et al., 2009a; Barry et al., 2007), that is arousal level drops from open eyes to closed eyes. Most of the previous ASSR studies investigating the effect of arousal level evaluated ASSR in sleep, the main finding being diminished ASSR amplitude with stimulation frequencies up to 70Hz (Cohen et al., 1991; Jerger et al., 1992; Linden et al., 1985) and there are no reports where the modulating effect of eyes closed vs. eyes open on ASSRs is examined. However, Pockett and Tan (2002) showed an increase of power of the ASSR during lower arousal state-drowsiness (Pockett and Tan, 2002).

In the current study the order of eyes open and eyes closed runs was not counterbalanced and the total closed eyes period lasted about 30 minutes. Such a design was selected to surely achieve lower arousal level in closed eyes condition-especially second run of closed eyes. Based on the baseline EEG alpha power changes, the change in arousal level following the current experimental manipulation was successful: as suggested by Barry et al. (2005, 2007), increase in arousal is marked by a global decrease in alpha power, whereas the specific regional activity is associated with task processing (Barry et al., 2007; Barry et al., 2005). Alpha power was significantly larger in low arousal closed eyes condition. Even more, it was significantly larger during second runs, meaning that alpha power was larger in the second run of closed eyes and open eyes as compared to the first runs (section 6.1.3.). The subjects

reported that they entered a relaxed resting state and felt sleepy during the closed eyes condition but did not fall asleep. However, it appears that the extent to which arousal level is diminished did not affect any measure applied. Only total intensity is enhanced by closing eyes (Fig. 6.1) – this effect was repeated when Paired T- Test was performed on unfocused closed eyes and unfocused open eyes data from Experiment II "Arousal/activation and attention" part.

The WTav -total intensity measure - reflects both phase-locked and non-phase locked activity. In a very recent work Tcheslavski and Beex (2010) assessed differences in the background EEG gamma rhythm phase synchrony between the two experimental conditions of eyes closed and eyes open. For the closed eyes condition gamma phase synchrony was higher than for the open eyes condition (Tcheslavski and Beex, 2010). The larger gamma power during closed eyes can be also seen in the study by Barry et al. (2007). In the current study where effects of closed/open eyes on ASSRs were tested, gamma power was larger in closed eyes condition in line with above mentioned observations (although it was largest over parietal region and no significant differences were obtained for frontal and central areas). Thus the total larger and more synchronized gamma power might contribute to the observed result of enhanced total intensity of 40Hz ASSRs in closed eyes condition.

The diminished arousal level did not affect the avWTs and ITPCs of ASSRs and correspondingly these measures did not differ between closed and open eyes states in Experiment II, "Arousal/activation and attention" part. However, the WTav was larger in closed eyes condition. The different modulation of the measures might be explained by their different nature: Evoked amplitude and phase precision co-variate and are mostly measures of the phase resetting of oscillations; the total intensity, like the power measure, includes amplitude modulation which are not phase aligned, i.e. induced amplitude changes. The present results indicate that arousal modulation with open/closed eyes conditions affects the amplitude of the oscillations, and not the phase alignment.

As widely accepted, the main contributor to the amplitude of a cortical recorded evoked potential is the amount of synchronization of firing pyramidal cells. The number of cells that fire depends on the firing mode of cortical pyramidal neurons. These cells fire either in the tonic or bursting mode, depending on the level of general arousal: tonic firing is typical for alert wakefulness; burst firing is characteristic of drowsiness (Coenen, 1995). It is easier to recruit a larger number of cells when cells are in the bursting mode than in the tonic firing mode.

On the other hand, Castro-Alamancos and Oldford (2002) showed in rats that sensory-evoked responses were suppressed in the neocortex by activating the brainstem reticular formation and during natural arousal (Castro-Alamancos and Oldford, 2002). Sensory suppression occurred at the thalamocortical connection and was a consequence of the activity-dependent depression of thalamocortical synapses caused by increased thalamocortical tonic firing during arousal. Although gamma oscillations can be generated by cortex cell without input from thalamus (Cunningham et al., 2004), the thalamic input to 40Hz rhythm generation is well known (Sukov and Barth, 2001).

Recently, increased ASSR was found in Alzheimer patients and this was interpreted as the lack of inhibition in cortical auditory processing (Osipova et al., 2006). Several studies have shown that power and phase precision of 40Hz ASSR is diminished in schizophrenic patients as compared to healthy controls (Brenner et al., 2009; Kwon et al., 1999; Light et al., 2006). This could also point to the changes in inhibition perhaps mediated by dysregulated NMDA-modulated GABAergic activity as this is suggested to be basic to the deficient binding capacity of pyramidal cell networks involved in gamma rhythm generation (Kwon et al., 1999; Light et al., 2006). We speculate that the inhibitory balance between the thalamic and cortical processes is changed during low arousal closed eyes state and differs from that in sleep, modulating sensory processing and gamma rhythm generation.

9.1.2. ERPs

The P1 amplitude increased with diminishing arousal level. No effects on N1 and P2 amplitudes were found, suggesting that general arousal affects preattentive auditory stimulus processing and not attentional allocation processes.

Our finding of lower amplitudes in higher arousal condition and higher amplitudes in lower arousal condition is in line with several previous observations. First, P1 amplitude was modulated by the level of arousal in a similar manner as ASSRs. This finding is in line with results of Pockett and Tan (2002). They have showed not only increased ASSRs amplitudes but increased auditory transient response amplitudes during drowsy state (Pockett and Tan, 2002). However, our finding is contradictory to earlier observation of Cardenas et al. (1997), who addressed the effect of varying alertness on P1 amplitude and did not find any differences between low arousal level and high arousal level periods while subjects were sitting with open eyes. However, they did not use closed eyes condition (Cardenas et al., 1997).

The larger amplitude of P1 may be result of more bursting cells during low arousal condition as discussed above for ASSRs (Coenen, 1995; Grootens et al., 2008). The assumption of P1 amplitude sensitivity to the level of arousal is supported by the observed effects of caffeine, theophylline and nicotine on P1 amplitudes (Ghisolfi et al., 2002; Ghisolfi et al., 2006; Knott et al., ; Knott et al., 2009). These compounds are known to increase arousal level (Barry et al., 2009b; Barry et al., 2005; Kaplan et al., 1993; Rusted and Trawley, 2006; Sawyer et al., 1982). Thus elevated arousal level upon consumption is associated with lower P1 amplitudes. On the contrary, neuropleptic medication, which decreases dopaminergic neuronal transmission and decreases arousal, increases the amplitude of the P1 auditory-evoked response (Moxon et al., 2003). As well, cocaine abused subjects show decreased P1 amplitudes (Boutros et al., 2000; Boutros et al., 2002; Fein et al., 1996). Jensen et al (2007) when studying effect of escitalopram (a highly selective serotonin reuptake inhibitor) have found increased P1 amplitudes. They attributed the

effect to a slight change in state, such as mild drowsiness that might have occurred with escitalopram (Jensen et al., 2008).

Increased P1 amplitudes in mild cognitive impairment identified individuals who will subsequently convert to dementia (Golob et al., 2007; Irimajiri et al., 2005). Similar relationships have been identified in HIV-1 infection, correlating with indices of disease progression (Schroeder et al., 1994). Both dementia and HIV infection cause marked reduction in arousal level (Edman et al., 2003; Polich et al., 2000). Grootens et al. (2008) found significantly higher P1 amplitudes in the borderline personality disorder group compared to the comparison group (Grootens et al., 2008). Grootens et al. (2008) related their finding to changed arousal level of the subjects as compared to controls (Grootens et al., 2008).

Our results suggest that the amplitude, measured as total intensity of 40Hz ASSRs and P1 potential amplitude are modulated by the general arousal level– larger total intensity of 40Hz ASSRs and P1 amplitude in low arousal states. Phase alignment is not influenced by the arousal level. This observation promotes supplementary assessment of arousal in ASSRs and ERPs studies, where amplitudes of the response are primer measures.

9.2. Counting vs. Unfocused state vs. Distraction: Attention or arousal/activation?

9.2.1. 40Hz ASSR

Focused attention vs. distraction task

It was predicted that highest levels of 40Hz ASSR avWT and ITPC would be observed during counting, i.e. focused on targets and focused on non-targets conditions, when attention was focused on the stimuli. The ITPC and avWT of 40Hz ASSR during focused attention (i.e. paying direct attention to stimuli) were significantly higher as compared to reading and performing the

search task where attention to stimulation was reduced. This finding is congruent with several previous studies that compared enhanced attention conditions and reduced attention conditions and showed 40Hz ASSR amplitude increment when paying attention to stimulation (Ross et al., 2004; Saupe et al., 2009; Skosnik et al., 2007). Both Ross et al. (2004) and Saupe et al. (2009) observed modulation of the amplitude of ASSRs by intermodal attention, i.e. attention to the sounds in their studies led to a significant enhancement of the ASSR (Saupe et al., 2009). In the study of Gander et al. (2007) enhancement of ASSR amplitude was observed when subjects attended to the task stimuli, but attention had no detectable effects on ASSR phase (Gander et al., 2007).

