Longitudinal high-density EEG study of looming in full-term and preterm infants

by

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Acknowledgments

Abstract

Electroencephalogram (EEG) was used on full-term and preterm infants at the age of 5 months and again at the age of 12 months to investigate brain electrical activity as a function of looming visual stimuli on a collision course. Analyses of visual evoked potential (VEP) were performed on EEG data recorded with a 128-channel sensor array. VEP results showed that prematurity affects the brain processing of looming visual stimuli under three different accelerations. Preterm infants at the age of 12 months responded to the looming visual stimuli early in the looming sequence on average -1.12 s before virtual contact as compared to 12-month-old full-term infants who responded on average -0.43 s before contact. In addition, full-term infants at the age of 12 months had switched to a strategy based on time, whereas preterm infants at the age of 12 months still appeared to be using a strategy based on visual angle. The results further showed that with growing age, both full-term and preterm infants showed significantly shorter VEP durations for all three looming conditions compared to when they were 5 months old. Furthermore, it was found that full-term and preterm infants at the age of 5 months showed larger negative brain activity in the Oz electrode compared to when they were 12 months old. However, full-term infants at the age of 12 months showed larger negative brain activity in the Pz electrode than at the age 5 months, but preterm infants did not show increased brain activity in the Pz electrode with age. Preterm infants showed poorer performance in looming visual motion perception that seems to be related to impairments of the dorsal visual stream which is specialized in processing visual motion. As the infants grow older they will face tasks with higher demands and complexity. The small deviations shown at an early age could possibly be a predictor of later visual processing disabilities.

1. Introduction

It is important for both animals and humans to respond quickly when a danger is approaching them. While this response may vary between species, the main function is to detect whether or not an object is approaching on a direct collision course and how imminent is the collision. An object on a direct collision course will project and expand image with a rapid growth on the retina, this is referred to as looming (Schiff, Caviness, & Gibson, 1962).

A wide range of studies on avoidance behavior to a looming stimulus in both non-mammals and mammals has been carried out. Studies on animals like the fly (Holmqvist, 1994), locust (Rind & Simmons, 1997), pigeon (Sun & Frost, 1998), monkey (Zeki, 2004; Zeki, 1974) and other animals show that middle temporal (MT) and medial superior temporal area (V5) (Zeki, 2004) are active to detect and respond to looming movement. The eye's perception of visual information is reproduced from the lateral geniculate nucleus (LGN) to primary cortex. The information in primary cortex is divided by two interconnected pathways, either through a dorsal stream towards the parietal lobe or through a ventral stream towards the temporal lobe. The dorsal stream is involved in the processing of movement while the ventral stream is involved in object recognition (Dougherty, Koch, Brewer, Ficher, Modersitzki, & Wandell, 2003; Holliday & Meese, 2005; Morrone, Tosetti, Monntanaro, Fiorentini, Cioni, & Burr, 2000; Shirai & Yamaguchi, 2004).

In early infancy, the processing of motion stimuli is mostly localized in the occipito-parietal areas in the left and right visual cortex (van der Meer, Fallet & van der Weel, 2008; van der Weel & van der Meer, 2009). Different studies on infants and adults show that motion engages peripheral magno cells (M cells) within the occipital cortex (Molloy, Doyle, Makrides, & Anderson, 2012) and also neurons in the dorsal stream of the cortex and its surrounding areas (V1, V2, MT +) are activated by motion stimuli (Braddick, Birtles,

Wattam-Bell, & Atkinson, 2005; Cheng, Fujita, Kanno, Miura, & Tanaka, 1995; Rosander, Nyström, Gredebäck, & Hofsten, 2007).

While navigating in the environment, the visual world streams around observers in a pattern which reflects their motion. These optic flow fields combine the effects of all observers' movements in three-dimensional space to provide visual information that can guide self-motion, stabilize posture, and reveal the structure of the environment (Gibson, 1986). Adults (Royden, Crowell, & Banks, 1994) and infants both can perceive optic flow, but infants can only discriminate large changes in heading during navigation (Gilmore, Baker, & Grobman, 2004).

Visual evoked potential (VEP) is a measuring technique that can help researchers to study the neural basis of motion perception in the millisecond (ms) time scale through electroencephalography (EEG) (Kubová, Kuba, Spekreijse, & Blakemore, 1995). The VEP amplitude represents the number of post-synaptic response from main cortical pyramidal neurons to changes in afferent visual activities (Pfurtscheller & Lopes da Silva, 1999).

Motion perception is different in the infant and adult brain. A high density EEG study investigated and compared brain electrical activity in response to perception of structured optic flow and random visual motion in infants and adults. This study showed significant differences in N2 component of VEP for optic flow versus random visual motion. Infants showed larger amplitudes as compared to adults. They also showed longer latencies especially for random visual motion as compared to adults (van der Meer, Fallet, & van der Weel, 2008). These findings suggested that lack of self-produced locomotor experience in young infants, results in less efficient detection of optic flow.

Some studies support the fact that the human brain starts developing before birth and a lot of developments take place during the first year of life. The first three months of human life are

important for stabilizing gaze on a visual flow field with head and body movements (Von Hofsten & Rosander, 1996). Because the infant brain during the first year of life undergoes rapid changes and also doubles in weight. There is strengthening of some connections and pruning of other connections. This strengthening and pruning of neural pathways might be a developmental change that affects processing speed and efficiency as a more restricted brain area gets activated due to neural pathway specialization (Johnson, 2000). Synaptic maturation (Grieve, Emerson, Fifer, Isler, & Stark, 2003) of the visual cortical areas (Dubois, Dehanene-Lambertz, Cointepas, Le Bihan, & Hertz-Pannier, 2008) and myelin sheaths over the axon can increase the speed of electrical impulse propagation from soma to dendrites of the neurons. During early maturational stages until adulthood synaptic maturation and myelination could affect the visual processing speed with age (Webb, Long, & Nelson, 2005).

In daily life, visual information is used for motor actions and predicting future events. It becomes more important when an object is approaching on a collision course. A magneto-encephalography (MEG) study in humans has shown significantly larger brain activity in response to looming visual stimuli as compared to other motion sequences (Holliday & Meese, 2005).

Experiments on infants regarding looming stimuli and defensive behavior (Bower, Broughton, & Moore, 1971; Schiff et al., 1962; Yonas, Pettersen, & Lockman, 1979) were at their peak in the 1960's and 1970's. Experimenters caught an interest in looming stimuli and investigated how soon infants could respond to motion stimuli. Recent studies also investigated infants' defensive behavior in response to a virtual looming stimulus heading towards them. The stimulus was programmed to loom symmetrically towards the infant's face so the infant would get an experience of a visual collision (Kayed & van der Meer, 2007; van der Meer, Svantesson, & van der Weel, 2012).

