

Warmer ears, warmer brain?

Lateralized tympanic membrane temperature in relation to
asymmetric frontal cortical activity in infants

Applied Neuroscience in Education and Child Studies

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Abstract

Emotions have motivating characteristics: either you approach or withdraw in reaction to emotional stimuli. This approach-withdrawal system has been the focus of emotion studies for several decades now. Asymmetric frontal cortical activity has been widely used in these studies, using electroencephalogram (EEG). Two separate neural circuits are involved in different emotional reactions. The first is involved in emotional reactions that are more positive and elicit goals to approach or engage in a situation, and the second is involved in emotional reactions that are more negative and elicit goals to withdraw from a situation. Recently, another measurement has been hypothesized to be related to emotion, and particularly the approach-withdrawal system, namely tympanic membrane temperature (TMT). TMT would be a powerful instrument to use as an alternative for EEG, particularly in infant research. However it is not yet clear if a relationship between EEG and TMT exist, which is analysed in this study. A relationship between those two measurements means that TMT might be of use in emotion studies. This is of great importance, because TMT is less invasive and can be easily used in research. The study population consisted of 18 infants of 10 months old. The current study used EEG to measure asymmetric frontal cortical activity and TMT was measured using an ear thermometer. The findings suggest that no relationship can be found between TMT and EEG, indicating that TMT does not measure the same thing as EEG. Hence, the hypothesis that TMT is a cheaper and easier replacement for EEG measurements cannot be endorsed in light of present data.

Keywords

Emotions – Approach-Withdrawal – EEG – Asymmetry – Tympanic Membrane Temperature

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Emotions, Frontal Cortical Activity and Tympanic Membrane Temperature

The role of asymmetric frontal cortical activity in emotion has been the subject of interest for several decades. Asymmetric frontal cortical activity has been related to the Approach-Withdrawal system, which states that behaviour is motivated by emotions: a person will either approach or withdraw from certain stimuli. The measure that has been most frequently used to measure the asymmetric frontal cortical activity is the electroencephalogram (EEG). Unfortunately, EEG is expensive, time-consuming, inconvenient for use in real world situations and it requires specialized knowledge. Recently, researchers have been beginning to use another technique in examinations of hemispheric lateralization, namely tympanic membrane temperature (TMT). This measure is less invasive and can be easily used. TMT would be a powerful instrument to use as an alternative for EEG, particularly in infant research. The use of TMT in these examinations is quite new in the field of emotion and frontal cortical activity, hence not every aspect of TMT has been examined. Before TMT can be proposed as an alternative for EEG in emotion studies, a clear relationship between TMT and EEG activity has to be found. The current study will test whether there is a relationship between TMT and asymmetric frontal cortical activity, measured using EEG. Finding a clear predictive relationship between TMT and frontal asymmetry would benefit both research and clinical practice.

Defining Emotion: Object, Affect and Mood

Emotion is a concept that has been difficult to pinpoint and define. Plutchik (as cited in Gazzaniga, Ivry & Mangun, 2002) gives an estimation of at least 90 different definitions of emotion that are generated by psychologists. Evidently, psychologists have not find an agreeable definition for emotion. Gray (2007) defines emotion as “a subjective feeling that is mentally directed toward some object” (p. 213). According to this definition, emotion has two components, namely object and feeling. The object of an emotion (a person, thing or event) is something that is in some way very important to the individual who experiences the emotion. It is perceived as the cause of the emotion. Sometimes the object is one’s self or one’s own behaviour, like shame, embarrassment, pride and guilt. These emotions are called self-conscious emotions.

The second component of emotion, feeling, is referred to as affect by some psychologists (Gray, 2007). Affect can vary along two dimensions. One dimension has to do with the degree of pleasantness (pleasure and displeasure) and the other dimension has to do with the degree of mental and physical arousal (activation and deactivation). The emotion of individual experiences depends on both the object as the feeling. For instance, the feeling of pleasure can be experienced as the emotion pride when the object is the self, but also as the emotion love when the object is someone else.

Sometimes emotional feelings are not attached to objects, but instead, experienced as free-floating (Gray, 2007). In that case, feelings are commonly referred to as moods. Moods can colour all aspects of one's life, behaviour and thought and it can last for hours, days or even longer.

Motivational Character of Emotion and Affective Style

Emotions possess motivating qualities. To motivate is to set in motion. In psychology, motivation is a reference to the entire constellation of factors that cause an individual to act in a specific way at a specific time (Gray, 2007). In 1959 Schneirla proposed the approach-withdrawal continuum or dichotomy, based on the motivating characteristics of emotions. This primary motivational system applies for most behaviour (Schneirla, 1959). Starting from the first few months of life, emotions guide the behaviour of the infant: an infant will either escape or explore people and things in the world. So, two systems are distinguished: the approach system and the withdrawal system. Specific types of positive affect will activate the approach system and this will consequently facilitate approach behaviour (Davidson & Irwin, 1999). The withdrawal system has been described as a system that is involved in negative emotions related to withdrawal and facilitating the withdrawal of an organism from sources of aversive stimulation. Moreover, it organizes appropriate responses to cues of threat, e.g. fear and disgust.

One of the most striking characteristics of emotion is how diverse individuals can be in their emotional reaction or in reaction to emotional stimuli (Gazzaniga, Ivry, Mangun, 2002). Everyone may react negatively when hearing bad news. However, some people may overcome quickly and look at the bright side of the situation, but other people may worry for days and sink into a depression. Thus, individuals can be motivated differently by emotions. This difference in their reaction to emotional stimuli is the state of a person and the difference in their dispositional mood, or temperament, is called the trait of a person (Davidson, 1998). The overall definition for these variations among individuals is affective style.

Brain Mechanisms of Emotion and Motivation

The brain is the centre for experiencing emotions. The main focus of research on the brain's emotional system are the amygdala and the prefrontal cortex (PFC; Gray, 2007; see Figure 1). The amygdala is part of the limbic system, which plays a role in regulating basic drives and emotions (Gray, 2007). The amygdala is a cluster of nuclei buried deep, underneath the cerebral cortex, in the temporal lobe. It receives stimulus input from all sensory systems of the body and it evaluates continuously and rapidly this input. When these assessments suggest that some sort of reaction is called for, the amygdala alerts the brain and body. Information is quickly routed to the amygdala, before it reaches the PFC, which is involved in thinking and reasoning. When information about learned threats is assessed by the amygdala, a cascade of subsequent processing in the amygdala and beyond is initiated (LeDoux,

2012). Specifically, the amygdala triggers behavioural, autonomic and endocrine responses in the effort to cope with the danger. Moreover, a global state of readiness results from projections of the amygdala to the cortex, in which attention, perception, memory retrieval and memory formation are all facilitated. The amygdala is involved in a variety of emotional tasks, ranging from fear conditioning to social responses (Gazzaniga, Ivry, Mangun, 2002). It might be inaccurate to state that the amygdala is only involved in responding to negative or fear stimuli, but it does appear to be particularly sensitive to these types of stimuli.

