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Influence of Physiological Changes of Glycemia on VEPs and Visual ERPs

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Summary

Since hypoglycemia is known to influence cognitive functions, we checked whether the physiological changes in glycemia (after fasting or exertion) can explain the rather high intra-individual variability of event-related potentials (ERPs). Besides the ERPs to "change in coherence of a moving pattern" with reaction time (RT) recording, binocular pattern reversal VEPs and motion-onset VEPs (to linear and radial motion) were also examined in 14 healthy subjects prior to and after 24-h fasting that decreased glycemia from 5.3 to 3.9 mmol/l on the average. We only found one significant change in the latencies and amplitudes of VEPs and ERPs (with no change of RT). The N160 peak in the motion-onset VEPs to radial (expansive) motion (EM-VEPs) showed a larger amplitude at lower glycemia. For evaluation of the exertion influence, we tested glycemia prior to and after 90 min long exercise – bicycle ergometry with the load set to 2 W/kg in women and 2.5 W/kg in men (average age-related values for W170/kg index). The changes of glycemia to exertion were, however, less distinct than those to fasting. We conclude that in healthy subjects the glycemia decrease due to 24-h fasting or intensive time-limited exercise never reaches the critical value to change the VEP, ERPs and RTs.

Key words

Hypoglycemia • Visual event related potentials • Cognitive potentials • Evoked potentials • Fasting • Exertion

Introduction

Event related potentials (ERPs) reflect the mental processes connected with recognition of "occasional" stimuli and "frequent" ones. From the clinical point of view, this type of evoked potentials represents a promising approach especially to psychiatric disorders evaluation – the specific changes of ERPs (mainly the P300 latency prolongation) were described e.g. in various types of dementia (mainly in Alzheimer´s disease), schizophrenia (for review see Polich 1991), depression (Bange and Bathien 1998) and even in multiple sclerosis (Polich *et al.* 1992) and migraine (Evers *et al.* 1998). However, the routine diagnostic use of ERPs encounters the problem of relatively very high inter-individual variability, which causes apparent difficulties in the evaluation of differences between groups (e.g. patients vs. normal subjects). The inter-individual variability can result from various factors (for review see Polich and Herbst 2000) and the estimation of

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ERPs thus seems to be more reliable when the intra-individual changes (like the medication effects) are tested. Nevertheless, not even the individual ERP parameters are absolutely stable – they become prolonged with age (Knott *et al.* 2003) and can be theoretically influenced by body function fluctuations (temperature, fatigue, food intake, circadian rhythm etc.) (for review see Polich and Herbst 2000). Fatigue (measured in "time on task" testing) was shown not to alter the ERPs (Polich *et al.* 2000) and we have shown that the circadian rhythm also does not cause the ERP intra-individual variability (Kubová *et al.* 2002). Another reason for the variations of ERP parameters could be hypoglycemia; the latency delay and amplitude decrease of late components of ERPs to auditory stimuli were described e.g. by Jones *et al.* (1990). However, studies reporting the hypoglycemia effect on ERPs were so far based on insulin-induced lowering of plasma glucose only. We were therefore interested whether similar ERP changes can also result from a physiologically induced hypoglycemia following simple fasting or excessive body activity, i.e. situations which can easily occur in patients.

Methods

Subjects

Fourteen healthy drug-free subjects (7 women and 7 men, mean age 28±11 years, all right-handed) with normal visual acuity (corrected if needed) participated in the fasting experiments and another age-matched group of 14 subjects (7 women and 7 men, mean age of 26±7 years) was included in the physical exertion experiments.

The examination of patients and control subjects were planned and confirmed in advance by the Ethical Committee of our Faculty of Medicine and they were performed with the full consent of the subjects.

Experimental designs

Glycemia measurement and the set of VEP and ERP tests were performed first after a normal breakfast and then after 24 h of complete fasting (fasting experiments) or prior to and after 60 min bicycle ergometry (exertion experiments). The load was set to 2 W/kg in women and 2.5 W/kg in men, which are the average values fitting well to normal data for W 170/kg index (W170 = working capacity at a pulse rate of 170 beats/min) of moderately trained subjects of this age (Seliger and Bartůněk 1977). Glycemia was tested by means of the Glucocard II GT-1630 digital test meter.

Electrophysiological testing

The same set of VEPs and ERPs was tested in each subject from the "fasting" experimental group; the electrophysiologic investigation was not performed in the "exertion" group since the glycemia changes were less distinct in this group (see the Results).

