Development of anticipatory gaze in low risk premature infants born before 32nd gestational week.

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Short title: gaze in premature infants

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Key words: smooth pursuit, preterm infants, prematurity, visual development, infant brain

ABSTRACT

Aim: to investigate the impact of premature birth on visual functioning in a group of 37 infants, born in 27 to 32 we and diagnosed as being without pathological complications. **Methods**: Eye-and head movements were measured in a tracking situation at 2 and 4 months of corrected age, in accordance with our earlier studies of fullterms (1,2). Accuracy of gaze, proportion of smooth eye movements, head movements and saccades were calculated.

Results: Between 2 and 4 months of age both fullterms and premature infants improved their ability to smoothly anticipate a moving object. However, at both occasions, the preterms had less proportion smooth pursuit than the fullterms. Furthermore, the smooth eye movements lag relative to the object motion was higher in the preterm group. The groups did not differ with respect to gaze, head movements or saccades.

Conclusion: In early infancy, the developmental delay in the preterms was around 2 months. Postnatal experience did not influence the ability- (This paper is a part of the LOVIS study).

Key words: smooth pursuit, premature infant, saccades, infant development, vision

Introduction:

The visual system is the single sensory system that is not exposed to patterned stimuli prenatally. At birth, however, the infant's visual system is comparably mature (3). Newborn human infants fixate both stationary and moving objects and are attracted by facial stimuli (4). The ability to perceive directed motion and track small moving objects with smooth pursuit (SP) eye movements develops rapidly from about 6 weeks of age to adult-like performance at around 12-16 weeks of age. During the same period, the cortical activation of the temporal-occipital-parietal junction (corresponding to MT-MST) increases dramatically (5). This area is crucial for motion perception. At 4 months of age the visual system functions rather similar to that in adults in many respects. What are the prerequisites for this early and extensive maturation? One main reason is the comprehensive cellular maturation patterns that take place prenatally from week 28 to 40, continuing after birth (6,7). Huttenlocher (8) reported histological data of synaptogenesis in area 17 in the human fetus. Synaptic profiles were demonstrated at 28 gestational weeks , and from that time they increased 3 times until birth. The second significant increase took place between 2 and 4 months postnatally, coinciding with the maturation of lateral geniculate pathways (9). Similar results were found by Sauer et al (10). They emphasize that the visual cortex has the highest growth rate as compared to the cerebrum and cerebellum, resulting in a 50% increase of the volume of v17 area at birth. Becker et al (11) examined dendritic development in layers 3 and 5 of the visual cortex and found that branching in layer 5 developed earlier than in layer 3, and the increase was most marked between weeks 36 and 40 weeks gestation. (layer 5 max at 4 month postnatal age). At around birth the glial proliferation peaks (12). The growth rate is influenced by several agents. One intrauterine factor is the IGF factors that have been shown to have a strong impact, as promoting synaptogenesis and stimulating myelin production (13,14,15,16). Premature birth at this time, before 32 we gestational age (GA), will interrupt the exposure to growth factors . Furthermore, hypoxia induced a decrease in the synaptogenesis after premature birth, (17). Thus, the vulnerability of the rapidly maturing visual system may result in a slowing down of oligodendroglia differentiating and myelination, as a consequence of premature birth. For example, Shah et al (18) concluded from MR examinations of premature infants of GA 24 to 33 weeks that the occipital lobe was reduced at term. Another study by Limperopoulus et al (19) demonstrated that the late cerebellar growth decreased by premature birth. Reduced cerebellar volumes in late childhood of preterms has been shown elsewhere (xxxx).

Bassi et al (20) performed diffusion tractography in a group of preterms and found a correlation between white matter microstructure in the optical radiations and visual function: the impairment increased with the degree of focal lesions.

Taken together, the cited studies show that the premature change from intrauterine environment in infants without neurological sequelae give rise to extensive anatomical impairments that have considerable neuroanatomical and possible neurofunctional consequencies.