In the study by Skosnik et al. (2007) 20Hz and 40Hz trains were used in an oddball discrimination task: click trains were presented binaurally and participants had to count targets (20% of the stimuli) and the enhancement of the response was observed selectively in response to 40 Hz clicks defined as targets (Skosnik et al., 2007). On the other hand, in the very first paper Linden et al. (1987) compared the effect of attending to the stimulus (counting intensity increments and frequency changes) and not attending (reading) and found no significant differences in amplitudes and phases of the response between conditions. Recently this was supported by de Jong et al (2009), who did not find any significant effect of intermodal attention on ASSR amplitude (de Jong et al., 2009).

Distraction task vs. unfocused attention

Studies comparing the effect of unfocused attention during resting and distraction tasks on ASSRs are sparse. There are only two research reports that employed an unfocused condition comparable to our study. Both Linden et al. (1987) and Alegre et al. (2008) did not find any difference in the energy and phase-locking of the response around 40 Hz between the unfocused closed eyes condition and reading condition (Alegre et al., 2008; Linden et al., 1987). In the present study 40Hz ASSR was larger and more precise in

unfocused closed eyes condition as compared to reading and the search task – this was obtained in two separate experiments (Experiment I and Experiment II). Moreover, in the current study we have found higher evoked amplitude and phase precision during unfocused open eyes condition as compared to reading and performing the search task.

This is congruent with observation made by Kallai et al.(2003) for auditory evoked 40Hz response (Kallai et al., 2003). The evoked 40-Hz response in their study was more prominent while subjects were lying awake in bed than while they were sitting and reading. Kallai et al. (2003) have interpreted this observation as a confirmation of earlier proposals connecting the evoked 40-Hz response to attentional mechanisms, especially selective attention (Kallai et al., 2003).

The effect of arousal/activation cannot be separated when some task is performed. This is best explained on the example of Experiment I. Closed eyes condition and reading condition were shown to be different both by arousal and activation levels: during closed eyes condition both arousal and activation were diminished as compared to reading. All the participants of Experiment I stated that they entered a relaxed resting state and felt sleepy during the low arousal/activation closed condition. The lower arousal level in the low arousal condition was confirmed by the lower percentage of beta power (Cardenas et al., 1997; Regan, 1989). Along with a globally higher alpha power in low arousal/activation closed eyes condition, we observed a slight trend for theta and beta powers to be larger over frontal region in high activation level condition. The involvement of both theta and beta in reading has previously been shown: frontal theta and beta are increased during semantic task completion (Fitzgibbon et al. 2004; Spironelli et al. 2008). Taking into account the suggestion of Barry et al. (2005, 2007) that increase in arousal is marked by a global decrease in alpha, whereas the specific regional activity is associated with task processing (Barry et al., 2007; Barry et al., 2005) we speculated that both modulation of activation level together with

change in arousal level following the current experimental manipulation was successful.

Focused attention vs. unfocused state

It was hypothesized that during unfocused closed eyes and unfocused open eyes conditions when subject had no task to perform, intermediate levels of evoked amplitude, phase precision and total intensity would be obtained as compared to counting (focused on target and focused on non-target), reading and performing the search task conditions. On the contrary, the phase precision and evoked power obtained during unfocused closed eyes and unfocused open eyes conditions in the present study were not different from those obtained in focused on target and focused on non-target conditions. Recently, Lazzouni et al. (2009) showed that attention to carrier frequency change in stimulation enhances the amplitude of the right hemisphere ASSRs for dichotic stimulation as compared to open eyes no-task condition. However, they evaluated ASSRs only from temporal sensors (Lazzouni et al., 2009) and we analysed the frontal response. We have not found more studies with a comparable experimental set-up and this is the first time to show that ASSRs might be insensitive to the effect of direct attention.

ERPs

We did not find any effects of attention on P1 and P2 amplitudes. We showed that N1 potential amplitude was modulated by distraction tasks with largest amplitudes obtained in unfocused open eyes condition. Thus, arousal/activation and attention levels modulate processes that trigger the allocation attention to the stimulus.

The lack of modulation by task on P1 and P2 amplitudes is in line with several previous studies (Yee and White, 2001; Jerger et al., 1992; Rosburg et al., 2009; White and Yee, 1997). The N1 modulation by task in the current study is congruent with previous findings. Based on previous reports, we expected N1 to be largest in stimuli counting task condition, when direct

attention was paid to the stimulation (Parasuraman, 1980; Woldorff and Hillyard, 1991). However, unexpectedly the largest N1 amplitude was obtained during no-task unfocused open eyes condition. Moreover it was significantly larger as compared to stimuli counting and performing a search task conditions. Similarly as in our study, Lavoie et al. (2008) compared several experimental conditions and did not observe the largest N1 during the counting condition. Lavoie et al (2008) explained their finding as the counting task being the most boring part of the experiment and it is possible that subjects' interest and alertness drifted. Indeed, in our experiment subjects counted tone pairs that were delivered in a regular order and this did not require high memory, or attentional load, or high activation level. Although, Lavoie et al (2008) did not find any effect of distraction task (reading or watching a movie) on amplitudes of any ERP component as compared to counting and sitting without any task with eyes open, the prestimulus noise level was significantly affected by the task in their experiment, being lowest in reading and watching a movie conditions (Lavoie et al., 2008).

Previously, effects of prestimulus ongoing EEG on N1 wave were shown. Haig and Gordon (1998) showed that alpha synchronicity at the moment of stimulus has a significant influence on the resulting N1 amplitude: the more synchronized is alpha activity the more pronounced is the N1 wave (Haig and Gordon, 1998a; Haig and Gordon, 1998b). On the other hand, Krause et al. (1996, 1997, 2000) has showed that involvement in cognitive or memory tasks causes event related desynchronization of alpha, whereas simple listening to stimuli results in event-related alpha synchronization (Krause et al., 1996; Krause et al., 1997; Krause et al., 2000). Taken the two facts into account, it appears logical that N1 amplitude is largest at the state of simple listening to the stimuli as compared to the task conditions.

9.2.2. Generalization

It is widely accepted that the gamma response serves a broad range of physiological functions: early phase-locked gamma being primarily a

manifestation of sensory processing and late non-phase-locked gamma resembling cognitive processes (Karakas et al., 2001). It has been shown, that early sensory gamma response is modulated via top-down processes (Karakas et al., 2006; Karakas and Basar, 1998). We speculate that competing modulating factors might be at work to produce the current result profile of the gamma ASSR modulation by the task.

Firstly, ASSRs were more expressed when direct attention was paid to stimulation as compared to performing distraction task. Focused attention on stimuli (focused on target and focused on non-target) showed largest significant effect at 400-500ms that is in line with observation made by Ross et al (2004) (Ross et al., 2004). Effect was similarly expressed when subjects counted both 40 Hz and 20 Hz stimuli. At 500-600ms 40Hz ASSR was enhanced only in focused on target condition, when 40Hz stimuli were targets, probably due to the physiological significance of 40Hz target stimuli (Fig. 6.3). This is supported by the study of Skosnik et al. (2007) who showed the exceptional processing of 40Hz stimuli (Skosnik et al., 2007).

Next, phase precision and evoked amplitude were lower during reading and the search task as compared to unfocused closed eyes and unfocused open eyes conditions. During reading and the search task ASSRs were significantly lower as compared to unfocused closed eyes state at the time intervals of 400-500ms, 500-600ms and 700-800ms intervals (however this trend remained during whole stimulation period) and during unfocused open eyes state at time intervals of 500-600ms (as compared to reading and the search task) and 700-800ms (as compared to the search task) (Fig. 6.3). Lower ITPCs and avWTs of the ASSRs during these distraction tasks could reflect a sensory cortical inhibition to promote stimulus-unrelated task performance. This interpretation is based on the assumption that the strong attention focus required by the difficult visual task does not allow subjects to process the irrelevant auditory input; whereas during no task, attentional resources are available for this purpose (Muller-Gass et al., 2006).

Subsequently, the high level of phase synchronized gamma activity evoked in the unfocused closed eyes and unfocused open eyes conditions without any task could be a manifestation of increased focal cortical activity due to induced shifts of involuntary attention. This could be substantiated by the fact that the measures did not differ between focused attention (focused on target, focused on non-target) and unfocused attention (unfocused closed eyes, unfocused open eyes) conditions (Table 6.1 and Figure 6.2). However, it has been shown in visual modality that involuntary attention does not augment gamma band activity as contrasted to voluntary attention (Landau et al., 2007). Also, most notably, during the condition in which subjects were not required to perform any specific task, their attention might have been focused on internal thoughts (instead of re-directed to the to-be ignored auditory stimuli) (Muller-Gass et al., 2005).

Furthermore, we assume that the easy counting tasks (focused on target and focused on non-target) were closer in arousal/activation level to the state of doing nothing – unfocused closed eyes and unfocused open eyes (lower arousal/activation level), than the effortful visual task – reading and the search task (higher arousal/activation level). It is substantiated by the fact that total intensity (and slightly phase precision and evoked power) of 40Hz ASSRs also was increased with the lower arousal level. Moreover, the pattern of background alpha power supports this notion.