Transient pattern-reversal visual evoked potentials (P-VEPs) were acquired with high contrast (96 %) square-wave black and white checkerboard (element size 40'). Two variants of motion-onset VEPs (M-VEPs) were used. The first consisting of linear motion (random order of fundamental directions, velocity 10 deg/s) of low contrast (10 %) isolated checks (40' check size and 120' check-to-check distances). Second motion stimulus consisted of low contrast (10 %) gray concentric frames with increasing size and motion velocity (centrifugal expansion) from the center (fixation point) towards the periphery (respecting the size of the retinal perception fields and sensitivity differences to motion velocity across the retina). Both moving stimuli had the same timing – 200 ms of motion was followed by 1 s inter-stimulus interval (stationary pattern). In motion-onset VEPs, the latency and amplitude of the N160 peak was evaluated (this peak was shown to represent the main motion-related component of this VEP type – Kuba and Kubová 1992).

Visual stimuli for cognitive EPs consisted of four low contrast (10 %) 40' checks placed centrally in the visual field on the grey background (L = 12.6 cd/m^2). The checks were either displayed in the stationary mode (for inter-stimulus interval of 1-3 s) or they moved at a velocity of 10°/s for 200 ms. In a pseudo-random order the upper and lower rows of checks moved either both in the same direction (left or right) – "coherent" motion or they moved in opposite directions – "non-coherent" motion. This type of stimulus was introduced in an earlier article from our laboratory (Kuba *et al.* 1998). The oddball paradigm was applied for recording of cognitive responses. The target stimulus was the non-coherent motion and subjects were asked to press a hand-held button immediately when they recognized it. This was used for an off-line evaluation of the chosen reaction time. The ratio of the target (rare) and non-target (frequent) stimuli was 1:3. The latencies and amplitudes of the P300 peak were estimated in cognitive EPs.

All visual stimuli were generated (using our own software (Kremláček *et al.* 1998) and AutoDesk Animator, USA) on the 21"Monitor ViewSonic with the vertical frequency of 70 Hz. The stimulus field subtended

45 x 35 deg at viewing distance of 0.5 m. The average luminance was 17 cd/m^2. Correct fixation of the center of the stimulus field was monitored *via* an infra-red CD camera.

Recording and data analysis

Standard recordings included pseudounipolar derivations (with the right ear lobe reference) from the midline Oz, Pz, Cz and Fz and also from Ol and Or (5 cm to the left and right from the Oz position). These lateral recording sites were used, since N160 motion-onset specific peak is mostly lateralized (irrespective of the dominant hemisphere) towards the temporo-occipital cortex (Kuba and Kubová 1992). For further evaluation, the data were taken from the lead with the maximum response (typically Oz in P-VEPs and Ol or Or in M-VEPs).

Each stimulus condition was repeated at least twice and the average value of the latencies and amplitudes was evaluated. Forty single VEPs (440 ms epochs with sampling frequency 500 Hz) were averaged. In ERP altogether 80 sweeps were recorded (1000 ms epochs), 20 target and 20 non-target responses were averaged. In case of artificial contamination (most frequently by electric activity related to eye blinking), the whole recording was repeated. To verify a possible contamination of motion related VEPs by eye movements, both horizontal and vertical electro-oculograms were also recorded in three subjects (electrode placement on outer canthi and above and below the right eye). No significant eye movement related activity was found.