Considering the rapid prenatal growth of essential parts of the visual system, and thus its vulnerability, it is important to measure the visual function of premature infant's early in life (21,22,23). The ability to track a small moving target in an anticipatory way is well studied in term infants (1,2), in adults (see eg, 24), and in monkeys (eg 25). An extensive neural network is involved in such an oculomotor task: frontal eye field, posterior parietal cortex, MT+ area, DLFPC, basal ganglia, thalamus, superior colliculus and cerebellum. In a recent review, Luna et al (26) describes the system as "a neuroscientific tool to examine relationship between brain and behavior". Furthermore, eye movements are critical for keeping a tight coupling between perception and action. As the oculomotor system is an action system that functions very early in life its control must develop early as well. Disturbances or lesions affecting the oculomotor neural network may on one hand indicate more severe impairment and on the other a less precise neural control of the eye movements. In adults the primary visual pathway for motion stimuli passes the V1 area to the MT and MST areas for further processing in the dorsal stream. A parallel processing may take place (27) when the visual occipital areas are bypassed. It has been shown by EEG (5) and concluded from behavior data (23, 28) that this bypass is functioning until 2 months of age. At 4 months of age the primary visual pathway V1-MT/MST is maturing rapidly.

It is concluded that the ability to smoothly anticipate a moving object may be a sensitive measure of a premature infant's developmental status and of the functioning of the visual system. Then, the proportion of smooth pursuit of a moving object (1) at 2 and 4 months of age respectively can be compared between full terms (FT) and very prematures (VPT). In fullterm infants a rather consistant increase in smooth pursuit accuracy ("gain") is found between 2 and 4 months of age. At 5 -6 months of age it is close to that in adults.

In the present study we measured eye-and head tracking in a group of very preterm infants at two occasions, 2 and 4 months corrected age. Only infants without diagnosed IVH, SGA, BPD or ROP were included (see below). The group was a low-risk subgroup of the LOVIS group, that consists of the major part of infants born < we 32 in Uppsala 2004-2007 (referens till posters?). The main purpose was to assess the degree of SP between 2 and 4 months of age in the FT and VPT groups, and to compare these data with the clinical outcome. The design of the study was similar to that presented earlier (1). The infant was presented with a small moving object that moved according to a sinusoidal motion pattern or with constant velocity and with two amplitudes, resulting in 4 conditions.

Methods:

Subjects: The selection of subjects has been described elsewhere (ref). Briefly, parents were contacted during their stay at the Neonatal unit at Uppsala Academic Hospital, and asked to participate in this longitudinal study ("LOVIS"). The study was approved by the Ethics committes at the Uppsala University.

In the present study the parents were contacted at around the time of 2 and 4 months corrected age respectively. Totally 44 infants were selected, based on the criteria of lack of ROP, BPD, IVH, PVL, and SGA. ROP was diagnosed by inspection, IVH and PVL by ultrasound. There was no NEC in the group. Neurological and ophtalmological follow-up of this group has been described elsewhere (ref). Seven infants were excluded due to technical problems or fuzziness, and in the remaining group were 17 girls and 20 boys. The average birth weight of the female group was 1412 g (SD=55), and in the male group 1388 g (sd= 64). The average GA was 29.7 (range 27.3 to 31.9) in the female and 29.7 (range 27 to 31.9) in the male group respectively. In the control groups, parents were contacted via the State Tax Authority System. Thirt-five infants were contacted, (boys-girls) and 3 were excluded due to inattention and technical problems.

Procedure: Upon arrival to the lab (Dep. of Psychology, Uppsala University) the parent was informed of the study and the procedure, and signed a consent form in accordance with the Helsinki Declaration 1964. For participation, the parent received a gift cerificate to a local toy store or to a bookshop, 10€. The VPT infants were seen at average ages 9.8 and 19.2 weeks corrected age, and the FT infants at 8.4 and 17.9 weeks respectively. Below, the two occasions are called 2 and 4 months respectively.