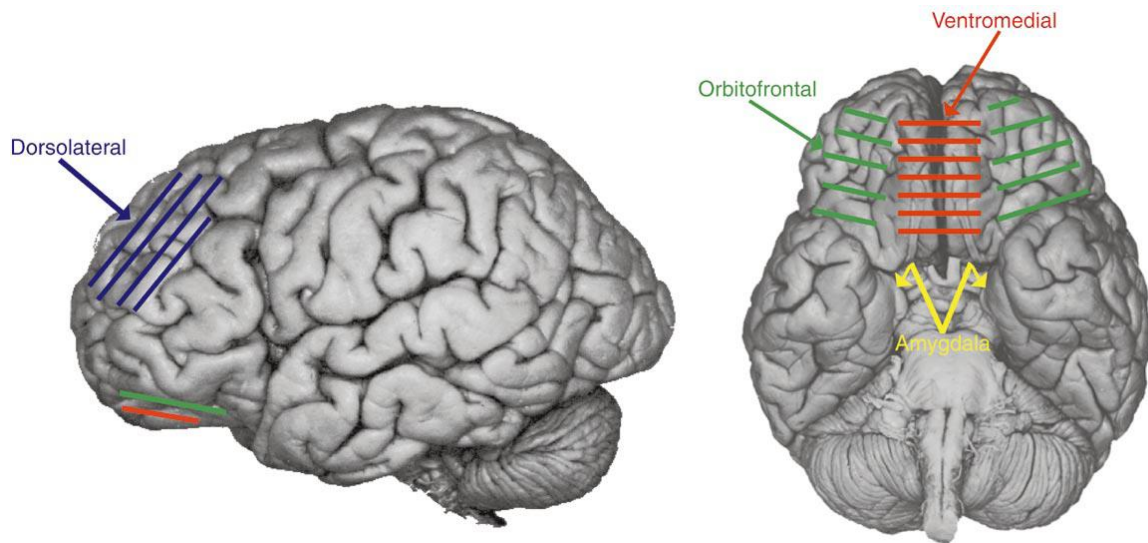


Figure 1. Topography of the brain systems amygdala and prefrontal cortex. The subdivisions of the prefrontal cortex, which are involved in affective processing, are the dorsolateral (blue), orbitofrontal (green) and ventromedial (red) prefrontal cortex. (Adapted from Davidson & Irwin, 1999.)

While the amygdala is essential for automatic emotional responses, the prefrontal cortex is essential for the full conscious experience of emotions and for acting in deliberate, planned ways, based on these feelings (Gray, 2007). Evidence for this stems from observations of people who were undergoing a prefrontal lobotomy. This operation disconnects the prefrontal area from the rest of the brain and usually freed people from their disturbed emotional feelings. As a negative side effect, the lobotomy also left them unable to organize and plan their lives effectively (Valenstein, 1986). Goal-oriented behaviour, like organizing and planning your life, allow us to interact in the world in a purposive manner (Gazzaniga, Ivry & Mangun, 2002). A plan of action has to be formulated, one that counts in past experiences and is adaptive to the current environment. To be flexible and adaptive, to switch from one plan to another, requires a means for monitoring the success of our ongoing actions. These operations are commonly referred to as executive functions. Executive functions are the

operations that serve to control and regulate information processing across the brain, in which the prefrontal cortex plays a role.

The connections between the PFC and other brain regions clarifies why the prefrontal cortex plays this crucial role in goal-directed behaviour. The prefrontal cortex consists of a massive network that connects the brain's motor, perceptual and limbic regions (Goldman-Rakic, 1995). There exist extensive projections from the other regions of the cortex, the parietal, temporal and occipital cortices, to the prefrontal cortex (Gazzaniga, Ivry & Mangun, 2002). In addition, subcortical structures and various brainstem nuclei project indirectly to the prefrontal cortex via thalamic connections. Either directly or indirectly, almost all cortical and subcortical areas influence the prefrontal cortex. Not only does the PFC receive projections, it also reciprocate connections to most structures, and also to the premotor and motor areas. From these neuroanatomical considerations follows that the prefrontal cortex appears to be in an excellent position to coordinate processing across wide regions. For example, input from the amygdala and from the somatosensory cortex, provides the PFC with information about the amygdala's assessment of the emotional stimulus and the body's state of arousal. All these projections will facilitate the emotional processing and regulation, and goal-oriented behaviour, based on these feelings (Gray, 2007).

Brain Lateralization and Emotion

The right and the left side of the brain (the right and left hemisphere) uniquely contribute to emotion. This lateralization of the brain was first suggested in 1939 by Goldstein. He described observations of individuals who had experienced damage to the left or right anterior cortex. Research that followed verified the findings that the right hemisphere appears to be more involved in negative emotions, whereas left appears to be more in involved in positive emotions.

First studies used the Wada test. Researchers injected amytal into one of the internal carotid arteries. Amytal is a barbiturate derivative, which is a drug that acts as a central nervous system depressant and is used as a sedative. In this way they were able to suppress the activity of one hemisphere. Injecting amytal into the left carotid artery produced a depressed, catastrophic reaction, consisting of crying, guilt, complaints, worries and pessimistic statements. Injections into the right carotid artery produced an euphoria, characterized by a lack of apprehension, smiling, laughing, mimicry, and a sense of well-being (e.g., Rossi & Rosadini, 1967). The mood changes associated with the amytal injection represent the release from inhibitory influence on the contralateral hemisphere of the injected side (Silberman & Weingarter, 1986). In other words, by injecting amytal in the left carotid arteries, the left hemisphere is not able to inhibit the right hemisphere, which causes the right hemisphere to stay active. Considering the injection evoked depressive feelings, these data suggest that the right side is more strongly involved in negative emotions. Similarly, injection into the right carotid arteries stopped the right from inhibiting the left side. Because the amytal infection caused an

euphoric feeling, it is suggested that left hemisphere is more strongly associated in positive emotions. Studies of patients with neurological damage have further confirmed the findings of brain lateralization. Damage to the left can lead to severe depression (Demaree, Everhart, Youngstrom & Harrison, 2005), whereas damage to the right side results in opposite symptoms of mania (Bearden, Hoffman, & Cannon, 2001).