Blinking of the eye is an indication that the infant is aware of the optical collision (Yonas et al., 1977). Behavioral studies on blinking the eyes to optical collisions in infants have shown that as infants grow older, they switch to using a more advanced timing strategy and judge the upcoming collisions more accurately (Kayed & van der Meer, 2007). Timing of defensive blinking should be accurate to protect the eyes from things that are about to hit the face. If the blinking occurs too early then the eyes will reopen before the collision has taken place. On the other hand, if the blinking occurs after the collision, the sensitive cornea of the eyes could be injured. Thus, defensive blinking needs to be accurately timed. Kayed and van der Meer (2007) suggested that infants initially used a visual angle or velocity strategy and then switched to a strategy based on time at the age of seven months. The findings also showed that infants who used strategies based on visual angle and velocity showed a significantly higher number of late defensive blinks than infants who used a strategy based on time (Kayed & van der Meer, 2007).

Another study on infants reported that looming-related brain activity localized in visual cortex is characterized by theta oscillations between 5-11 months of age. A very prominent developmental trend was found where infants at the age of 5-7 months were not able to differentiate between the loom's three different accelerations and also used about twice as long to process the looming information, but infants at the age of 10-11 months showed very short and separated brain waves (van der Weel & van der Meer, 2009).

Preterm labor is that which begins prior to the completion of 37 weeks of pregnancy, and its causes are not fully understood. Delivering a baby between 32-34 weeks poses even more risks to learning and behavioral problems because brain development is incomplete and preterm babies are more likely to show developmental delay. But most development of the brain takes place during the first year of life (Dougherty et al., 2003). As mentioned above, at the time of birth preterm infants' brain development is still incomplete and they could

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therefore face more risks during coming years. Inder et al. (2005) identified important volumetric changes in the brain structures of preterm infants, in the grey matter of cortex and in the cerebral white matter, with an increase in the cerebrospinal fluid volume. Therefore, developmental researchers highlight the developmental disorders related to prematurity and seek to understand the mechanisms of brain and behavioral development.

Preterm infants face lots of challenges when they come into this world. The first few months are very difficult for them and they have to struggle for survival, and during later months they need to catch up with their full-term peers. Studies have shown that preterm infants are at more risk of facing difficulty at school than infants born at term (Wolke & Meyer, 1999). At the age of two, preterm infants have shown a clear trend in performing poorer on a memory task as compared with full-term infants (Woodward, Edgin, Thompson, & Inder, 2005). The survival rates of preterm babies have increased due to the advancement in neonatal health care. Yet, preterm infants also face a high risk of cerebral injury or white matter damage causing problems in motor development and possibly cognitive and behavioral problems as well (Inder, Warfield, Wang, Hüppi, & Volpe, 2005).

Docosahexaenoic acid (DHA), an omega 3 long chain polyunsaturated fatty acid is present in high concentrations in cerebral cortex and retina (Neuringer, Connor, Lin, Barstad, & Luck, 1986). This fatty acid is highly important for vision. During the last three months of pregnancy, a significant amount of DHA is transferred from mother to fetus. Thus, infants who are born < 32 weeks have insufficient amount of DHA and have higher rates of refractive errors, strabismus, amblyopia, and deficient visual perception skills (Cioni et al., 2000; Downie, Jakobson, Frisk, & Ushycky, 2003; O'Connor et al., 2001). DHA is an important component for synthesis of brain tissue, metabolism of neurotransmitters, cellular differentiation and synaptogenesis (Sabel, Lundqvist-Persson, Bona, Petzold, & Strandvik, 2009). Several studies suggested that preterm infants show a particular vulnerability of dorsal

stream processing (Atkinson & Braddick, 2007; Jakobson, Frisk, & Downie, 2006). MRI studies have shown that M cells need a high amount of polyunsaturated fatty acids to preserve their membrane flexibility crucial for allowing physiological processes, which underlie M cells transient sensitivity important for detecting motion (Stein, 2001). Thus damaged or inefficient M cells in the magnocellular pathway and dorsal stream may affect processing and functions within and related to these pathways.

Understanding the functional development of the infant brain is important today, since it may help us to provide preterm infants with an early diagnosis and open the way to better therapeutic interventions. In this project the aim was to understand the brain development in early infancy by focusing on the effects of prematurity in response to looming visual stimuli via high-density EEG. It was investigated whether there are any significant differences in timing of looming-related brain responses through a longitudinal perspective by testing preterm and full-term infants at the age of 5 months and again at the age of 12 months.

2. Method

2.1 Participants

Preterm and full-term normally developing infants were recruited for the study. In the preterm group, ten healthy infants (7 boys, 3 girls) were recruited with the help of the pediatrician in charge of the Neonatal Intensive Care Unit at St. Olav's University Hospital (Trondheim, Norway). The recruitment criteria were: born at ≤ 33 weeks of gestation with birth weight ≥ 1000 grams and absence of any major problems such as brain damage, retinopathy of prematurity (ROP), or any other serious prenatal problems. But, experimenters were unaware of the infants' birth history, neurological status, and any other events related to their stay in hospital.

In the full-term group, ten healthy infants (5 boys, 5 girls) born at term were recruited through local newspaper birth announcements or parents showing an interest and making voluntary contact with the laboratory. According to parental report, all infants were developing normally.

Preterm and full-term infants were matched according to their age in full months. For valid matching, age corrected for prematurity was calculated for the preterm babies. However, one of the preterm babies was not comfortable during the second testing at the age of 12 months and we were unable to collect any data on him. As a result, he had to be excluded from the longitudinal analysis. So, his matched full-term control was excluded automatically. Thus, longitudinal analyses were performed on 18 infants in total for both testing sessions.

Both groups were tested twice (longitudinal design). For the first testing the mean age of preterm infants was 4 months and 26 days (SD=6.9) with a range of 139-158 days, followed by the second testing with the same preterm infants at the mean age of 12 months and 18 days (SD=18.6) with a range of 356-414 days. The mean age of the full-term infants at the first

testing session was 4 months and 16 days (SD=21.3) with a range of 111-164 days and for the second testing the same full-term infants was tested at the mean age of 11 months and 20 days (SD=13.9) with a range of 333-371 days. At the first testing session none of the infants had any crawling experience, while at the second testing session all infants had crawling experience from the age of 9 months, except for three of the preterm infants who had not started crawling or pulling to stand at the time of the second testing, and were bottom shuffling instead.

Electroencephalogram (EEG) as a psychological procedure causes no physical harm or pain to participants. Before the experiment, parents gave their informed and signed consent and they were free to withdraw from the study at any time. The Norwegian Regional Ethics Committee and the Norwegian Data Services for Social Sciences approved the study. This study was conducted in the Developmental Neuroscience Laboratory at the department of Psychology (NTNU) Trondheim, Norway.