The measurements were as described earlier (1). For motion measurement of the object and the head of the subject, a Proreflex system (Qualisys,Gothenburg, Sweden) was used with passive markers, 3 on the skull and 1 on the object. The system was sampling at 240 Hz in synchrony with the horizontal eye movements measurements (electrooculographics, EOG). Two skin electrodes (Beckman) were attached to the outer canthi, and a ground on the forehead. (See Figure 1). The infant was seated in a special infant chair that gave support to the body while permitting arm-and leg movements, and head movements as well. In front of the infant (50 cm) a small object (a happy face, 8° visual angle) attached to a small video camera could be moved horizontally. Its motion was realized by a motor, controlled so that two amplitudes and two motion types (triangular and sinusoid) at 0.25 Hz were obtained. The amplitudes were approximately 10 and 20 degrees in each direction respectively. Each trial was 20 s: the first one was for calibration of the EOG, then the trials of 2 amplitudes and motion types followed in random order. After these 5 trials further trials were run, focused on the functioning of the vestibular and vestibular-ocular responses, and the ability to represent motion behind occluder. However the data from these trials were not included in the present paper.

Data analysis: The data for the head , the object and the eye was transferred to rotation angles (1). The data analysis of eye calibration and tracking was performed in FYSTAT (Umeå University) or MATLAB (Mathworks Inc) environment. Statistics was performed in SPSS.

Tracking characteristics

The tracking measures used to assess the quality of the VPT infants visual functions were Proportion of smooth pursuit (PRSP), Saccades per second, Head gain and Gaze gain. Gain was calculated with Fourier analysis (FFT). The PRSP was the ratio of smooth pursuit gain and raw eye movement gain. The number of saccades was estimated from the velocity profile of the raw eye movement data. As found in earlier studies using the present method, a threshold of 40 deg/s was then chosen. The difference between object and head was calculated defined as head slip. Timing between the head slip and the eye was estimated by cross-correlation.

Statistical analysis

Multiple regression

37 VPT infants and 32 FT infants were included in the statistical analysis. 21 of the VPT and 19 of the FT were measured at both 2 and 4 months of age. 5 VPT and 9 FT at were measured at 2 months only, and 11 VPT and 3 FT at 4 months.

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The outcome was the various tracking variables measured during the experiment for infant *i* at time *t*. The model investigates the relationship between the 2 (corrected) age groups(*Age)* , between VPT and FT (*Group*), and interaction effects between *Group* and *Age.*

Results:

General: Most VPT infants were interested and attentive during the trials. As every trial lasted 20 s, the measurements were completed in less than 2 minutes. The set up worked well and the observing parent could observe their child during the whole session. In figure 2 examples of an VPT infant (born we??) eye-and head tracking are shown. It is observed that the eyes track the object with mainly saccades at this age. (mera)

Gaze tracking

The gain of gaze did not differ between the VPT and the control children. In one case there is an interaction between VPT/FT and age (Table 1). In the sinus motion condition with large amplitude, the VPT group has a gain that is lower than the FT group at 2 months. At four months of age, the VPT group has caught up. A closer inspection of the gaze gain shows that the standard deviation in the VPT group is significantly larger (M= 0.25) than in the FT group (M=0.18), $t(14)$= 2.837, $p<0.05$.

Smooth pursuit tracking

The VPT had less proportion of smooth pursuit compared to FT group for all 4 experimental conditions. At 4 months the infants had more PRSP than at 2 months. In one condition, the triangular with small amplitude, an interaction effect between the two was found. The VPT PRSP increased more between 2 and 4 months compared to the FT. (see table 2 & figure 3)

Saccadic tracking

The VPT group had significantly more saccades than the FT group when they were looking at the sinus motion with small amplitude (table 3). The same trend was shown for the triangular motion with large amplitude. No difference was obtained between 2 and 4 months of age in any group.

Head tracking

No significant difference was found between the VPT and FT groups concerning how much they moved their heads during tracking. However, for all 4 motion types head gain was larger at 4 months than at 2 months (table 4).

Timing

The cross correlation between SP and head slip shows that the lag decreases between 2 and 4 months for the sinusoidal motion patterns. Furthermore, the timing for the FT group is more precise than for the VPT group. (figure 4)

Discussion

In the present study the velocity 0.25 Hz was chosen. Our earlier studies indicate that this velocity is fast enough to keep the attention level high, and low enough to allow tracking. Quite a few VPT infants, however, failed to look attentively at the age of 2 months.