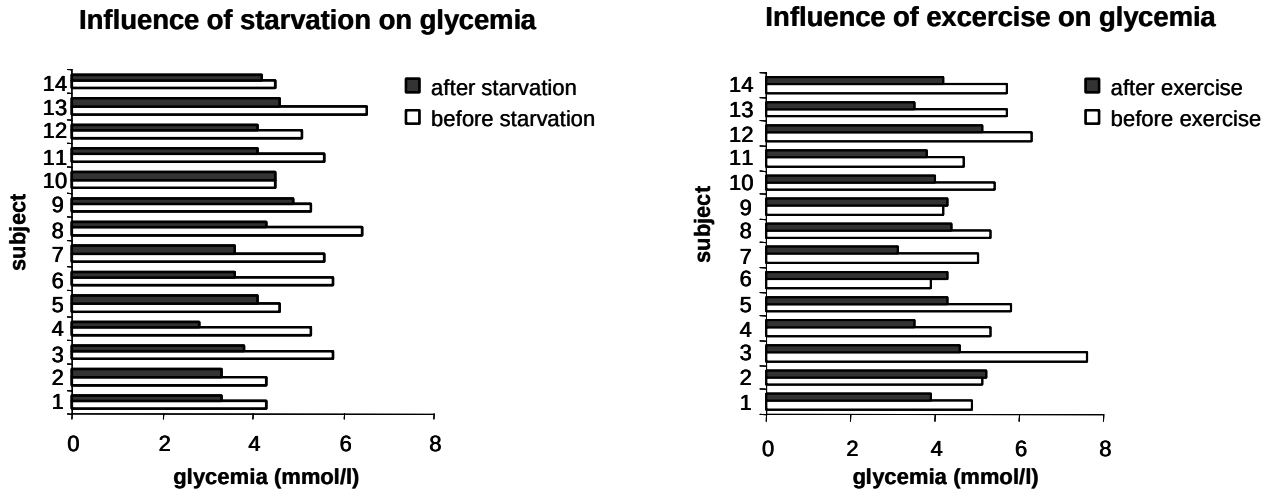

Fig. 1. Individual changes of glycemia to fasting and exertion (both groups n = 14 subjects). Grey part of the background represents normal physiological range of glycemia (3.1- 6.7 mmol/l).

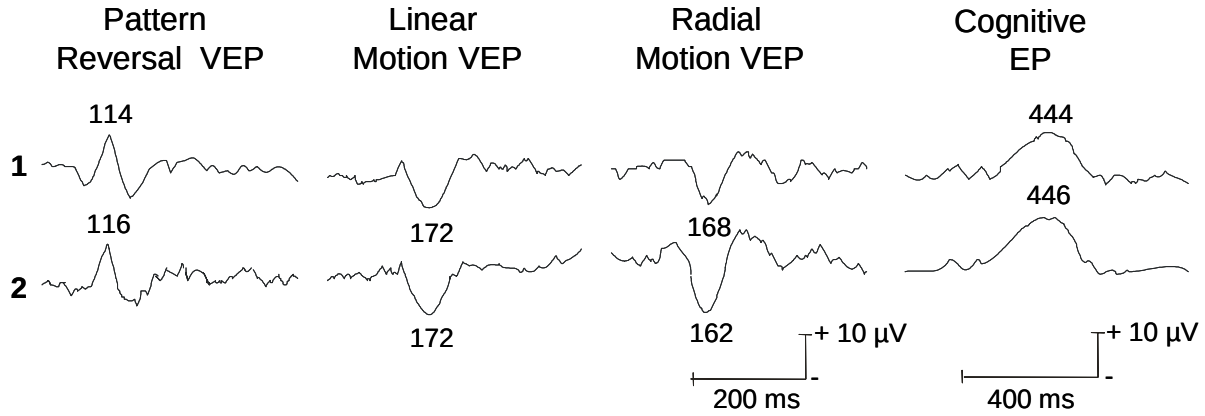

Fig. 2. Results of electrophysiological examination in one subject from the „fasting"group (the person with the deepest glycemia drop – to 2.8 mmol/l - was selected). 1 – set of EPs obtained before fasting, 2 – set of EPs obtained after 14 hours of fasting. Note two different scales – one for the pattern-reversal and both motion VEPs, second for the cognitive EPs.

Results

Fasting experiment

The 24-h fasting period caused a significant lowering of glycemia ($p<0.001$) with the average decrease of glycemia of 1.3±0.7 mmol/l (mean ± S.D.). Nevertheless, in all but one subject, the individual value of blood glucose after fasting remained within the physiological range (3.3-6.1 mmol/l) (Fig. 1). Whilst the glycemia prior to starving was about the same in men and women, the fasting influenced women's glycemia to a much greater extent ($p<0.001$). The average decrease of glycemia in men was 1.0 mmol/l, in women 1.6 mmol/l.

As to the electrophysiological findings, the set of tested EPs is presented in Figure 2. We did not find any difference in the latencies of any of the tested peaks to both pattern-reversal, motion-onset and cognitive stimuli prior to and after a period of fasting (Fig. 3). No change in amplitudes was found in the pattern-reversal and cognitive EPs, however, fasting caused a significant ($p<0.01$) increment in the N2 peak of the VEPs to an "expanding" motion stimulus (Fig. 3).