2.2 Apparatus

E-prime (Psychology Software Tools) was used to generate the stimuli. The looming stimulus was projected on a white screen (108 cm wide, 70.5 cm high) which was placed 80 cm away from the infants, through a video projector (ASK M2) (see Figure 1A). During the experiment, infants' attention and behavior was monitored with two digital cameras. Tobii x50 was used to record the gaze of both eyes of infants by picking up their corneal reflection.
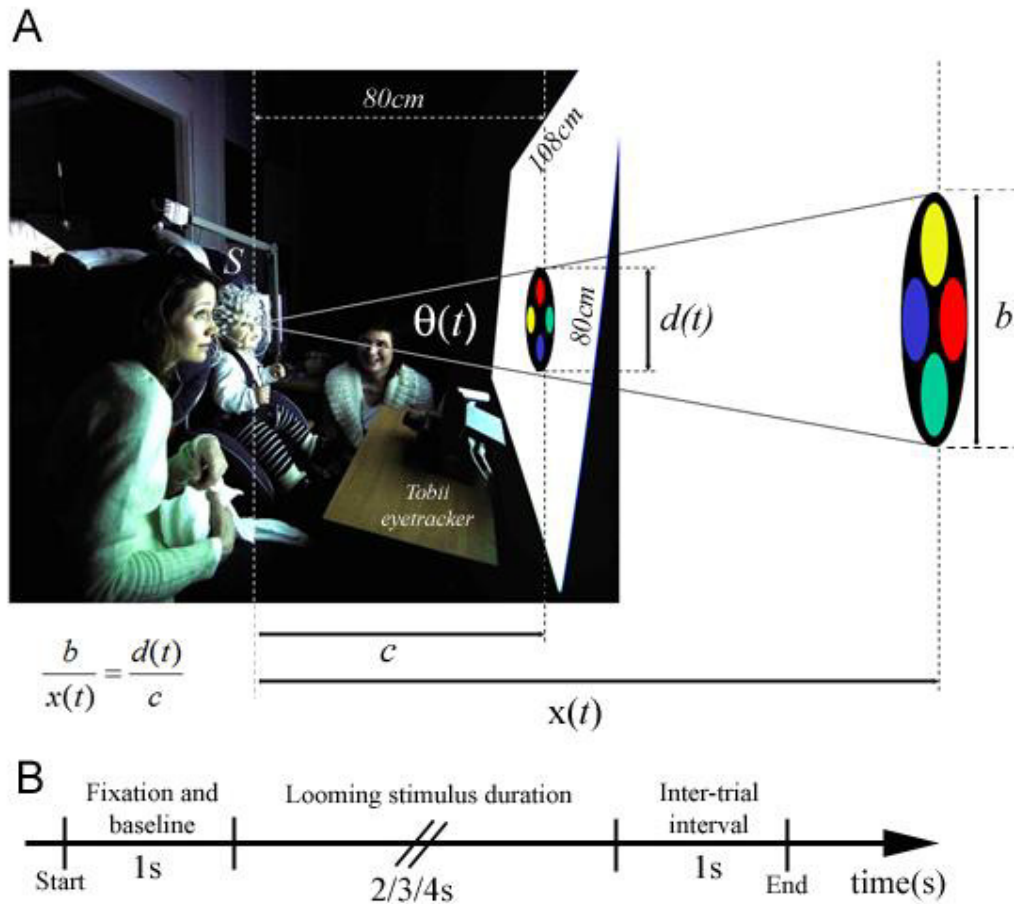
Figure 1. *Experimental setup with a diagram of the stimulus configuration. (A) Each infant was shown a virtual object of a flat circle approaching on a direct collision course. The looming stimuli simulated an object coming from far way approaching for duration of 2, 3 and 4 seconds under three different constant accelerations (-21.1 m/s², -9.4 m/s² and -5.3 m/s²). The stimulus approached the infant as the image on the screen grew, and the loom stopped when the image filled the entire screen leaving a blank screen. (B) Timeline showing the approach of the looming disc and time duration during the phase of fixation, looming, and inter-trial interval from start to end in a single trial.*

To record EEG activity, a high density 126 channel Geodesic Sensor Net (GSN) 200 (Tucker, 1993) was used. Across the head surface, arrays of 12g Ag sponge sensors were evenly distributed. Net Station software on a Macintosh computer was used to record the amplified EEG signals, using a sampling rate of 500 Hz with a low pass filter of 100 Hz and 0.1 Hz high pass filter. To ensure an optimal signal-to-noise ratio, all electrode impedance was kept under 50 kΩ as recommended for the high-input-impedance EGI amplifiers (Budai,

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Contento, Locatelli, & Comi, 1995; Ferree, Luu, Russell, & Tucker, 2001; Picton et al., 2000). For off-line analyses, data were stored on a hard disk.

2.3 Stimuli

The stimulus consisted of an image of a flat black circle disc with four colored (red, green, blue, and yellow) rotating inner circles of equal size. The rotating object was shown on a white projection screen with constant angular velocity of 300 degrees per second (see Figure 1). The radius of the inner circles was 1/3 of the radius of the outer circle. To get an experience of a visual collision, the stimuli were programmed to loom towards the infant with different accelerations, and finally coming up to the infant's face. The virtual object appeared on the screen at a virtual distance of 43.1 m and stayed there at its minimum size for 1 s. It expanded during the looming phase and finally reached its largest size and then disappeared, leaving a blank screen for 1 s. The virtual object initially had a visual angle of 5° (diameter of 6.5 cm) and expanded to a maximum visual angle of 131° (diameter of 350 cm). It moved towards the infant under three different conditions with constant acceleration over a period of 2s (-21.1 m/s²), 3s (-9.4 m/s²) and 4s (-5.3 m/s²). The virtual object had the same visual angle at the beginning and at the end of the approach, independent of the virtual object's approach time. It moved over the same virtual distance (43.1 m – 0.80 m) at a constant acceleration in all three conditions (see Figure 1B). The three looming conditions with a fourth reversed condition appeared on the screen in random order. The reversed condition was introduced as a control condition simulating the circular disc moving away from the infant and turned out to be great in (re)capturing the infant's interest.

2.4 Procedure

Parent(s) arrived with their infant some time prior to the experiment. At that time, the experimenter informed the parent(s) about the experiment and also played with the baby so

that the baby became familiar, relaxed, and settled down before the experiment. In the process, an assistant measured the infant's head circumference for the correct size selection of the GSN 200. To optimize electrical conductivity, the appropriate electrode net was soaked in a saline electrolyte. After that, the net was mounted on the infant's head while he/she was sitting on the parent's lap. The infant's attention was distracted from the net with the help of small sounding toys and soap bubbles. After mounting the net, the infant was positioned in front of the screen while sitting on the parent's lap (5-month-olds) or placed in a baby car seat (12-month-olds) with one parent present in the experimental room, to avoid any stress an absent parent may have on the infant. An assistant was also present in the experimental room, to help the infant focus on the screen. The net was connected to the amplifier and the impedance of the electrodes was checked.

The experimental room was separated by a transparent glass partition from a control room where two assistants controlled the computers necessary for the data acquisition. An experimental session started immediately after calibrating the infant's eye movement in virtual space to the Tobii x50. In both groups of infants, each infant completed between 40-80 trials. If an infant lost interest in the stimuli or started fussing, the experiment was ended. Each infant was tested individually and testing sessions lasted for about 20-30 minutes. Each session consisted of an optic flow, looming, and occlusion experiment. The looming experiment was usually conducted after the optic flow experiment, about 5 minutes into an experimental session.