The main result was that the VPT infants produced less accurate smooth pursuit than the FT. The smooth pursuit requires sensitivity for motion direction. Atkinson et al (21) and Birtles et al (22) showed that this is less in preterms of similar GA and age.

One question in studies of premature infant's abilities is whether there is an effect of experience, or if the development proceeds according to a conceptual age schedule. For visual stimuli, experience may influence development: Bourgeois et al (29) examined synaptogenesis in the visual cortex in Rhesus monkeys that were 3 weeks preterm and exposed to light from birth. At 1 month of age the size and type of the synapses were significantly different from the controls. It was, however, not reported to what extent these changes influenced function. Weinacht (30) estimated visual acuity and binocular vision in prematures (born we 33) at 4 and 14 weeks corrected age. They did not find any increase in ability that could be related to experience. Hunnius et al (31) showed that gaze shifting in a competitive and a non competitive task respectively favored VPT infants to FT. The VPT group was very similar to the present one ; the authors concluded that the VPT infants hade more experience. This conclusion does not seem to be valid for the present study. However, the moving stimuli (the happy face) is different from the stationary objects in Hunnius et al study. This could indicate that the dorsal pathway (moving stimuli) is more vulnerable than the ventral (static stimuli)(31). This conclusion is supported by data from Atkinson et al (21) who found that the orientation VEP in preterm's (28 we) younger than 12 months of age is less pronounced than in controls. Furthermore, Birtles et al (22) found that at 2 and 4 months of age the motion direction sensitivity is smaller in the preterm group.

Although none of the VPT infants were diagnosed as ROP, it cannot be ruled out that other types of visual dysfunctions were present that influenced motion tracking. Such ophtalmological complications have been found up to 10 years of age in preterm infants (32)

At birth FT infants have a disposition for perceiving biological motion, (33) which is composed by sinusoidal components. In an earlier study of the development of smooth pursuit of an object moving according to a sinus or a triangular (constant velocity) pattern, (1) the lag at 2 months of age was smaller for the former than the latter. At 5 months, however, the lag had decreased further. The conclusion was that at the younger age a velocity-based extrapolation process gave better prediction of the sinusoidal object motion-the constant velocity gave no clues. However, its periodicity could facilitate a prediction. The VPT infants had greater lags than the FT infants.

The current findings of differences between VPT and FT infants concerning PRSP and its time lag may be interpreted as follows.

The fact that the gain of SP was acceptable, although lower, at 2 months of corrected age but significantly lower than in the controls at 4 months confirm earlier results by Dubowitz et al (23). In their study of visual function in preterms, the subcortical pathway was suggested to dominate up to 48 weeks GA, based on VEP. Here we also obtain differences at 2 months. Although the VPT group had no focal type of lesions affecting visual pathways no diagnosis concerning the more diffuse damage could be conducted, as MRI was not available. However, we conclude that if the SP gain in VPT is less than in FT at 2 months of age the V1 area might be without impairment, and more probably, the subcortical pathway could be suboptimally functioning. At 4 months of age the LGN-V1---MT/MST pathway is functioning less well due to delayed cellular maturation, or diffuse PVL. For a group of premature infants at young age, Mercuri et al (34) conclude that lesions involving basal ganglia will have more severe effects on visual functioning than occipital lobe lesions.

A less well functioning smooth pursuit is compensated with head movements and saccades so that the gaze will target the moving object. However, the ability to predict the object motion is severely impaired. What are the possible consequences? One may be impaired …Further studies of SP at 10 months of age are in progress.

In the near future, the presented data will be included in a review of the total LOVIS premature group study. This will include clinical data as well as MRI at 2-4 years of age (??et al xxxx). However, a decreased smooth pursuit has been shown to remain at school age (35).

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Abbreviation list

Figure captions

Figure 1: set up

Figure 2: Examples of registrations of eye-and head movements in an infant, born xx we at an age of 9 weeks (corrected).