The Laterality of the Prefrontal Cortex and Affective Style

As described above, the prefrontal cortex plays a crucial role in emotion. More specifically, research findings suggest that the prefrontal cortex contributes to the affective style of an individual (Davidson, 1998, 2002, 2004). Affective style is defined as the individual differences in reacting to emotional stimuli (state) and the differences in dispositional mood or temperament (trait; Davidson, 1998), which are related to the motivational characteristic of emotion. Davidson and colleagues (e.g. Davidson, 1998) attempt to understand the neural systems related to affective style. They examined the effect of lateralized brain function on affective style by measuring electrical activity from the scalp using EEG. Neural activity is an electrochemical process (Gazzaniga, Ivry & Mangun, 2002). Although the electrical potential produced by a single neuron is minute, when large populations of neurons are active together, they produce electrical potentials large enough to be measured by electrodes placed on the scalp. A change in voltage corresponding to the difference in potential between the signal at a recording electrode and that at a reference electrode is measured. EEG provides a continuous recording of overall brain activity and has proved to have many important clinical applications. Different frequency bands, derived from EEG, are associated with different behavioural states, among others the alpha frequency band. Alpha power has been found to be more reliably related to cognitive task performance compared with power in other frequency bands (Davidson, Chapman, Chapman & Henriques, 1990). It is assumed that emotion conditions differ reliably on measures of alpha power asymmetry and therefore, it has been the measure most consistently obtained in studies of EEG asymmetry. The alpha band is inversely related to neural activation (Davidson, Ekman, Saron, Senulis & Friesen, 1990).

The region of interest in the study of Davidson and colleagues is the anterior portion of the scalp. They found relative differences in the laterality of the EEG activity during rest. Some individuals show relatively more activity in the anterior right hemispheres, whereas others show relatively more activity in the anterior left hemispheres. These differences appear to be related to affective style and the motivational characteristic of emotion. There may be two separate neural circuits involved in different emotional reactions, i.e. the approach-withdrawal circuits are lateralized. The left prefrontal cortex is involved in emotional reactions that are more positive and elicit goals to approach or engage in a situation, and the right prefrontal cortex is involved in emotional reactions

that are more negative and elicit goals to withdraw from a situation. This explains the depressive symptoms in individuals with damage to the right. These symptoms are related to withdrawal behaviour (Demaree, Everhart, Youngstrom & Harrison, 2005). And it explains in turn the symptoms of individuals with symptoms of mania. These symptoms are related to decreased right frontal arousal, which produces a tendency to approach behaviour, for example goal-directed activities and excessive involvement in pleasurable activities that have the potential for painful consequences (Bearden, Hoffman, & Cannon, 2001). Moreover, positive emotional stimuli are likely to elicit approach behaviour, whereas negative emotional stimuli will elicit withdrawal or avoidance. Evidence for this theory is stated below.

Trait. In a study of Tomarken and colleagues (1992) subjects were given the Positive and Negative Affect Scale (PANAS). Subjects were asked to rate the extent to which a series of positive and negative traits describe their personality. The ones who rated themselves as a more positive affective person showed relatively greater EEG activity in the left midfrontal region than the ones who showed relatively greater EEG activity in the right midfrontal region. An opposite pattern was found when looking at the self-ratings for negative personality traits. In conclusion, this study demonstrates that individuals who describe themselves as having a more positive temperament showed more left frontal activity, whereas individuals who describe themselves as having a more negative temperament showed more right frontal activity.

State. The same difference in relative EEG activity of the right and left frontal lobe has been related to how the brain responds to emotional stimuli. Davidson (1995) measured the activity of the two hemispheres while the subjects watched short video clips, which evoked either positive or negative emotional reactions. The EEG activity measured during these video clips was compared to that where the subjects watched a neutral video clip, the baseline condition. Results showed that more neural activity over the left frontal lobe was observed when the subjects watched the positive video clips, in comparison to the negative video clips. And there was a large increase in activity over the right frontal lobe while subjects watched the more negative video clips.

In another compelling study of Davidson and Fox (1989) infants' EEG activity was measured before they were separated from their mothers. They found that infants whose prior EEG measures showed a right sided frontal activity were most likely to cry when the mother left the room, in comparison to infants with a more dominant left sided frontal activity. These results suggest that frontal asymmetry in brain activity may be linked to affective style from a very early age.

Anger. The findings discussed so far affirm the assumption of the involvement of the frontal regions of the hemispheres in emotional processing. Most negative emotions elicit withdrawal behaviour (e.g., fear or disgust) and most positive emotions elicit approach behaviour (e.g., happiness or amusement). However, anger is a negative emotion that can elicit either approach or withdrawal behaviour. Harmon-Jones and colleagues have conducted several studies in which they concluded that

anger is significantly correlated with both decreased right-anterior cortical activity and increased left-cortical activity (see for review Harmon-Jones, Gable & Peterson, 2010). Greater relative left activity occurred when the anger inducing information was presented in a way in which the participant could act against (approach) it, but not when this action was impossible.

In summary, the approach-withdrawal model postulates that emotions related to approach and withdrawal behaviours are processed in the left- and right-anterior brain regions, respectively. All individuals are capable of approach and withdrawal emotional reactions, but the magnitude and frequency of these different types of reactions may be related to the relative baseline asymmetry in an individual's right and left frontal activity.

EEG Asymmetry as Marker for Psychopathology

There is a fairly consistent pattern of EEG asymmetry in relation to approach and withdrawal motivation. Decrease of left frontal activity have been found in people with a lack of approach behaviour and depressive symptomatology (Demaree, Everhart, Youngstrom & Harrison, 2005). Moreover, adults with clinical depression and adults who were depressed and currently in remission showed this pattern of right frontal asymmetry (Henriques & Davidson, 1991). On the other hand, symptoms of mania appear to be related to decreased right frontal activity, which produces a tendency to approach behaviour, for example goal-directed activities and excessive involvement in pleasurable activities that have the potential for painful consequences (Bearden, Hoffman, & Cannon, 2001). Due to these associations EEG asymmetry has been given a considerable amount of attention in developmental research. One purpose of developmental research is to find biomarkers that show reliable associations with children's vulnerability to emotional and behavioural problems (Peltola et al., 2014). EEG asymmetry may be a phenotypic marker of the child's vulnerability to negative emotionality and risk for psychopathology (Davidson, 1998). The pattern of asymmetric frontal cortical activity may reveal a person's core biases, or tendencies, which hold consequences for their emotional behaviour. One's affective style may in part determine one's risk for certain affective disorders. A large amount of research has shown that right-sided frontal asymmetry may be an endophenotype of a trait-like withdrawal motivation associated with internalizing psychopathology, such as depression and anxiety (Allen & Cohen, 2010).

Asymmetric frontal cortical activity is influenced by the early social environment. A considerable amount of research has shown an association between maternal depression and EEG asymmetry in infants and young children (for a review see Field & Diego, 2008), in which a greater right frontal EEG asymmetry in infants has been related to depressed mothers (Dawson, Frey, Panagiotides, Osterling, & Hessler, 1997; Diego, Field, Jones, & Hernandez-Reif, 2006). Other types of psychosocial risk factors, among others, maternal caregiving, parental alcohol dependence and child

maltreatment, have not yet been investigated extensively (Peltola et al., 2014). And while some studies have shown strong effects of psychosocial risk factors in identifying the extent and direction of children's frontal asymmetry, others have found no significant differences between children who experience high or low risks for psychopathology.