The effect of arousal/activation level is supported by the results of Pockett and Tan (2002), who reported increased ASSRs in drowsy condition (in fact, sitting with closed eyes after a consumption of some alcohol) as contrasted to reading or writing (Pockett and Tan, 2002). Recently, Fischer at al. (2008) showed in their study investigating the effect of self-selected stimulation on compensation of attentional demands to the task, that cortical excitability was higher in the condition with self-chosen stimulation (i.e. arousal level optimal for subjects' performance) than in conditions with high or low stimulation (that required compensatory processes to achieve optimal arousal level) (Fischer et al., 2008).

Under currently used experimental conditions lower arousal/activation level was achieved during the counting and unfocused state and the most phase aligned 40Hz ASSR was observed in these conditions as contrasted to the high arousal/activation distraction task performance. In line with 40Hz ASSR, N1 amplitude was modulated by arousal/activation level – higher amplitudes obtained in lower arousal/level conditions with unfocused attention to stimulation. Thus, the difference between focused attention condition, unfocused state and the distraction task is the result of the counterbalancing effects of the attention and the achieved optimal (lower) arousal/activation level. The current results suggest an important improvement of the practical use of ASSRs - in cases where ASSRs are applied to investigate the ability to generate synchronized high frequency cortical activity a "distraction" task is not favourable.

9.3. 20Hz ASSRs

The 20Hz ASSR deserved to be discussed separately. Periodical 20Hz stimulation leads to the electroencephalographic (EEG) entrainment (Figure 4A) with a clear peak identifiable at 20Hz in the power spectrum (Kwon et al., 1999; Pastor et al., 2002; Skosnik et al., 2006). However, conventional spectral analysis of 20Hz SSR distinguished an additional peak at 40Hz (Kwon et al., 1999; Pastor et al., 2002; Skosnik et al., 2006). The present time-frequency analysis of wavelet transformed 20Hz ASSR response also resulted in the identification of two components: a component within 20Hz frequency range and a component peaking at 40Hz (Figure 4.4. and Figure 4.5). This time-frequency pattern of 20Hz ASSR was recently observed by Light et al. (2006) and Spencer et al. (2008) and it was observed in the present work. Additional activity around 40Hz in response to 20Hz stimulation has previously been considered a harmonic of the signal (Kwon et al., 1999; Light et al., 2006; Pastor et al., 2002; Spencer et al., 2008). By definition, the harmonic is a single

oscillation whose frequency is an integral multiple of the fundamental frequency (i.e. 40Hz is a higher order harmonic of 20 Hz oscillation). However, several findings support the notion that 40Hz activity elicited by 20Hz stimulation is of physiological significance.

First of all, we have shown that phase locking and evoked amplitude of 20Hz ASSR-related 40Hz activity were higher in the low activation state, whereas 20Hz ASSR did not differ between conditions. The latter is in line with Linden et al. (1985), who did not find any difference between aroused and sleep states in the amplitude and phase of 20Hz ASSR assessed by FFT but they did not evaluate 40Hz activity (Linden et al., 1985). The divergent modulation of 20Hz and 40Hz activities is supported by the finding of Spencer et al. (2008), who demonstrated decreased phase locking and decreased evoked power of 40Hz harmonics in the first episode psychosis, whereas 20Hz ASSR was not affected in their study (Spencer et al., 2008). In a report of ASSR in schizophrenics and healthy subjects Light et al. (2006) did not find any difference in phase locking and evoked power of 20Hz ASSR (Light et al., 2006). Yet, they did not evaluate the 40Hz harmonic, which is apparent in Figure 3 of the report (Light et al., 2006). The figure shows that the 40Hz harmonic is weaker in the schizophrenic patient group (Light et al., 2006). Secondly, the direction of modulation of 20Hz ASSR-related 40Hz activity corresponds to our recent results on 40Hz stimulation: we have shown increase of evoked amplitude and phase precision of the 40Hz ASSR during a low activation state compared to a high activation, reading condition (Griskova et al., 2007). Again, it corresponds to studies both by Light et al. (2006) and Spencer et al. (2008) where the impairment of 20Hz ASSR-related 40Hz activity in patient groups was of the same direction as the change in actual 40Hz ASSR (Light et al., 2006; Spencer et al., 2008). This observation was repeated when paired T-Test was performed on the data from Experiment II, "Arousal/activation and attention" part.

Interestingly, the phase-locked 40Hz activity was more prominent than 20Hz activity in the present study. The same relationship has been observed

before as it is evident in the time-frequency maps both in Figure 1 of Spencer et al. (2008) and Figure 3 of Light et al. (2006): 20Hz ASSR-related 40Hz activity is more pronounced than actual 20Hz ASSR.

Notably, significant modulation by the level of activation was apparent only for phase-locked activity (amplitude and phase), whereas no significant effects were obtained for total (both non-phase-locked and phase-locked) intensity, although there was a trend for larger 20Hz ASSR amplitude in high activation condition. In contrast to phase-locked measures, total intensity of 20Hz ASSR was larger compared to 20Hz ASSR-related 40Hz activity. This may be explained by slightly larger frontal beta power in the high activation level condition.

There is a limited number of studies analyzing task effects on responses to 20Hz stimulation (Muller et al., 2009; Skosnik et al., 2007). Skosnik et al. (2009) reported that 20Hz ASSR and 20Hz ASSR-related 40Hz activity did not change either when 20Hz stimuli were targets or non-targets in their study. On the other hand, Muller et al. (2009) demonstrated effect of auditory spatial selective attention selectively on the power of 20Hz ASSR. The 20 Hz responses were significantly enhanced in the left hemisphere when subjects had to attend to the right ear and the 20 Hz responses were significantly suppressed in the left hemisphere when subjects had to attend to the ipsilateral left ear (Muller et al., 2009). We have shown that 20Hz ASSR and 20Hz ASSR-related 40Hz activity are differently modulated by reading and closed eyes conditions: 20Hz ASSR-related 40Hz activity was significantly larger in closed eyes condition and there were no effect on 20Hz ASSR. Although we did not show significant effect of task on 20Hz ASSR-related 40Hz activity when comparing six tasks (focused on target, focused on non target, unfocused closed eyes, unfocused open eyes, reading and the search task) in our study, paired T-test comparison of unfocused closed eyes condition and reading revealed significantly larger phase precision during unfocused closed condition. The divergent modulation of 20Hz ASSR and 40Hz ASSR shown in this and previous studies (Bidet-Caulet et al., 2007; Griskova et al., 2009;

Liegeois-Chauvel et al., 2004; Muller et al., 2009; Skosnik et al., 2007) supports the notion of different generators of ASSRs around 40 Hz compared to the 20 Hz response.

20Hz ASSR and 20Hz ASSR-related 40Hz activity are distinctly modulated by the arousal/activation level: arousal/activation level has no impact on 20Hz ASSR, however 20Hz ASSR-related 40Hz activity is higher in low arousal/activation condition similarly as 40Hz ASSR. The modulation of 20Hz ASSR-related 40Hz activity by the level of activation and disease might point to a physiological nature of this activity beyond a mere periodic effect in relation to the 20Hz activity and urges for further careful assessment of experimental effects on 20Hz ASSR-related 40Hz activity.

9.4. Methodological considerations

We have observed frontally distributed ASSR activity, maximal at Fz electrode. The highest gamma power values in the inter-stimulus period were obtained over parietal region, especially for closed eyes condition and reading and searching task conditions (Figure 6.1). Recently it has been shown that induced gamma is closely related to saccadic eyes movement. The timing of significant difference between eyes closed and eyes open condition in Experiment II "Arousal" part corresponds to the timing of saccades (100-200ms). However, topography of the response (strict frontal distribution with maximum at Fz, Figure 6.1) suggests its origin not due to the eyes dipoles. Moreover, as shown by Matsue et al (1986) the intensity of saccades is significantly less in closed eyes condition as compared to eyes-fixated condition. We suppose that the effect of muscular noise on frontal ASSRs is sparse.

Picton et al. (2003) suggested that the power of the ASSR occasionally could be increased during drowsiness due to activity in the postauricular muscles recorded from a mastoid reference (Picton et al., 2003b). The

increased amplitude of ASSRs in Experiment I was not an effect of muscular noise. Apart from detection of the focal ASSR component, the noise component was also extracted by the decomposition from multichannel (64 channels) EEG data. The avWT of ASSR was higher in the low arousal closed condition, whereas noise component was much more prominent in the high arousal condition (Figure 4.1). The topography of both focal ASSR component and noise component showed that postauriculal muscles had no impact on the frontally distributed ASSR component (Figure 4.1). In the ITPC analysis, the noise component did not emerge as, due to the nature of ITPC measure, the amplitude information is removed leaving only phase information and all epochs are given the same weight. Thus, a noisy epoch with high amplitude activity only influences the measure by a possible phase degree shift divided by the number of epochs (n=122). When noisy channels (the outer range) were removed from analysis of 20Hz ASSR response, no noise component was detected (Figures 4.3 and 4.4). This was also true for 9EEG channel data of Experiment II. Although, the higher inter-stimulus gamma power over parietal region probably was of muscular origin due to slightly different head position in closed eyes condition (more reclined) and during reading and task performance (more leaning) in comparison to open eyes and counting conditions, the total intensity of 40Hz ASSR, that was significantly larger during closed eyes condition in Experiment II, was frontally distributed (Figure 6.1). The total intensity is a measure that is strongly affected by noise and if so, the noise would appear in parietal channels with high background gamma level.

