NEURODEVELOPMENTAL RETARDATION, AS ASSESSED WITH MAGNETOENCEPHALOGRAPHY AND ELECTROENCEPHALOGRAPHY, ASSOCIATED WITH PERINATAL DIOXIN EXPOSURE

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¶ in memoriam: Henk Spekreijse passed away in 2006.

Introduction

Perinatal exposure to Dutch "background" dioxin levels was rather high in the years 1987-1991, as was the case in Western Europe in general. In retrospect many health disturbances have been noted following perinatal exposure during this period. Effects were seen on thyroid function[1], lung function[2], puberty[3], but also on neuromotor development[4]. We therefore hypothesized that exposure during the sensitive perinatal period may also result in permanent damage to the brain.

Neurodevelopment

During the first trimester of pregnancy all neurons in the brain are formed. During the second and third trimester, especially around thirty weeks of gestational age, the growth and development of the brain takes place, characterised by the forming of dendrites, connecting the neurons, synapses and by the start of glial myelinisation. Structures in the brain necessary to process visual and auditory signals, for instance for language development, are then formed. This process proceeds, albeit somewhat slower than prenatally, during the first year of life, and still slower thereafter, up until a dolescence.

Amsterdam-Zaandam study

In 1987 in the Amsterdam-Zaandam region a longitudinal study on the effects of background exposure to dioxins was started. The dioxin exposure was measured in breastmilk shortly after birth. Out of a group of 120 mothers and their children 44 mothers intended to breastfeed for at least two months and these mother-baby pairs were included in the study. The concentrations of dioxins in breastmilk was used as a measure for the prenatal exposure to dioxins. Lactational exposure was calculated as the concentration measured in breastmilk multiplied by the amount of breastmilk the baby consumed during the period of breastfeeding. In this study only the 17 dioxins (PCDDs) and furans (PCDFs) that accumulate in man were measured, and not the PCBs. Levels in breastmilk ranged from 8.74 to 88.8 ng TEQ dioxin (PCDD + PCDF)/kg milkfat. In the group of 44 breastfed children signs of enhanced neuromotor maturation were found at the age of 2½ years in relation to prenatal dioxin exposure. It was hypothesised that this may be due to the thyroxine-agonistic action of dioxins[4].

Materials and methods

In order to assess the neurophysiological development of our Amsterdam-Zaandam cohort, born during the period 1987-1991, we made use of magnetoencephalography (MEG) and electroencephalography (EEG) and standardized intelligence and behavioural questionnaires.

MEG and EEG

Magnetoencephalography (MEG) was at the time of testing a new non-invasive technique of brain function

imaging. With a time resolution of milliseconds and spatial resolution of a few millimetres, it provides objective information about neurological functioning of the human brain. MEG mapping is performed by recording minuscule changes in the magnetic field just outside the head using many dozens of magnetic sensors. The changes in the magnetic field are due to changes of current densities effected by strong coherent neuronal activity. In contrast to electroencephalography (EEG), MEG is not hampered by volume conduction of the various cerebral tissues or the electrical impedance of the skull. This generally results in a better spatial resolution of MEG as compared to EEG. In addition to MEG recordings, we simultaneously recorded the EEG using six leads. This had a two-fold purpose: to facilitate the interpretation of the MEG data by comparing them to the data obtained with the more familiar EEG technique, and in many conditions (sensory evoked responses, epileptic activity) both techniques have proved to be supplementary rather than complementary. The latter is dependent on the strength, position and current-direction of the source(s) which account for the activity. A source with a current perpendicular to the skull surface is difficult to detect with the MEG technique, but a source with a current parallel to the skull can easily be detected.

It is generally known that spontaneous brain activity is affected by neurological disorders, such as tumors, brain hemorrhages, encephalopathies, many drugs and toxins and even hypoxia, hyperoxia and hypercapnia. Alpha activity is, in healthy subjects of all ages, the most prominent spontaneous activity. This is generally so, however healthy subjects may lack an alpha rhythm. Since the alpha frequency of the spectral alpha peak is indicative of resting brain activity, a quantitative analysis of the alpha frequency and amplitude was performed to search for a possible dioxin related effect.

The performance of the peripheral pathways and the primary cortical processing can be established to quantify the elicited responses to sensory stimulation. In humans the most prominent sensory system, in terms of cortical involvement, is the visual system. Dominance in the processing of visual stimuli lies with the processing of moving visual stimuli, or motion stimuli. The N2 components (comprising N2a and N2b) are the specific components seen after a motion stimulus.