Tympanic Membrane Temperature and Affective Style

Not only EEG has been used as a technique in frontal asymmetry and affective style studies. Kagan (1994) was first to demonstrate a link between temperature of the forehead and affective style. Shy, inhibited preschool children had a cooler right forehead after unfamiliar events. The left and right side differ in temperature, which can be explained by sympathetically controlled circulatory asymmetries (Rimm-Kaufman & Kagan, 1996). A greater right frontal EEG activation and a greater cooling of the right forehead are both related to this sympathetic blood flow. Asymmetries in this blood flow might serve as a marker for a pattern of neural asymmetry, which in turn may be related to the processing of emotion.

More evidence to support a relation between behavioural and biological aspects of affective style is found in studies in tympanic membrane temperature (TMT). This membrane is part of the hearing organ, which vibrates in response to sound waves. In humans it forms the eardrum, between the outer and middle ear. (Vollandri, Puccio, Forte & Carmignani, 2010). In a study of Boyce, Higley, Jemerin, Champoux and Suomi (1996) preliminary evidence is provided which suggests the presence of asymmetries in the temperatures of the left and right tympanic membranes in eight-year-old children. Children's warmer left and cooler right TMT was a predictive factor for troubled behaviour and maladaptive responses to the environment. Other researchers concluded the same: warmer left TMT is associated with negative emotions (Jones, Osmond, Godfrey & Phillips, 2011). However, a few years later, Boyce and colleagues (2002), and Gunnar and Donzella (2004) found the opposite from a sample of children, age ranged 4.5 to 8 years old. Both studies found evidence for a correlation between relatively warmer left tympanic membranes and positive affectivity or socially competent behaviours. Children with warmer right tympanic membranes, relative to cooler left, show more affectively negative and problematic behaviours. All in all, it can be said that there are incongruent findings considering affective style and TMT.

Current Research: EEG versus TMT

Both EEG and TMT asymmetries are said to provide phenotypic markers of the child's vulnerability to negative emotionality and risk for psychopathology. Presently, the most frequently used measurement in cortical activity and emotion research is EEG. This technology allows studying neural activity in detail, but it has several limitations that restrain its use, particularly in studies of children. The limitations include limited access, motion intolerance, expense, and an environment that

may stress the participants. Additionally, it demands a lot from the experimenters. It is time-consuming and the data analysis procedures are technically challenging. Therefore, Cherbuin and Brinkman (2005) have called for a cost-effective and simple measurement. The measurement that has been used increasingly lately, TMT, might be a very good alternative. As mentioned before, TMT appears to be related to aspects of temperament and bio-behavioural reactivity (Boyce et al., 2002). This relationship may be due to carotid blood flow. Differences in carotid blood flow are correlated with variations in cortical blood flow (Rothoerl, Schebesch, Woertgen & Brawanski, 2003), and increase in cortical blood flow is associated with cerebral activity (Gur et al., 1994). Carotid arteries have two branches, namely an internal and an external carotid artery. The internal carotid artery supplies blood to most of the cortex, while blood to the tympanic membrane is supplied by a network of veins branching from both internal and external carotid arteries (Gray, 1991). Therefore, it is likely that changes in cortical activity that are associated with changes in carotid blood flow are also associated with changes in blood flow to the tympanic membrane. Neurosurgical studies support this hypothesis. It shows that cortical temperature is correlated with tympanic membrane temperature (Schuman, Suhr, Gesseln, Jantzen & Samii, 1999). For these reasons, TMT might be a promising candidate for use in measuring lateralised cortical activity. Due to its simple use, it is a perfect instrument for large scale applications. This will benefit developmental research to biomarkers that show reliable associations with children's vulnerability to emotional and behavioural problems. Peltola and colleagues (2014) found that many studies contributing to the combined effect size of psychosocial risk were highly underpowered. The estimated power of .39 is substantially lower than the ideal threshold of .80, which is a risk for not finding true effects and, moreover, a risk for finding false effects. More studies with larger sample sizes are needed for finding biomarkers that show reliable associations with children's vulnerability to emotional and behavioural problems. TMT is a simple measurement that can be used in large scale applications. For that reason, finding a relationship between TMT and affective style would greatly benefit developmental research.

Although it is said that EEG and TMT might measure the same thing, the relationship between hemispheric activity and TMT, however, is by no means straightforward (Propper & Brunyé, 2013). There are conflicting hypotheses that can explain the physiological mechanisms underlying the change in TMT as a function of cortical activity. The review of Propper and Brunyé (2013) shows a division of two groups with different results, leading to different predictions for the relationship between TMT and affective style. The first studies show that increased TMT on one versus the other side is associated with ipsilaterally increased hemispheric activity (e.g. Boyce et al., 2002; Gunnar & Donzella, 2004). And the second group of studies show that increased TMT on one versus the other side is associated with ipsilaterally decreased hemispheric activity (Boyce et al., 1996; Helton, 2010).

So, it is not yet clear if, for example, increased left TMT is associated with right or left sided frontal activity and in turn, is related to withdrawal or approach.

A clear relationship between TMT and hemispheric activity has not yet been found (Propper & Brunyé, 2013). This study will test whether there is a relationship between TMT and frontal asymmetric activity. Moreover, it is not clear whether TMT is specifically associated with frontal brain activity or more central and posterior activity as well, which will be tested. Finding a clear predictive relationship between TMT and frontal asymmetry would benefit both research and clinical practice. From a research perspective, this finding would give rise to a simple and cost beneficial measurement for research on the role of cortical activity in emotion and motivational orientation. Moreover, TMT is less time-consuming and easier to use than EEG. Therefore, TMT is a powerful tool for large scale application in asymmetric frontal cortical studies. In conclusion, research has not yet reached consensus about the direction of the relationship between TMT and cortical activity. And to my knowledge, this is the first on cortical activity and TMT in infants. This study will contribute to the research on cortical activity and affective style.

Methods

Participants

Participants were recruited from a database on births of infants in the area of Leiden. Letters to parents of potential participants were sent out and they were asked to return a prepaid response letter to the research project if they would like their child to participate.

Twenty-nine healthy, full term ten-month-old infants, of which 17 were boys and 12 were girls, participated in this study. They ranged in age between 9.70 and 10.93 months ($M = 10.12$, $SD = 0.27$). Of these, 18 infants provided useable data. The attrition rate was due to insufficient EEG data caused by artefacts in the EEG from eye and body movements, and insufficient TMT measurements. Thus, 18 infants aged 9.70 to 10.93 months ($M = 10.10$, $SD = .28$) represent the final sample included in this analysis. The final sample consists of 11 boys and 7 girls. Table 1 gives the descriptive statistics of the total study population.

Ethical approval for this study was obtained from the Ethics Committee of the Faculty of Social and Behavioural Sciences of Leiden University.

Procedures

The session consisted of several procedures. EEG was recorded at rest (baseline) and while the infant watched a video clip, the temperature of the tympanic membrane was measured, and the eyes of the infant was tracked while he/she was watching a video clip. Additionally, mother and infant played freely. Parents were given a fully-detailed description of the procedure and the aim of the research,

and all their questions were answered. When everything was clear to the mother, her consent was obtained. The data of the baseline and TMT are used in this study.