2.5 Data analysis

Brain Electrical Source Analysis (BESA) research software version 5.3 was used to analyze the raw EEG data. Same methods were used to analyze the data for both testing sessions. Recordings were segmented with the Net Station software as an initial pre-processing step

and then exported as raw files to BESA for further analyses. By visual inspection, bad channels and trials contaminated with artifacts from body/head movement were discarded. None of the participants had more than 10% of the channels defined as bad. Notch filter was set at 50 Hz to remove 50 Hz lines interference from the recorded data. Low cut-off filter (high band pass) was set at 1.6 Hz to remove slow drift in the data while a high cut-off filter (low band pass) was set at 30 Hz. A reference free montage showing EEG at 27 standard electrodes was used.

A VEP is the post synaptic firing pattern of neurons at a particular time in relation to a particular visual stimulus (Webb et al., 2005). It represents the changes in the brain's electrical activity at a high temporal resolution by using high density EEG. For typical looming related VEP responses, the main occipital and parietal electrodes were specifically observed for the three different looming conditions. Highly prominent VEP peaks were marked at electrode site Oz based on earlier studies investigating visual evoked potentials (Di Russo, Martínez, Sereno, Pitzalis, & Hillyard, 2002; van der Meer et al., 2012). These VEP peaks provided information about the looming-related brain activity at the selected brain region. This activity could also be visualized by a 3D mapping of a build-up and a decline of voltage activity in the visual cortex on a trial-by-trial basis (see Figure 2).
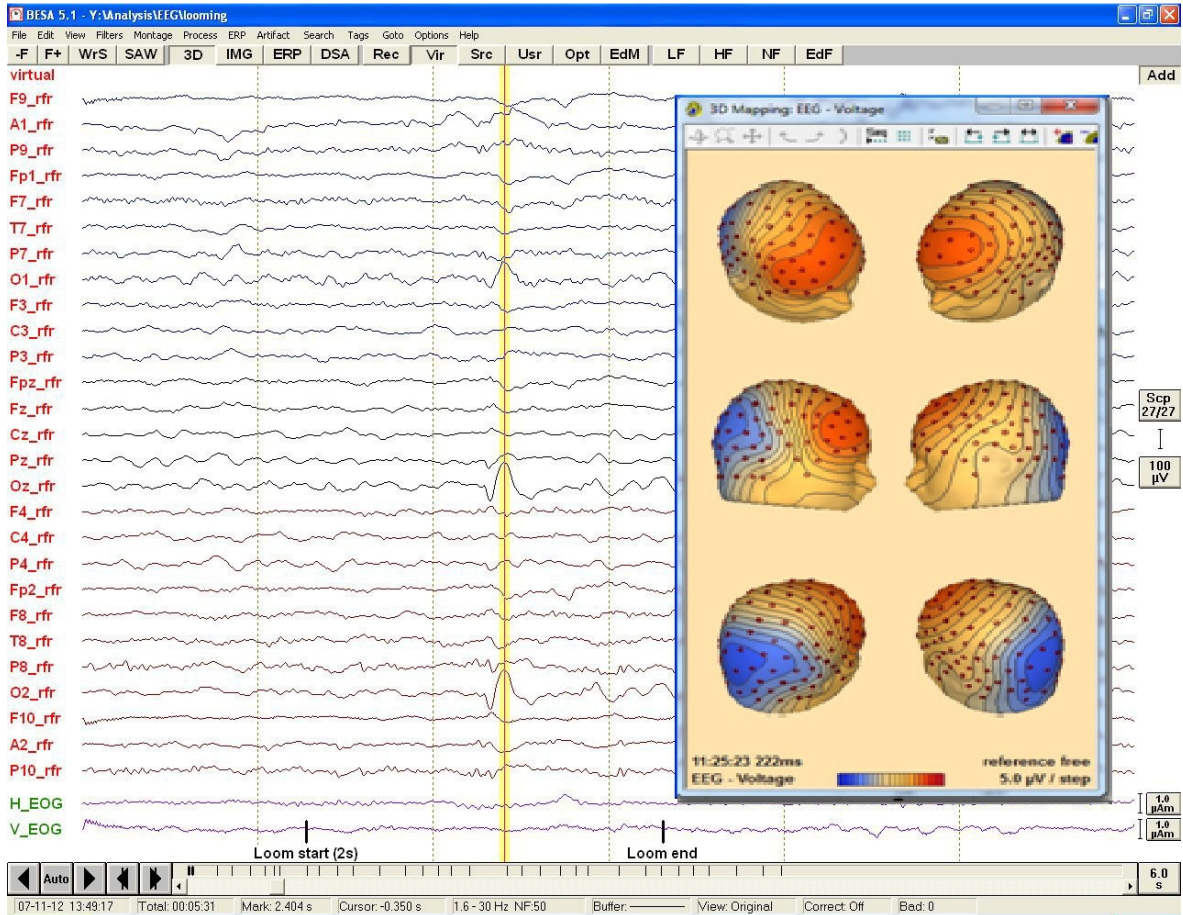
Figure 2: *EEG data analysis in BESA showing a typical looming VEP response in the middle of the looming sequence. A reference free montage of 27 electrodes is shown. Prominent VEP peak activity at site Oz in a 2 s looming trial is viewed as a direct response to the looming stimulus. A 3D voltage mapping shows high activity in the visual cortex.*

When a number of consecutive looming peaks were seen in a trial, only one peak was selected that showed increased brain activity in 3D mapping window and was near to the stimulus end (see Figure 2). Trials in which looming responses were not prominent or occurring too late (after the end of the loom) were excluded from further analysis.

2.5.1 Timing of VEP responses

In this study, we investigated which strategy infants used to time their looming-related brain response. This can be illustrated by plotting the looming stimulus at an arbitrary point in time

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during its looming phase. The graph shows the expected pattern when the brain response is initiated by either one of the three information variables (see Figure 3).
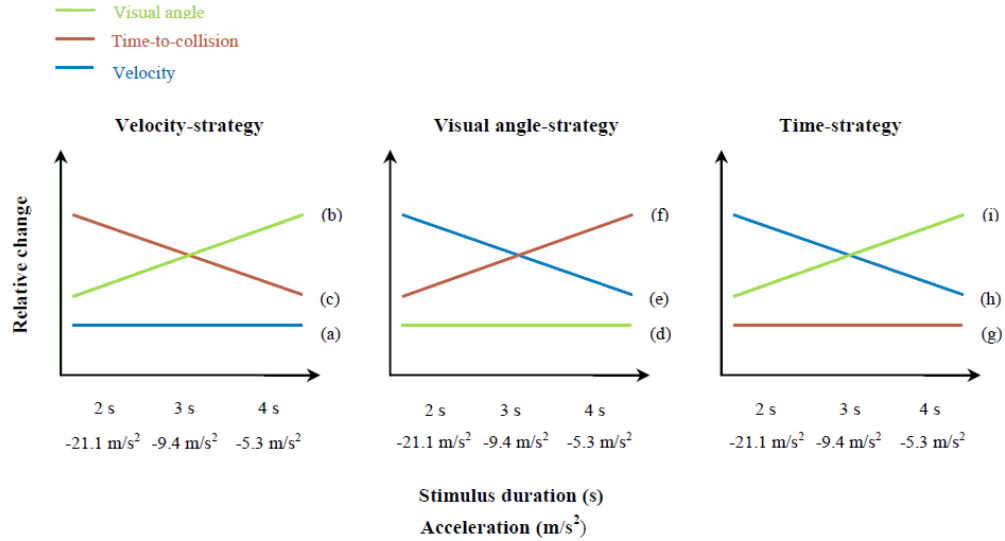