10. CONCLUSIONS

1) The lower arousal level increases the total neuronal resources (measured as total intensity) leaving the proportion of phase-aligned neuronal recourses (measured as evoked amplitude and phase precision) of the 40Hz ASSR unchanged.
2) The differences between tasks varying in arousal/activation and attention level are the result of the counterbalancing effects of the attention and arousal/activation induced processes:
    - the lower arousal/activation level increases the proportion of phase-aligned neuronal resources (measured as evoked amplitude and phase precision) of the 40Hz ASSRs and 20Hz ASSR-related 40Hz activity leaving the total amount of neuronal resources (measured as total intensity) of the oscillation unchanged;
    - the lower attention level paid to the stimuli reduces the proportion of phase-aligned neuronal resources (measured as evoked amplitude and phase precision) of the 40Hz ASSRs leaving the total amount of neuronal resources (measured as total intensity) of the oscillation unchanged.
3) Neither total amount of neuronal resources (measured as total intensity) nor the proportion of phase-aligned neuronal resources (measured as evoked amplitude and phase precision) of the 20Hz ASSR are sensitive to the levels of arousal, activation and attention.
4) The lower arousal level increases the neuronal resources (measured as the amplitude of P1 wave) for preattentive auditory stimulus processing.
5) The lower arousal/activation and attention levels enhance processes that trigger allocation of attention to the stimulus, as reflected by N1 wave amplitude.

11. REFERENCES

Alegre M, Barbosa C, Valencia M, Perez-Alcazar M, Iriarte J, Artieda J. 2008. Effect of reduced attention on auditory amplitude-modulation following responses: a study with chirp-evoked potentials. J Clin Neurophysiol 25(1):42-47.

Arnfred SM, Hansen LK, Parnas J, Morup M. 2008. Regularity increases middle latency evoked and late induced beta brain response following proprioceptive stimulation. Brain Res 1218:114-131.

Arnfred SM, Morup M, Thalbitzer J, Jansson L, Parnas J. 2010. Attenuation of beta and gamma oscillations in schizophrenia spectrum patients following hand posture perturbation. Psychiatry Res.

Barry RJ, Clarke AR, Johnstone SJ, Brown CR. 2009a. EEG differences in children between eyes-closed and eyes-open resting conditions. Clin Neurophysiol 120(10):1806-1811.

Barry RJ, Clarke AR, Johnstone SJ, Brown CR, Bruggemann JM, van Rijbroek I. 2009b. Caffeine effects on resting-state arousal in children. Int J Psychophysiol 73(3):355-361.

Barry RJ, Clarke AR, Johnstone SJ, Magee CA, Rushby JA. 2007. EEG differences between eyes-closed and eyes-open resting conditions. Clin Neurophysiol 118(12):2765-2773.

Barry RJ, Clarke AR, Johnstone SJ, McCarthy R, Selikowitz M. 2009c. Electroencephalogram theta/beta ratio and arousal in attention-deficit/hyperactivity disorder: evidence of independent processes. Biol Psychiatry 66(4):398-401.

Barry RJ, Rushby JA, Wallace MJ, Clarke AR, Johnstone SJ, Zlojutro I. 2005. Caffeine effects on resting-state arousal. Clin Neurophysiol 116(11):2693-2700.

Bidet-Caulet A, Fischer C, Besle J, Aguera PE, Giard MH, Bertrand O. 2007. Effects of selective attention on the electrophysiological representation of concurrent sounds in the human auditory cortex. J Neurosci 27(35):9252-9261.

Bourdon B 1985. Observations comparatives sur la reconnaisance, la discrimination et l'assoication. Revue Philosophique, 40, 153-185.

Bonhomme V, Plourde G, Meuret P, Fiset P, Backman SB. 2000. Auditory steady-state response and bispectral index for assessing level of consciousness during propofol sedation and hypnosis. Anesth Analg 91(6):1398-1403.

Boutros N, Campbell D, Petrakis I, Krystal J, Caporale M, Kosten T. 2000. Cocaine use and the mid-latency auditory evoked responses. Psychiatry Res 96(2):117-126.

Boutros NN, Gelernter J, Gooding DC, Cubells J, Young A, Krystal JH, Kosten T. 2002. Sensory gating and psychosis vulnerability in cocaine-dependent individuals: preliminary data. Biol Psychiatry 51(8):683-686.

Bregman AS, Levitan R, Liao C. 1990. Fusion of auditory components: effects of the frequency of amplitude modulation. Percept Psychophys 47(1):68-73.

Brenner CA, Kieffaber PD, Clementz BA, Johannesen JK, Shekhar A, O'Donnell BF, Hetrick WP. 2009. Event-related potential abnormalities in schizophrenia: a failure to "gate in" salient information? Schizophr Res 113(2-3):332-338.

Burton MJ, Cohen LT, Rickards FW, McNally KI, Clark GM. 1992. Steady-state evoked potentials to amplitude modulated tones in the monkey. Acta Otolaryngol 112(5):745-751.

Cardenas VA, Gill P, Fein G. 1997. Human P50 suppression is not affected by variations in wakeful alertness. Biol Psychiatry 41(8):891-901.

Castro-Alamancos MA, Oldford E. 2002. Cortical sensory suppression during arousal is due to the activity-dependent depression of thalamocortical synapses. J Physiol 541(Pt 1):319-331.

Coenen AM. 1995. Neuronal activities underlying the electroencephalogram and evoked potentials of sleeping and waking: implications for information processing. Neurosci Biobehav Rev 19(3):447-463.

Cohen LT, Rickards FW, Clark GM. 1991. A comparison of steady-state evoked potentials to modulated tones in awake and sleeping humans. J Acoust Soc Am 90(5):2467-2479.

Conti G, Santarelli R, Grassi C, Ottaviani F, Azzena GB. 1999. Auditory steady-state responses to click trains from the rat temporal cortex. Clin Neurophysiol 110(1):62-70.

Coull JT. 1998. Neural correlates of attention and arousal: insights from electrophysiology, functional neuroimaging and psychopharmacology. Prog Neurobiol 55(4):343-361.

Cunningham MO, Whittington MA, Bibbig A, Roopun A, LeBeau FE, Vogt A, Monyer H, Buhl EH, Traub RD. 2004. A role for fast rhythmic bursting neurons in cortical gamma oscillations in vitro. Proc Natl Acad Sci U S A 101(18):7152-7157.

Davis PA. 1939. Effects of the acoustic stimuli on the waking human brain. J Neurophysiol 2: 494-499

de Jong R, Toffanin P, Harbers M. 2009. Dynamic crossmodal links revealed by steady-state responses in auditory-visual divided attention. Int J Psychophysiol.

Delorme A, Makeig S. 2004. EEGLAB: an open source toolbox for analysis of single-trial EEG dynamics including independent component analysis. J Neurosci Methods 134(1):9-21.

Dimitrijevic A, John MS, Picton TW. 2004. Auditory steady-state responses and word recognition scores in normal-hearing and hearing-impaired adults. Ear Hear 25(1):68-84.

Dolphin WF, Mountain DC. 1992. The envelope following response: scalp potentials elicited in the Mongolian gerbil using sinusoidally AM acoustic signals. Hear Res 58(1):70-78.

Duffy E. 1957. The psychological significance of the concept of arousal or activation. Psychol Rev 64(5):265-275.

Edman A, Brunovsky M, Sjogren M, Wallin A, Matousek M. 2003. Objective measurement of the alertness level in dementia. Dement Geriatr Cogn Disord 15(4):212-217.

Fein G, Biggins C, MacKay S. 1996. Cocaine abusers have reduced auditory P50 amplitude and suppression compared to both normal controls and alcoholics. Biol Psychiatry 39(11):955-965.

Fischer T, Langner R, Birbaumer N, Brocke B. 2008. Arousal and attention: self-chosen stimulation optimizes cortical excitability and minimizes compensatory effort. J Cogn Neurosci 20(8):1443-1453.

Franowicz MN, Barth DS. 1995. Comparison of evoked potentials and high-frequency (gamma-band) oscillating potentials in rat auditory cortex. J Neurophysiol 74(1):96-112.

Gander PE, Bosnyak DJ, Wolek R, Roberts LE. 2007. Modulation of the 40-Hz auditory steady-state response by attention during acoustic training. International Congress Series 1300:37-40.