Tympanic Membrane Temperature. Immediately after the mother signed the consent form, left and right TMT were taken, using Braun ThermoScan, model IRT 4520, ear thermometer, with probe cover (one per subject), while the infant was sitting on the mothers lap. Immediately after this first measurement, left and right TMT were taken for a second time. All temperature recordings were made in concordance with the manufacturer's recommendations.

EEG recordings. Infants were seated on the lap of their mother, who were seated in a comfortable chair. The mothers were told to interact with their child as little as possible and to try to keep the infant's hands from the EEG net. They were facing Lego blocks on top of a table, at a distance of approximately 100 centimetres. EEG was recorded while the infant looked for two minutes at the experimenter who was building towers of Lego blocks.

Table 1

Descriptive statistics of the total study population (N = 18)

		<i>M</i>	<i>SD</i>	n	%
Age infants (Months)		10.10	0.28		
Gender infants	Boys			11	61.1
	Girls			7	38.9
Age parents (Years)	Mother	32.88	4.99		
	Father	35.65	8.08		
Education mother	Lower vocational/partial secondary education			1	5.9
	Secondary education			2	11.7
	Higher education first stage			8	47.1
	Higher education second stage			6	35.3
Education father	Lower vocational/partial secondary education			1	5.9
	Secondary education			5	29.4
	Higher education first stage			3	17.6
	Higher education second stage			8	47.1
Nationality	Dutch			16	94.1
	Different			1	5.9
Family situation	Both biological parents			16	94.1
	One biological parent			0	0
	One biological parent and one stepparent			1	5.9
	One or two foster or adoptive parents			0	0
	Alternating between two households			0	0
	Different			0	0

Measures

Tympanic Membrane Temperature. To obtain TMT asymmetry scores, referred to as Δ TMT, the averaged right and left sided temperature, was calculated. This averaged right temperature was then subtracted from the averaged left sided temperature. Positive Δ TMT scores reflected a warmer left TM and more negative Δ TMT scores reflected a warmer right tympanic membrane.

EEG recordings and data reduction. Infant's EEG was acquired using a 128 channel hydrocel geodesic sensor net (Electrical Geodesics, Inc., Eugene, OR, USA) and amplified using a NetAmps300 amplifier. The sampling rate was 250 Hz with a low-pass filter of 125Hz. Figure 2 shows electrode groupings used for frontal asymmetry analysis at baseline. Cz was used as the reference electrode. Data were offline processed using Brain Vision Analyzer (BVA) 2.0 software (Brain Products). EEG data was exported to BVA with a high-pass filter with a cut-off of .3 Hz. Low-pass filtering was done with a high cut-off of 30Hz and 48dB/octave.

Segmentation. In order to compare the left and right activity at rest, the baseline period (i.e. the infant watching the experimenter building Legos) was extracted from the continuous EEG recording. This was done by creating segments limited by start and end markers. Particular events were marked using Net Station EGI software. The markers that were made for the baseline are called 'baseline begin' and 'baseline end', respectively as start and end markers. After the segmentation of the baseline, the baseline was divided into shorter overlapping segments of 512 data points (2048 ms) with a 50 percent overlap. By dividing the baseline in shorter segments, a more realistic and accurate result can be found in the process of computing the Fourier Transformation (FT). Instead of taking the complete baseline at once, the FT will be computed over smaller segments and then these results will be averaged. Before continuing to the FT, artefacts were rejected.

Artefacts. EEG recordings may contain, besides the desired brain electrical activity, non-cerebral contributions to the observed signal. These include artefactual contributions of the scalp, and artefacts generated by eye movements and blinks (Gratton, 1998). The eyes are ion-filled imperfect spheres. The cornea is positively charged, whereas the retina is negatively charged. These charged spheres create electrical fields when the eyes move that will be observed as artefacts. Data were checked for artefacts using BVA. First segments were rejected based on the contaminations of artefacts of eye movements. The electrodes around the eyes were selected (namely 1, 8, 14, 21, 25 and 32). In this way BVA will highlight these channels on changes in waveforms of 100 μ V in 50ms and on low-activity, namely 0.5 μ V in 100ms. The highlighted segments were checked and removed or kept manually. Secondly, individual channels were removed from segments. Individual electrodes from segments were removed in case the EEG waves looked abnormal.

The percentage of the total segments that were left after the rejection of the artefacts were checked for every infant. The criterion for keeping the infant in the analyses was that the remaining segments should consist of a minimum of 40% of the segments at the beginning. Infants whose

baseline data that did not fulfil this criterion were excluded from this study. From the 26 infants 18 infants remained.

Fourier Transform. After the artefact rejection, the Fourier Transform was performed for an analysis of the EEG power spectra. EEG waveforms consist of periodic signals, which can be decomposed into a series of sine and cosine functions of various frequencies (Allen, Coan, Nazarian, 2004). A periodic signal repeats at uniformly spaced intervals of time. EEG signals are not really periodic, because the repetition of features is not spaced at uniform intervals. However, this periodic signal can be approximated by selecting short epochs of EEG waveforms. In this way, small segments of data can be analysed that have features that repeat in a highly similar way at other points in the waveform. The waveform will be decomposed into a voltage by frequency spectral graph (μV^2), a power spectrum. The epoch length will determine the frequency resolution of the Fourier, in this study a resolution of 0.5 Hz was used. The power spectrum of this study's interest, was the alpha power band, which has a domain range of 6-9 Hz in infants. Power in the alpha band is inversely related to activation (Davidson, Ekman, Saron, Senulis & Friesen, 1990). Hanning window was selected to be used in the FT. Windowing was used to avoid creating artefactual frequencies in the power spectrum derived from the FT (Allen, Coan, Nazarian, 2004).

Natural logarithm. To normalize the data distribution, natural logarithm (ln) of the power values was calculated. In this study left and right alpha activity of both frontal, central and posterior sites were obtained. Therefore, these ln-transformed values were averaged across sets of electrodes. Each set represents a different region of interest: left-frontal (20, 23, 24, 26, 27, 28, 33, 34), right-frontal (2, 3, 116, 117, 118, 122, 123, 124), left-central (30, 36, 37, 40, 41, 42, 45, 46), right-central (87, 93, 102, 103, 104, 105, 108, 109), left-posterior (52, 53, 58, 59, 60, 61, 64, 65), right-posterior (78, 85, 86, 90, 91, 92, 95, 96). Figure 2 displays the division of the electrodes. Cronbach's alpha for the different sites was .99 for both left- and right frontal, and .97 for both left- and right-central, and both left- and right-posterior, which can be interpreted as excellent (George & Mallery, 2003).