Figure 3: Proportion smooth pursuit in the PT and VPT groups

Figure 4 :Timing between smooth pursuit and head slip in the PT and VPT groups

	(1)	(2)	(3)	(4)	(5)
			Outcome: Gaze		
	Sinus Small	Sinus Large	Triangul. Small	Triangul. Large	Mean
Group	0.059	-0.195***	0.034	-0.038	-0.094
	(0.062)	(0.066)	(0.083)	(0.065)	(0.056)
Age	0.127*	0.112	0.073	0.000	0.064*
	(0.067)	(0.071)	(0.086)	(0.065)	(0.059)
Group*Age	-0.048	0.161*	-0.184	0.055	0.052
	(0.090)	(0.093)	(0.117)	(0.090)	0.079
Observations	86	95	88	85	104
# Infants	58	61	57	55	64
R-square	0.075	0.225	0.043	0.010	0.077

Note: The dependent variable is gain of gaze for the four different movements that was presented for the children. Group is whether the children are in the VPT or FT group. Age is if the child is measured at 2 or 4 months of age. Standard errors in parentheses, * significant at 10%; ** significant at 5%; *** significant at 1%

	(1)	(2)	(3)	(4)	(5)
			Outcome: proportion smooth pursuit		
	Sinus Small	Sinus Large	Triangul. Small	Triangul. Large	Mean
Group	-0.254***	-0.188***	-0.311***	-0.279***	-0.222***
	(0.056)	(0.061)	(0.061)	(0.060)	(0.048)
Age	0.189***	0.216***	0.123*	0.183***	0.189***
	(0.062)	(0.066)	(0.063)	(0.060)	(0.051)
Group*Age	0.087	0.018	0.271***	0.088	0.089
	(0.084)	(0.088)	(0.087)	(0.083)	(0.068)
Observations	88	94	92	89	106
# Infants	60	61	61	58	66
R-square	0.387	0.307	0.405	0.408	0.412

Note: The dependent variable is proportion of Smooth Pursuit for the four different movements that was presented for the children. Group is whether the children are in the VPT or FT group. Age is if the child is measured at 2 or 4 months of age. Standard errors in parentheses, * significant at 10%; ** significant at 5%; *** significant at 1%

	(1)	(3)	(5)	(7)	(9)
			Outcome: Saccades		
	Sinus Small	Sinus Large	Triangul. Small	Triangul. Large	Mean
Group	0.405**	0.019	0.294	0.445*	0.227
	(0.199)	(0.179)	(0.251)	(0.246)	(0.176)
Age	-0.150	-0.114	-0.078	0.032	-0.014
	(0.221)	(0.188)	(0.225)	(0.249)	(0.182)
Group*Age	-0.169	0.044	-0.334	-0.505	-0.183
	(0.285)	(0.244)	(0.340)	(0.330)	(0.240)
Observations	80	87	81	77	95
# Infants	56	59	59	55	63
R-square	0.094	0.008	0.046	0.073	0.028

Note: The dependent variable is saccades for the four different movements that was presented for the children. Group is whether the children are in the VPT or FT group. Age is if the child is measured at 2 or 4 months of age. Standard errors in parentheses, * significant at 10%; ** significant at 5%; *** significant at 1%

	(1)	(2)	(3)	(4)	(5)
			Outcome: Head gain		
	Sinus Small	Sinus Large	Triangul. Small	Triangul. Large	Mean
Group	0.008	-0.012	-0.026	0.001	-0.022
	(0.052)	(0.059)	(0.069)	(0.066)	(0.051)
Age	0.162***	0.195***	0.156**	0.236***	0.202***
	(0.057)	(0.063)	(0.071)	(0.066)	(0.53)
Group*Age	-0.024	-0.031	-0.105	-0.115	-0.078
	(0.077)	(0.083)	(0.097)	(0.091)	(0.072)
Observations	86	94	90	88	104
# Infants	57	61	59	58	65
R-square	0.155	0.174	0.080	0.174	0.186

Note: The dependent variable is head gain for the four different movements that was presented for the children. Group is whether the children are in the VPT or FT group. Age is if the child is measured at 2 or 4 months of age. Standard errors in parentheses, * significant at 10%; ** significant at 5%; *** significant at 1%