Garey J, Goodwillie A, Frohlich J, Morgan M, Gustafsson JA, Smithies O, Korach KS, Ogawa S, Pfaff DW. 2003. Genetic contributions to generalized arousal of brain and behavior. Proc Natl Acad Sci U S A 100(19):11019-11022.

Ghisolfi ES, Prokopiuk AS, Becker J, Ehlers JA, Belmonte-de-Abreu P, Souza DO, Lara DR. 2002. The adenosine antagonist theophylline impairs p50 auditory sensory gating in normal subjects. Neuropsychopharmacology 27(4):629-637.

Ghisolfi ES, Schuch A, Strimitzer IM, Jr., Luersen G, Martins FF, Ramos FL, Becker J, Lara DR. 2006. Caffeine modulates P50 auditory sensory gating in healthy subjects. Eur Neuropsychopharmacol 16(3):204-210.

Golob EJ, Irimajiri R, Starr A. 2007. Auditory cortical activity in amnestic mild cognitive impairment: relationship to subtype and conversion to dementia. Brain 130(Pt 3):740-752.

Grootens KP, van Luijtelaar G, Miller CA, Smits T, Hummelen JW, Buitelaar JK, Verkes RJ. 2008. Increased p50 gating but intact prepulse inhibition in borderline personality disorder. J Neuropsychiatry Clin Neurosci 20(3):348-356.

Gutschalk A, Mase R, Roth R, Ille N, Rupp A, Hahnel S, Picton TW, Scherg M. 1999. Deconvolution of 40 Hz steady-state fields reveals two overlapping source activities of the human auditory cortex. Clin Neurophysiol 110(5):856-868.

Hackett TA, Preuss TM, Kaas JH. 2001. Architectonic identification of the core region in auditory cortex of macaques, chimpanzees, and humans. J Comp Neurol 441(3):197-222.

Haig AR, Gordon E. 1998a. EEG alpha phase at stimulus onset significantly affects the amplitude of the P3 ERP component. Int J Neurosci 93(1-2):101-115.

Haig AR, Gordon E. 1998b. Prestimulus EEG alpha phase synchronicity influences N100 amplitude and reaction time. Psychophysiology 35(5):591-595.

Harada J, Aoyagi M, Suzuki T, Kiren T, Koike Y. 1994. A study on the phase spectral analysis of middle latency response and 40-Hz event-related potential in central nervous system disorders. Acta Otolaryngol Suppl 511:34-39.

Hari R, Pelizzone M, Makela JP, Hallstrom J, Leinonen L, Lounasmaa OV. 1987. Neuromagnetic responses of the human auditory cortex to on- and offsets of noise bursts. Audiology 26(1):31-43.

Harmony T, Hinojosa G, Marosi E, Becker J, Rodriguez M, Reyes A, Rocha C. 1990. Correlation between EEG spectral parameters and an educational evaluation. Int J Neurosci 54(1-2):147-155.

Hebb DO. 1955. Drives and the C.N.S. (conceptual nervous system). Psychol Rev 62(4):243-254.

Hong LE, Summerfelt A, McMahon RP, Thaker GK, Buchanan RW. 2004. Gamma/beta oscillation and sensory gating deficit in schizophrenia. Neuroreport 15(1):155-159.

Howard MA, Volkov IO, Mirsky R, Garell PC, Noh MD, Granner M, Damasio H, Steinschneider M, Reale RA, Hind JE, Brugge JF. 2000. Auditory cortex on the human posterior superior temporal gyrus. J Comp Neurol 416(1):79-92.

Irimajiri R, Golob EJ, Starr A. 2005. Auditory brain-stem, middle- and long-latency evoked potentials in mild cognitive impairment. Clin Neurophysiol 116(8):1918-1929.

Yee CM, White PM. 2001. Experimental modification of P50 suppression. Psychophysiology 38(3):531-539.
James W. 1890. Origin of Right-handedness. Science 16(406):275.
Jensen KS, Oranje B, Wienberg M, Glenthoj BY. 2008. The effects of increased serotonergic activity on human sensory gating and its neural generators. Psychopharmacology (Berl) 196(4):631-641.
Jerger K, Biggins C, Fein G. 1992. P50 suppression is not affected by attentional manipulations. Biol Psychiatry 31(4):365-377.
Kallai I, Harsh J, Voss U. 2003. Attention to external stimuli during wakefulness and sleep: evoked 40-Hz response and N350. Psychophysiology 40(6):955-966.
Kaplan J, Fredrickson PA, Renaux SA, O'Brien PC. 1993. Theophylline effect on sleep in normal subjects. Chest 103(1):193-195.
Karakas S, Arikan O, Cakmak ED, Bekci B, Dogutepe E, Tufekci I. 2006. Early gamma response of sleep is sensory/perceptual in origin. Int J Psychophysiol 62(1):152-167.
Karakas S, Basar-Eroglu C, Ozesmi C, Kafadar H, Erzengin OU. 2001. Gamma response of the brain: a multifunctional oscillation that represents bottom-up with top-down processing. Int J Psychophysiol 39(2-3):137-150.
Karakas S, Basar E. 1998. Early gamma response is sensory in origin: a conclusion based on cross-comparison of results from multiple experimental paradigms. Int J Psychophysiol 31(1):13-31.
Karmos G, Lakatos P, Pincze Z, Rajkai C, Ulbert I. 2002. Frequency of gamma activity is modulated by motivation in the auditory cortex of cat. Acta Biol Hung 53(4):473-483.
Knott V, Millar A, Fisher D, Albert P. Effects of nicotine on the amplitude and gating of the auditory P50 and its influence by dopamine D2 receptor gene polymorphism. Neuroscience 166(1):145-156.
Knott VJ, Bolton K, Heenan A, Shah D, Fisher DJ, Villeneuve C. 2009. Effects of acute nicotine on event-related potential and performance indices of auditory distraction in nonsmokers. Nicotine Tob Res 11(5):519-530.
Krause CM, Lang AH, Laine M, Kuusisto M, Porn B. 1996. Event-related EEG desynchronization and synchronization during an auditory memory task. Electroencephalogr Clin Neurophysiol 98(4):319-326.
Krause CM, Porn B, Lang AH, Laine M. 1997. Relative alpha desynchronization and synchronization during speech perception. Brain Res Cogn Brain Res 5(4):295-299.
Krause CM, Viemero V, Rosenqvist A, Sillanmaki L, Astrom T. 2000. Relative electroencephalographic desynchronization and synchronization in humans to emotional film content: an analysis of the 4-6, 6-8, 8-10 and 10-12 Hz frequency bands. Neurosci Lett 286(1):9-12.
Krishnan GP, Hetrick WP, Brenner CA, Shekhar A, Steffen AN, O'Donnell BF. 2009. Steady state and induced auditory gamma deficits in schizophrenia. Neuroimage 47(4):1711-1719.
Kuwada S, Anderson JS, Batra R, Fitzpatrick DC, Teissier N, D'Angelo WR. 2002. Sources of the scalp-recorded amplitude-modulation following response. J Am Acad Audiol 13(4):188-204.
Kuwada S, Batra R, Maher VL. 1986. Scalp potentials of normal and hearing-impaired subjects in response to sinusoidally amplitude-modulated tones. Hear Res 21(2):179-192.

Kwon JS, O'Donnell BF, Wallenstein GV, Greene RW, Hirayasu Y, Nestor PG, Hasselmo ME, Potts GF, Shenton ME, McCarley RW. 1999. Gamma frequency-range abnormalities to auditory stimulation in schizophrenia. Arch Gen Psychiatry 56(11):1001-1005.

Landau AN, Esterman M, Robertson LC, Bentin S, Prinzmetal W. 2007. Different effects of voluntary and involuntary attention on EEG activity in the gamma band. J Neurosci 27(44):11986-11990.

Lavoie BA, Hine JE, Thornton RD. 2008. The choice of distracting task can affect the quality of auditory evoked potentials recorded for clinical assessment. Int J Audiol 47(7):439-444.

Lazzouni L, Ross B, Voss P, Lepore F. Neuromagnetic auditory steady-state responses to amplitude modulated sounds following dichotic or monaural presentation. Clin Neurophysiol 121(2):200-207.

Liegeois-Chauvel C, Lorenzi C, Trebuchon A, Regis J, Chauvel P. 2004. Temporal envelope processing in the human left and right auditory cortices. Cereb Cortex 14(7):731-740.

Light GA, Hsu JL, Hsieh MH, Meyer-Gomes K, Sprock J, Swerdlow NR, Braff DL. 2006. Gamma band oscillations reveal neural network cortical coherence dysfunction in schizophrenia patients. Biol Psychiatry 60(11):1231-1240.

Linden RD, Campbell KB, Hamel G, Picton TW. 1985. Human auditory steady state evoked potentials during sleep. Ear Hear 6(3):167-174.

Linden RD, Picton TW, Hamel G, Campbell KB. 1987. Human auditory steady-state evoked potentials during selective attention. Electroencephalogr Clin Neurophysiol 66(2):145-159.

Lindsley DB. 1968. Neurophysiological basis of attention. Dev Med Child Neurol 10(2):250.