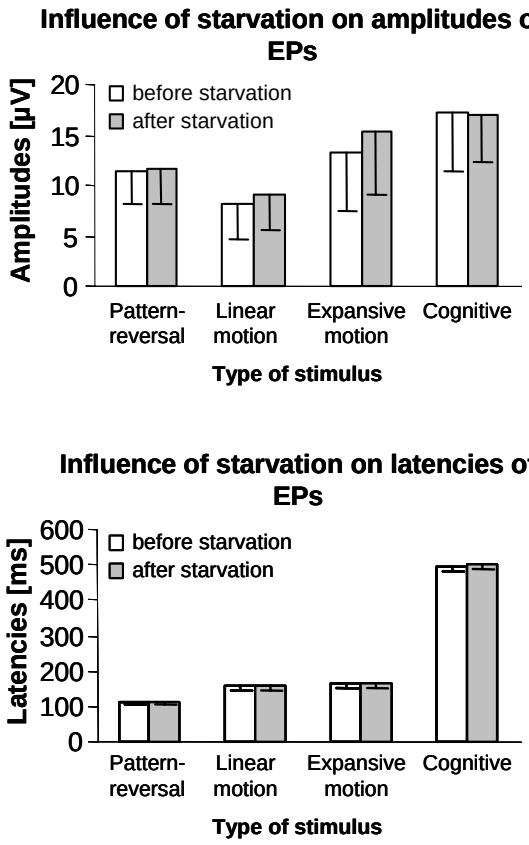

Fig. 3. Comparison of amplitudes and latencies of tested EP types before and after fasting. SDs are depicted downwards only. ** represents significance - $p<0.01$.

The reaction time (RT) comparison has shown that the group averages of all tested RT types (we estimated the minimum, maximum RT and its median) remained without any significant change after fasting, although this was quite different in individual subjects.

Exertion experiment

Sixty minutes of exercise also significantly lowered the glycemia ($p<0.001$), but the average decrease was lower than that in the fasting experiment (1.1±0.7 mmol/l) and the glycemia values stayed always in within the normal physiological range. In three subjects, the glucose blood level even increased after ergometry (Fig. 1). Hence, there was no need to perform the electrophysiological testing in this group of subjects.

Discussion

It is generally well known that the blood level of glucose is carefully maintained at a stable level that is necessary for the normal function of the whole organism and especially of the nervous system. In case of hypoglycemia, which occurs in normal subjects most often from prolonged fasting or excessive body exertion, the person affected shows a set of characteristic subjective and objective signs. With the use of the hyperinsulinemic glucose clamp technique it was found that the autonomic symptoms (anxiety, palpitations, sweating, irritability and tremor) began at a glucose level of 3.2 mmol/l and the neuroglycopenic symptoms (hunger, dizziness, tingling, blurred vision, difficulty of thinking and faintness) as well as deterioration in cognitive function tests occurred when plasma glucose dropped below 2.8 mmol/l (Mitrakou *et al.* 1991). The earliest cognitive dysfunctions were noted at glycemia of about 2.8 to 3.1 mmol/l (Jones *et al.* 1990, Snorgaard *et al.* 1991, Blackmann *et al.* 1992, Fanelli *et al.* 1994). The data suggest that at glycemia around 3 mmol/l the mental activities which are relatively undemanding are often unaffected, while the performance of more complex tasks deteriorates (for review see Heller and MacDonald 1996).

Our experiments have shown that 24-hour fasting does not lead to any change of latencies of all tested evoked potentials (pattern-related, motion-related as well as cognitive), which is the parameter almost exclusively estimated in clinical practice. The reason is quite obvious – the hypoglycemia, although eliciting the unpleasant subjective feelings in all cases, never fell (except of one subject) below 2.8 mmol/l, i.e. the value

reported to be the level of the onset of cognitive dysfunctions (e.g. Jones *et al.* 1990). Since the 60-min lasting intensive exercise caused an even less distinct change of glycemia than fasting, it seems evident that both limited fasting and exercise (a situation occurring in patients who were subjected to electrophysiological testing) elicited in healthy (non-diabetic) subjects regulatory mechanisms capable of maintaining glycemia in the physiological range. For this reason we can conclude that the physiological fluctuations of glycemia are not responsible for the intra-individual variability of the cognitive evoked potentials. Very probably much longer fasting or extreme exertion could influence the glycemia in a more distinct way; however, we do not expect such activity in patients who are tested in an electrophysiological laboratory.

Surprisingly, we found that the amplitude of VEPs to centrifugal motion increased significantly after fasting. This can be perhaps related to the known fact that mild hypoglycemia (in the range between 3.0 and 3.8 mmol/l) makes the CNS more excitable, because this degree of hypoglycemia seems to facilitate neuronal activity (Guyton 1980). In comparison with centripetal motion that represents a strong warning signal, the centrifugal motion serves more for allocation of attention. We can speculate that the change of overall activity due to hypoglycemia can explain the above mentioned finding.

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