	(1)	(2)	(3)	(4)	(5)
	Outcome: Cross correlation to head slip				
	Sinus Small	Sinus Large	Triangul. Small	Triangul. Large	Mean
Group	-122.68**	-186.43**	-121.27	-70.49	
	(56.99)	(85.33)	(79.58)	(67.80)	
Age	141.80**	260.49***	83.95	115.02	
	(61.38)	(90.21)	(80.76)	(69.52)	
Group*Age	-12.34	-8.04	-107.41	-55.07	
	(82.97)	(120.06)	(112.30)	(96.53)	
Observations	83	88	83	81	
# Infants	56	59	54	53	
R-square	0.188	0.232	0.119	0.089	

Note: The dependent variable is proportion of Smooth Pursuit for the four different movements that was presented for the children. Group is whether the children are in the VPT or FT group. Age is if the child is measured at 2 or 4 months of age. Standard errors in parentheses, * significant at 10%; ** significant at 5%; *** significant at 1%

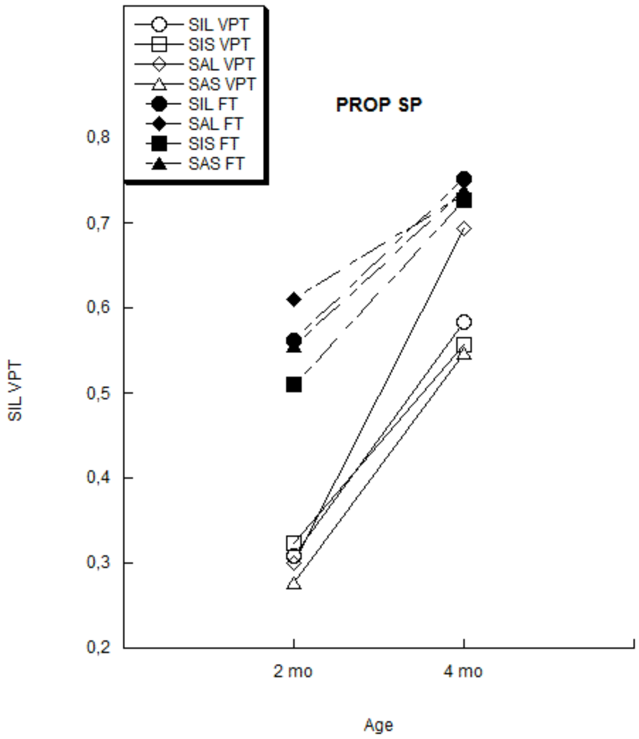

Figure: 3. Means for PRSP

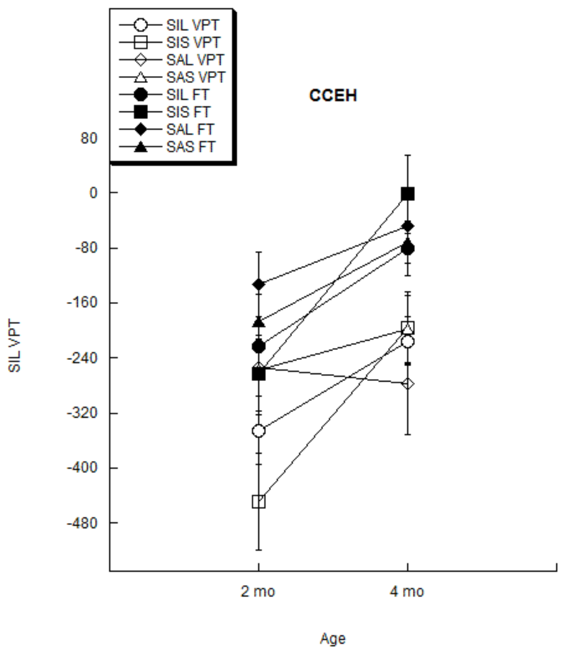

Figure 4: Mean and SEM of velocity time shifts between the smooth pursuit and head slip for the different motion functions and amplitudes at the different age levels1