Lucertini M, Verde P, De Santis S. 2002. Human auditory steady-state responses during repeated exposure to hypobaric hypoxia. Audiol Neurootol 7(2):107-113.

Malmo RB. 1959. Activation: a neuropsychological dimension. Rass Giuliana Med No 11:86-114.

Martin BA, Tremblay KL, Stappels DR. 2007. Principles and application of cortical auditory evoked potentials. Auditory evoked potentials– basic principles and clinical application;edts Burkard RF, Eggermont JJ, Don M, pp. 482-507.

McCormick LH. 2003. ADHD treatment and academic performance: a case series. J Fam Pract 52(8):620-624, 626.

Mesulam MM. 1981. A cortical network for directed attention and unilateral neglect. Ann Neurol 10(4):309-325.

Michel CM, Seeck M, Landis T. 1999. Spatiotemporal Dynamics of Human Cognition. News Physiol Sci 14:206-214.

Moller AR. 1974. Responses of units in the cochlear nucleus to sinusoidally amplitude-modulated tones. Exp Neurol 45(1):105-117.

Morup M, Hansen LK, Herrmann CS, Parnas J, Arnfred SM. 2006. Parallel Factor Analysis as an exploratory tool for wavelet transformed event-related EEG. Neuroimage 29(3):938-947.

Moruzzi G, Magoun HW. 1949. Brain stem reticular formation and activation of the EEG. Electroencephalogr Clin Neurophysiol 1(4):455-473.

Mountcastle VB. 1978. Brain mechanisms for directed attention. J R Soc Med 71(1):14-28.

Moxon KA, Gerhardt GA, Adler LE. 2003. Dopaminergic modulation of the P50 auditory-evoked potential in a computer model of the CA3 region of the hippocampus: its relationship to sensory gating in schizophrenia. Biol Cybern 88(4):265-275.

Muller-Gass A, Stelmack RM, Campbell KB. 2005. "...and were instructed to read a self-selected book while ignoring the auditory stimuli": the effects of task demands on the mismatch negativity. Clin Neurophysiol 116(9):2142-2152.

Muller-Gass A, Stelmack RM, Campbell KB. 2006. The effect of visual task difficulty and attentional direction on the detection of acoustic change as indexed by the Mismatch Negativity. Brain Res 1078(1):112-130.

Muller N, Schlee W, Hartmann T, Lorenz I, Weisz N. 2009. Top-down modulation of the auditory steady-state response in a task-switch paradigm. Front Hum Neurosci 3:1.

Naatanen R, Picton T. 1987. The N1 wave of the human electric and magnetic response to sound: a review and an analysis of the component structure. Psychophysiology 24(4):375-425.

Naatanen R. 1992. Event-related potentials and automatic information processing. Attention and brain functions, pp. 102-210.

O'Donnell BF, Hetrick WP, Vohs JL, Krishnan GP, Carroll CA, Shekhar A. 2004. Neural synchronization deficits to auditory stimulation in bipolar disorder. Neuroreport 15(8):1369-1372.

Oken BS, Salinsky MC, Elsas SM. 2006. Vigilance, alertness, or sustained attention: physiological basis and measurement. Clin Neurophysiol 117(9):1885-1901.

Osipova D, Pekkonen E, Ahveninen J. 2006. Enhanced magnetic auditory steady-state response in early Alzheimer's disease. Clin Neurophysiol 117(9):1990-1995.

Pantev C, Elbert T, Makeig S, Hampson S, Eulitz C, Hoke M. 1993. Relationship of transient and steady-state auditory evoked fields. Electroencephalogr Clin Neurophysiol 88(5):389-396.

Pantev C, Roberts LE, Elbert T, Ross B, Wienbruch C. 1996. Tonotopic organization of the sources of human auditory steady-state responses. Hear Res 101(1-2):62-74.

Parasuraman R. 1980. Effects of information processing demands on slow negative shift latencies and N100 amplitude in selective and divided attention. Biol Psychol 11(3-4):217-233.

Pastor MA, Artieda J, Arbizu J, Marti-Climent JM, Penuelas I, Masdeu JC. 2002. Activation of human cerebral and cerebellar cortex by auditory stimulation at 40 Hz. J Neurosci 22(23):10501-10506.

Perry RJ, Hodges JR. 1999. Attention and executive deficits in Alzheimer's disease. A critical review. Brain 122 (Pt 3):383-404.

Picton TW, John MS, Dimitrijevic A, Purcell D. 2003a. Human auditory steady-state responses. Int J Audiol 42(4):177-219.

Picton TW, John MS, Purcell DW, Plourde G. 2003b. Human auditory steady-state responses: the effects of recording technique and state of arousal. Anesth Analg 97(5):1396-1402.

Picton TW. 2007. Audiometry using auditory steady-state responses. Auditory evoked potentials– basic principles and clinical application;edts Burkard RF, Eggermont JJ, Don M, pp. 441-462.

Plourde G. 1996. The effects of propofol on the 40-Hz auditory steady-state response and on the electroencephalogram in humans. Anesth Analg 82(5):1015-1022.

Plourde G, Baribeau J, Bonhomme V. 1997. Ketamine increases the amplitude of the 40-Hz auditory steady-state response in humans. Br J Anaesth 78 (5):524-529

Plourde G, Boylan JF. 1991. The auditory steady state response during sufentanil anaesthesia. Br J Anaesth 66(6):683-691.

Plourde G, Picton TW. 1990. Human auditory steady-state response during general anesthesia. Anesth Analg 71(5):460-468.

Plourde G, Villemure C, Fiset P, Bonhomme V, Backman SB. 1998. Effect of isoflurane on the auditory steady-state response and on consciousness in human volunteers. Anesthesiology 89(4):844-851.

Pockett S, Tan SM. 2002. The auditory steady-state response is not a suitable monitor of anesthesia. Anesth Analg 95(5):1318-1323, table of contents.

Polich J, Ilan A, Poceta JS, Mitler MM, Darko DF. 2000. Neuroelectric assessment of HIV: EEG, ERP, and viral load. Int J Psychophysiol 38(1):97-108.

Ponton C, Eggermont JJ, Khosla D, Kwong B, Don M. 2002. Maturation of human central auditory system activity: separating auditory evoked potentials by dipole source modeling. Clin Neurophysiol 113(3):407-420.

Posner MI. 1980. Orienting of attention. Q J Exp Psychol 32(1):3-25.

Posner MI. 1987. Cognitive neuropsychology and the problem of selective attention. Electroencephalogr Clin Neurophysiol Suppl 39:313-316.

Posner MI, Cohen Y, Rafal RD. 1982. Neural systems control of spatial orienting. Philos Trans R Soc Lond B Biol Sci 298(1089):187-198.

Posner MI, Petersen SE, Fox PT, Raichle ME. 1988. Localization of cognitive operations in the human brain. Science 240(4859):1627-1631.

Posner MI, Snyder CR, Davidson BJ. 1980. Attention and the detection of signals. J Exp Psychol 109(2):160-174.

Poulsen C, Picton TW, Paus T. 2009. Age-related changes in transient and oscillatory brain responses to auditory stimulation during early adolescence. Dev Sci 12(2):220-235.

Pribram KH, McGuinness D. 1975. Arousal, activation, and effort in the control of attention. Psychol Rev 82(2):116-149.

Pribram KH, McGuinness D. 1992. Attention and para-attentional processing. Event-related brain potentials as tests of a model. Ann N Y Acad Sci 658:65-92.

Rebert CS. 1978. Neuroelectric measures of lateral specialization in relation to performance. Electroencephalogr Clin Neurophysiol Suppl(34):231-238.

Regan D. 1989. Human Brain Electrophysiology: Evoked Potentials and Evoked Magnetic Fields in Science and Medicine. New York: Elsevier.

Reite M, Teale PD, Neumann R, Davis K. 1987. Localization of a 50 msec latency auditory evoked field component. Electroencephalogr Clin Neurophysiol Suppl 40:487-492.

Rickards FW, Tan LE, Cohen LT, Wilson OJ, Drew JH, Clark GM. 1994. Auditory steady-state evoked potential in newborns. Br J Audiol 28(6):327-337.

Rif J, Hari R, Hamalainen MS, Sams M. 1991. Auditory attention affects two different areas in the human supratemporal cortex. Electroencephalogr Clin Neurophysiol 79(6):464-472.

Robbins TW. 1997. Arousal systems and attentional processes. Biol Psychol 45(1-3):57-71.

Robbins TW, Everitt BJ. 1996. Neurobehavioural mechanisms of reward and motivation. Curr Opin Neurobiol 6(2):228-236.