Figure 3: *A theoretical graph of the looming stimulus' velocity, visual angle, and time-to-collision as a function of the duration and acceleration of the looming sequence. A velocity-strategy implies that as the acceleration of the loom decreases, (a) the velocity of the loom is kept relatively constant, while the loom's (b) visual angle increases, and (c) time-to-collision decreases. A visual angle-strategy implies that as the acceleration of the loom decreases, (d) the visual angle of the loom is kept relatively constant, while the loom's (e) velocity decreases and (f) time-to-collision increases. A time-strategy implies that as the acceleration of the loom decreases (g) the loom's time away from the virtual collision is kept relatively constant, while the loom's (h) velocity decreases and (i) visual angle increases. For simplicity the lines are drawn as straight lines.*

A time-strategy is defined as a constant brain response when the looming object reaches a specific time before the optical collision. As the acceleration decreases, the loom's time-to-collision is held relatively constant (see Figure 3g), while the loom's velocity would decrease (see Figure 3h) and the visual angle would increase (see Figure 3i).

This information variable is not dependent on knowledge of object size or speed so the usage of a time-strategy would always leave the same amount of time for the brain response. The

time-strategy is considered to be more efficient strategy that is less prone to errors compared to the visual angle strategy and velocity strategy (Kayed et al., 2008; Kayed & van der Meer, 2000, 2007).

Duration of time from start to end of each VEP was explored in depth in Oz electrode, to detect any differences in time duration for looming-related VEP responses between full-term and preterm infants (see Figure 4).
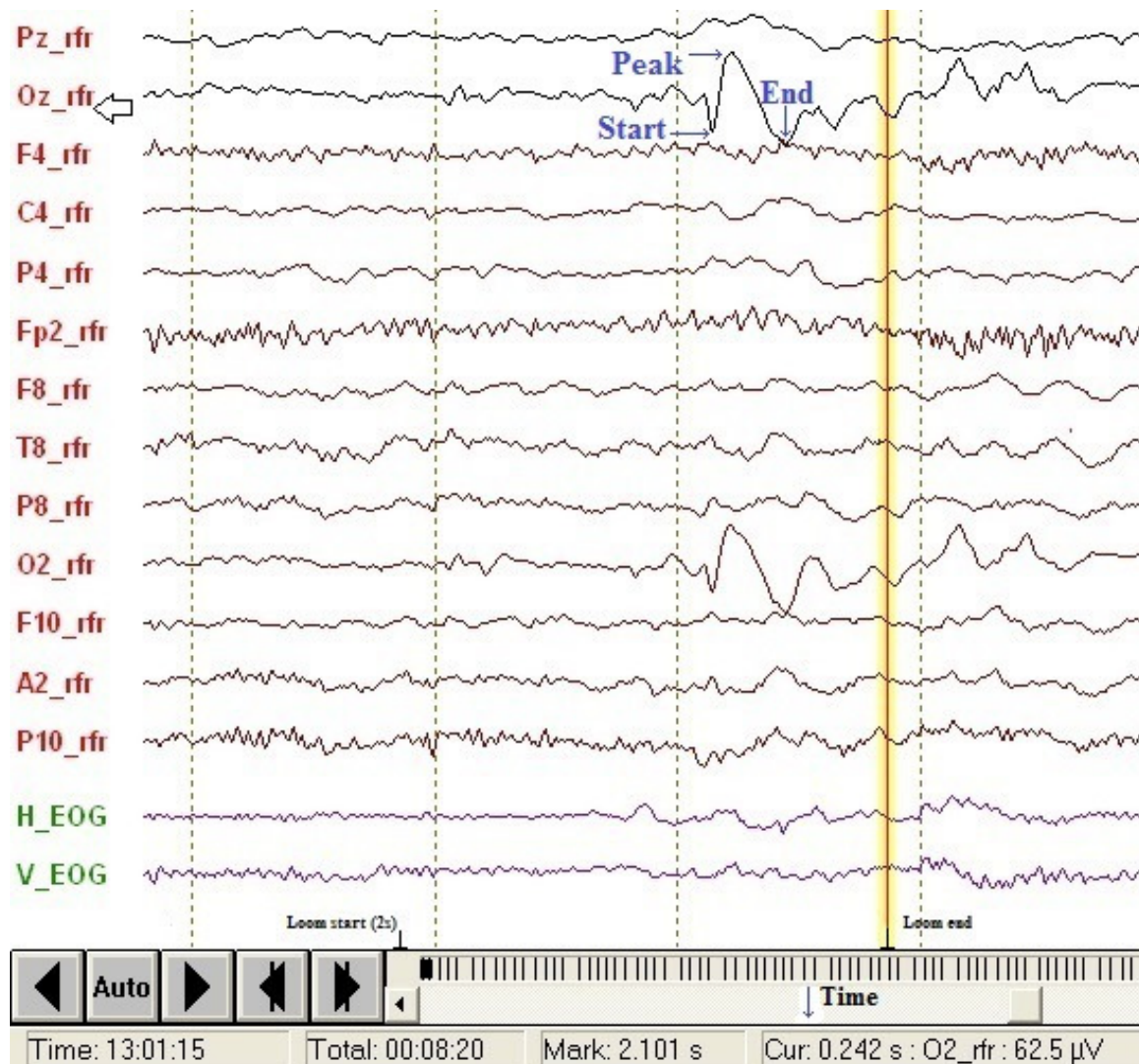

Figure 4: *Marking and timing of individual VEP in raw EEG data, showing how the start, peak and end of the looming-related VEP were marked manually in Oz electrode in a single trial. Corresponding timing data were recorded from the lower tab.*

2.5.2 VEP analysis at the electrode level

Each individual infant's data were averaged over whole EEG files and interpolated to 81 standard electrodes after re-referencing to an artificial reference calculated from the average potentials over the scalp such that individual averages were obtained. The individual averages for both groups of infants in each of the two sessions were then combined into grand averages. The grand average VEP amplitudes for two posterior electrodes Oz and Pz in full-term and preterm infants were recorded for further analysis.

3. Results

3.1 VEP responses

The analyses were performed on an average of 46 trials (SD = 3.6, range = 41-52) for full-term infants at the age of 5 months, 43 trials (SD = 10.1, range = 23-53) for full-term infants at the age of 12 months, 32 trials (SD = 7.3, range = 20-40) for preterm infants at the age of 5 months, and 31 trials (SD = 7.2, range = 25-43) for preterm infants at the age of 12 months. The number of trials was distributed equally among the three looming conditions. Looming-related VEP responses were observed in the occipital and parietal areas, especially in Oz and Pz some time before the loom would have made contact with the infant.

Five-month-old full-term infants showed their looming-related VEP responses on average -1.77 s (SD = 0.19) before contact and with age their looming-related VEP responses occurred closer to the virtual collision, on average at -0.43 s (SD = 0.08). Preterm infants at five months showed their looming-related VEP responses on average -1.61 s (SD = 0.08) before contact and with age their looming-related VEP responses occurred closer to the virtual collision, on average at -1.12 s (SD = 0.17).