Asymmetry score. An index of lateralisation was computed as a measure of asymmetry. This was done separately for frontal and parietal sites by subtracting left alpha activity from right alpha activity ($\ln(\text{Right}) - \ln(\text{Left})$ alpha power). These difference scores result in a one-dimensional scale that represents the relative activity of the right and left hemispheres. A zero value means that there is no difference in activity over left and right sites, thus no asymmetry. A positive value on this scale, on the other hand, means greater alpha power over right than over left, which represents a greater relative left cortical activity. Likewise, a more negative value represents a greater relative right cortical activity.

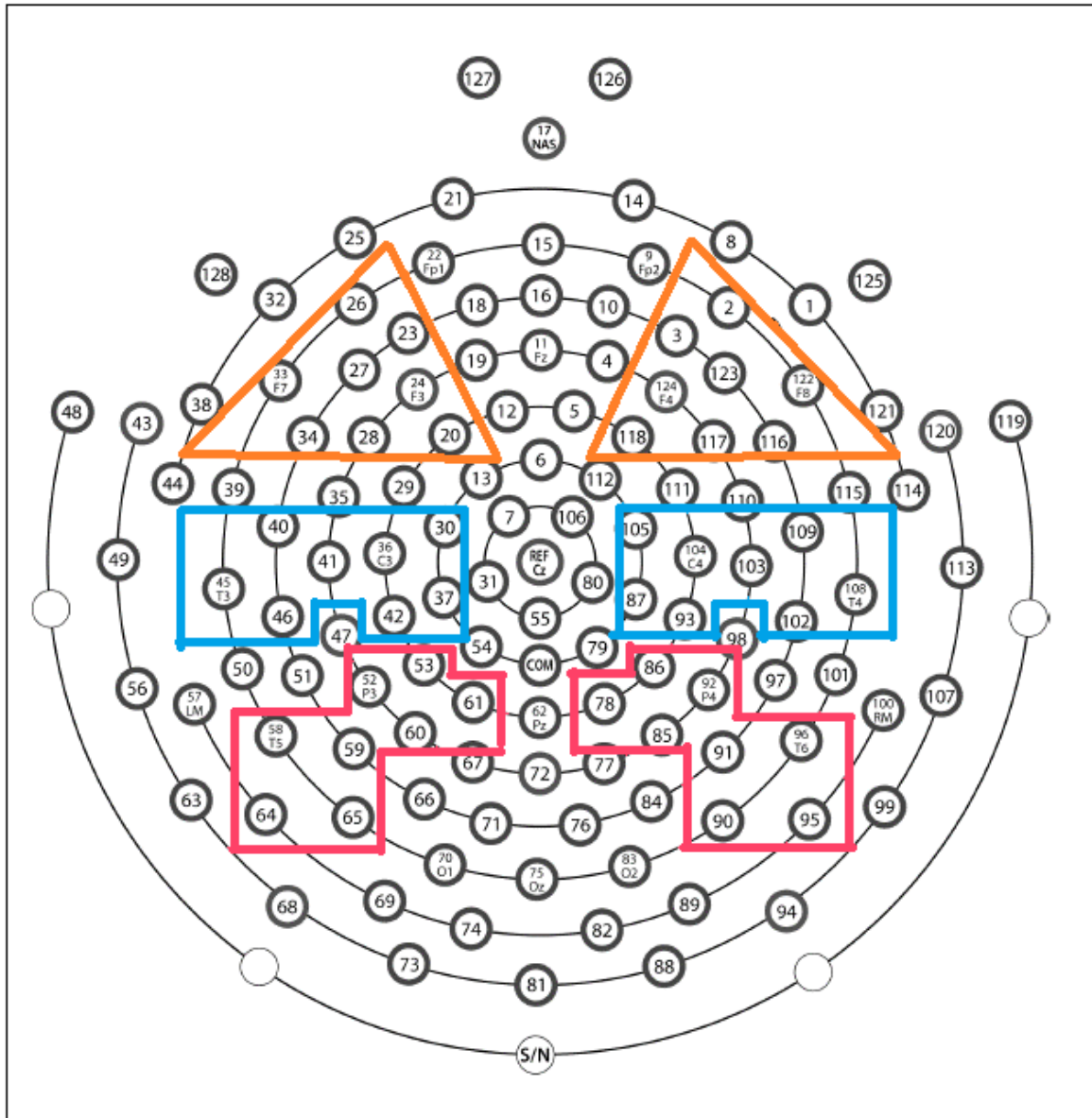


Figure 2. Division of the electrodes. Each set represents a different site: left-frontal (20, 23, 24, 26, 27, 28, 33, 34), right-frontal (2, 3, 116, 117, 118, 122, 123, 124), left-central (30, 36, 37, 40, 41, 42, 45, 46), right-central (87, 93, 102, 103, 104, 105, 108, 109), left-posterior (52, 53, 58, 59, 60, 61, 64, 65), right-posterior (78, 85, 86, 90, 91, 92, 95, 96).

Statistical Analyses

Statistical analyses were carried out with IBM SPSS Statistics 20. The relation between the variables Δ TMT and frontal, central and posterior asymmetry scores were analysed. To evaluate this relation, three repeated measures analyses of covariance (RM-ANCOVA) were performed. The first analysis was computed with frontal, central and posterior asymmetry scores as within-subjects (dependent) variables and Δ TMT as covariate. The variables of the first analysis consist of difference scores. Difference scores are less reliable and have larger standard error of measurements than a single score (Harvill, 1991). For this reason it was chosen to compute two more analyses with frontal, central and posterior alpha scores on the left side as within-subjects variables and TMT on the left side as covariate, and alpha scores on the right side as within-subjects variables and TMT on the right side as covariate. For all variables the level of measurement was interval.

Data inspection. Before analysing the data, the data was inspected on irregularities. First the distribution of the variables was inspected by looking at histograms with a normal curve, a Q-Q plot, and the standardised skewness and kurtosis. Moreover, Z-scores were computed for raw scores. To comply with the assumptions of a normality, the value of the standardised skewness and kurtosis has to be between -3 and +3.

After inspecting the distribution, data were investigated for outliers. Z-scores were computed for raw scores. Higher values than 3 and lower values than -3 were considered as outliers. Viewing a box plot showed whether the data consisted of outliers.

The variables were inspected solely individually, but the relation between variables were inspected as well. Correlations were computed for TMT of the left ear and frontal, central and posterior alpha scores on the left side; for TMT of the right ear and alpha scores on the right side and for the difference score of TMT and frontal, central and posterior asymmetry scores. Cohen (1992) holds the following criteria for correlations in social sciences: $r = .10$ is small, $r = .30$ is medium and $r = .50$ is large.

Results

The relationship between tympanic membrane temperature and asymmetric cortical activity was analysed. First, striking features of the data-inspection are given and descriptive statistics of the variables are briefly discussed. Following this, the findings resulting from the repeated measures analyses of covariance will be reported. The purpose of the analyses was to see if the asymmetric cortical activity differed over the different locations (parietal, central and posterior) and if this was related to tympanic membrane temperature.