Rogers LJ. 1991. Determination of the number and waveshapes of event related potential components using comparative factor analysis. Int J Neurosci 56(1-4):219-246.
Romani GL, Williamson SJ, Kaufman L. 1982. Tonotopic organization of the human auditory cortex. Science 216(4552):1339-1340.
Rosburg T, Trautner P, Elger CE, Kurthen M. 2009. Attention effects on sensory gating--intracranial and scalp recordings. Neuroimage 48(3):554-563.
Ross B, Herdman AT, Pantev C. 2005. Stimulus induced desynchronization of human auditory 40-Hz steady-state responses. J Neurophysiol 94(6):4082-4093.
Ross B, Picton TW, Herdman AT, Pantev C. 2004. The effect of attention on the auditory steady-state response. Neurol Clin Neurophysiol 2004:22.
Rusted JM, Trawley S. 2006. Comparable effects of nicotine in smokers and nonsmokers on a prospective memory task. Neuropsychopharmacology 31(7):1545-1549.
Santarelli R, Carraro L, Conti G, Capello M, Plourde G, Arslan E. 2003. Effects of isoflurane on auditory middle latency (MLRs) and steady-state (SSRs) responses recorded from the temporal cortex of the rat. Brain Res 973(2):240-251.
Santiago-Rodriguez E, Harmony T, Bernardino M, Porras-Kattz E, Fernandez-Bouzas A, Fernandez T, Ricardo-Garcell J. 2005. Auditory steady-state responses in infants with perinatal brain injury. Pediatr Neurol 32(4):236-240.
Saupe K, Widmann A, Bendixen A, Muller MM, Schroger E. 2009. Effects of intermodal attention on the auditory steady-state response and the event-related potential. Psychophysiology 46(2):321-327.
Sawyer DA, Julia HL, Turin AC. 1982. Caffeine and human behavior: arousal, anxiety, and performance effects. J Behav Med 5(4):415-439.
Scherg M, Von Cramon D. 1986. Evoked dipole source potentials of the human auditory cortex. Electroencephalogr Clin Neurophysiol 65(5):344-360.
Schoonhoven R, Boden CJ, Verbunt JP, de Munck JC. 2003. A whole head MEG study of the amplitude-modulation-following response: phase coherence, group delay and dipole source analysis. Clin Neurophysiol 114(11):2096-2106.
Shagass C. 1976. An electrophysiological view of schizophrenia. Biol Psychiatry 11(1):3-30.
Shinn JB, Musiek FE. 2007. The auditory steady state response in individuals with neurological insult of the central auditory nervous system. J Am Acad Audiol 18(10):826-845.
Skosnik PD, Krishnan GP, Aydt EE, Kuhlenshmidt HA, O'Donnell BF. 2006. Psychophysiological evidence of altered neural synchronization in cannabis use: relationship to schizotypy. Am J Psychiatry 163(10):1798-1805.
Skosnik PD, Krishnan GP, O'Donnell BF. 2007. The effect of selective attention on the gamma-band auditory steady-state response. Neurosci Lett 420(3):223-228.
Spencer KM, Salisbury DF, Shenton ME, McCarley RW. 2008. gamma-Band Auditory Steady-State Responses Are Impaired in First Episode Psychosis. Biol Psychiatry.
Stapells DR, Linden D, Suffield JB, Hamel G, Picton TW. 1984. Human auditory steady state potentials. Ear Hear 5(2):105-113.

Stapells DR, Makeig S, Galambos R. 1987. Auditory steady-state responses: threshold prediction using phase coherence. Electroencephalogr Clin Neurophysiol 67(3):260-270.

Steinschneider M, Reser DH, Fishman YI, Schroeder CE, Arezzo JC. 1998. Click train encoding in primary auditory cortex of the awake monkey: evidence for two mechanisms subserving pitch perception. J Acoust Soc Am 104(5):2935-2955.

Steinschneider M, Volkov IO, Noh MD, Garell PC, Howard MA, 3rd. 1999. Temporal encoding of the voice onset time phonetic parameter by field potentials recorded directly from human auditory cortex. J Neurophysiol 82(5):2346-2357.

Sukov W, Barth DS. 2001. Cellular mechanisms of thalamically evoked gamma oscillations in auditory cortex. J Neurophysiol 85(3):1235-1245.

Sussman E, Ritter W, Vaughan HG, Jr. 1998. Attention affects the organization of auditory input associated with the mismatch negativity system. Brain Res 789(1):130-138.

Sussman E, Ritter W, Vaughan HG, Jr. 1999. An investigation of the auditory streaming effect using event-related brain potentials. Psychophysiology 36(1):22-34.

Swanepoel D, Erasmus H. 2007. Auditory steady-state responses for estimating moderate hearing loss. Eur Arch Otorhinolaryngol 264(7):755-759.

Szalda K, Burkard R. 2005. The effects of nembutal anesthesia on the auditory steady-state response (ASSR) from the inferior colliculus and auditory cortex of the chinchilla. Hear Res 203(1-2):32-44.

Tcheslavski G, Beex AA. 2010. Effects of smoking, schizotypy, and eyes open/closed conditions on the γ_1 rhythm phase synchrony of the electroencephalogram. Biomedical Signal Processing Control 5 (2): 164-173

Vaughan HG, Jr., Ritter W. 1970. The sources of auditory evoked responses recorded from the human scalp. Electroencephalogr Clin Neurophysiol 28(4):360-367.

White PM, Yee CM. 1997. Effects of attentional and stressor manipulations on the P50 gating response. Psychophysiology 34(6):703-711.

Wienbruch C, Paul I, Weisz N, Elbert T, Roberts LE. 2006. Frequency organization of the 40-Hz auditory steady-state response in normal hearing and in tinnitus. Neuroimage 33(1):180-194.

Woldorff MG, Hillyard SA. 1991. Modulation of early auditory processing during selective listening to rapidly presented tones. Electroencephalogr Clin Neurophysiol 79(3):170-191.

Yerkes RM, Dodson JD. 1908. The relation of strength of stimulus to rapidity of habit-formation. J Comparative Neurol Psychol 18:459–482.

12. PUBLICATIONS

Griskova I, Morup M, Parnas J, Ruksenas O, Arnfred SM. 2007. The amplitude and phase precision of 40 Hz auditory steady-state response depend on the level of arousal. Exp Brain Res 183(1):133-138.

Griskova I, Morup M, Parnas J, Ruksenas O, Arnfred SM. 2009. Two discrete components of the 20 Hz steady-state response are distinguished through the modulation of activation level. Clin Neurophysiol 120(5):904-909.

Paper in preparation

Griskova-Bulanova I, Ruksenas O, Dapsys K, Maciulis V, Arnfred SM. Distraction task rather than focal attention modulates gamma activity associated with auditory steady-state responses.

Conference proceedings:

1.
2. Vasiliauskaite D, GRIŠKOVA-BULANOVA I. The Comparative Analysis of Steady-state Response Generation Model Data and Real Data. Proceedings of National scientific-practical conference „Virtual Instruments in Biomedicine“, Klaipėda, Lithuania, 2010.05.21, p.91-94.
3. GRIŠKOVA-BULANOVA I, Paškevič J, Rukšėnas O., Mačiulis V. Arousal-dependent modulation of auditory P50 potential. Proceedings of 13th International conference „Biomedical Engineering“, Kaunas, Lithuania, 2009. 10. 23-24, p.116-118.
4. GRISKOVA I, Morup M, Parnas J, Ruksenas O, Arnfred SM. The time-frequency analysis of steady-state responses evoked by periodical acoustic stimulation. Proceedings of 12th International conference „Biomedical Engineering“, Kaunas, Lithuania, 2009. 10. 23-24, p. 145-148.

5. GRIŠKOVA I, Rukšėnas O. Noninvaisive analyses methods of electrical brain responses. Proceedings of the Fifth Scientific Conference of Faculty of Natural Sciences, Vilnius, Lithuania, 2008.09.26, p.46-56.

Conference abstracts

1. GRIŠKOVA-BULANOVA I, Paškevič J, Iljinychas I, Rukšėnas O, Mačiulis V. Task-dependent modulation of auditory evoked P50 potential. Proceedings of 1st Lithuanian neuroscience association scientific conference, Vilnius, Lithuania, 2009.11.20, p. 19.
2. GRISKOVA I, Morup M, Ruksenas O, Arnfred SM. The Phase Precision of 40 Hz Steady-state Response in awake and drowsy conditions. Proceedings of the Fourth Scientific Conference of Faculty of Natural Sciences, Vilnius, Lithuania, 2006.10.23-24, p. 251-252.
3. GRISKOVA I, Ruksenas O, Dapsys K, Hoppner J. The Effects of 10Hz Transcranial Magnetic Stimualtion on the Resting EEG Power Spectrum. Proceedings of the Fourth Scientific Conference of Faculty of Natural Sciences, Vilnius, Lithuania, 2006.10.23-24, p. 253-254.
4. GRIŠKOVA I, Dapšys K, Rukšėnas O, Šiurkutė A, Mačiulis V. Effects of electroconvulsive therapy on P300 potential in schizophrenic patients. Proceedings of the Congress of Laboratory Medicine, Vilnius, Lithuania, 2006.05.18-20, p. 25.
5. GRIŠKOVA I, Rukšėnas O, Dapšys K, Höppner J. Repetitive transcranial magnetic stimulation effect on EEG power spectra. Proceedings of the Congress of Laboratory Medicine, Vilnius, Lithuania, 2006.05.18-20, p. 25.