Cognitive functioning of the brain was assessed by analyzing the evoked responses, obtained by applying a visual oddball paradigm. In such a set-up there is a continuous presentation of a visual stimulus, which is infrequently and at random replaced by a deviating stimulus, the oddball. The subject has some task dictated by the appearance of each oddball. We hypothesized a reduced cognitive function as a result of perinatal dioxin exposure. By identifying and evaluating the individual components of the response, measurements of their latency time and amplitude can be made. Visual oddball activity have been strongly associated with the occurrence of the N200 (after approximately 200 ms) and P300 (latency approximately 300 ms) response components, the latter being further subdivided in P3a and P3b. In the EEG responses, the N200 is an occipital negative component, the P3a (not quantified) is a broadly distributed fronto-central positivity and the P3b is a more parietal positive component. N200 has been associated with the deviating appearance of the oddball with respect to the frequent (common) stimulus, and P3b has been associated with the task.

Resuming, the visual stimulus (here the motion stimulus) inherently tests the sensory function of the brain, and the oddball stimulus indicates the task performance capability of the cerebrum. We chose to evaluate the above mentioned components due to their high specificity for brain activity in sensory, and visual task performance, and because of extensive experience in this field, thereby giving a reliable evaluation of cerebral functioning. The technical approach has been previously published[5].

Results and discussion

The Amsterdam-Zaandam study group are 38 children (age 7-12 years) selected because of an optimal pregnancy and all breast[1]. A statistical correction was made for the numbers of (sub)tests performed, in order to correct for chance findings. This was done according to the formula: $n\alpha = 0.05$, where n is the number of tests and α the level of significance (p value).

The standardised Teacher Report Form for children 4-18 years of age revealed a significant increase in social problems ($p<0.001$, slope 0.03± SE 0.006 unit versus ng) and aggressive behaviour ($p=0.001$ slope 0.03±0.01) in relation to postnatal dioxin exposure (corrected significance level $p=0.005$), and an increase in thought problems ($p=0.005$ slope 0.009±0.003). The postnatal exposure also showed a significant increase in relation to the total ($p=0.003$ slope 0.12±0.04) and total externalisation ($p=0.002$ slope 0.04±0.01) scores. The outcomes were corrected for maternal educational status. The teachers were blinded to the dioxin exposures of the children and to the outcomes of the previous studies.

The standardised Child Behaviour Check List for children 4-18 years of age revealed a significant increase in social problems in relation to both prenatal ($p<0.001$, slope 0.09±0.02 unit versus ng/kg**)** and postnatal ($p=0.001$, 0.02±0.005 unit versus ng) dioxin exposure (significance level $p=0.005$). The prenatal exposure was related to increasing anxious/depressed feelings ($p=0.002$ slope 0.106±0.03) and a tendency towards increasing aggressive behaviour ($p=0.06$) was seen in relation to postnatal exposure. The internalised behaviour ($p=0.007$ slope 0.20±0.07) and total behaviour score ($p=0.016$ slope 0.47±0.19) showed a relational tendency with prenatal exposure. The postnatal exposure showed a tendency towards increasing total externalisation ($p=0.075$) scores. The outcomes were corrected for maternal educational status.

Linear regression showed increased social problems, thought problems and aggressive behavior as reported by teachers, in relation to postnatal dioxin exposure. Increased anxious/depressed feelings as reported by parents was associated with prenatal exposure and increased social problems were seen in relation to both exposures pre- and postnatal. Neuromotor examination showed no abnormalities between higher and lower exposed children. The WISC-R was not significantly different, but behavior was.

The neurophysiological measurements demonstrate that an increasing prenatal exposure resulted in an increased (delay) latency of 13 milliseconds in the N2b EEG component between the higher and lower exposed children. The normal latency is 140 milliseconds, which would indicate an almost 10 % delay in the higher exposed children. A similar delay was found in the odd ball measurements, N200 and P300, together with an amplitude decrease of 12 %[5]. These last two measurements are indicative of cognitive functioning.
In a recent paper Woodley et al. describe a decline in general intelligence estimated from a meta-analysis of the slowing of simple reaction time in the last 100 years. It seems plausible that dioxin exposure in Western Europe has further contributed to that loss.

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