Data Inspection

Among the 18 infants included in this study, one infant's scores were deviant from the other infants. The Z-scores of this infant were outstanding for the variables frontal and central asymmetry score, and frontal, central and posterior alpha score on the left and right ($z = -2.91, -2.30, 2.93, 2.10, 2.77, 2.49, 2.76$ and 2.86 , respectively). Moreover, the infant's scores had an influence on the normal distribution. The standardised skewness and kurtosis did not fall between -3 and $+3$ when this infant was included. So although the values of the Z-scores did not comply with the criteria for an outlier, it was chosen to continue with a dataset without this infant, because of the influence of his scores on normality.

The generated skewness and kurtosis showed that, overall, the variables were normally distributed (Table 2). The variable that stands out was TMT of the right ear, with a relatively high kurtosis ($z_{\text{kurtosis}} = 3.22$). The TMT of the left and right ear were both right skewed, the rest of the variables were left skewed. The sample size could be the reason for the abnormal distribution and because it was approaching normal distribution, it was chosen to continue with the analyses.

Correlations between TMT and cortical activity were measured. TMT of the left and right ear are both not significantly correlated to the left or right frontal, central or posterior alpha scores. Moreover, the difference score of TMT was not significantly correlated to frontal, central or posterior asymmetry score. Central alpha asymmetry was significantly correlated with frontal and posterior alpha asymmetry (respectively, $r = .63, p < .01$ and $r = .54, p = .05$), whereas frontal alpha asymmetry was not significantly related to posterior alpha asymmetry (respectively, $r = .18, p = .48$). The significant correlation between central alpha asymmetry and posterior alpha asymmetry were interpreted as large (Cohen, 1992).

Descriptive Statistics

A first investigation of the data revealed the separate means on the TMT of the left and right ear and the generated difference score. Moreover, it revealed the alpha scores for activity of the left and right side on frontal, central and posterior sites, and the alpha asymmetry score for all locations. The general characteristics of the variables are shown in Table 2.

The mean TMT of the left and right ear were 36.53 and 36.69 degrees Celsius, respectively. On average, infants' left and right tympanic membrane temperature differed 0.16 degrees Celsius. Average values for frontal, central and posterior asymmetry scores were -0.03 ($SD = 0.25$), -0.03 ($SD = 0.35$) and 0.07 ($SD = 0.29$). These scores mean that the left side was less active for the frontal and central sites, whereas for the posterior site it is true that the left side was more active than the right (see Table 2 for alpha scores on the left and right side).

Table 2
Descriptive statistics of the distribution of variables (N = 17)

	Min	Max	<i>M</i>	<i>SD</i>	<i>Z</i> _{skewness}	<i>Z</i> _{kurtosis}
TMT left ear	35.55	37.25	36.53	0.45	-1.04	0.21
TMT right ear	35.70	37.30	36.69	0.35	-2.29	3.22
Δ TMT	-0.55	1.10	0.16	0.37	0.98	1.58
Alpha score left frontal	0.21	2.50	1.21	0.68	1.24	-0.33
Alpha score right frontal	0.39	2.88	1.18	0.76	2.18	0.39
Alpha score left central	0.25	2.74	1.16	0.69	2.04	0.94
Alpha score right central	0.50	2.12	1.12	0.53	0.91	-0.95
Alpha score left posterior	0.70	2.87	1.67	0.64	0.49	-0.39
Alpha score right posterior	0.81	2.53	1.74	0.60	0.07	-1.45
Frontal asymmetry score	-0.47	0.63	-0.03	0.25	1.55	2.10
Central asymmetry score	-0.86	0.38	-0.03	0.35	-2.38	1.42
Posterior asymmetry score	-0.56	0.55	0.07	0.29	-0.04	0.28

Note: Z_{skewness} = skewness/standard error;
 Z_{kurtosis} = kurtosis/standard error

Relationship Between TMT and Asymmetric Cortical Activity

To give an answer to the question whether tympanic membrane temperature is related to frontal, central or posterior asymmetric cortical activity, a repeated measures analysis of covariance was executed. Frontal, central and posterior asymmetric cortical activities were the within-subjects in this analysis, whereas TMT was the covariate. Assumptions of normality and of independent observations were assumed. Another core assumption in the RM-ANCOVA procedure is that of sphericity. Sphericity checks whether the variance/covariance matrix of the observed data follows a particular pattern, usually identified as one with equal variances in the diagonal and equal covariance in the off-diagonal elements. If sphericity is observed the analysis provides a powerful test about repeated measures. In order to test this assumption Mauchly's test was inspected. The test was not significant, $W = .83$, $\chi^2(2) = 2.66$, $p = .26$, which means that the matrix had equal variances and covariances.

Table 3 summarizes the results of the repeated measures analysis of covariance. It follows that there was no significant difference in asymmetry scores across the three locations, $F(2, 30) = 1.84$, $p = .18$. There was no significant main effect of Δ TMT, $F(2, 30) = 1.84$, $p = .18$. TMT did not have an

Table 3

Repeated measures analysis of variance: within-subjects variables: Location asymmetry score; covariance: Δ TMT (N=17)

Effect	<i>MS</i>	<i>df</i>	<i>F</i>	<i>p</i>
Location	0.09	2	1.84	.18
Δ TMT	0.00	1	0.01	.91
Location x Δ TMT	0.06	2	1.16	.33
Error Location	0.10	30		
Error Δ TMT	0.18	30		

effect on the asymmetry scores. Moreover, the covariate TMT, did not significantly interact with asymmetry score, $F(2, 30) = 1.16, p = .33$. The differences in asymmetry scores across the three locations did not depend on the difference in tympanic membrane temperature

To obtain a more accurate answer to the question whether cortical activity is related to tympanic membrane temperature, two other repeated measures analyses of covariance were executed. In these analyses the relationship between left tympanic membrane temperature and left cortical activity, as well as the relationship between right tympanic membrane temperature and right cortical activity were investigated. Frontal, central and posterior alpha scores were the within-subjects in this analysis, whereas TMT was the covariate. Assumptions of normality and of independent observations were assumed. Mauchly's test for sphericity was not significant for the left side, $W = .75, \chi^2(2) = 4.06, p = .13$, which means that the matrix had equal variances and covariances. The Mauchly's Test was significant for right side, $W = .65, \chi^2(2) = 6.01, p = .05$, which means that the matrix had no equal variances and covariances. For this reason the Greenhouse-Geisser-corrected F-values were reported. Table 4 and 5 show the results of the repeated measures analyses of covariance. On both the left and right side no significant differences in cortical activity across frontal, central and posterior were found, $F(2, 30) = 0.43, p = .66$ and $F(1.48, 22.24) = 2.01, p = .17$, respectively. The tympanic membrane temperature on both the left and right side had no effect on the alpha scores, $F(1, 15) = 0.01, p = .93$ and $F(1, 15) = 0.23, p = .64$, respectively. Moreover, no interaction effect was found for both sides, $F(2, 30) = 0.36, p = .70$ and $F(2, 30) = 1.87, p = .18$, respectively. This means that differences in alpha scores across the three locations did not depend on the difference in tympanic membrane temperature.