A 2 (group: full-term and preterm) x 2 (age: 5 and 12 months) x 3 (loom condition: 2, 3, and 4 s) repeated measures ANOVA was performed on averaged looming-related peak activation. Adjustment for multiple comparisons was made by Bonferroni correction. The results showed a main effect of age, $F(1, 16) = 10.87$, $P < 0.005$, indicating that with age, both preterm and full-term infants responded at smaller values of time-to-contact across the three loom speeds, as well as a two-way interaction effect of age and group, $F(1, 16) = 89.56$, $P < 0.005$, indicating that at 12 months full-term infants responded significantly closer to time-to-contact than preterm infants (see Figure 5).

The results further showed a three-way interaction effect of age, group, and looming condition, $F_{(2, 32)} = 6.22$, $P < 0.005$, indicating that looming-related responses occurred closer to contact (i.e., later in the looming sequence) with increasing age and with increasing loom speeds, with the exception of the full-term infants at the age of 12 months who showed their averaged looming-related responses at a fixed time-to-contact of about -0.430 s, irrespective of loom speed (see Figure 5).
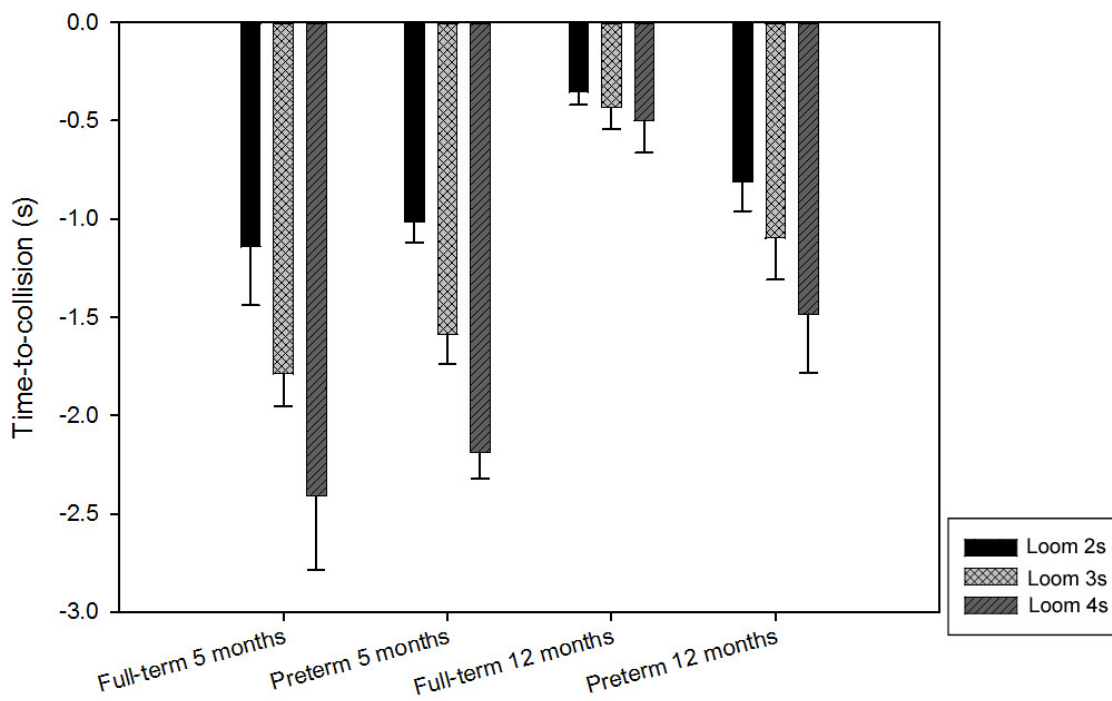

Figure 5: *Averaged looming-related VEP peak activation (including SD bars) for the three looms at the age of 5 months and at the age of 12 months in both full-term and preterm infants. With increasing age both full-term and preterm infants responded at smaller values of time-to-contact across the three loom speeds, but full-term infants at the age of 12 months responded significantly closer to time-to-contact than preterm infants. Also, looming-related responses occurred closer to contact in the looming sequence with increasing age and with increasing loom speeds, but only full-term infants at the age of 12 months showed their averaged looming-related responses at a fixed time-to-contact of about -0.430 s irrespective of loom speed.*

3.2 VEP duration

Averaged VEP duration in the Oz electrode ($VEP_{start} - VEP_{end}$) for looming-related peaks at 5 months for both full-term and preterm infants, 0.28 s (SD = 0.05) was significantly longer than averaged VEP duration at 12 months for full-term and preterm infants, 0.23 s (SD = 0.03), irrespective of loom speed.

Another 2 (group: full-term and preterm) x 2 (age: 5 and 12 months) x 3 (loom condition: 2, 3, and 4 s) repeated measures ANOVA was performed on averaged VEP duration. The results showed a main effect of age on averaged VEP duration, $F(1, 16) = 6.16$, $P < 0.05$, indicating that averaged VEP duration at 12 months was significantly shorter than averaged VEP duration at 5 months for both full-term and preterm infants, irrespective of loom speed. Results further showed a three-way interaction effect of age, group, and looming condition, $F(2, 32) = 8.03$, $P < 0.005$, indicating that only the full-term infants at the age of 5 months showed increasing VEP duration for looming-related peaks with decreasing loom speed (see Figure 6).
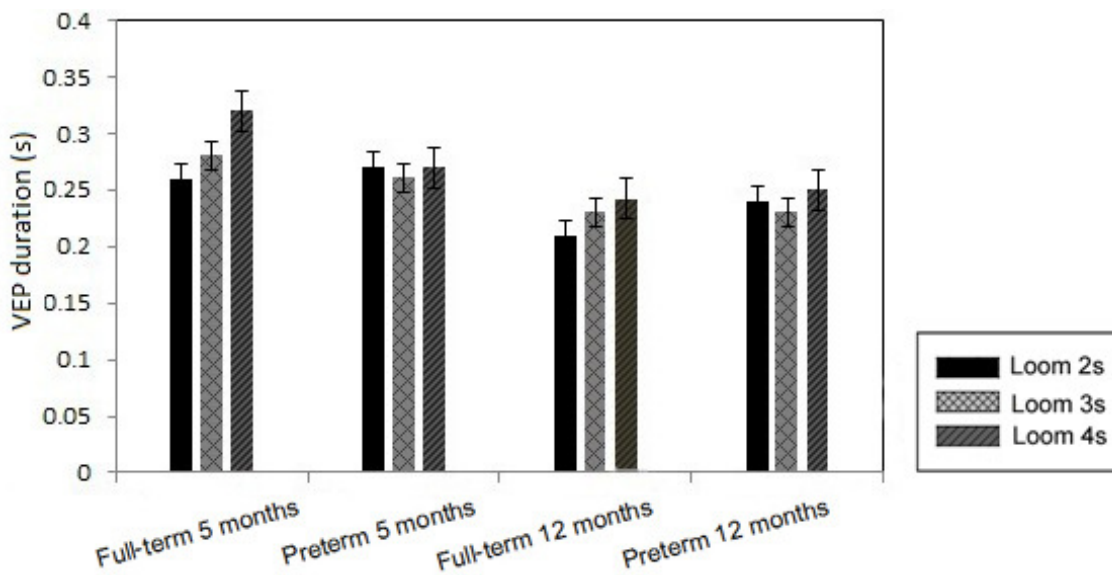

Figure 6: *Averaged VEP duration in Oz electrode for looming-related peaks, $VEP_{start} - VEP_{end}$ (including SD bars) for the three looms at the age of 5 months and at the age of 12 months for both*

full-term and preterm infants. VEP duration for looming-related peaks across looms at the age of 5 months was significantly longer than averaged VEP duration at the age of 12 months for both full-term and preterm infants.