Conference presentations

Oral:

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3. GRISKOVA I, Morup M, Panas J, Ruksenas O, Arnfred SM. The time-frequency analysis of steady-state responses evoked by periodical acoustic stimulation. 12th International conference „Biomedical Engineering“, Kaunas, Lithuania, 2009.10.23-24.
4. GRIŠKOVA I, Rukšėnas O. Noninvaisive analyses methods of electrical brain responses. The Fifth Scientific Conference of Faculty of Natural Sciences, Vilnius, Lithuania, 2008.09.26.
5. GRISKOVA I, Arnfred SM. An Electrophysiological Approach to Investigations of Sensory Dysfunction in Schizophrenia. International school „The Impact of Current Developments in the Neurosciences on the Concept of Psychiatric Disease”, Bonn, Germany, 2007.10.2-9.
6. GRISKOVA I. Precision of auditory steady state response during arousal and drowsiness. Conference “Status on research projects“, Hvidovre hospital, Copenhagen, Denmark, 2006.04.24

Posters:

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2. GRISKOVA I, Morup M, Ruksenas O, Arnfred SM. The Phase Precision of 40 Hz Steady-state Response in awake and drowsy conditions. The Fourth Scientific Conference of Faculty of Natural Sciences, Vilnus, Lithuania, 2006.10.23-24.
3. GRISKOVA I, Ruksenas O, Dapsys K, Hoppner J. The Effects of 10Hz Transcranial Magnetic Stimualtion on the Resting EEG Power Spectrum. The Fourth Scientific Conference of Faculty of Natural Sciences, Vilnius, Lithuania, 2006.10.23-24.
4. GRISKOVA I, Morup M, Parnas J, Arnfred SM. The Phase Precision of Auditory Steady-state Response Elicited by 20Hz Stimulus Repetition Rate. PENS Training Centre "Imaging Brain Functions: From Molecules to Mind", Lausanne and Geneva, Switzerland, 2006.09.4-24.
5. GRIŠKOVA I, Dapšys K, Rukšėnas O, Šiurkutė A, Mačiulis V. Effects of electroconvulsive therapy on P300 potential in schizophrenic patients. Congress of Laboratory Medicine, Vilnius, Lithuania, 2006.05.18-20.
6. GRIŠKOVA I, Rukšėnas O, Dapšys K, Höppner J. Repetitive transcranial magnetic stimulation effect on EEG power spectra. Congress of Laboratory Medicine, Vilnius, Lithuania, 2006.05.18-20.

Publications not included in dissertation:

1. Veršinskas R, GRIŠKOVA I, Razgauskas E, Račiukaitytė K, Jankauskas D, Jocys Č, Motuzas R, Marksienė E, Vaičiulis K, Žukauskas G, Liausėdas A. (2010) Agresijos požymiai įvairiose Lietuvos žmonių grupėse. Sveikatos mokslai, 2, 3034-3046.(in Lithuanian)
2. GRISKOVA I, Arnfred S. (2008) An electrophysiological approach to investigations of sensory dysfunction in schizophrenia. Poiesis and Praxis, DOI 10.1007/s10202-008-0063-1

3. GRISKOVA I, Ruksenas O, Dapsys K, Herpertz S, Hoppner J. (2007) The effects of 10 Hz repetitive transcranial magnetic stimulation on resting EEG power spectrum in healthy subjects. Neuroscience Letters, 419(2): 162-167.
4. GRISKOVA I, Hoppner J, Ruksenas O, Dapsys K. (2006) Transcranial magnetic stimulation: the method and application. Medicine (Kaunas), 42(10):798-804.
5. GRISKOVA I, Dapsys K, Andruskevicius S, Ruksenas O. (2005) Does electroconvulsive therapy (ECT) affect cognitive components of auditory evoked P300? Acta Neurobiologiae Experimentalis, 65:73-77.
6. GRISKOVA I, Dapsys K, Ruksenas O, Kaukenas R, Maciulis V. (2005) Association between changes of ERPs and physiological response to ECT treatment: a pilot study. Laboratory Medicine, 4(28):3-6.
7. GRISKOVA I, Dapsys K, Ruksenas O, Siurkute A, Maciulis V. (2005) Effects of electroconvulsive therapy on auditory information processing in treatment-resistant schizophrenic patients: P300 potential study. Laboratory Medicine, 3(27):14-17.
8. GRISKOVA I, Buchmann J, Ruksenas O, Dapsys K, Hoppner J. (2005) Application of transcranial magnetic stimulation in motor cortex excitability studies: brief review. Laboratory Medicine, 4(28):11-16.
9. GRISKOVA I, Dapsys K, Ruksenas O, Korostenskaja M. (2004) Mechanisms of action of electroconvulsive therapy. Theory and Practice in Medicine, 1(37):90-91.

Book chapters:

GRISKOVA I. Changes assessed by EEG after TMS. In Repetitive transcranial magnetic stimulation (rTMS) in the treatment and rehabilitation of central nervous diseases. Edited by Mally J. Budapest, Eurobridge Co, 2009, p.77-93.

GRISKOVA I, composite author. Sleep and anxiety. Edited by Verbitsky EV. Rostov-on-Don, SSC RAS Publishers, 2008, p. 222-231. (in Russian)

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14. CURRICULUM VITAE

Personal data

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Education

2006- Present: pursuing Ph.D. in Biophysics, Vilnius University, Vilnius, Lithuania, Mentor: Osvaldas Ruksenas (Habil. Dr. Biomed. Sci., Professor); scientific advisor Sidse M. Anfred (Dr.)
2004-2006 MSc in Neurobiology, Vilnius University, Vilnius, Lithuania,
2000-2004 BSc in Molecular biology, Vilnius University, Vilnius, Lithuania,

Academic awards

1) 2006-2010: Research scholarships for doctoral students from Lithuanian Science and Study Foundation.
2) 2007: Gerbert Rüf Stiftung scholarship for research visit at Functional Brain Mapping Laboratory, Neurology Clinics, University Hospital of Geneva, Geneva, Switzerland.
3) 2006: CIRIUS scholarship for research visit at Cognitive Research Unit, Hvidovre hospital, Copenhagen university hospital, Copenhagen, Denmark.
4) 2005: ERASMUS scholarship for research visit at Neurophysiology Laboratory in the Centre of Nervous Disease, Department of Psychiatry, Rostock University, Rostock, Germany.
5) 2005: Lithuanian Science Academy award for Student scientific work.

Scientific interests

Electoencephalogaphy, event-related potentials, transcranial magnetic stimulation

Positions held

2008-present: Lecturer, Mykolas Romeris University, Vilnius, Lithuania,
2007-present: Medical biologist, Republican Vilnius psychiatric hospital, Vilnius, Lithuania,
2007-2009 Junior research fellow, Vilnius University, Vilnius, Lithuania,
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Teaching experience

2007-present: lectures and conduction of laboratory exercises for the course „Psychophysiology“
2006-2008: conduction of laboratory exercises for the course „Human and animal physiology“
2007-2010: supervision of term papers and theses (undergraduate students)

Scientific visits

1) Traineeship at Kiev national Taras Sevchenko university, Faculty of Biology, Department of Human and animal physiology, Kiev, Ukraine, 2008.01.18-22
2) Fellowship at Functional Brain Mapping Laboratory, Neurology Clinics, University Hospital of Geneva, Geneva, Switzerland, 2007.04.01-2007.05.01
3) Traineeship at Cognitive Research Unit, Hvidovre hospital, Copenhagen university hospital, Copenhagen, Denmark, 2006.07, 2006.11.
4) Fellowship at Cognitive Research Unit, Hvidovre hospital, Copenhagen university hospital, Copenhagen, Denmark, 2006.02.01-2006.06.01
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Related experience

MOCCA (Model for Core Curricula with Integrated Mobility Abroad), project seminar, Napier University, Edinburg, Scotland, 2007.09.19 – 22

ESFA funded project „Biophysics: Modernization of Master and doctoral programs“, preparation of course “Biophysics of Sensory Systems”, 2007-2008

Summer schools and workshops

1) Ethical aspects of scientific research with humans, Vilnius University, Lithuania, 2007
2) Statistical analyses of scientific results: principles and practical application programs, Vilnius University, Lithuania, 2008
3) International school „International School on Neural Nets “Dynamic Brain”, Erice, Italy, 2007.12.5-12
4) International school „Advanced Methods in Biophysics“, Trakai, Lithuania, 2007.11.26-30
5) International school „The Impact of Current Developments in the Neurosciences on the Concept of Psychiatric Disease”, Bonn, Germany, 2007.10.2-9
6) International symposium „Symposium on imaging and simulation of human brain activity“, Split, Croatia, 2007.07.21-24.
7) PENS Training Centre “Imaging Brain Functions: From Molecules to Mind”, Lausanne & Geneva, Switzerland, 2006.09.04-24.
8) Workshop “EEG Generation and Interpretation”, University of Kuopio, Kuopio, Finland, 2006.08.28-31.