Table 4

Repeated measures analysis of variance: within-subjects variables: Location left alpha scores; covariance: TMT left ear (N =17)

Effect	<i>MS</i>	<i>df</i>	<i>F</i>	<i>p</i>
Location	0.01	2	0.43	.66
TMT Left	0.01	1	0.01	.93
Location x TMT Left	0.01	2	0.36	.70
Error Location	0.03	30		
Error TMT Left	1.38	15		

Table 5

Repeated measures analysis of variance: within-subjects variables: Location right alpha scores; covariance: TMT right ear (N =17)

Effect	<i>MS</i>	<i>df</i>	<i>F</i>	<i>p</i>
Location	0.16	1.48	2.01	.17
TMT Right	0.26	1	0.23	.64
Location x TMT Right	0.15	1.48	1.87	.18
Error Location	0.08	22.24		
Error TMT Right	1.16	15		

Conclusion and Discussion

EEG has been widely used in studies of emotions and affective style. TMT has been proposed as an alternative for EEG, because this measurement will be less invasive and easier to use than EEG. The current study tested whether there is a relationship between tympanic membrane temperature and hemispheric activity using EEG. Different regions of the hemisphere was examined, namely frontal, central and posterior sites. TMT was found to be unrelated to EEG activity in any of these locations. It seems that differences in tympanic membrane temperature do not reflect hemispheric activity. This means that TMT does not measure the same thing as EEG. Hence, the hypothesis that TMT is a cheaper and easier alternative for EEG measurements cannot be endorsed.

TMT and EEG activity can still both be related to emotional processing, but they might measure different neural circuits. The blood supply to the tympanic membrane is supported by the same vasculature that supports blood flow to the hypothalamus (Benzinger & Taylor, 1963; Chrousos & Gold, 1992). That might change the temperature change of the tympanic membrane, because the hypothalamus plays a central role in processing emotional information (Parr & Hopkins, 2000). Thus, when the blood flow to the hypothalamus changes, due to emotional processing, the blood flow to the

tympanic membrane changes. This might have been the case in other studies that tested the relationship between TMT and affective style.

The findings of this study also raise questions about the way the tympanic membrane temperature is related to affective style. The statements resulting from studies on tympanic membrane temperature and emotion-motivation should be interpreted with caution. Other studies found relationships between lateralized TMT and affect, but only limited evidence is given. Only three studies have examined children (age ranged between 3 to 8 years), which relied their behavioural outcome on parental reports of affect/motivational orientation. Of these three studies two came from the same laboratory and had opposite findings. The first study done in this laboratory (Boyce et al., 1996) found that warmer left TMT was associated with negative/withdrawal emotions, whereas the second study (Boyce et al., 2002) found the opposite, that warmer left TMT was associated with positive/approach emotions and warmer right TMT was associated with negative emotions. The opposite findings in these studies may result from different focus in behavioural outcomes. The second study focused on negative emotions and ‘problem’ behaviour, whereas the first study was more elaborated on negative behavioural outcomes, focusing on both negative emotions and behavioural withdrawal. Moreover, the first study included 18 children in the study and the second study included 37 children. This difference in sample size can have an influence on the findings, because a larger sample size will have more power and therefore, has an increased probability in finding true findings. The third study examining children’s emotions and TMT included 77 children (Gunnar & Donzella, 2004). They looked at the same negative and positive scales as Boyce and colleagues (2002) and also included the scale laughter and smiling. Overall, they found the same associations as Boyce and colleagues (2002). However, Gunnar and Donzella (2004) found different TMT asymmetry distributions. While they found a strong bias toward positive asymmetry, which supports the frontal EEG asymmetries, Boyce and colleagues (2002) found an almost equal distribution of asymmetry scores around zero.

Not only is there no consensus about TMT and affective style, but also the relation between asymmetric cortical activity and affection/motivation cannot yet be confirmed with confidence. Recent studies show that the apparent frontal asymmetry in relation to the tendencies to approach or withdraw is dependent on the milieu of the children, for example parental caregiving, and the affective characteristics of the children (Peltola, et al., 2014). So, the relation between TMT and affect, nor the relation between EEG asymmetry and affect are clear and thus relating TMT with hemispheric activity as a predictive factor for affective style is not firmly grounded.

Limitations and Implications for Future Research

First of all, the current research study has some strengths. To my knowledge, it is the first study to relate asymmetric cortical activity and TMT. The findings are interesting preliminary results to

continue with in future research studies. Moreover, the research assistants in the project group were all involved with the infants and the infants' mothers and did their best to make them feel at ease.

The findings of this study should be assessed with several shortcomings in mind. First, due to the small sample size the power was below the recommended criterion of .80. Small sample sizes are at risk for an increase of false positive findings as well as a decrease in finding true effects.

Second, the novelty and somewhat stressing experimental milieu may elicit stressful responses of the infant. Since some of the infants were crying while putting the EEG net on, this may have influenced the EEG baseline, as it may not truly reflect the baseline state of the infant. This also applies to the measurement of tympanic membrane temperature. Infants are not always at ease during this operation and the measured temperature may not reflect the actual baseline temperature of the infant. Moreover, only infants with practically artefact-free data were included. It is possible that the excluded infants differ from the infants in the current study sample, because the artefacts might be related to the difference in affective style. Future studies should increase the baseline period and/or provide variety in the way the baseline is measured to keep the infant interested. This will increase the number of infants that can be included. In this way a more representative sample might be gained.

Third, other factors may also influence the baseline of TMT and EEG. Two situational factors that appear to influence baseline asymmetric frontal activity, for example, are time of day and time of year (Harmon-Jones, Gable & Peterson, 2010). These two factors are related to basal cortisol levels and mood, which in turn can influence the asymmetric cortical activity and tympanic membrane temperature.

The above mentioned factors should be measured or controlled for in future studies. Moreover, future research should include measures of affective style as well. In this way it is possible to relate both tympanic membrane temperature and asymmetric cortical activity independently to affective style. Researchers can include the Laboratory Temperament Assessment Battery (lab-TAB, Gagne, Van Hulle, Aksan, Essex & Goldsmith, 2011). Lab-TAB consists of 3-5 minute episodes that reflect everyday situations. In these situations one can reliably observe differences in activity level, in the expression of emotion, in approach/avoidance and other instrumental behaviour, and in regulatory aspects of behaviour. It might be that the two variables influence affective style in a different manner. Including this in future research will lead to a better understanding of the involvement of these two variables.

TMT was found to be unrelated to EEG activity in any of these locations. It seems that differences in tympanic membrane temperature do not reflect hemispheric activity. This means that TMT does not measure the same thing as EEG. Hence, the hypothesis that TMT is a cheaper and easier alternative for EEG measurements cannot be endorsed.

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