3.3 VEP amplitudes in Oz and Pz

The grand average VEP amplitudes for two posterior electrodes Oz and Pz in full-term and preterm infants at the age of 5 months and 12 months are shown in Figure 7. These occipital and parietal electrodes showed the highest looming-related peaks of the grand average VEPs.

A 2 (group: full-term and preterm) x 2 (age: 5 and 12 months) x 2 (electrode: Oz, Pz) repeated measures ANOVA was performed on averaged peak VEP activation. The results showed a main effect of age, $F(1,16) = 49.31$, $P< 0.01$, indicating that amplitude activity decreased with growing age. A significant two-way interaction of age and cortical area was found, $F(1,16) = 67.29$, $P< 0.01$, indicating that irrespective of group of infants, amplitude activity in cortical area electrode Oz decreased significantly with growing age, whereas amplitude activity in cortical area electrode Pz did not change significantly with growing age. Further, a significant three-way interaction of group, age, and cortical area was found, $F(2, 32) = 23.18$, $P < 0.01$, indicating that amplitude activity in both cortical areas electrodes decreased with growing age in both groups of infants, except for activity in Pz for the full-term infants which increased significantly with growing age (see Figure 7).
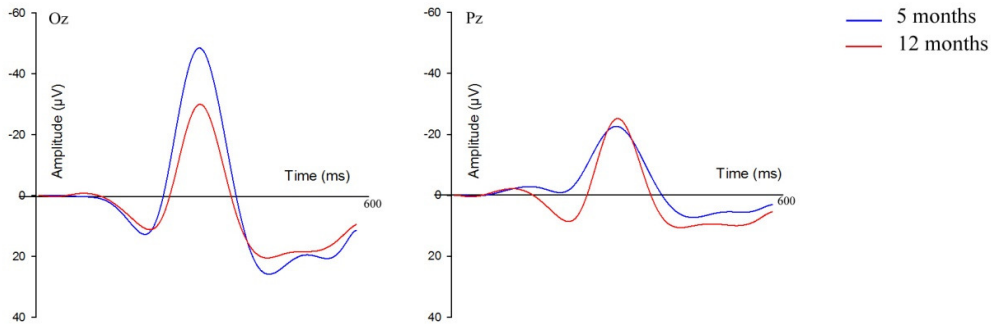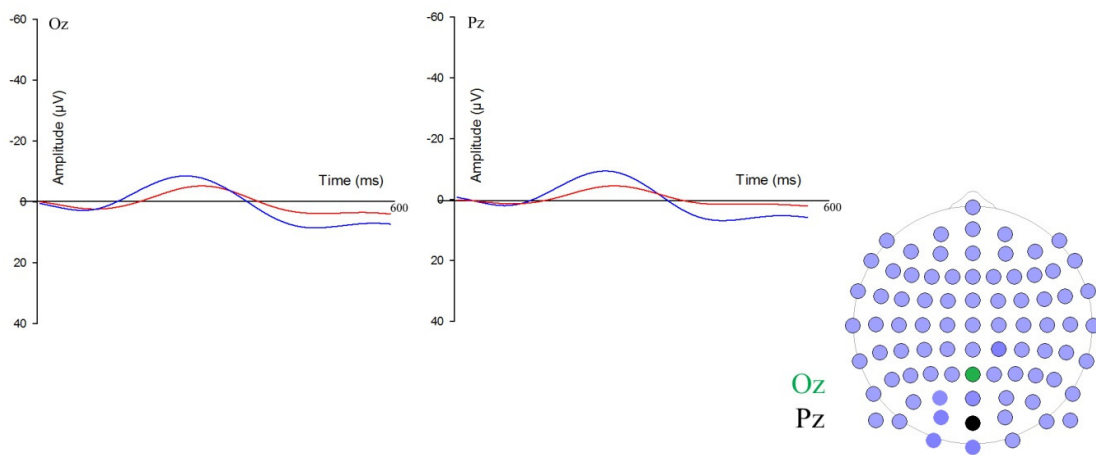
Figure 7: *Grand average looming-related VEP peaks for preterm and full-term infants at the age of 5 months (blue) and 12 months (red) at the two selected posterior electrodes Oz and Pz. The head drawing (nose up) shows the scalp localization of the 81 standard electrodes. The two electrodes of interest are indicated with colored filled circles: Oz (green) and Pz (black). With increasing age the amplitude at Oz and Pz electrodes decreased significantly for both full-term and preterm infants, except at electrode Pz where full-term infants at 12 months showed larger amplitudes than at 5 months.*

4. Discussion

In this longitudinal study, preterm infants at the age of 5 months and 12 months were compared with a group of full-term infants at the age of 5 months and 12 months on a looming task with high-density EEG. VEP analysis was used to investigate whether there are any differences between normally developing full-term infants and preterm infants. VEP responses in the looming sequence, VEP amplitude in Oz and Pz, and duration of looming-related VEP peaks were compared separately for full-term and preterm infants.

4.1 VEP responses

By investigating the looming-related VEP peaks, a significant improvement in VEP responses was observed in the development of full-term infants with growing age. Across loom speeds, full-term infants at the age of 5 months responded on average -1.77 s before virtual contact and with growing age they responded significantly closer to contact, on average at -0.430 s. This result supports earlier findings of a longitudinal study which compared full-term infants at 5-6 and 12-13 months with the same experimental paradigm (van der Meer et al., 2012).

Results further indicated that preterm infants at the age of 5 months showed their looming-related responses on average -1.61 s before virtual contact and with growing age they responded on average -1.12 s before contact. Preterm and full-term infants at the age of 5 months showed their looming-related brain responses early in the looming sequence when the looming object was still far from their faces. Preterm infants at 12 months of age responded at smaller values of time-to-contact, but they still showed their looming-related responses significantly earlier in the looming phase as compared to the full-term infants at 12 months.

One contributing factor that is possibly associated with this observed improvement is ongoing neurobiological development such as maturation of brain pathways and further processing of visual information (Johnson, 2000) that allows for efficient information processing as infants get older. Although most of the brain development occurs in utero, rapid cortical synapse formation and synapse overproduction occur for the first two postnatal years in most cortical areas (Casey, Giedd, & Thomas, 2000). For adaptation and plasticity of the brain, overproduction of synapses is important during maturation. Synaptic maturation increases through glucose metabolic levels which allow readily available neuronal energy for effective information transmission (Chugani, Müller, & Chugani, 1996).

Full-term infants at the age of 12 months showed their averaged looming-related responses at a fixed time-to-contact, irrespective of loom speed. On the other hand, preterm infants at the age of 12 months responded later in the looming sequence (i.e., closer to contact) with increasing loom speeds. These findings suggest that full-term infants at the age of 12 months had switched to a strategy based on time, whereas preterm infants at the age of 12 months still appeared to be using a strategy based on visual angle. According to Kayed and Van der Meer (2000, 2007), the time-strategy is considered to be a more efficient strategy that is less prone to errors. Full-term infants at 12 months of age showed their brain responses when the looming object reached a specific time before the optical collision, thereby allowing themselves the same amount of time to make a response independent of the loom's speed.

Prematurity poses incomplete brain development and impairments in cerebral development. Inder et al. (2005) identified important volumetric changes in the brain structures of preterm infants, in the grey matter of cortex and in the cerebral white matter, with an increase in the cerebrospinal fluid volume. The fact that prematurity always affects the white matter, even when the morphologic MRI findings appear to be normal, is demonstrated by studies using

diffusion tensor imaging (Counsell et al., 2008). A MRI study analyzing the regional brain volume in preterm infants compared to full-term infants found a decreased grey matter density in all cortical regions but the inferior occipital gray matter volumes are highly reduced in preterm infants (Peterson et al., 2003).

Atkinson and Braddick (2007) report on a number of studies that investigated the visual and visuo-cognitive development of preterm children. The studies show that three cognitive areas in particular are affected: selective attention, spatial function, and executive control. These all have links to the dorsal visual system. Motion perception is processed by the dorsal pathway (Braddick, 2003) and it is possible that this pathway is more vulnerable in preterm infants than the ventral pathway (Atkinson & Braddick, 2007) even in the absence of brain damage (Guzzetta et al., 2009). This has been shown previously as orientation-specific visual event related potential (VERP) in preterm infants. Preterm infants performed poorer in visual cortical tests and these infants are at high risk of neurodevelopmental delay at the age of 2 years (Atkinson et al., 2008).

Another factor could be an inadequate DHA, a long-chain of polyunsaturated fatty acid that increases in the brain and retina, especially during the last three months of pregnancy and the early postnatal months. Infants born prematurely are generally deprived of this essential fatty acid compared with full-term infants, which has been directly linked to cognitive development, visual attention, and visual recognition memory (Carlson & Werkman, 1996; O'Connor et al., 2001). According to MRI studies, M cells need a high amount of polyunsaturated fatty acids to preserve their membrane flexibility crucial for allowing physiological processes, which underlie M cells' transient sensitivity important for detecting motion (Stein, 2001). Thus damaged or inefficient M cells in the magnocellular pathway and dorsal stream may affect processing and functions within and related to these pathways.

4.2 VEP duration

In this study, full-term infants at the age of 5 months showed VEP responses not only earlier in the looming sequence, but also of a longer duration in Oz electrode compared to when they were 12 months old. This finding corresponds with the earlier finding of a longitudinal study which compared full-term infants at 5-6 and 12-13 months with the same experimental paradigm (van der Meer et al., 2012). In addition, preterm infants at the age of 5 months also showed VEP responses in Oz electrode of a longer duration compared to when they were 12 months old.

Shorter duration of looming-related VEP peaks with growing age in full-term and preterm infants could indicate an increase in speed of the processing of visual information (Dubois et al., 2005; Johnson, 2000; Webb et al., 2005). According to Johnson (2000), the infant brain during the first year of life undergoes rapid changes and also doubles in weight, suggesting that infants at the age of 5 months have less developed brains compared to at the age of 12 months. In our findings, shorter VEP duration with growing age could indicate a developmental trend.

A major contributing factor to this observed marked improvement could be myelin sheaths over the axons which serve to increase the speed of electrical impulse propagation from soma to dendrites of the neurons (Webb et al., 2005). An MRI study on preterm infants reported that myelination in most structures of the brain increased with increasing gestational age, and it is visible on MRI images within 4 weeks of gestational age (Counsell et al., 2002). During early maturational stages until adulthood, white matter fibers increase in volume and density due to the ongoing axonal myelination. Thus, myelination could affect the visual processing speed with age (Webb et al., 2005). Shorter duration of looming-related VEP peaks with

growing age in full-term and preterm infants could indicate that as infants get older they use less time to process the looms.

4.3 VEP amplitudes in Oz and Pz

In this study Oz (occipital) and Pz (parietal) electrodes showed the looming-related response in both infant groups. Full-term and preterm infants at the age of 5 months showed larger negative amplitude in occipital area as compared to when they were 12 months old. Furthermore, full-term infants at the age of 12 months showed larger negative brain activity in the parietal area than at the age 5 months, but preterm infants did not show increased brain activity in the Pz electrode with age. The VEP amplitude represents the number of synchronously excited neurons (Pfurtscheller & Lopes da Silva, 1999). So, it can be argued that higher amplitude means more synchronous signals elicited by excited neurons in a larger area.

Higher amplitude at the age of 5 months compared to at the age of 12 months for both full-term and preterm infants could be the result of an increase in skull thickness, because infant skull thickness increases most rapidly within the first year of life (Grieve et al., 2003). Despite of this, full-term infants showed increased negative brain activity in the Pz electrode at the age of 12 months.

Activation of neurons in dorsal visual pathways while processing motion information from V1 via MT area to the parietal cortex is observed in infants in response to motion stimuli (Braddick, Atkinson, & Wattam-Bell, 2003). It also corresponds with an earlier finding in adults, in which processing of motion stimuli is propagated from the dorsal stream towards the MT+ area (Holliday & Meese, 2005; Morrone et al., 2000; Shirai & Yamaguchi, 2004). Propagation towards parietal areas means that the processing of motion stimuli starts to influence more advanced areas of the brain such as parietal and temporal areas (Webb, 2005).

In addition, larger negative brain activity in the parietal area could be due to propagation of the processing of motion stimuli from the occipital towards the parietal area (van der Meer et al., 2012). However, in our findings preterm infants did not show increased brain activity in the Pz electrode with age, whereas full-term infants did. So, it could be argued that looming-related brain activity in preterm infants did not propagate towards more advanced areas of the brain as they got older.

In conclusion, the results from this study showed that with growing age full-term infants responded to the looming visual stimuli at a relatively constant time-to-collision close to virtual contact, while showing shorter duration of looming-related VEPs and propagation of VEP activation towards higher information processing areas in the visual pathway. On the other hand, preterm infants at 12 months did not time their looming-related brain activity as close to contact as full-term infants did and they varied the timing of their responses with loom speed. Also, preterm infants did not show propagation of VEP activation towards higher information processing areas in the visual pathway even though they did show shorter VEP duration for looming-related responses with growing age. This could be caused by impairments in cerebral development such as neuronal loss or impaired neuronal differentiation with a reduction in dendritic and axonal development, or an inadequate amount of DHA, a long-chain of polyunsaturated fatty acid.

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